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A microbial fuel cell integrated with an absorption column for the removal of hydrogen sulfide from anaerobic wastewater treatment

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ABSTRACT

The presence of hydrogen sulfide in biogas produced from the anaerobic treatment of organic wastewater containing high sulfate concentrations has been a key concern for this type of wastewater. This study investigates the performance of a two-compartment single-chamber microbial fuel cell (MFC) integrated with a packed absorption column in the removal of hydrogen sulfide produced from the anaerobic treatment of organic wastewater containing high sulfate concentrations (~3,000 mg/L chemical oxygen demand (COD) and ~300 mg/L sulfate). The first and the second compartments of the system functioned as an anaerobic bioreactor and an MFC, respectively. The anaerobic compartment removed COD and sulfate at removal efficiencies of 81.3% ± 4.5% and 100% ± 0.0%, respectively, whereas the MFC compartment removed hydrogen sulfide in the effluent and biogas. The hydrogen sulfide concentrations in the biogas decreased from 14,540 ± 620 to 153 ± 63 ppm after the installation of the MFC with a packed absorption column. A maximum power density of 8.0 mW/m^2 was achieved. The results of 16S rRNA gene clone libraries revealed the presence of a fermentative bacterium (*Streptococcus macedonicus*), a sulfur-oxidizing bacterium (*Halothiobacillus neapolitanus*), and an exoelectrogenic microorganism (*Klebsiella pneumoniae*) in the biofilm on the anode electrode.

Keywords: Microbial fuel cell; Hydrogen sulfide; Sulfate; Absorption column; Biogas

1. Introduction

Anaerobic wastewater treatment is considered one of the most suitable approaches for use in the treatment of high-strength organic wastewater due to its cost-effectiveness and the production of renewable energy in the form of biogas. However, organic wastewater from certain industries, for example, paper mills and the rubber, pharmaceutical, mining, and tannery industries, contains high amounts of sulfate [1]. The presence of high sulfate concentrations in anaerobic treatment systems can favor the growth of sulfate-reducing bacteria, which convert sulfate into hydrogen sulfide. Hydrogen sulfide has a nuisance odor and can lower the quality of biogas. Further biogas treatment is typically required before its use as fuel.

Microbial fuel cells (MFCs) were originally developed for the treatment of organic wastewater with simultaneous

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electricity generation [2]. In single-chamber air-breathing MFCs, organic compounds generally serve as electron donors on the anode, with electrochemically active bacteria biofilms being the catalysts, whereas oxygen in the air serves as the electron acceptor on the cathode [3]. These spontaneous electron transfers in MFCs result in the oxidation of organic compounds into CO_2, the reduction of O_2 into water, and electricity generation [4]. The electrochemical treatment of organic wastewater using MFCs has been extensively demonstrated in the literature [4–7]. More recently, MFCs have been applied for energy recovery in the treatment of organic wastewater containing sulfate as an alternative [8–11] to conventional anaerobic treatment methods, which have many hydrogen sulfide-related issues. Nevertheless, the energy recovery from MFCs is still less than that obtained from conventional anaerobic treatment [12]. Therefore, instead of using an MFC directly for the treatment of organic wastewater containing sulfate, this study intends to use an MFC for the purpose of hydrogen sulfide removal from effluent and biogas produced from the anaerobic treatment of organic wastewater containing sulfate. This approach can serve as an alternative technology for hydrogen sulfide removal and energy recovery from organic wastewater containing sulfate.

The removal of dissolved sulfide from wastewater with simultaneous electricity generation has been widely accomplished in previous studies with various types of MFC configurations, ion exchange membranes, and electrode materials [8,9,13,14]. However, the application of MFCs for the removal of hydrogen sulfide gas is limited, with only one previous study using an MFC for the control of malodorous hydrogen sulfide compounds generated from domestic wastewaters using a two-chamber MFC operated under batch conditions [15]. To the authors' knowledge, there has been no research using MFCs for the removal of hydrogen sulfide from the biogas produced from anaerobic wastewater treatment. The development of such a process will further expand the applications of MFCs in biogas treatment. In this study, we have proposed a novel continuous-flow MFC system with the integration of an absorption column for hydrogen sulfide removal from both the aqueous phase and biogas. The first compartment functioned as an anaerobic bioreactor, while the second compartment functioned as an MFC, with an absorption column installed to enhance the mass transfer of hydrogen sulfide gas into the aqueous phase. The main objective of this study was to investigate the performance of the proposed novel system in hydrogen sulfide removal from both the aqueous phase and biogas. In addition, the microbial communities on the anode electrode and suspended microorganisms in the anaerobic compartment were analyzed using 16S rRNA gene clone libraries.

2. Materials and methods

2.1. Operation of a two-compartment single-chamber MFC integrated with a packed absorption column

A two-compartment single-chamber air-breathing MFC with internal dimensions of 15.0 cm × 25.5 cm × 20.0 cm (width × length × height) was used in this study (Fig. 1). The working volume of each compartment was 2.0 L. The first compartment functioned as an anaerobic bioreactor, whereas the second compartment functioned as an MFC for hydrogen sulfide removal from the effluent and biogas from the anaerobic compartment. Synthetic wastewater was continuously fed into the system at a flow rate of 2.0 L/d. The system was operated at room temperature (28°C–34°C). The pH values in the system were in the range of 6.8–7.2 throughout MFC operation. The anaerobic compartment, which was initially seeded with 10,000 mg mixed liquor suspended solids (MLSS)/L anaerobic sludge from a starch industrial wastewater treatment system, was operated alone for 109 d (results shown in Fig. S1) before starting the MFC operation. During MFC operation, the effluent from the anaerobic compartment flowed continuously into the MFC compartment. An activated carbon cloth with a size of 5 cm × 5 cm (Zorflex Knit FM50K, Calgon Carbon Corporation, USA) was used as the anode electrode, and a wet-proofed carbon cloth loaded with 0.5 mg/cm² Pt (Fuel Cell ETC, USA) was used as the cathode. The anode and the cathode were connected by a titanium wire. A proton-exchange membrane (PEM) (Nafion N117) was hot-pressed to the cathode. Silver mesh (Fuel Cell Materials, USA) was attached to the cathode to improve the electron transfer. Mixing within each compartment was gently provided by a magnetic stirrer.

After 20 d of MFC operation, a cylindrical absorption column with a diameter of 5 cm and height of 15 cm was installed in the second (MFC) compartment to enhance the mass transfer of hydrogen sulfide gas into the aqueous phase (Fig. 1). The absorption column was packed with 200 mL of propylene plastic media (porosity = 0.30) (Fig. S2). Effluent from the MFC compartment was recirculated to the absorption column at a flow rate of 36 L/d (25 mL/min), which is the minimum flow rate that can create small droplets (172 ± 8.2 μm in diameter) through the water distribution device used in this study. In addition, after 35 d of

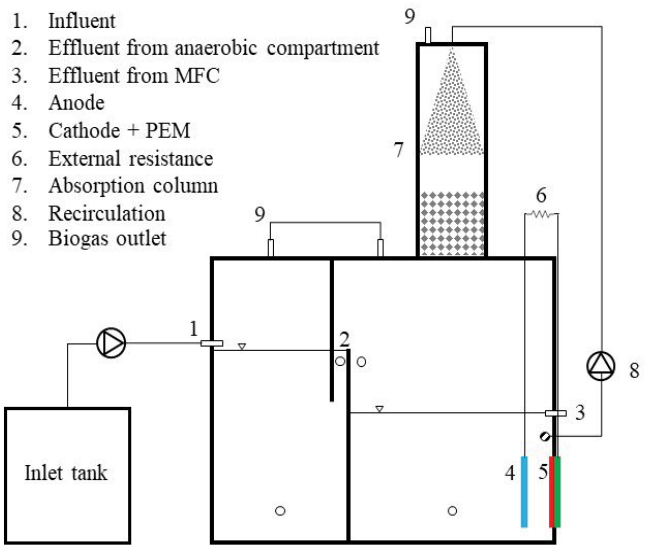

Fig. 1. Two-compartment single-chamber air-breathing MFC with an absorption column. (1) Influent, (2) effluent from anaerobic compartment, (3) effluent from MFC, (4) anode, (5) cathode + PEM, (6) external resistance, (7) absorption column, (8) recirculation, and (9) biogas outlet.

MFC operation, the anode and the cathode-PEM assembly were replaced. After 60 d of MFC operation, the anode was replaced again.

