5-8-2014

Treating Wastes with Microbial Fuel Cells (MFCs) and Microbial Electrolysis Cells (MECs)

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Recommended Citation
Li, Yan, "Treating Wastes with Microbial Fuel Cells (MFCs) and Microbial Electrolysis Cells (MECs)" (2014). Master's Theses. 589. http://digitalcommons.uconn.edu/gs_theses/589

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Treating Wastes with Microbial Fuel Cells (MFCs) and Microbial Electrolysis Cells (MECs)

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B.S., Jilin University, 2012

A Dissertation
Submitted in Partial Fulfillment of the Requirements for the Degree of Master of Science at the University of Connecticut 2014
Treating Wastes with Microbial Fuel Cells (MFCs) and Microbial
Electrolysis Cells (MECs)

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University of Connecticut
2014
Acknowledgements

Thanks to my committee:

Baikun Li   Ph.D
Timothy Vadas   Ph.D
Yu Lei   Ph.D

Thanks to my laboratory:

Carlo Santoro   Ph. D
Udayarka Karra   Ph.D
Bingchuan Liu   Ph.D
Zhiheng Xu   Ph.D
Secil Tutar   MS

And for the help in some part of my research:

Yining Wu   Ph.D
Lichun Zhang   Ph.D
Sampada Puranik   Ph.D
Hongwei Luan   Ph.D
Guoxian Huang   Ph.D
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Abstract

Two typical oxidized-status metals (Fe (III) and Cr (VI)) were studied as electron acceptors on cathodes in single chamber microbial fuel cells (SCMFCs) to explore novel sustainable technology for metal treatment in wastewater. The batch-mode tests indicated that the voltages of SCMFCs steadily increased with Fe(III) concentrations (10mg L-1, 30mg L-1, and 50mg L-1) and Cr (VI) concentrations (1mg L-1, 3mg L-1, and 10mg L-1). The maximum power density and current density reached at 658.34mW m-2 and 0.46mA cm-2 at 50mg L-1 of Fe(III), and reached at 419.31mW m-2 and 0.27mA cm-2 at 10 mg L-1 Cr (VI). The conversion efficiency of Cr (VI) and Fe (III) were high (>89%), and coulombic efficiency ranged 23-100% at different metal concentrations. Cr (VI) concentration of 10 mg L-1 started to irreversibly inhibit SCMFCs. In addition, the open circuit potentials (OCPs) of anodes and cathodes well reflected the organic substrate removal in anode and metal reduction on cathode. Cathode liner sweep voltammetry (LSV) clearly showed the electrochemical activity increased with metal concentrations, and the cathode of Fe (III) had better LSV performance than Cr (VI). This study demonstrated high power generation of SCMFCs with metals as electron acceptors, and revealed the great potential of expanding MFCs for diverse waste treatment.
Chapter 1: Introduction
Wastewater containing diverse organic substances (e.g. fatty acid, carbohydrate, protein) and heavy metals (e.g. chromium, copper, and cadmium) has caused severe environmental pollution. The organic pollutants are removed by aerobic treatment traditionally but it consumes large amounts of electrical energy for aeration. However, wastewaters are increasingly being paid more and more attention as a renewable resource for the production of electricity, fuels and chemicals. So far, anaerobic digestion has been proven as the only method to extract the energy from wastewater.

Microbial fuel cell (MFC) is a promising biotechnology capable of converting organic substrates in wastewaters (e.g. domestic wastewater, swine wastewater, leachate, and urine) to electricity [1-8]. The electrogenic microorganisms colonized on the anode surface degrade organic substrates and generate electrons, which then transfer to the cathode through external circuit and complete reduction reactions [9]. The transfer of electrons obtained from an electron donor to the anode electrode is occurred either through direct contact, nanowires, or mobile electron shuttles in the anode compartment. During electron production protons are also produced in excess. The protons migrate through the cation exchange membrane into the cathode chamber. In the meanwhile, the electrons flow from the anode through an external resistance to the cathode where they react with final electron acceptors (e.g. oxygen) and protons. Besides, most effective electricigens (e.g. Geobacteraceae, Shewanella) on the anode are proven to be Fe(III) reducers[32].
Two MFC configurations, two-chamber MFCs (2C-MFCs) and single-chamber MFCs (SC-MFCs) have been extensively studied [10-15]. Anode and cathode chambers are separated by membranes (e.g. proton exchange membrane) in 2CMFCs [10], while membranes are removed in SCMFCs and both anode and cathode are contacted with anodic solution (wastewater), with oxygen as cathodic electron acceptor [11,16,17]. Due to the removal of membranes between anode and cathode, SCMFCs have lower internal resistance and higher power generation than 2CMFCs [16]. But oxygen could penetrate through cathode and diffuse into anode solution of SCMFCs, which leads to lower coulombic efficiency than 2CMFCs [9,16].

For the selection of materials of electrodes, anodic materials must be conductive, biocompatible, and chemically stable in the reactor solution. The most versatile electrode material is carbon available as compact graphite plates, rods, or granules, carbon cloth, carbon brush, etc. For the cathode, electrode should also be conductive and chemically stable, usually, carbon materials are also adopted in cathodes for the transferring the electrons from the external circuit to the final electron acceptors in the cathode solution.

Oxygen from air has been widely used as the final electron acceptor on cathodes in SCMFCs, due to its abundance and high redox potential \( E_{\text{mfc}}=0.805 \text{V}, \ pO_2=0.2, \ pH=7 \) [9]. However, to fully explore SCMFCs as an electro-bio-chemical system to treat wastes, it is important to utilize anode/cathode reactions for oxidizing/reducing diverse contaminants in wastewater. Until now, many studies have focused on the removal of organic substances in anaerobic anodes of 2CMFCs and SCMFCs [1-8, 15]. For election
acceptance reactions on cathodes, denitrification (nitrate reduction), fumarate and chlorinated compound reduction were examined in 2CMFCs [18-21]. In fact, metals (e.g. Cr$^{6+}$, Mn$^{7+}$, and Fe$^{3+}$) could also be used as electron acceptors on cathodes, and thus being removed from wastewater [22-24]. But until now, metal reduction has only been studied in 2CMFCs to prevent the transfer of metals from cathode to anode, and to eliminate the potential inhibition on anodic electrogenic bacteria. Besides, the internal resistance ($R_{\text{in}}$) of 2CMFCs is higher than that of SCMFCs, and the cost is high due to membranes and complicated two-chamber structure. Therefore, in order to enhance power generation and simplify MFC configuration for scale up application, it is critical to explore metal reduction in SCMFCs.

