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Nitrogenous heterocyclic compounds degradation in the microbial fuel cells

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ABSTRACT

Growing energy needs and concerns about environmental pollution have stimulated increased interest in the research and application of microbial fuel cell (MFC) systems. The objective of this study was to investigate possible electricity production with nitrogenous heterocyclic (N-heterocyclic) compounds degradation in the MFCs. Two-chamber MFCs were designed and inoculated with anaerobic sludge acclimated for several months. The experiments were conducted to test the potential for biodegradation of refractory organic matters and electricity generation using representative N-heterocyclic compounds such as pyridine, quinoline and indole. A maximum voltage of 524 mV, 494 mV, 413 mV (based on an external resistance of 1000 Ω), and the corresponding maximum power densities of 228.8 mW m^{-2}, 203.4 mW m^{-2}, 142.1 mW m^{-2} were obtained from pyridine, quinoline, and indole, respectively. Meanwhile, the maximum degradation efficiency of these substrates and COD (chemical oxygen demand) removal were up to 90% and 88%, respectively. The metabolic intermediate products were detected by GC/MS analyses of the anode solution. These results indicated that N-heterocyclic compound may be used as the MFC fuel in practical applications of wastewater treatment.

Keywords: Nitrogenous heterocyclic compounds; Degradation; Microbial fuel cell; Electricity generation

1. Introduction

Nitrogenous heterocyclic (N-heterocyclic) compounds are of immense concern from point view of environment and have potential application in manufacturing of dyestuffs, pesticides, agrochemicals and disinfectants. Due to their toxicity, mutagenicity and carcinogenicity, they constitute a danger for natural biogenic environment and severe odour potential (Kaiser et al., 1996). Furthermore, most of N-heterocyclic compounds are difficult to be degraded by microorganisms under aerobic and anaerobic conditions, and also have an adverse impact on the conventional biological wastewater treatment system for their toxicity to microbial communities. The researches on the effective treatment techniques of organic wastewater containing N-heterocyclic compounds are increasingly focused and become a challenge in the environmental protection domain. Existing technologies for N-heterocyclic compound treatment can be divided into three categories: chemical, physical, and biological methods. Many of these processes entail high operational costs, strict operational regulations, or a large area of land for treatment. Thus one new promising solution is to link wastewater treatment with energy recovery to offset operational costs.

Microbial fuel cell (MFC) has recently gained a remarkable increase in interest and attention. It is reported that MFCs have been operated successfully using many types of organic matter to generate electricity, including readily biodegradable compounds (glucose, acetate or butyrate), complex carbohydrates (cellulose, cysteine) and wastewater streams (brewery wastewater, swine wastewater) (Chaudhuri and Lovley, 2003; Liu et al., 2005; Logan et al., 2005; Min et al., 2005a,b; Ren et al., 2007; Wen et al., 2009). However, Many MFC studies have aimed to enhance energy recovery, while few studies have focused on the removal of contaminants during lower levels of electric-

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Received 15 April 2010; Received in revised form 19 October 2010; Accepted 27 October 2010
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doi:10.1016/j.psep.2010.10.006

ity production. There are many constraints related to making MFCs effective in industrialized applications and MFC could not supply enough electron donors to the actual need of electrical current. The development of MFCs using recalcitrant pollutants as fuels is still in its infancy and needs to be further studied.

Special sources of the organic material and the type of reactor have resulted in different power outputs from the MFCs. In laboratory and field experiments, 661 mW m^{-2} (maximum power density) was generated with acetate, but 371 ± 10 mW m^{-2} with a food processing wastewater and a maximum of 146 ± 8 mW m^{-2} with domestic wastewater in the similar single-chambered MFC (Liu et al., 2004, 2005; Oh and Logan, 2005). Significant improvements in the performance of MFCs have been made in recent years. However, information about electricity generation in MFCs based on recalcitrant pollutants is limited. The recent reports demonstrated the feasibility of using MFCs to produce electricity with co-substrates (glucose–quinoline, glucose–pyridine, glucose–indole) as the fuels and degradation of the organics simultaneously (Zhang et al., 2009, 2010; Luo et al., 2010), although the organic matters may not do a major contribute to electricity generation. They also reported that the MFCs were fed with the readily biodegraded organics of glucose to inoculate the biofilm. These toxic and refractory N-heterocyclic compounds often coexist or exist solely in the coking wastewater. Therefore, the co-substrate as the fuels has a great inhibition from the point view of application.