The synthetic wastewater used in this study resembled organic wastewater containing sulfate at a chemical oxygen demand (COD):SO_4^{2-} ratio of 10:1. The synthetic wastewater consisted of glucose (3,000 mg COD/L), Na_2SO_4 (300 mg SO_4^{2-}/L), NH_4Cl (58 mg N/L), Na_2HPO_4 (11.32 mg P/L), NaCl (150 mg Na/L), KCl (300 mg K/L), $CaCl_2$ (150 mg Ca/L), $MgCl_2$ (160 mg Mg/L), $CoCl_2$ (22.4 μg Co/L), $NiCl_2$ (2.32 μg Ni/L), $ZnCl_2$ (36 μg Zn/L), $CuCl_2$ (23.2 μg Cu/L), $MnCl_2$ (23.2 μg Mn/L), and H_3BO_4 (20 μg Bo/L). Alkalinity was added using $NaHCO_3$ (2,500 mg/L as $CaCO_3$) [10].

2.2. Roles of abiotic processes in hydrogen sulfide removal and electricity generation

In addition to the main MFC, another abiotic fuel cell was operated to investigate the roles of abiotic hydrogen sulfide oxidation in the cathode and electricity generation. The second compartment of the main MFC was used as an abiotic fuel cell after the installation of a new anode and cathode-PEM assembly. Synthetic wastewater containing only hydrogen sulfide at a concentration of 80 mg S/L, which was approximately the same as the effluent sulfide concentration from the first compartment of the main MFC, was fed directly into the abiotic fuel cell. The hydrogen sulfide and sulfate concentrations were monitored, as was the voltage across the electrodes.

2.3. Analytical measurement

The water samples from the MFC were collected and analyzed for COD, sulfate, and dissolved sulfide concentrations using the closed reflux method (5220C) [16], the turbidimetric method (4110E) [16], and an ion-selective electrode (PerfectION™ combination silver/sulfide electrode, Mettler Toledo, USA), respectively. Alkalinity and volatile fatty acids were measured using the titration method (2320B) [16]. The pH was monitored with a pH meter (In Lab® Expert Pro-ISM electrode, Mettler Toledo, USA). The hydrogen sulfide gas in 50 mL biogas samples was trapped as sulfide ions in 10 mL of sulfide anti-oxidation buffer (SAOB). The SAOB solution consisted of 2 M NaOH, 0.2 M ascorbic acid, and 0.2 M disodium ethylenediaminetetraacetic acid (EDTA) (dihydrate) [17]. Then, the dissolved sulfide was measured using an ion-selective electrode (Perfect ION™ combination silver/sulfide electrode, Mettler Toledo, USA) and converted into hydrogen sulfide gas concentrations. The measurement of hydrogen sulfide gas concentrations was performed in triplicate, resulting in a standard deviation below 5% of the measured values. The surface of the anode electrode removed from the MFC on day 61 was analyzed using scanning electron microscopy with energy dispersive X-ray spectroscopy (SEM [JSM-6400] with EDX).

2.4. Electrical measurements

The open-circuit voltage (OCV) and voltage across the electrodes were measured over time during MFC operation by a multimeter (Fluke 115, Washington, U.S.A.). Current density and power density curves were calculated from the voltages and external resistances.

2.5. Microbial community analysis

A biofilm sample from the anode electrode and a sludge sample from the anaerobic compartment were collected for microbial community analysis on day 61 and at the end of MFC operation, respectively. The DNA samples were extracted using a DNA FastDNA® SPIN Kit (MP Biomedicals, USA). The 16S rRNA genes of bacteria and archaea were amplified using polymerase chain reaction (PCR) with universal primers for bacteria, 8F (5′-AGAGTTTGATCCTGGCTCAG-3′) and 1492R (5′-GGTTACCTTGTTACGACTT-3′) [18], and universal primers for archaea, 21F (5′-TTCYGGTTGATCCYGCCRGA-3′) and 1492R (5′-GGTTACCTTGTTACGACTT-3′) [19], respectively. The PCR products were gel-purified using NucleoSpin® Gel and PCR clean-up (Macherey-Nagel, Germany). Clone libraries of 16S rRNA genes were constructed using pGEM®-T Easy Vectors I (Promega, USA) for ligation and Blue Supercompetent cells (Agilent Technologies, USA) for transformation. Plasmid inserts of white colonies were amplified by PCR and then submitted for DNA sequencing to First BASE Laboratories, Malaysia. The sequences were compared with the 16S rRNA gene database of the National Center for Biotechnology Information (NCBI) using BLASTN. Phylogenetic trees were constructed in MEGA7 software using the neighbor-joining method with bootstrap replicates of 500.

3. Results and discussion

3.1. Performance of the two-compartment single-chamber MFC integrated with the packed absorption column

The concentrations of COD, sulfate, and hydrogen sulfide in the two-compartment single-chamber air-breathing MFC are shown in Fig. 2. From the results (Figs. 2a and b), COD and sulfate were primarily removed in the anaerobic compartment, with the COD removal of 2,385 ± 139 mg/L and the sulfate removal of 303 ± 14 mg/L. According to these results, 8.6% of the electron equivalents from COD removal was estimated to be used in sulfate reduction in the anaerobic compartment. The remaining 91.4% of the electron equivalents from COD removal was expected to be used for methane production, assuming that sulfate was the single electron acceptor in the system and that the biomass growth yields under anaerobic conditions were negligible. According to this electron equivalent estimation, methanogens appeared to be the dominant group of microorganisms responsible for COD removal, which was in agreement with previous research reporting the predominant methanogenic activities at the COD:sulfate ratio of 10:1 [20,21]. In contrast, only small amounts of COD and sulfate were removed in the MFC compartment since virtually no sulfate remained in the effluent of the anaerobic compartment, and H_2S removal appeared to outcompete COD removal in the MFC compartment, as COD and H_2S were competitive electron donors in the MFC compartment. The overall removal efficiencies for COD and sulfate in the two-compartment