The pollution of heavy metals has gained worldwide attention due to their toxicity, difficult disposal, and accumulation in the living organisms. Therefore, treatment of wastewater contaminated by heavy metals is an important environmental issue. Several methods have been developed to treat wastewater contaminated by heavy metals, including ion exchange, chemical precipitation, electrolysis, and reverse osmosis. However, most of these methods required high operational and maintenance costs, and generated toxic sludge. Therefore, treating metal wastes with MFC would be an innovative method to reduce metals and harvest power at the same time.

In order to determine the possibilities of treating metals in SCMFCs, this study targeted the reduction of two typical metals, chromium (Cr) and iron (Fe) in SCMFCs. Hexavalent chromium is normally present in wastewaters from electroplating, pigment,
and lumber industries [25], and has posed a serious risk to human, animals, and environment due to its high solubility, high toxicity, and potential carcinogenicity [26]. The accumulation of Cr(VI) in living tissues throughout food chain causes many serious health problems. Numerous traditional physical, chemical, and biological processes have been used to reduce Cr(VI) to Cr(III) [27,28], but the problems of excessive chemical usage, high energy consumption, and toxic waste sludge treatment have lowered treatment efficiency and increased the operational costs.

In terms of Fe(III), it is abundantly present in the biosphere and some anaerobic bacteria (e.g. *Geobacter* species, *Shewanella* species, *Desulfovibrio vulgaris*, *Thermotoga maritima*) can utilize it as the terminal electron acceptor for their growth [29-31,44], which made it an ideal electron acceptor in MFCs. Most effective current-producing anaerobic microorganisms (e.g. *Geobacteraceae*, *Shewanella*) on the anode were usually Fe(III) reducers[32], and thus adding Fe(III) in electrolytes is expected to promote the growth of anaerobic microorganisms and improve the electron producing capabilities of MFCs. Moreover, both Cr and Fe have high standard potentials ($E_0=1.33V$ for Cr from Cr(VI) to Cr(III), $E_0=0.77V$ for Fe from Fe(III) to Fe(II), vs SHE), which could increase power generation in MFCs.

Microbial fuel cell is one of the bioelectrochemical systems(BESs) which uses microorganisms to catalyze an oxidation and reduction reaction on an anodic or cathodic electrode, respectively. A BES is called a microbial fuel cell(MFC) if electricity is harvested from the circuit and the Gibbs free energy change of the corresponding reaction
is negative. However, when the Gibbs free energy change of the overall reaction is positive and extra electrical power need to be input, then it is regarded as a microbial electrolysis cell (MEC). Hydrogen production is conducted in MECs widely since this requires much lower voltage than the theoretical value of 1.2V required for water electrolysis to get hydrogen and when compared with fermentative hydrogen production, the MEC process can utilize a variety of organic materials.

A lot of heavy metals, such as Cu$^{2+}$, Hg$^{2+}$, and Cr$^{6+}$, could be reduced with MFCs to metal elements or ions with lower chemical valences to reduce the metal pollution. However, other metals (e.g. Ni$^{2+}$, Cd$^{2+}$, Zn$^{2+}$) could not use MFC because of their negative standard potentials, which make the difference between the anodic and cathodic potentials too small to drive the electrons transferred from the anode to cathode. Therefore, in these situations, MEC is applied to reduce the metals although this process consumes electricity to support MEC. On account of these situations, MFC-MEC combined system is designed to solve the energy problems. MFC could produce power when reducing oxygen or heavy metals and the energy recovered are stored in the capacitors and later to be used to support the MEC system to continuously reduce heavy metal pollutions. Besides, different metal solutions are designed to be injected together in the cathode chamber and some of them are reduced in the function of MFC and some of them are reduced in the function of MEC when the power from MFC is discharged. In order to realize this idea, metal reduction in the MFC should be realized first to determine how much power a MFC could produce and how many MFCs should be connected together to power a MEC.
Therefore, there were four main tasks in this study. First, the relationship between power generation and concentration of Cr(VI) and Fe(III) in the SCMFCs was explored, and the maximum Cr (VI) concentration that bacteria could tolerate was determined. Second, the conversion efficiencies and coulombic efficiencies of Cr\(^{6+}/Cr^{3+}\) and Fe\(^{3+}/Fe^{2+}\) in SCMFCs were calculated for the efficiency of using metals rather than oxygen as electron acceptors. Third, important biochemical and electrochemical parameters (e.g. pH, open circuit potential (OCP), and R\(_{in}\)) were measured in anode and cathode of SCMFCs to determine the metal impacts. Finally, linear sweep voltammetry (LSV) was performed on cathodes to characterize the electrochemical activity of Cr (VI) and Fe (III).
Chapter 2: Materials and methods

2.1 MFC and electrode materials
Membraneless SCMFCs made of plexiglass bottles (length: 9.5cm, diameter: 4.5cm, and total volume of 150ml) were used in this study. Carbon brush (length: 4cm, diameter: 4cm, Mill-Rose Carbon Fiber Brush Anode) fixed on a titanium wire was used as the anode electrode, and plugged into the SCMFC anode chamber. Carbon cloth (effective area: 3cm², 30%wt polytetrafluoroethylene (PTFE), ETek) was used as the cathode and inserted into the glass extension on the lower part of SCMFCs (Figure 1). The water-side of cathodes was loaded with 0.5mg cm⁻² Pt as the cathodic catalyst, and the air-side was coated with three layers of PTFE [16]. Pt was adopted to accelerate the inoculation stage during which electrogenic bacteria would grow on the anode preferentially and steadily and biofilm on the cathode. However, without Pt catalyst, it would take twice as long as time for the bacteria growth on electrodes of SCMFCs, which would greatly affect the operation time of SCMFCs. After inoculation, the air-side of cathodes was covered with a gasket to prevent oxygen leading to the water-side (Figure 1), so that oxygen could not act as the electron acceptor on cathodes, and only metals were electron acceptors.

2.2 Inoculation and SCMFC operation
The SCMFCs were inoculated with municipal wastewater taken from the influent of the Wastewater Treatment Facility at the University of Connecticut, which contained sufficient microorganisms for MFCs. Sodium acetate (20 mmol L⁻¹) was added during batch mode inoculation to provide sufficient carbon substrates for bacterial growth. During inoculation stage, Pt air-cathode was adopted to accelerate the growth of electrogenic bacteria on the anode and the biofilm growth on the cathode. After 7 days,
the voltages of SCMFCs reached 0.25 V at the external resistance \( R_{\text{ext.}} \) of 500\( \Omega \), and stabilized during one-month inoculation. Wastewater was then replaced with phosphate buffer solution (PBS, 50mmol L\(^{-1}\), pH 6.5) in anode chamber, which contained \( \text{NH}_4\text{Cl} \) 0.31g, KCl 0.13g, \( \text{NaH}_2\text{PO}_4 \cdot 2\text{H}_2\text{O} \) 5.618g, \( \text{Na}_2\text{HPO}_4 \cdot 12\text{H}_2\text{O} \) 6.155g, 1ml trace mineral solution and 1ml trace vitamin per liter solution [33]. Sodium acetate (20mmol L\(^{-1}\)) was added as carbon source for microorganisms. \( \text{K}_2\text{Cr}_2\text{O}_7 \) and \( \text{FeCl}_3 \cdot 6\text{H}_2\text{O} \) solution were also injected to SCMFCs, respectively, to provide Cr(VI) and Fe(III) as electron acceptors. Then the air-side of cathode was sealed with gasket to prevent the diffusion of oxygen from the air. SCMFCs were sparged with \( \text{N}_2 \) for 10 min to remove the residual dissolved oxygen to prevent the oxygen inhibition to electrogenic bacteria on anodes and the competition with Cr (VI) and Fe (III) as electron acceptors on cathodes. All the experiments were carried out at 30°C in duplicate at each cycle, and at least two cycles for each metal concentration.

Three concentrations of Cr(VI) (1mg L\(^{-1}\), 3mg L\(^{-1}\), and 10mg L\(^{-1}\)) and Fe(III) (10mg L\(^{-1}\), 30mg L\(^{-1}\), and 50mg L\(^{-1}\)) were examined individually in batch-mode SCMFCs. Previous studies showed that 1mg L\(^{-1}\) of Cr (VI) was the threshold inhibition concentration for microorganisms in wastewater [34]. Cr (VI) concentrations higher than 1 mg L\(^{-1}\) were tested in this study to simulate wastewaters with higher amounts of metals, and to find out whether MFCs could treat higher Cr (VI) concentrations. Fe (III) is not toxic to microorganisms, and the concentrations selected in this study represented the Fe (III) content in diverse conditions (e.g. wastewater, aquatic sediments, and aquifers) [35].

Since the concentrations of Cr(VI) (1mg L\(^{-1}\), 3mg L\(^{-1}\), and 10mg L\(^{-1}\)) were lower than...
Fe(III) (10mg L-1, 30mg L-1, and 10mg L-1), a cycle of Cr(VI) was about five days until the voltage dropped below 0.050V ($R_{\text{ext}}.500\Omega$), while a cycle of Fe(III) was a week. Thus, the power production of SCMFCs was studied at 2 days (48hrs) in a cycle when the reactions actively proceeded. In addition, because both Cr (VI) and Fe(III) were completely removed on the 5th day (120 hrs) after being added in SCMFCs, the metal conversion efficiency (%) was measured after 120hrs. Moreover, to elucidate whether adsorption or sedimentation also contributed to metal removal in SCMFCs, a control test was conducted on SCMFCs added with Cr (VI) and Fe (III) without being connected with external circuit throughout the test period.

2.3 Analysis and calculations

2.3.1 Power generation
The overall performance of an MFC could be evaluated through power output. Power was often normalized to some characteristic of the reactor in order to make it possible to compare power output of different systems. The choice of the parameter that was used for normalization depended on application, as many systems were not optimized for power production. The power output was usually normalized to the projected cathode or anode surface area.

The voltage across $R_{\text{ext}}$ (500Ω) was continuously recorded every 0.5h using a Keithley 2700 data logging system. Power curve measurement was conducted with different $R_{\text{ext}}$ (10-2940 Ω) and the voltage over each $R_{\text{ext}}$ was measured with a multimeter (Radioshack digital multimeter) until the reading stabilized after 5 minutes. A power curve described the power density as the function of the current density. The current density was
calculated according to $I=V/(R_{ext} \times A)$, and the power density was calculated according to $P=V^2/(R_{ext} \times A)$, where $A$ is the effective area of the cathode.

### 2.3.2 The anodic and cathodic open circuit potentials (OCPs), pH, internal resistance ($R_{in}$) and coulombic efficiency

The cell electromotive force was a thermodynamic value that did not take into account internal losses. The open circuit potential was the cell voltage that could be measured after some time in the absence of current. Theoretically, the OCP should approach the electromotive force. In practice, however, the OCP was substantially lower than the cell electromotive force, due to various potential losses. The open circuit potentials (OCPs) of anode and cathode were measured using a potentiostat (Gamry P600) with Ag/AgCl (+197mV vs SHE) as the reference, anode or cathode as the working electrodes, and Pt wire as the counter electrode. pH was measured using a portable pH meter (Thermo Fisher Scientific Orion 3-star). Since $V=OCV - R_{in} \times I$, $R_{in}$ was calculated as the slope of V-I curves generated from the polarization measurement excluding the higher voltages, which were the activation loss region [17].

The coulombic efficiency was defined as the ratio of total coulombs actually transferred to the anode from the substrate, to maximum possible coulombs if all substrate removal produced current. In this experiment, the coulombic efficiency was calculated as the ratio of the current flowing across SCMFCs and the theoretical current based on the total Cr(VI) and Fe(III) fed into the system.

$$CE = \frac{M \int_0^t l \, dt}{nFV(C_o - C_t)} \times 100\%$$
Where \( M \) is the molecular weight (52 for Cr and 56 for Fe); \( I \) is the current(A); \( n \) is the number of electrons accepted by metals on cathodes (3 for Cr (Cr\(^{6+}\) to Cr\(^{3+}\)), and 1 for Fe (Fe\(^{3+}\) to Fe\(^{2+}\)); \( F \) is Faraday’s constant \((9.64853\times10^4 \text{C mol}^{-1})\); \( V \) is the liquid volume of SCMFCs (L); and \( C_0 \) is the initial concentration(g L\(^{-1}\)) of metals fed into system and \( C_t \) is the concentration(g L\(^{-1}\)) after time \((t)\).

Microscopy observation of biofilm growth and metal deposition on the cathode was carried out with Scanning Electron Microscope (SEM) (Joel6335F). The chemical elements on the cathode surfaces were analyzed using the Energy-dispersive X-ray spectroscopy (EDX). Prior to SEM observation, the cathode samples were taken from SCMFCs and treated as previously described [36].

2.3.3 Linear sweep voltammetry (LSV)
The linear sweep voltammetry was a voltammetric method where the current at a working electrode was measured while the potential between the working electrode and a reference electrode was swept linearly in time. Oxidation or reduction of species was registered as a peak or trough in the current signal at the potential at which the species began to be oxidized or reduced. The electrochemical analysis of cathodes of SCMFCs was performed using linear sweep voltammetry (LSV) in a three-electrode configuration, with Ag/AgCl as the reference, cathodes as the working electrodes, and Pt wire as the counter electrode. The potentiostat (Gamry P600) was used to perform the LSV tests at a scan rate of 0.25mV s\(^{-1}\)and scanned from the cathode OCP to -0.45V.
2.3.4 Cr(VI) and Fe(III) measurement
Colorimetric standard methods with a spectrophotometer (Varian Cary 50 Bio UV-visible spectrophotometer) were used to measure Cr (VI) and Fe (II) concentrations. Specifically, 1,5-diphenycarbazide method was used for the concentration of soluble Cr (VI) by filtering the samples through 0.2 µm membrane syringe filters (Fisherbrand nylon) [37]. Fe (II) concentration was determined using 1,10-phenanthrolin colorimetric method [37], and Fe(III) was then calculated from the difference between total Fe concentration and Fe(II) concentration.