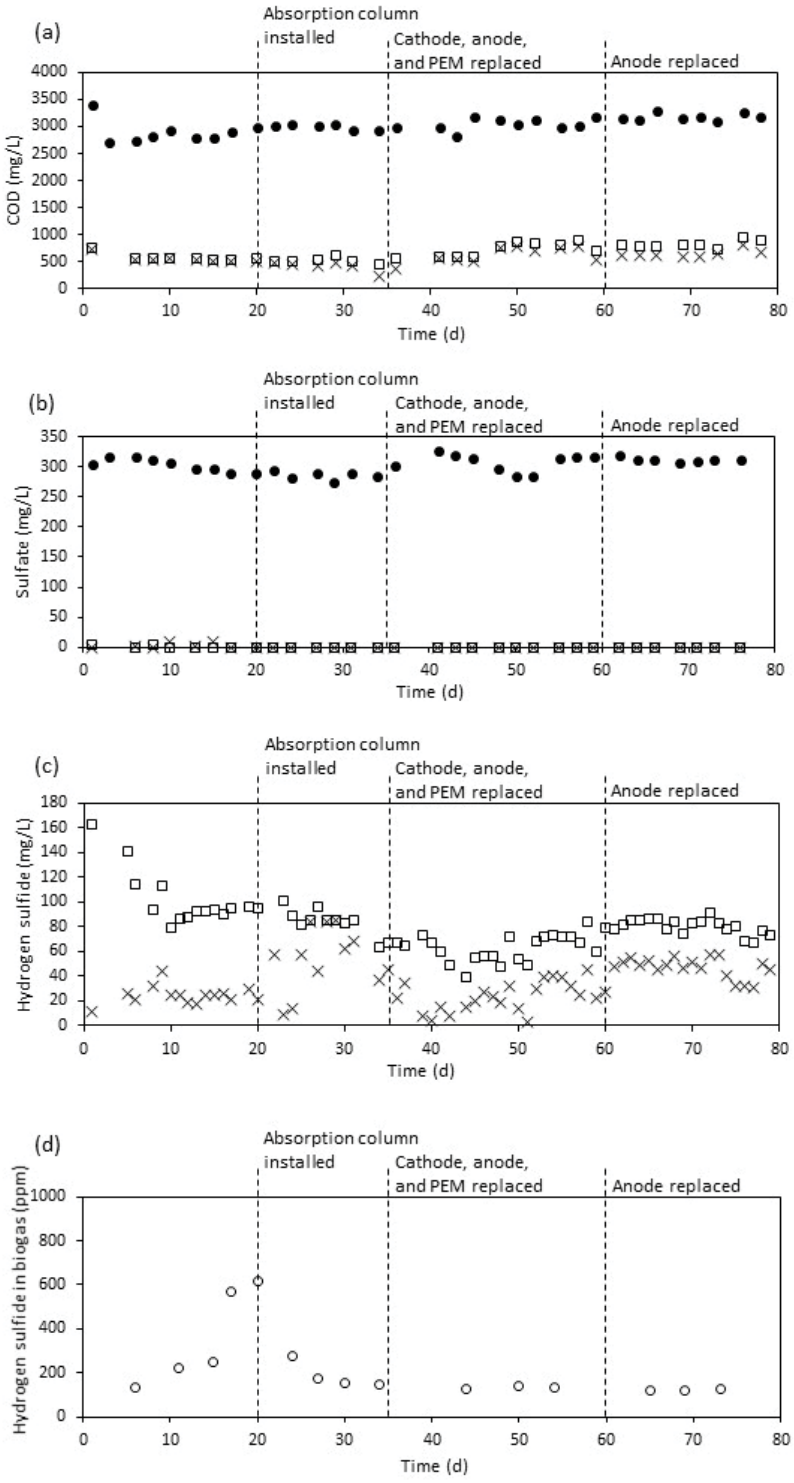

Fig. 2. Concentrations of (a) COD, (b) sulfate, and (c) hydrogen sulfide in the influent (•), the effluent of the anaerobic compartment (□), and the effluent of the MFC (×), and (d) hydrogen sulfide concentrations in the biogas (○).

MFC before the installation of the packed-bed column were 81.2% ± 1.4% and 99.9% ± 0.4%, respectively.

On the other hand, hydrogen sulfide was generated in the anaerobic compartment but removed in the MFC compartment, with a removal efficiency of 75.1% ± 7.9%. Deposits of elemental sulfur, which is a potential product of sulfide oxidation, were found at very low amounts (1% by weight) on the anode electrode after its usage in the MFC (Fig. S3). This result was in contrast to the results of previous studies on MFCs used to treat organic wastewaters containing high sulfate concentrations (COD:sulfate ratio of 0.67–6), which found higher accumulations of elemental sulfur on the anode

electrodes, but this discrepancy could be due to the higher COD:sulfate ratio used in this study (COD:sulfate ratio of 10) compared to those in previous studies [10,11].

The average gaseous hydrogen sulfide concentration was 14,542 ± 623 ppm in the biogas produced from the sole operation of the anaerobic compartment (day 97–105) before MFC operation. Then, during MFC operation without the packed absorption column, the average gaseous hydrogen sulfide concentration in the biogas decreased to 357 ± 219 ppm. As estimated from these gaseous hydrogen sulfide concentrations, the removal efficiency was approximately 97.5%, which was comparable to that in a previous study by Eaktasang et al. [15] reporting the $H_2S_{(g)}$ removal efficiency of 96.4% in a two-chamber MFC operated under batch conditions. The results (Fig. 3) indicated that the MFC compartment without the packed absorption column was able to effectively remove gaseous hydrogen sulfide from the biogas.

After the installation of the packed absorption column after 20 d of MFC operation, the removal efficiencies of COD and sulfate remained approximately the same at 81.3% ± 4.5% and 100% ± 0.0%, respectively. However, the aqueous hydrogen sulfide concentration in the MFC compartment increased from 24.4 ± 7.4 to 38.2 ± 19.8 mg/L (Fig. 2c), resulting in a decrease in the hydrogen sulfide removal efficiency in the aqueous phase from 75.1% ± 7.9% to 49.7% ± 21.7%. This observation was likely due to the increase in the mass transfer of hydrogen sulfide from the gas phase into the aqueous phase after the installation of the packed absorption column. Furthermore, the gaseous hydrogen sulfide concentration in the biogas decreased from 357 ± 219 to 153 ± 63 ppm after the installation of the packed absorption column (Figs. 2d and 3). Although the concentration fluctuation was so large that the decrease in gaseous hydrogen sulfide concentration was not statistically significant, the gaseous hydrogen sulfide concentration was maintained at lower levels (<280 ppm) throughout operation after the packed absorption column was installed (Fig. 2d).

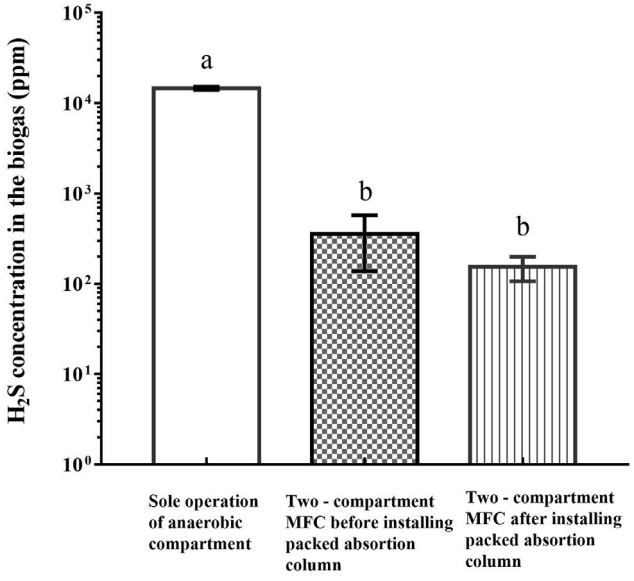

Fig. 3. Gaseous hydrogen sulfide concentrations (ppm) in the biogas produced from the sole operation of the anaerobic compartment and the two-compartment MFC before and after installing the packed absorption column.

In terms of electricity generation, the OCV of the MFC slightly decreased and became stable in the range of 558–605 mV after 7 d of operation. In contrast, the voltage across the electrodes at an MFC external resistance of 1,000 ohms increased, with a maximum voltage of 105 mV observed on day 8 of operation (Fig. 4). Then, the voltage was stable within the range of 50–60 mV. The polarization (I–V) curves and power density curves constructed on days 1, 8, 14, and 21 are shown in Fig. 5. The maximum power density was 8.0 mW/m^2, which was observed on day 8 of operation at an external resistance of 3,000 ohms. According to the polarization curves, which appeared to be straight lines, the activation losses and ohmic losses that are the two types of voltage losses existing in the system cannot be distinguished. On day 8, lower voltage losses with a higher maximum power density were achieved compared to those on day 1, which could have been due to the biofilm formation of exoelectrogenic microorganisms on the anode electrode that could lower the activation losses of the system. A similar observation has been reported in Rabaey et al. [8]. In contrast, after day 8, the voltage losses gradually increased, along with the decrease in maximum power densities, which could have been caused by (1) the deterioration of the electrodes and PEM over time [10,11] and/or (2) the biofilm formation of non-exoelectrogenic microorganisms on the anode electrode reducing the active surface areas and blocking the electron transfers.

The replacement of the anode and cathode-PEM assembly did not appear to affect the performance in terms of COD, sulfate, and sulfide removal in the system (Fig. 2). However, the replacement of the cathode-PEM assembly improved the OCV of the system, though only for a short period of time (Fig. 4). The deterioration of electrodes in MFCs treating high-sulfate wastewater (COD:sulfate ratio of 0.67–6) has been previously reported [10,11] with much more severity than that observed in this study, which was likely due to greater elemental sulfur accumulation on the anode electrodes at lower COD:sulfate ratios.

3.2. Roles of abiotic processes in hydrogen sulfide removal and electricity generation

The roles of abiotic processes in hydrogen sulfide removal and electricity generation were investigated using the abiotic fuel cell. Fig. 6 shows the concentrations of hydrogen sulfide and sulfate in the abiotic fuel cell. The hydrogen sulfide removal efficiency was 98.5% ± 0.44%. Sulfate was observed as the product of sulfide oxidation at a ratio of 0.47 ± 0.095 mol of SO_4^{2-}–S produced per mole of H_2S–S removed. An additional product could be elemental sulfur. Previous studies have reported that the abiotic oxidation of hydrogen sulfide in fuel cells resulted in either sulfate [22] or elemental sulfur [23]. The results suggest the important role of abiotic processes in hydrogen sulfide removal and electricity generation in the main two-compartment single-chamber MFC, which is similar to previous observations for MFCs treating organic wastewater containing high sulfate concentrations [11].