2.3.5 RAPD test
In order to test the diversity of microbial community in the system, RAPD(random amplified polymorphic DNA) test was conducted during the SCMFC experiments. The carbon brush of the anode and the biofilm on the cathode of SCMFC operated with Cr(VI), Fe(III), and traditional SCMFC without any metal solution(oxygen as electron acceptor, control) were examined. The DNA of these biofilm were extracted with soil DNA isolation kit(MO BIO Laboratories, Carlsbad). Then the extracted DNA were amplified by RAPD using 4 different random primers respectively. The sequences of the primers were: 1-5'GGCCAACGCGGCC3', 2-5'CCTGCAGCAGA 3', 3-5'CMCGYCRSCA 3', 4-5'CAGCAGCAGCAG 3'(Invitrogen). Amplification conditions were performed in a total volume of 25µl, containing template 2.5 µl, primer 2.5 µl, Master Mix 12.5 µl(AmpliTaq Gold 360 Master Mix, Applied Biosystems), 360GC enhancer 1.25 µl(AmpliTaq Gold 360 Master Mix, Applied Biosystems), and water 1.25 µl(Fisherbrand) and was carried out with the following conditions: single cycle of first denaturation at 94 °C for 10min, 45 circles containing 1min second denaturation at 95 °C, 1min annealing at 45 °C, and 1min extension at 72 °C, and one circle of final extension at
72°C for 10min. Amplification products were analyzed by electrophoresis in 2% agarose gels and detected by staining with SYBR safe(Invitrogen). The bands were compared among the same primers for anodes and cathodes and the total bands showed up with the same DNA template and 4 primers were added together to determine the diversity of the microbial community on the electrode. With more bands, it was assumed to be more diversity.
Chapter 3: Results and discussion

3.1 Power generation of SCMFCs fed with Cr (VI) and Fe (III)
Batch-mode SCMFCs had stable voltage generation for both Cr (VI) and Fe (III) after 15-20 hrs initial lag period (Figure 2). The SCMFCs fed with Fe (III) (10-50mg L-1) had higher voltage and current generation than those fed with Cr (VI) (1-10mg L-1) (Figures 2 and 3). The maximum power generation of SCMFCs was 658 mW m-2 at the Fe (III) concentration was 50mg L-1 and was 419 mW m-2 at the Cr (VI) concentration of 10mg L-1 (Figures 3). It should be noted that 10mg L-1 Cr(VI) was beyond the bacterial tolerance capability, and the voltage of SCMFCs became low (<10 mV) in the 2nd cycle.

For the SCMFCs fed with Fe (III), power density steadily increased with Fe (III) concentrations (Figure 3a). Higher concentration of Fe(III) led to higher power density, due to lower $R_{in}$, higher ionic strength, and higher electron transfer capacity [17,38]. Besides, Fe is an excellent terminal electron acceptor for anaerobic chemitrophic microorganisms [30]. SCMFCs reached the highest current density at the $R_{ext}$ of 10Ω (the lowest $R_{ext}$ used in this study) (Figure 3a), since low $R_{ext}$ increased the current production. Like power density, the current density increased with Fe (III) concentrations, and reach 0.46mA cm-2 at 50mg L-1.

For the SCMFC fed with Cr(VI), power density increased with Cr (VI) concentration. However, due to the toxicity of Cr (VI), the high power generation was only achieved in the 1st cycle (Figure 3b), while stable power generation still continued in the 2nd cycle at Cr (VI) concentrations of 1 mg L-1 and 3 mg L-1. The highest current density was achieved at the $R_{ext}$ of 10Ω, with 0.27mA cm-2 for 10mg L-1, 0.24mA cm-2 for 3mg L-1,
and 0.23mA cm-2 for 1mg L-1. These results demonstrated that before reaching to toxic level, increasing metal concentrations enhanced the power generation of SCMFCs. Metal concentrations are closely correlated with bacterial metabolic activities [30,34], especially for anaerobic chemotrophic microorganisms with slow growth rate and using oxidized metal ions as electron acceptor to obtain energy [30]. Cr (VI) of 1 mg L-1 was found toxic for microorganisms [34], but this study showed that SCMFCs had stable power generation at Cr (VI) 1--3 mg L-1, and even 10 mg L-1, indicating that electrogenic bacteria could still produce power even at high toxic metal concentrations. In addition, the maximum power generation of SCMFCs in this study was higher than 2CMFCs fed with CrO_7^{2-} (55.5 mW m-2 and 123.4 mA/m² at 80mg L-1 [39], and 150mW m-2 and 0.04mA cm-2 at 200mg L-1 [23]). This clearly demonstrated that even though 2CMFCs tolerated higher metal concentrations than SCMFCs, SCMFCs had much higher power generation than 2CMFCs.

3.2 Reduction of metals in SCMFCs
The conversion efficiency (%) of Cr (VI) and Fe (III) were examined after 5-day (120 hr) treatment (Table 1), during which the voltages of SCMFCs were sustainably high. SCMFCs had high conversion efficiency (>89.0±1.0%) for both metals, which indicated that using electrons generated from anaerobic anodic reactions to reduce Cr (VI) and Fe (III) on cathodes was quite efficient. In addition, the oxygen redox potential (ORP) of -420 -- -450mV (data not shown) in SCMFCs demonstrated that there was no oxygen present in anode chamber, and the reduction of metals was responsible for electron acceptance on cathode. It should be noted that the conversion efficiency of Cr (VI) increased with concentrations, which might be related with the measurement methods.
that spectrophotometers can only measure Cr (VI) concentration as low as 0.1mg L-1, thereby, the conversion efficiency at low concentration (Cr (VI) 1 mg L-1) might be underestimated. The maximum removal percentage of Cr (VI) (98.7% at 10 mg L-1) was higher than phy-chemical processes, however, phy-chemical processes consumed shorter time (92% for electrocoagulation at 20mg L-1 during 50min[40], 91% for biosorption at 30mg L-1 during 120min[41], 97% for electrocoagulation and electroflotation at 10mg L-1 during 75min[42], and 96% for nanofiltration at 17 mg L-1 during 900min[43]). This indicated that using SCMFCs to reduce metals is an effective bioprocess but much time consuming than traditional phy-chemical processes.