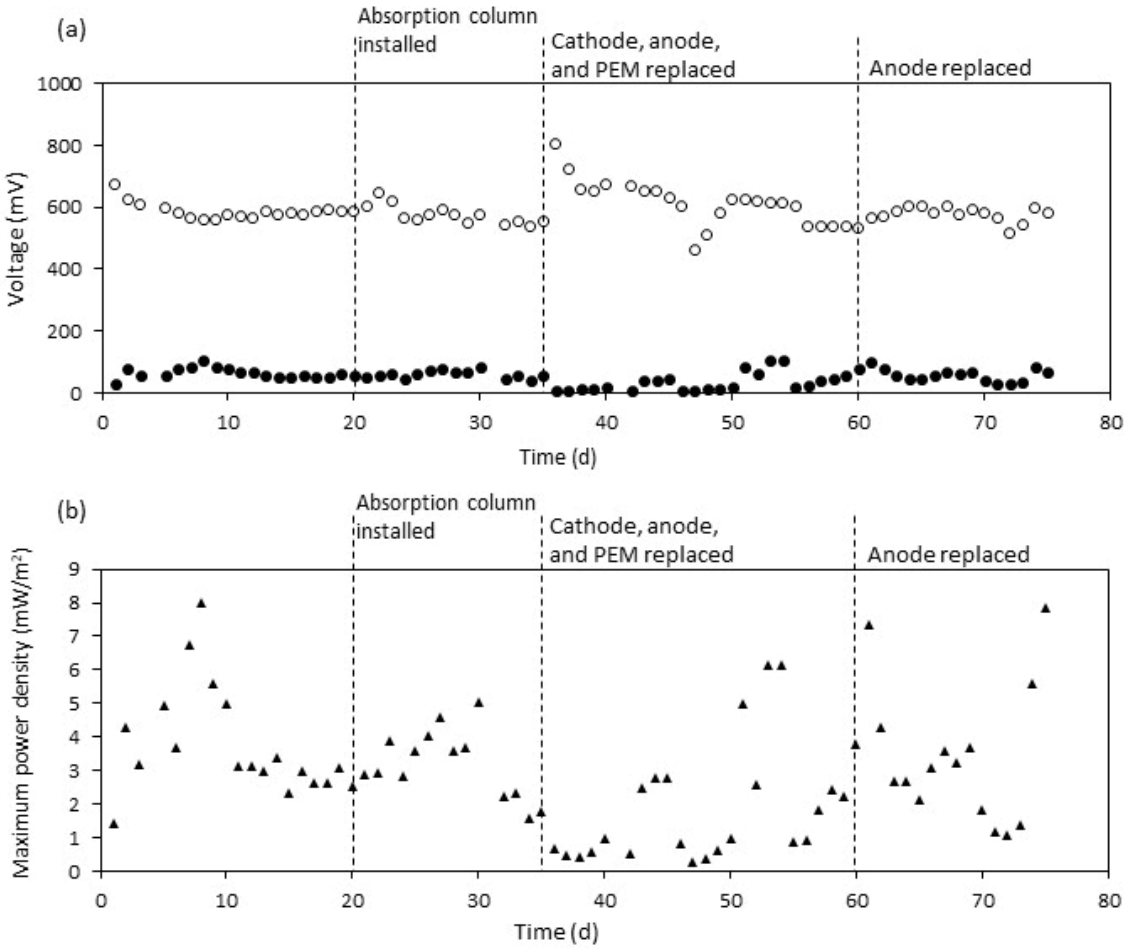

Fig. 4. (a) OCV (○) and voltage at an external resistance of 1,000 Ω (•) and (b) maximum power density (▲) of the MFC.

3.3. Microbial community analysis

The 16S rRNA gene clone library results for bacteria and archaea in the sludge from the anaerobic compartment are summarized in Fig. 7. Phylogenetic trees of the 16S rRNA gene clone libraries are shown in Fig. S4. From the results of the 16S rRNA gene clone library of bacteria (Fig. 7a), *Streptococcus macedonicus*, which is a fermentative bacterium, was found at the highest relative abundance (38% of total clones) in the anaerobic compartment. *Desulfovibrio vulgaris* and *Thermodesulfovibrio yellowstonii*, which are sulfate-reducing bacteria, were observed at relative abundances of 9% and 4% of total clones, respectively. In addition, *Syntrophomonas curvata*, which is a syntrophic bacterium associated with methanogens, were found at a relative abundance of 4% of total clones. From the results of the 16S rRNA gene clone library of archaea (Fig. 7b), *Methanosaeta concilii*, which is an acetate-utilizing methanogen, was the predominant archaea in the anaerobic compartment (83% of total clones). Other methanogens, including *Methanospirillum hungatei JF-1*, and *Methanosaeta thermophila*, were also observed.

Fig. 8 shows the microbial community in the biofilm attached to the anode electrode, and the phylogenetic trees of the 16S rRNA gene clone libraries are shown in Fig. S5. *S. macedonicus* was the predominant bacteria at a relative abundance of 48% of total clones. *Halothiobacillus neapolitanus*, which is a sulfur-oxidizing bacterium, was observed at a relative abundance of 12% of total clones, suggesting the occurrence of sulfur oxidation at the anode by sulfur-oxidizing bacteria. The presence of sulfur-oxidizing bacteria on the anode electrodes of MFCs treating sulfide wastewater or high-sulfate wastewater has previously been reported [10,11,13].

Moreover, *Klebsiella pneumoniae*, which is an exoelectrogenic microorganism [24,25], was found in the biofilm on the anode electrode at a relative abundance of 20% of the total clones. A *K. pneumoniae* biofilm was previously used in an MFC for the treatment of starch and glucose to generate electricity [24]. In addition, *K. pneumoniae* has been reported to be capable of producing an electrochemically active compound that serves as an electron mediator in MFCs. For the archaea on the anode electrode (Fig. 8b), the 16S rRNA gene clone library revealed a microbial community similar to that in the sludge in the anaerobic compartment. Methanogens, including *M. concilii*, *M. hungatei JF-1*, and *M. thermophila*, were observed. These results are in agreement with those of previous research that revealed the occurrence of methanogens (e.g., *Methanosaeta*) on the