Mechanisms other than electron reduction on cathodes could also contribute to the removal of Cr (VI) and Fe(III) in SCMFCs. In the control tests of SCMFCs without being connected with external circuit, about 88% of conversion efficiency was observed at the Cr (VI) of 1mg L-1 (Table 2). There were two possible reasons for metal removal in the control tests. First, a wide range of aerobic and anaerobic bacteria (e.g. *Pseudomonas dechromaticans, Bacilli, Clostridia*) could reduce Cr (VI) to Cr (III). Second, some species (e.g. *P. aeruginosa*) could adsorb heavy metals like Cr (VI) [39] and metals could also directly adsorb on the electrodes. However, the conversion efficiency of control tests decreased at higher concentration of Cr (VI) (63% at 3mg L-1 and 28% at 10mg L-1) (Table 2), which was in the opposite trend of the operated SCMFCs (93% at 3mg L-1 and 98% at 10mg L-1) (Table 1). This clearly showed that SCMFCs effectively reduce Cr (VI) and harvest energy simultaneously, while metal removal was inhibited in the control tests operated at anaerobic environment alone. In terms of Fe, when the Fe (III)
concentration was lower than 30mg L-1, the conversion efficiencies in the control tests (98% at 10mg L-1 and 92% at 30mg L-1) (Table 2) were similar to those of the operated SCMFCs (97% at 10mg L-1, and 93% at 30mg L-1) (Table 1), mostly due to anaerobic bacterial metabolisms and adsorption, since most active electrog enic microorganisms in MFCs are Fe-reducing bacteria [30,32]. However, at high Fe (III) concentration (50mg L-1), the conversion efficiency was only 79% in the control tests, much lower than that of the operated SCMFCs (94 %), indicating that the rate of anaerobic digestion was much slower than that of electrochemical reactions. This is a clear evidence that SCMFCs have higher metal removal efficiency than traditional anaerobic processes, and can utilize metal reduction to harvest electricity.

3.3 Changes of electrodes surface properties during metal removal in SCMFCs
Biofilm growth on cathodes normally occurred in SCMFCs treating wastewater [6,45,46]. It had been found that biofilms on cathodes facilitated electron acceptance and enhanced power generation [6,39,46], while the high extents of cathodic biofilm growth would lower the cathode OCPs [6,36]. Compared with the clean Pt-loaded weaving carbon cloth (Figure 4a) from SEM pictures, the cathodes in SCMFCs treating metals clearly had biofilms and metal crystals (Figure 5b and 5c), which indicated the attached bacteria could carry out metal reduction on cathodes. The elements on the cathodes analyzed using EDX showed the Pt peak on the clean Pt-loaded cathodes (Figure 5a), while Cr and Fe peaks appeared on the cathodes treating metals (Figure 5b and 5c), indicating the metal deposition on cathodes. Previous studies found that metal deposition(e.g. Cr, Fe, Ca, Cu) led to cathode fouling, blocked the contact between catalysts (e.g. Pt used in this study) and electrolyte solution in MFCs, increased R_in, and decreased power generation
Therefore, cathode surfaces needed to be cleaned periodically to remove metal deposition.

For the anodes, comparing with the clean carbon brush (Figure 6a), bacteria attachment and some substrate deposition on the carbon brush could be observed from Figure 6b and 6c. From the EDX results (Figure 6), carbon peaks were predominant in the graphs but there were still other element peaks when treating with metals, indicating that Cr, Fe and the components (Na, P, Ca, etc.) of the solution were adsorbed on the carbon brush. The Cr(5-6KeV) and Fe peaks (6-8KeV) were not obvious comparing with those from cathode EDX (Figure 5b and 5c), indicating that most of these metals were deposited on the cathodes rather than anodes. The element Au in the EDX was from the gold sputtering for samples to make it more conductive.

3.4 Electrochemical characteristics of metal reduction in SCMFCs

The reduction of Cr (VI) and Fe (III) on cathode was determined using the anode/cathode half-cell reactions (Eq. 1-4). The predominant species in the half-cell reactions were determined generally by E-pH diagram assuming that the concentrations of all the species in the system were 1M (Figure 4). The calculation of the theoretical maximum electromotive force (EMF) (Eq.5) was conducted based on the stoichiometric relationships between Cr (VI) and acetate / Fe(III) and acetate. In theoretical calculation, the molar concentration of acetate oxidized (the organic substrate in anode) should be 0.125 times of Fe(III) and 0.375 times for Cr(VI) (Eq.1- 4). Therefore, under specific conditions (pH 7, 1bar and 298.15K), the theoretical electrode reduction potential of 1M of Fe (III) was 0.492V (vs. SHE) and a complete oxidation of 0.125M of acetate has a
reduction potential of \(-0.281\text{V(vs. SHE)}\), theoretically. The combination of these two half reactions generate a total EMF of \(0.773\text{V (vs. SHE)}\) (Eq. 5). Following the same calculations, the reduction potential of \(1\text{M Cr (VI)}\) is \(0.696\text{V(vs. SHE)}\), the reduction potential of complete oxidation of \(0.375\text{M sodium acetate}\) is \(-0.278\text{V(vs. SHE)}\), and hence the total EMF for \(\text{Cr(VI)}\) is \(0.974\text{V(vs. SHE)}\).

For SCMFCs containing Fe(III):

\[
\begin{align*}
\text{Anode:} & \quad 2\text{HCO}_3^- + 9\text{H}^+ + 8\text{e} \rightarrow \text{CH}_3\text{COO}^- + 4\text{H}_2\text{O} \quad (1) \\
E_{\text{an}} & = -0.281\text{V vs SHE} \quad (\text{HCO}_3^- = 0.25\text{M, CH}_3\text{COO}^- = 0.125\text{M, pH=7})
\end{align*}
\]

\[
\begin{align*}
\text{Cathode:} & \quad \text{Fe}^{3+} + 2\text{H}_2\text{O} + \text{e} \rightarrow \text{Fe(OH)}_2 + 2\text{H}^+ \quad (2) \\
E_{\text{cat}} & = 0.492\text{V vs SHE} \quad (\text{Fe}^{3+} = 1\text{M, pH=7})
\end{align*}
\]

For SCMFCs containing Cr(VI):

\[
\begin{align*}
\text{Anode:} & \quad 2\text{HCO}_3^- + 9\text{H}^+ + 8\text{e} \rightarrow \text{CH}_3\text{COO}^- + 4\text{H}_2\text{O} \quad (3) \\
E_{\text{an}} & = -0.278\text{V vs SHE} \quad (\text{HCO}_3^- = 0.75\text{M, CH}_3\text{COO}^- = 0.375\text{M, pH=7})
\end{align*}
\]

\[
\begin{align*}
\text{Cathode:} & \quad \text{Cr}_2\text{O}_7^{2-} + 8\text{H}^+ + 6\text{e} \rightarrow 2\text{Cr(OH)}_3 + \text{H}_2\text{O} \quad (4) \\
E_{\text{cat}} & = 0.696\text{V vs SHE} \quad (\text{Cr}_2\text{O}_7^{2-} = 0.5\text{M, pH=7})
\end{align*}
\]