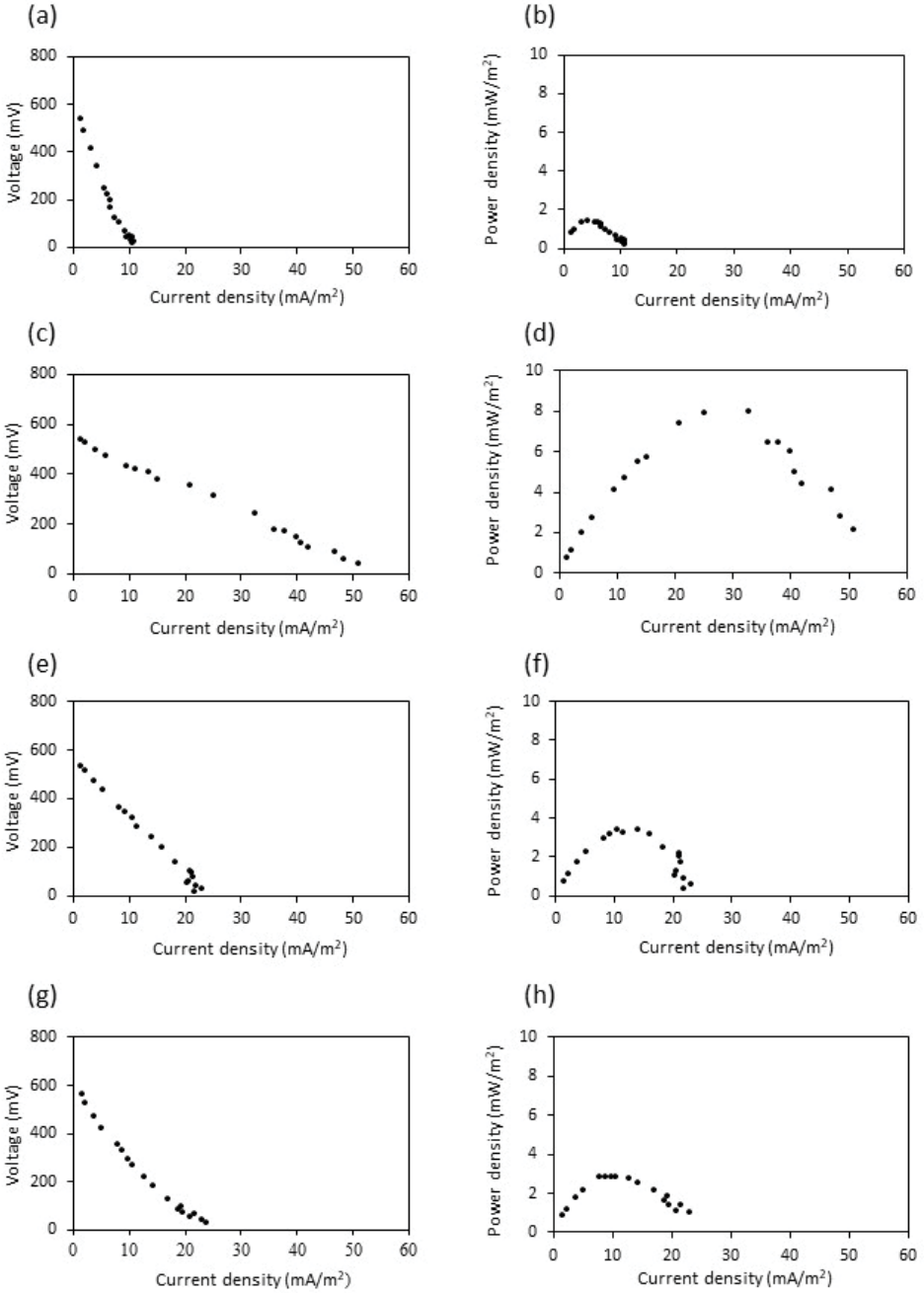

Fig. 5. Polarization curves on (a) day 1, (b) day 8, (e) day 14, and (g) day 21 and power density curves on (c) day 1, (d) day 8, (f) day 14, and (h) day 21.

anode electrodes [5,11]. Methanogenesis has been considered to be a major cause of electron loss in MFCs, which requires controlling approaches for suppression to increase the power production [26,27].

3.4. Possible mechanisms in the two-compartment single-chamber MFC

In the anaerobic compartment, the results of microbial community analysis clearly suggest the cooccurrence of fermentation, methanogenesis, and sulfate reduction processes, which result in COD and sulfate removal. These processes are generally expected in the anaerobic treatment of organic wastewater containing high sulfate concentrations at a COD:sulfate ratio of 10:1.

For the MFC compartment, the abiotic oxidation of hydrogen sulfide appears to play an important role in sulfide removal and electricity generation in the MFC. In addition, the presence of a sulfur-oxidizing bacterium (*H. neapolitanus*) on the anode electrode suggests the involvement of a microbially mediated sulfide oxidation process. In general, hydrogen sulfide oxidation by sulfur-oxidizing

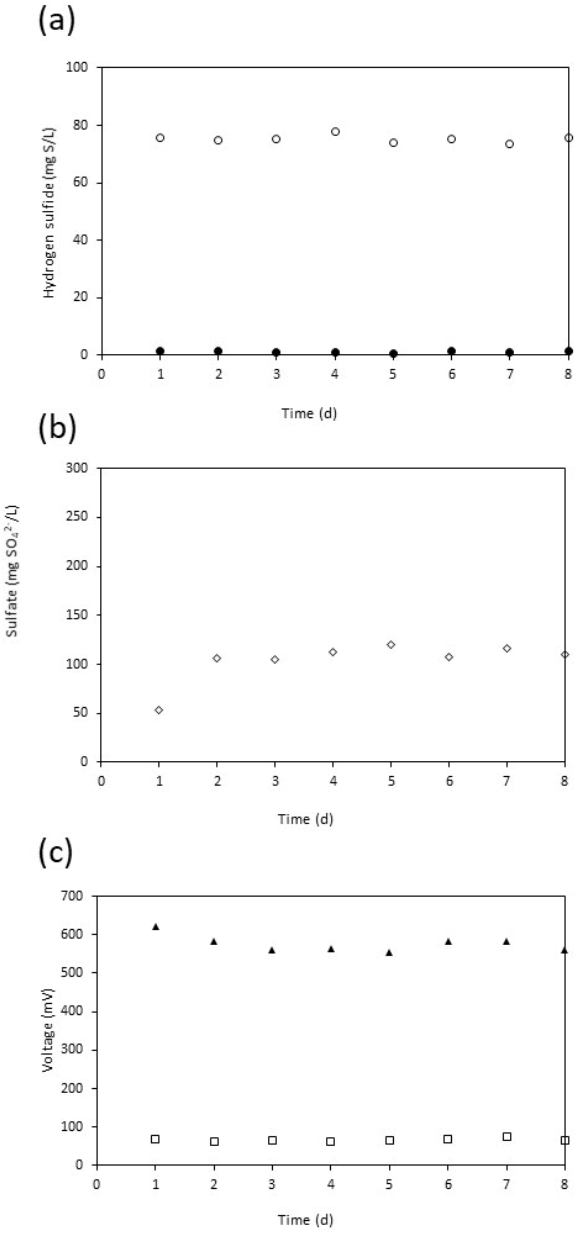

Fig. 6. Concentrations of (a) hydrogen sulfide in the influent (○) and effluent (•), (b) sulfate in the effluent (◊), and (c) the OCV (▲) and voltage at an external resistance of 1,000 Ω (□) in the abiotic fuel cell.

bacteria generates sulfate [28], and the abiotic oxidation of hydrogen sulfide in the abiotic fuel cell also resulted in sulfate (Fig. 6b). However, sulfate was not found as a product in the MFC compartment. A possible explanation is that since the synthetic wastewater fed into the two-compartment MFC contained an organic compound, sulfate reduction, which requires organic compounds, could also occur in the MFC compartment. This process could convert sulfate back to hydrogen sulfide, resulting in very low sulfate concentrations in the system. On the other hand, despite only small amounts removed, COD removal in the MFC compartment, was likely to occur by (1) exoelectrogenic microorganisms (*K. pneumoniae*), which can simultaneously produce electricity, (2) methanogenesis, which can contribute to electron loss in the MFC, and/or (3) sulfate reduction with sulfate produced from hydrogen sulfide oxidation.

3.5. Limitations and suggestions for future research

The results of this study demonstrate that it is technically feasible to apply an MFC integrated with an absorption column for the simultaneous removal of hydrogen sulfide in effluent and biogas from anaerobic wastewater treatment. However, the designed configurations (e.g., the size of each compartment and absorption column, the size of the fuel cell apparatus, and the types of water distribution devices in the absorption column) and operating parameters (e.g., the HRT and recirculation flow rate in the absorption column) still have not been optimized in this study. These factors could essentially affect the system performance and construction cost. For example, a longer HRT could increase the treatment efficiencies in both compartments, but it could also contribute to larger volumes and construction costs. A higher recirculation flow rate in the absorption column generally results in larger droplets, which could lower the absorption efficiency. Nevertheless, optimized flow rates for recirculation in the absorption columns largely depend on the column design, water distribution devices, and scale of the system, which we have not been studied in the current work. Further research on these issues, including pilot-scale tests, is still required.

In addition, in this study, we tested the proposed system using synthetic wastewater containing a COD:sulfate ratio of 10:1 since anaerobic treatment with biogas production is achievable at this ratio [20,21,29], but further treatment of the biogas is still required to remove hydrogen sulfide. At COD:sulfate ratios of less than 10:1, failures of anaerobic wastewater treatment processes for methane production have been reported [29]. On the other hand, at high COD:sulfate ratios, hydrogen sulfide in the biogas is usually not an issue. Nevertheless, our study is a proof of concept, and the suitable range of COD:sulfate for the proposed system should be further studied. Moreover, to calculate the energy efficiency or coulombic efficiency of this system, all possible sulfur products (e.g., S_0/S_x^{2-}, $S_2O_3^{2-}$, and SO_4^{2-}) from sulfide oxidation in the MFC should be quantified in future studies.