The cell electromotive force \(E_{\text{emf}}\) was calculated as

\[
E_{\text{emf}} = E_{\text{cat}} - E_{\text{an}} \quad (5)
\]

The electrochemical measurement showed that the overall OCP \(~630 \text{mV}\) of the SCMFCs with Fe (III) was about 0.8 times as the theoretical values \(773 \text{mV}\) (Table 3) and for Cr(VI), the overall OCP \(~580 \text{mV}\) of the SCMFCs with Cr (VI) was about 0.6 time as the theoretical values \(974 \text{mV}\) (Table 3), indicating that electrochemical
reactions of electron transfer from the external circuit and metal reduction proceeded effectively on cathodes. It should be noted that the anodic OCPs of all SCMFCs tested were -487 ± 4mV vs Ag/AgCl (-290±4mV vs SHE) (Table 3), which was around the theoretical value of sodium acetate oxidation (~ -284mV vs SHE). This elucidated that the electrogenic bacteria growing on the anode effectively generate electrons from anaerobic degradation of sodium acetate in electrolyte and were not affected by the metals added in SCMFCs.

The internal resistance ($R_{in}$) of SCMFCs fed with Cr (VI) (708-749 Ω) was higher than those with Fe(III) (393-507 Ω), and dropped with metal concentrations (Table 3), since higher metal concentrations decreased the ohmic losses by increasing the solution conductivity and ionic strength [10]. Due to Cr (VI) concentrations (1-10 mg L-1) much lower than Fe (III) concentrations (10-50 mg L-1), the corresponding $R_{in}$ of Cr(VI) were high. The $R_{in}$ of SCMFCs in this study was higher than those (30-300Ω) with air cathode [17][49]. There might be three reasons. First, the cathode area (carbon cloth, 3 cm$^2$) was much smaller than anode (carbon brush), which limited the reduction reactions on cathodes. Previously studies found that cathode rather than anode is the limiting factor for power generation of SCMFCs [49]. Second, the precipitation of Cr (III) or Fe (II) (e.g. $Cr(OH)_3$, $Fe(OH)_2$, $Fe(OH)_3$) and thick biofilms on cathodes (Figures 5b and 5c) would impede the continuous electron transfer and increase $R_{in}$. Third, the concentration of PBS solution (50mmol L-1) used in this study was lower than other SCMFC tests (200mmol L-1)[49].
The coulombic efficiency decreased substantially with the increase of metal concentrations (Table 3). At low concentrations (Fe 10 mg L\(^{-1}\), Cr 1 mg L\(^{-1}\)), the coulombic efficiency was higher than 100\%, which indicated that there should be other electron sinks present in SCMFCs, such as the residual trace oxygen in solution and other minerals (e.g. \(\text{Cu}^{2+}\), \(\text{MnO}_7^{4-}\)) in mineral solution. These extra electron sinks led to the overestimation of electron acceptance efficiency (coulombic efficiency value) at low metal concentrations. However, with the increase of the concentration of Cr(VI) and Fe(III), the function of these multiple sinks could be ignored since the residual oxygen would be consumed quickly and the amount of trace elements added were much lower than Cr(VI) and Fe(III) contents. In addition, other oxidation states of Cr lower than Cr(VI) (e.g., Cr(II), Cr(IV), and Cr(V)) could be the intermediate product existing in the SCMFC solution but they are unstable intermediates and Cr(III) was the most stable oxidation state\[54\], which acted as the main final electron acceptors in cathodic reactions. As for Fe, the standard reduction potential of Fe(III) to Fe was -0.04V (vs SHE), which could not happen simultaneously in SCMFCs. Therefore, the calculation of coulombic efficiency could be simplified by using Cr(VI) to Cr(III) or Fe(III) to Fe(II) as the electron acceptor and disregarding other contributors. At high concentrations (Fe 50 mg L\(^{-1}\) and Cr 10 mg L\(^{-1}\)), the coulombic efficiency values were 23.4\% and 25.6\%, respectively (Table 3). In contrast, the conversion efficiencies for Fe (III) and Cr (VI) remained high (> 93\%) at high concentrations (Table 1). This discrepancy revealed that some processes (e.g. anaerobic digestion, bacterial assimilation uptake, and adsorption) rather than electron acceptance reduction contributed to metal removal, but not electricity generation. It should be noted that the coulombic efficiency values calculated in this study was based
on metal reduction on cathodes, unlike those based on organic substrate degradation on anodes [3, 11, 16]. The coulombic efficiency values obtained (23-100%) was much higher than those with air cathodes (13-27%) [3, 11, 16], which clearly verified that the easily dissolved metals (e.g. Cr and Fe) have much lower mass transfer resistances for cathodic reactions than oxygen diffusion from air, and metal reductions had lower overpotentials than oxygen reduction reaction [48].

pH of SCMFCs dropped (6.3-6.4) after 120 hrs SCMFC operation (Table 3), due to the proton generation from sodium acetate degradation in anode (Eq. 1 and 3). The pH drop also indicated that sodium acetate (20mmol L-1) was sufficient for electrons needed by metal reduction on cathodes throughout batch-mode operation. Even though the reduction of Cr (VI) on the cathode consumed more protons than provided from anodic reactions (Eq. 4), the precipitation of Cr$^{3+}$ at pH 6-7 also produced protons (Eq. 6) [39] and avoided the proton shortage problem.

\[
2Cr^{3+} + 7H_2O \rightarrow 2Cr(OH)_3 + 6H^+ + H_2O \quad (6)
\]

3.5 Metal reduction activities on cathodes measured by LSV
The effects of metal concentrations to the cathode electrochemical performance were evaluated using LSV measurement. The current produced in LSV is the flow of electrons needed to support the active electrochemical processes at rates consistent with the potential [50]. The cathode at higher metal concentration had steeper wave slope of the I-V curves, and the highest electrochemical reaction rates achieved at the highest concentrations of Cr (VI) (10 mg L-1) and Fe (III) (50 mg L-1) (Figure 7). This indicated that higher concentration of metals accelerated the reduction reaction on cathodes, which
corresponded well with the lower $R_{in}$ at higher metal concentration (Table 3). In addition, because Cr (VI) existed as $Cr_2O_7^{2-}$ and Fe (III) existed as $Fe^{3+}$ or Fe(OH)$_3$, the mole concentration of Cr (VI) was almost half of Fe(III) at 10 mg L$^{-1}$, so that its electrochemical reaction rate was lower than Fe(III) (10 mg L$^{-1}$, Figure 7). Moreover, the relationship of electron flux ($J$) and current ($i$) could explain the LSV phenomenon (Eq.7) [50]. The current ($i$) is proportional to the flux of electroactive species (e.g. Cr (VI) and Fe (III) tested) transported to the electrode surface ($x=0$), $J_O(0,t)$. The flux is also proportional to the concentration gradient ($C_O(x,t)$) of oxidized species at the electrode surface. Here, $n$ is the number of electron exchanged in the reaction, $F$ is Faraday’s constant ($9.64853 \times 10^4$ C/mol), $A$ is the electrode surface area, and $D_O$ is the diffusion coefficient. Eq. 7 clearly showed that higher concentration possesses higher electrochemical reaction rate. Thereby, the SCMFCs fed with Fe (III) exhibited much higher electrochemical reaction rates than those with Cr (VI), which was the result of the higher concentration of Fe(III) than Cr(VI).