4. Conclusion

This study demonstrated the successful application of a two-compartment single-chamber MFC integrated with a packed absorption column for the treatment of organic wastewater containing high sulfate concentrations as well as for hydrogen sulfide removal in the effluent and biogas. The system removed COD and sulfate at removal efficiencies of 81.3% ± 4.5% and 100% ± 0.0%, respectively. In addition, hydrogen sulfide, which is the end product of sulfate reduction, was removed from both the aqueous and gaseous phases in the MFC compartment, resulting in a hydrogen sulfide concentration in the effluent of 38.2 ± 19.8 mg/L and in the biogas of 153 ± 63 ppm. According to the microbial community analysis using 16S rRNA gene clone

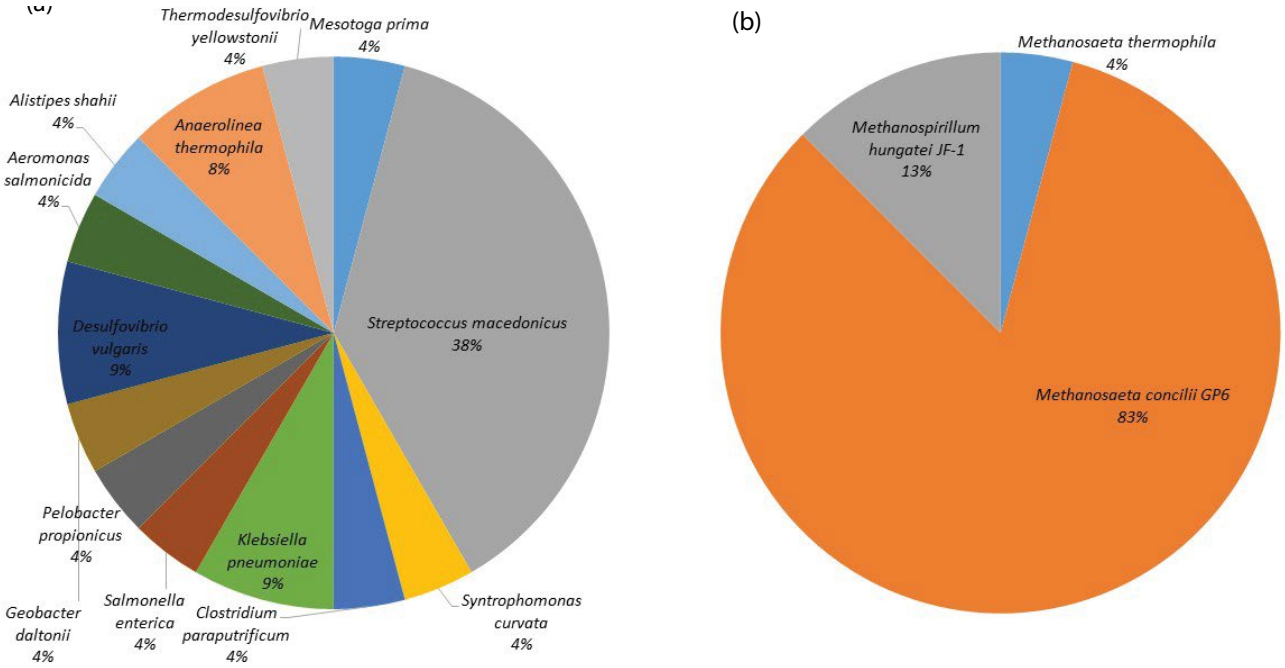

Fig. 7. Relative abundances in the 16S rRNA gene clone libraries of (a) bacteria (25 clones) and (b) archaea (24 clones) in the sludge in the anaerobic compartment.

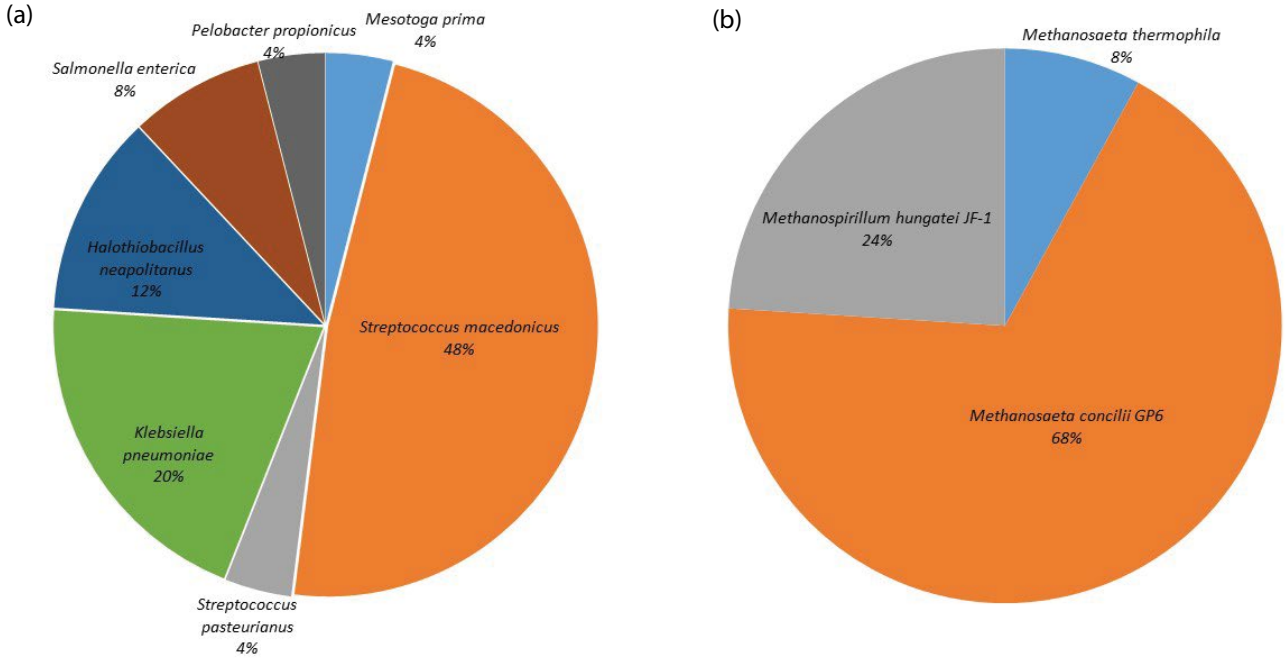

Fig. 8. Relative abundances in the 16S rRNA gene clone libraries of (a) bacteria (25 clones) and (b) archaea (25 clones) in the biofilm attached to the anode electrode.

libraries, *S. macedonicus*, *H. neapolitanus*, and *K. pneumoniae* were observed in the biofilm on the anode electrode.

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References

[1] P.N.L. Lens, F. Omil, J.M. Lema, L.W. Hulshoff Pol, Biological Treatment of Organic Sulfate-Rich Wastewater, P.N.L. Lens,

L.W. Hulshoff Pol, Eds., Environmnetal Technologies to Treat Sulfur Pollution: Principles and Engineering, IWA Publishing, London, 2000.
[2] H. Liu, R. Rmnarayanan, B.E. Logan, Production of electricity during wastewater treatment using a single chamber microbial fuel cell, Environ. Sci. Technol., 38 (2004) 2281–2285.
[3] H. Liu, S. Cheng, B.E. Logan, Production of electricity from acetate or butyrate using a single-chamber microbial fuel cell, Environ. Sci. Technol., 39 (2005) 658–662.
[4] Z. Du, H. Li, T. Gu, A state of the art review on microbial fuel cells: a promising technology for wastewater treatment and bioenergy, Biotechnol. Adv., 25 (2007) 464–482.
[5] Z. He, S.D. Minteer, L.T. Angenent, Electricity generation from artificial wastewater using an upflow microbial fuel cell, Environ. Sci. Technol., 39 (2005) 5262–5267.
[6] B. Min, J.R. Kim, S.E. Oh, J.M. Regan, B.E. Logan, Electricity generation from swine wastewater using microbial fuel cells, Water Res., 39 (2005) 4961–4968.
[7] S.E. Oh, B.E. Logan, Hydrogen and electricity production from a food processing wastewater using fermentation and microbial fuel cell technologies, Water Res., 39 (2005) 4673–4682.
[8] K. Rabaey, P. Clauwaert, P. Aelterman, W. Verstraete, Tubular microbial fuel cells for efficient electricity generation, Environ. Sci. Technol., 39 (2005) 8077–8082.
[9] K. Rabaey, K. Van de sompel, L. Maignien, N. Boon, P. Aelterman, P. Clauwaert, H.T. Pham, J. Vermeulen, M. Verhaege, P. Lens, W. Verstraete, Microbial fuel cells for sulfide removal, Environ. Sci Technol., 40 (2006) 5218–5224.
[10] A. Sangcharoen, W. Niyom, B.B. Suwannasilp, A microbial fuel cell treating organic wastewater containing high sulfate under continuous operation: performance and microbial community, Process Biochem., 50 (2015) 1648–1655.
[11] W. Niyom, D. Komolyothin, B.B. Suwannasilp, Important role of abiotic sulfide oxidation in microbial fuel cells treating high-sulfate wastewater, Eng. J., 22 (2018) 23–37.
[12] P.L. McCarty, J. Bae, J. Kim, Domestic wastewater treatment as a net energy producer – can this be achieved?, Environ. Sci., Technol., 45 (2011) 7100–7106.
[13] M. Sun, Z.H. Tong, G.P. Sheng, Y.Z. Chen, F. Zhang, Z.X. Mu, H.L. Wang, R.J. Zeng, X.W. Liu, H.Q. Yu, L. Wei, F. Ma, Microbial communities involved in electricity generation from sulfide oxidation in a microbial fuel cell, Biosens. Bioelectron., 26 (2010) 470–476.
[14] F. Zhao, N. Rahunen, J.R. Varcoe, A. Chandra, C. Avignone-Rossa, A.E. Thumser, R.C.T. Slade, Activated carbon cloth as anode for sulfate removal in a microbial fuel cell, Environ. Sci. Technol., 42 (2008) 4971–4976.
[15] N. Eaktasang, H.S. Min, C. Kang, H.S. Kim, Control of malodorous hydrogen sulfide compounds using microbial fuel cell, Bioprocess. Biosyst. Eng., 36 (2013) 1417–1425.
[16] APHA, AWWA, WEF, Standard Methods for the Examination of Water and Wastewater, 21st ed., American Public Health Association, American Water Works Association, Water Environment Federation, Washington, DC, 2005.
[17] ASTM, Annual Book of ASTM Standards, 1996, Pennsylvania, U.S.A., D4658–D4692.
[18] B. Boonchayaanant, P.K. Kitanidis, C.S. Criddle, Growth and cometabolic reduction kinetics of a uranium- and sulfate-reducing *Desulfovibrio*/*Clostridia* mixed culture: temperature effects, Biotechnol. Bioeng., 99 (2008) 1107–1119.
[19] S.R. Forschner, R. Sheffer, D.C. Rowley, D.C. Smith, Microbial diversity in Cenozoic sediments recovered from the Lomonosov Ridge in the Central Arctic Basin, Environ. Microbiol., 11 (2009) 630–639.
[20] Y. Hu, Z. Jing, Y. Sudo, Q. Niu, J. Du, J. Wu, Y. Li, Effect of influent COD/SO_4^{2-} ratios on UASB treatment of a synthetic sulfate-containing wastewater, Chemosphere, 130 (2015) 24–33.
[21] X. Lu, G. Zhen, J. Ni, T. Hojo, K. Kubota, Y. Li, Effecf of influent COD/SO_4^{2-} ratios on biodegradation behaviors of starch wastewater in an upflow anaerobic sludge blanket (UASB) reactor, Bioresour. Technol., 214 (2016) 175–183.
[22] W. Habermann, E.H. Pommer, Biological fuel cells with sulphide storage capacity, Appl. Microbiol. Biotechnol., 35 (1991) 128–133.
[23] P.K. Dutta, K. Rabaey, Z. Yuan, J. Keller, Spontaneous electrochemical removal of aqueous sulfide, Water Res., 42 (2008) 4965–4975.
[24] L. Zhang, S. Zhou, L. Zhuang, W. Li, J. Zhang, N. Lu, L. Deng, Micorbial fuel cell based on *Klebsiella pneumoniae* biofilm, Electrochem. Commun., 10 (2008) 1641–1643.
[25] B.E. Logan, Exoelectrogenic bacteria that power microbial fuel cells, Nat. Rev. Microbiol., 7 (2009) 375–381.
[26] K.J. Chae, M.J. Choi, K.Y. Kim, F.F. Ajayi, W. Park, C.W. Kim, I.S. Kim, Methanogenesis control by employing vaious environmental stress conditions in two-chambered microbial fuel cells, Bioresour. Technol., 101 (2010) 5350–5357.
[27] S. Bagchi, M. Behera, Methanogenesis suppression in microbial fuel cell by aluminium dosing, Bioelectrochemistry, 129 (2019) 206–210.
[28] M.T. Madigan, J.M. Martinko, J. Parker, Brock Biology of Microorganisms, 10th ed., Pearson Education Inc., Upper Saddle River, NJ, 2003.
[29] L.W. Hulshoff Pol, P.N.L. Lens, J. Weijma, A.J.M. Stams, New developments in reactor and process technology for sulfate reduction, Water Sci. Technol., 44 (2001) 67–76.

Supplementary information

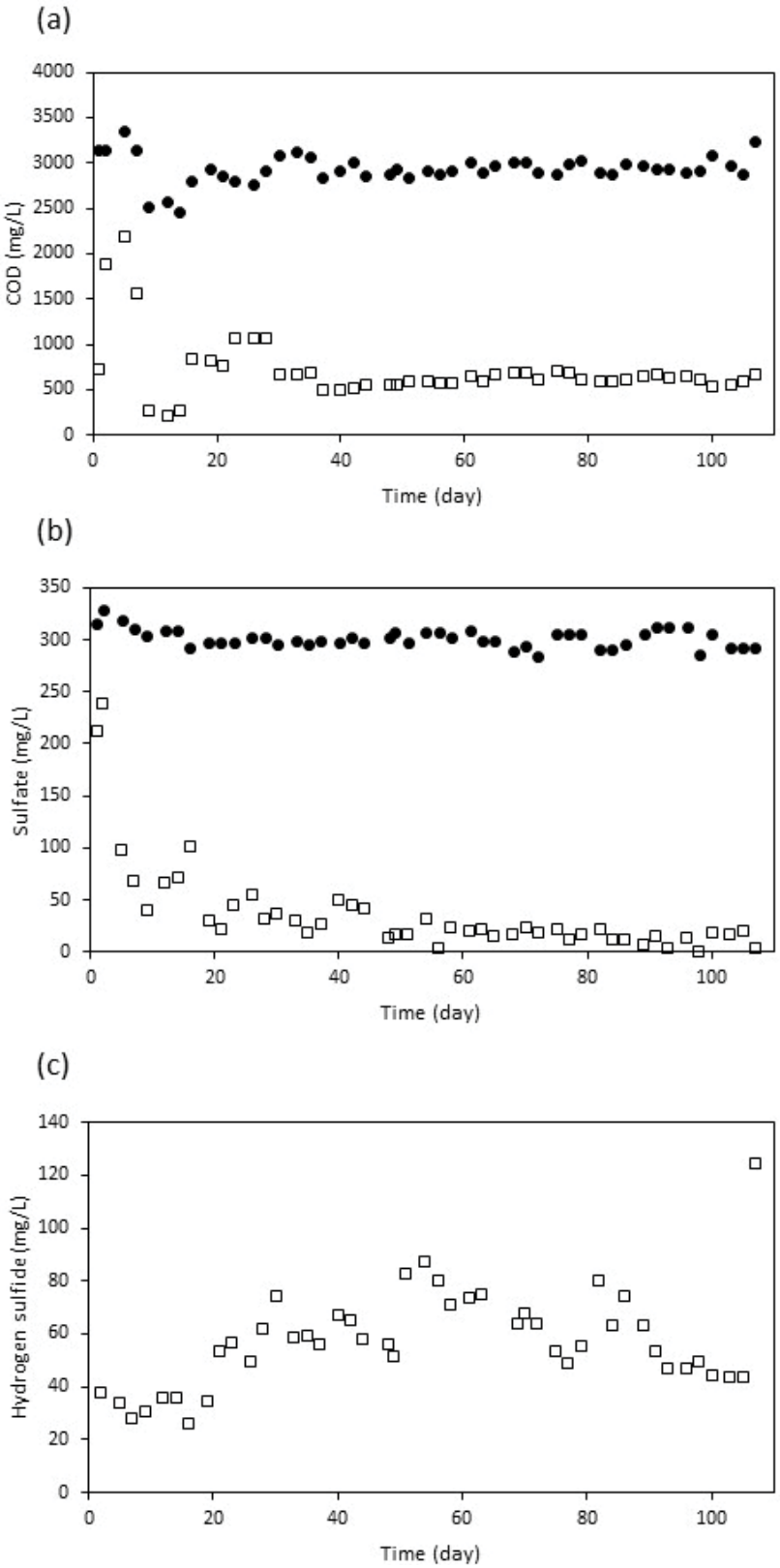


Fig. S1. Concentrations of (a) COD, (b) sulfate, and (c) hydrogen sulfide in the influent (•) and effluent (□) of the anaerobic compartment before MFC operation.