$$-J_O(0,t) = \frac{i}{nFA} = D_O \left[ \frac{\partial C_O(x,t)}{\partial x} \right]_{x=0}$$  \hspace{1cm} (7)

### 3.6 RAPD test

When comparing the total number of anode bands in different SCMFCs (Table 4) with RAPD test, the number with Cr(VI) as electron acceptor and O$_2$ as electron acceptor were similar (15 for Cr(VI) and 14 for O$_2$). The bands number and position in the gel were similar except the bands with first primer (Figure 8a), indicating the diversity could be observed when treating with the first random primer. For Fe(III), the total number of bands (10) (Table 4) was less than Cr(VI) and O$_2$ when comparing based on each primer,
indicating that although the diversity of community was less, the electrogenic bacteria were not affected or affected less by the high concentration of Fe(III) since SCMFC still produced power.

For cathode, Fe(III) had the most bands (20) (Table 4) among three cathodes and the difference was observed mainly with third and last primers (Figure 8b). But the overall number was similar indicating that bacteria growing on the cathode were not affected by the heavy metals greatly and in fact, for Fe(III), cathode the diversity was higher than other systems. Oxygen reduction possessed the highest redox potential (0.805V, at pH 7 and pO2 0.2) among these substrates and could produce higher power density (766mW m-2) [16] as electron acceptor than Cr(VI) and Fe(III) in our research. However, from the RAPD test results, the diversity was not significantly different from metal treated system, indicating that the power generation was not much affected by bacteria diversity and the electrogenic bacteria could still work to produce electrons with metal solution. RAPD test result could bring the primary idea of the diversity in the system, however, in order to clearly differentiate the bacteria species and for identification, the microbial community analysis of 16sRNA gene based clone library need to be conducted in the future to identify which kind of bacteria could tolerate the heavy metals and/or produce power in the system.

3.7 Significance of metal reduction in SCMFCs
This study, for the first time, applied the metal reduction in SCMFCs and expanded MFCs to treat diverse wastes. Stable power generation was achieved in SCMFCs at various concentrations of Cr (VI) and Fe (III), indicating that using oxidized-status metals as electron acceptors on cathode is a cost-effective approach to degrade toxic
metals and generate electricity. Compared with 2CMFCs, SCMFCs had much simpler structure, easy installation, and high power generation for future scale-up application in wastewater treatment. Moreover, because some metals (e.g. Fe and Mn) are widely distributed in natural water bodies and sediment, metal reduction on cathodes in this study reveals a great potential of applying benthic MFCs in underwater energy harvest [51]. However, several important issues should be solved for large-scale operation of metal reduction in MFCs. First, even though SCMFCs had higher power generation than 2CMFCs, they had lower tolerance of toxic metals than 2CMFCs [23,39]. Possible solution might be genetically transferring DNA from metal-tolerance bacteria to electrogenic bacteria to enhance the tolerance, or developing novel MFC configurations to minimize the impacts of toxic metals on anodic bacteria. Second, biofilm-based cathodes (biothodes) could be used to replace the cathodic catalyst (e.g. Pt) to effectively carry out cathodic metal reduction and electron acceptance. Cr-reducing bacteria had been studied as biocathodes in 2CMFCs [39,46,52,53]. Third, power generation of SCMFCs should be further enhanced by lowering $R_{in}$, increasing cathode area, using higher PBS concentration, and/or removing metal precipitation on the cathode. Fourth, the retention time of the batch-mode SCMFCs was 5 days in this study, which was much longer than the contact time of conventional physical-chemical processes ([40]-[43]). Continuous flow MFC tests should be conducted to determine the optimal HRT for anaerobic treatment of metals and provide the guidance for scale-up MFCs for real-world application.


**Chapter 4: Conclusion**

Metal reduction on cathodes in SCMFCs was studied at different concentrations of Cr (VI) and Fe (III). Metal removal efficiency and power generation were correlated. The inhibition of toxic Cr (VI) on anode/cathode was elucidated. Three major conclusions were drawn from this study.

First, metal concentrations posed two-side effects on SCMFCs performance. Higher metal concentration generally led to high power generation, but Cr (VI) at 10 mg L-1 started to inhibit power generation of SCMFCs. Fe (III) concentrations did not have negative impacts on SCMFCs.

Second, the conversion rates of \( \text{Cr}^{6+}/\text{Cr}^{3+} \) and \( \text{Fe}^{3+}/\text{Fe}^{2+} \) were high (>89%), and coulombic efficiency ranged at 23%-100%, indicating that metals were good electron acceptors on cathodes.

Third, the LSV tests demonstrated that cathode electrochemical activity increased with metal concentrations, which well corresponded with \( R_{in} \) of SCMFCs.
Chapter 5: Future research plan

First, the microbial community analysis of 16sRNA gene based cloning of anode and cathode biomass will be conducted in the future to identify the species on the cathode which might tolerate toxic metals and work as the catalyst in reducing metals. And comparison of the species between two metal reducing SCMFCs will be observed to determine whether the species growing on the different metal system is similar or totally different.

Second, MFC-MEC hybrid system as mentioned in the Chapter 1 will be conducted using the energy produced from MFC and continuously reducing other metals in MEC to fully explore metal reduction in these bioelectrochemical devices.
Table and Figure List

**Table 1.** Conversion efficiency (%) of Cr (VI) and Fe (III) in SCMFCs during 120hr treatment

**Table 2.** Conversion efficiency (%) of Cr(VI) and Fe (III) in the control SCMFCs without being connected with external circuit during 120hrs.

**Table 3.** Cathode OCPs, pH, R in, and coulombic efficiency for SCMFCs during 120hr treatment with different initial concentration of Fe(III) and Cr(VI).

**Table 4.** RAPD bands statistics of different anodes and cathodes treating with 4 different random primers.

**Figure 1.** Configuration of single-chamber microbial fuel cells (SCMFCs) with metal as the electron acceptors.

**Figure 2.** Voltage production over 48-hr operation at different concentrations of Fe(III) (a) and Cr(VI) (b) in SCMFCs.