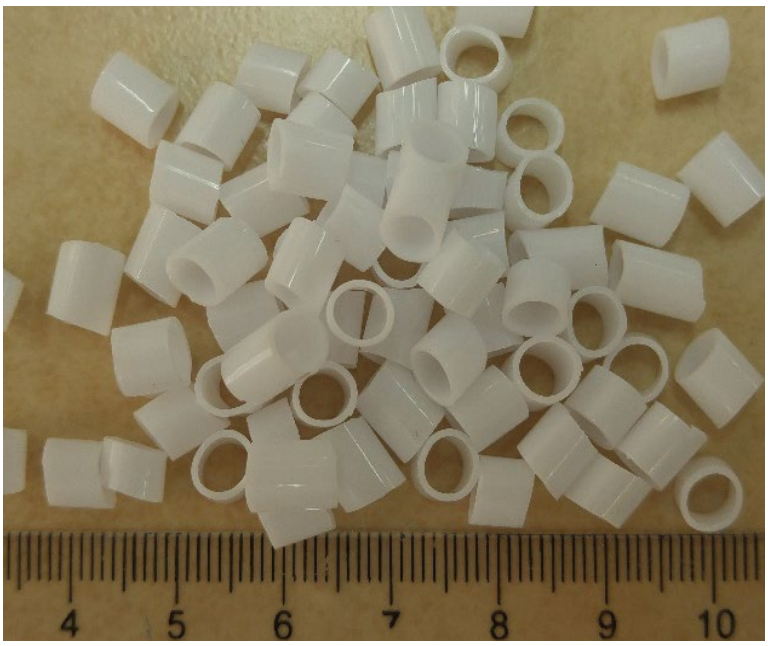
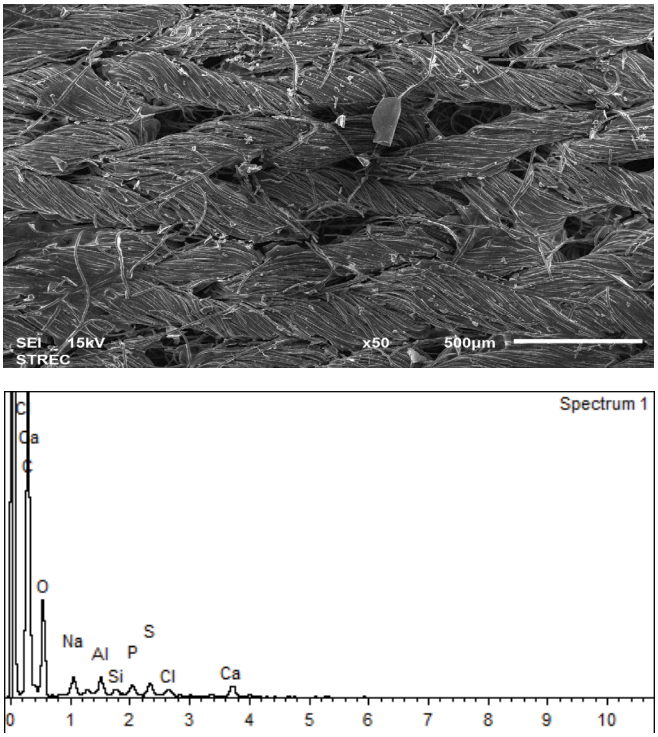


Fig. S2. Propylene plastic media used in the absorption column.



Elements	% by weight
C	65.33
O	28.26
Na	1.19
Al	0.93
Si	0.34
P	0.80
S	1.00
Cl	0.62
Ca	1.55

Fig. S3. SEM-EDX analysis of the surface of the anode electrode removed from the MFC on day 61.

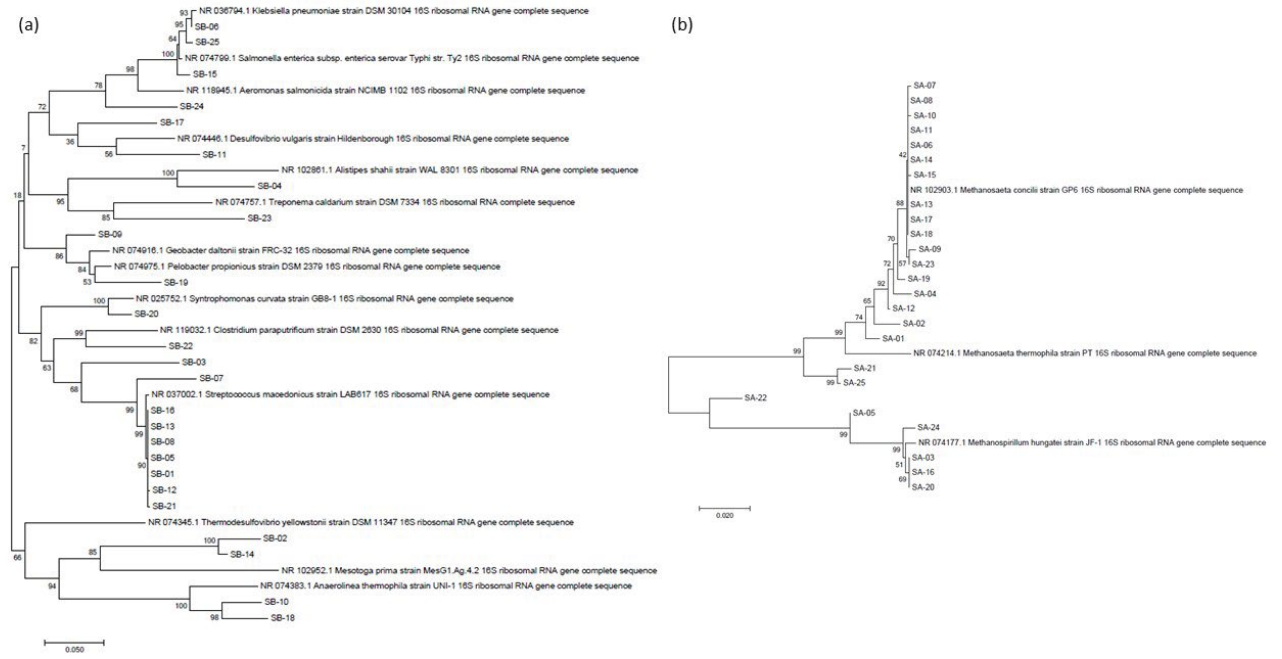


Fig. S4. Phylogenetic trees of the 16S rRNA gene clone libraries of (a) bacteria and (b) archaea in the sludge from the anaerobic compartment.

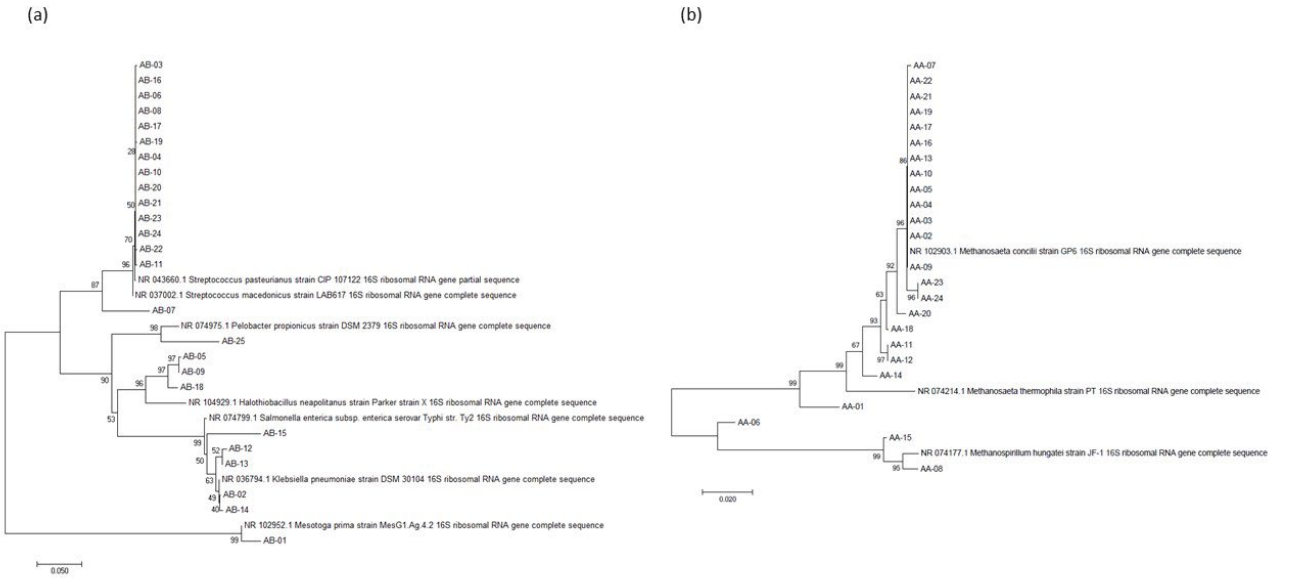


Fig. S5. Phylogenetic trees of the 16S rRNA gene clone libraries of (a) bacteria and (b) archaea in the biofilm attached to the anode electrode.