**Figure 3.** Voltage and power generated in the SCMFCs with different concentrations of Fe(III)(a) and Cr(VI)(b) when external resistance on the system is changed from 10-2940 Ω at 48h. (a) V 10 represented voltage at the concentration of 10mg L-1, V 30 represented voltage at the concentration of 30mg L-1, V 50 represented at the concentration of 50mg L-1, PD 10 represented power density at the concentration of 10mg L-1, PD 30 represented at the concentration of 30mg L-1, PD 50 represented at the concentration of 50mg L-1; (b) V 1 represented voltage at the concentration of 1mg L-1, V 3 represented voltage at the concentration of 3mg L-1, V 10 represented at the concentration of 10mg L-1, PD 1 represented power density at the concentration of 1mg L-1, PD 3 represented at the concentration of 3mg L-1, PD 10 represented at the concentration of 10mg L-1.)

**Figure 4.** E-pH diagram of Fe(a) and Cr(b). (For Fe(a), assuming that Fe^{3+}, Fe^{2+}, Fe(OH)_2, Fe(OH)_3, and Fe exist in the system and the concentration of each species was 1M: For Cr(b), assuming that Cr^{3+}, Cr(OH)_3, Cr_2O_7^{2-}, and Cr exist in the system and the concentration of each species was 1M.)

**Figure 5.** The SEM pictures and EDX spectrums of cathodes in SCMFCs before and after operation (a. clean Pt loaded cathode; b. cathode operated with Cr(VI) solution; c. cathode operated with Fe(III) solution).

**Figure 6.** The SEM pictures and EDX spectrums of anodes in SCMFCs before and after operation (a. clean carbon brush anode; b. anode operated with Cr(VI) solution; c. anode operated with Fe(III) solution).
Figure 7. Cathode liner sweep voltammetry (LSV) curves of different concentration of Fe(III) and Cr(VI) after 120hrs.

Figure 8. RAPD test results of different anodes(a) and cathodes(b) with 4 random primers.
<table>
<thead>
<tr>
<th>Initial Cr(^{6+}) concentration (mg L(^{-1}))</th>
<th>Final Cr(^{6+}) concentration (mg L(^{-1}))</th>
<th>Conversion efficiency (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.10±0.10</td>
<td>&lt;0.1</td>
<td>89.0±1.0</td>
</tr>
<tr>
<td>3.00±0.10</td>
<td>&lt;0.1</td>
<td>95.7±0.4</td>
</tr>
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<td>10.0±0.20</td>
<td>&lt;0.1</td>
<td>98.8±0.1</td>
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<tr>
<th>Initial Fe(^{3+}) concentration (mg L(^{-1}))</th>
<th>Final Fe(^{3+}) concentration (mg L(^{-1}))</th>
<th>Conversion efficiency (%)</th>
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<tr>
<td>10.0</td>
<td>0.28±0.08</td>
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<tr>
<td>30.0</td>
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<tr>
<td>50.0</td>
<td>2.72±0.92</td>
<td>94.6±1.8</td>
</tr>
</tbody>
</table>

**Table 1.** Conversion efficiency (%) of Cr(VI) and Fe (III) in SCMFCs during 120hr treatment
<table>
<thead>
<tr>
<th>Initial Cr$^{6+}$ concentration (mg L$^{-1}$)</th>
<th>Final Cr$^{6+}$ concentration (mg L$^{-1}$)</th>
<th>Conversion efficiency (%)</th>
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<td>2.60±0.70</td>
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<td>50.0</td>
<td>10.4±1.36</td>
<td>79.2±2.72</td>
</tr>
</tbody>
</table>

Table 2. Conversion efficiency (%) of Cr(VI) and Fe (III) in the control SCMFCs without being connected with external circuit during 120hrs.
Table 3. Cathode OCPs, pH, R<sub>i</sub>, and coulombic efficiency for SCMFCs during 120hr treatment with different initial concentration of Fe(III) and Cr(VI).

<table>
<thead>
<tr>
<th>Initial concentration(mg L-1)</th>
<th>Fe&lt;sup&gt;3+&lt;/sup&gt; 10</th>
<th>Fe&lt;sup&gt;3+&lt;/sup&gt; 30</th>
<th>Fe&lt;sup&gt;3+&lt;/sup&gt; 50</th>
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<th>Cr&lt;sup&gt;6+&lt;/sup&gt; 10</th>
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<tr>
<td>Anode potential (mV)</td>
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<td>Cathode potential (mV)</td>
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<td>163±3</td>
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<tr>
<td>(mV)</td>
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<td>651±5</td>
<td>582±3</td>
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Figure 1. Configuration of single-chamber microbial fuel cells (SCMFCs) with metals as the electron acceptors.
Figure 2. Voltage production over 48-hr operation at different concentrations of Fe(III) (a) and Cr(VI) (b) in SCMFCs.
Figure 3. Voltage and power generated in the SCMFCs with different concentrations of Fe(III)(a) and Cr(VI)(b) when external resistance on the system is changed from 10-2940 Ω at 48h. ((a)V 10 represented voltage at the concentration of 10mg L-1, V 30 represented voltage at the concentration of 30mg L-1, V 50 represented at the concentration of 50mg L-1, PD 10 represented power density at the concentration of 10mg L-1, PD 30 represented at the concentration of 30mg L-1, PD 50 represented at the concentration of 50mg L-1; (b) V 1 represented voltage at the concentration of 1mg L-1, V 3 represented voltage at the concentration of 3mg L-1, V 10 represented voltage at the concentration of 10mg L-1, PD 1 represented power density at the concentration of 1mg L-1, PD 3 represented at the concentration of 3mg L-1, PD 10 represented at the concentration of 10mg L-1.)
Figure 4. E-pH diagram of Fe(a) and Cr(b). (For Fe(a), assuming that Fe$^{3+}$, Fe$^{2+}$, Fe(OH)$_2$, Fe(OH)$_3$, and Fe exist in the system and the concentration of each species was 1M; For Cr(b), assuming that Cr$^{3+}$, Cr(OH)$_3$, Cr$_2$O$_7^{2-}$, and Cr exist in the system and the concentration of each species was 1M.)
Figure 5. The SEM pictures and EDX spectrums of cathodes in SCMFCs before and after operation (a. clean Pt loaded cathode; b. cathode operated with Cr(VI) solution; c. cathode operated with Fe(III) solution).
Figure 6. The SEM pictures and EDX spectrums of anodes in SCMFCs before and after operation (a. clean carbon brush anode; b. anode operated with Cr(VI) solution; c. anode operated with Fe(III) solution).
Figure 7. Cathode liner sweep voltammetry (LSV) curves of different concentration of Fe(III) and Cr(VI) after 120hrs.
Figure 8. RAPD test results of different anodes (a) and cathodes (b) with 4 random primers.
Reference