In the present study, we conducted different experiments to compare electricity generation and biodegradation of three representative N-heterocyclic compounds (pyridine, indole and quinoline) under the same operational conditions. The microbial communities in the anodic compartment were inoculated with anaerobic sludge acclimated for several months, which contribute to enhance the performance of MFC and removal efficiency of COD. Additionally, the one control experiment with carbon paper but no bacteria existing in solution was conducted to examine the adsorption effect of anode on N-heterocyclic compound. The other control experiment was current production and COD removal of the MFC without N-heterocyclic compounds at the anode solution. The aims of this study were to provide new information, and some theoretical guidance for further research about refractory organic compounds and how MFCs can be employed for degradation of these compounds during wastewater treatment.

2. Materials and methods

2.1. MFC construction and operation

Two-chambered MFCs are frequently used to examine electricity generation from different substrates, and/or microbial communities which arise during the biodegradation of specific compounds. Our experiments used MFCs made of polycarbonate, and had anodic and cathodic compartments separated by a proton exchange membrane (PEM, Nafion™ 117, Dupont Co. USA). The chambers were joined by a glass bridge containing a PEM held by a clamp between the flattened ends of the two glass tubes (inner diameter = 2.0 cm) fitted with a concave rubber gasket. Each chamber had a working volume of 250 mL. The anodic electrode was made of carbon paper (4.0 × 3.0 cm, E-Tek), while one side of the cathodic electrode, except as noted below, was coated with a catalyst (0.35 mg-Pt cm^{-2}). A copper wire was used to connect the circuit containing a resistance of 1000 Ω (unless stated otherwise), while at the same time a computer and multimeter (UNI-T 803) were used to record the voltage across the circle. The anode chamber was filled with substrate solution (pH = 7.0), and then purged with nitrogen for 15 min to keep anaerobic conditions. The cathode chamber was continuously aerated using an aquarium membrane pump to provide dissolved oxygen. Immediately after adding anolytes and catholytes, the MFC was hooked up to the data acquisition system to start monitoring the voltage generation. The anode solutions were mixed with a magnetic stir bar. All MFCs were operated simultaneously and kept in a water bath with a constant temperature of 30.0 ± 1.0 °C throughout the experiments.

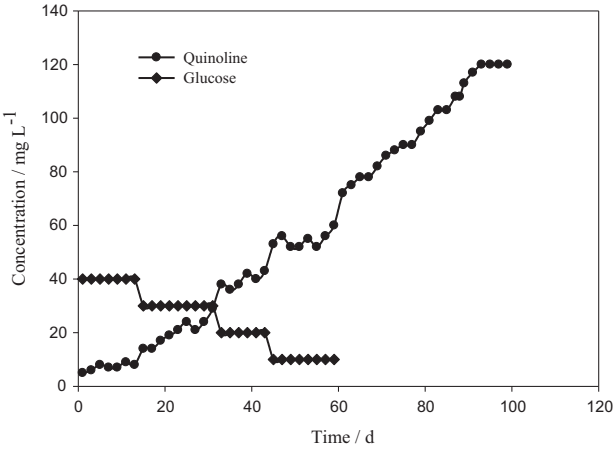

Fig. 1 – Dosing quantity of glucose and quinoline during the acclimation period of anaerobic sludge.

2.2. Sludge acclimation procedures

Since there are no known microorganisms that can both metabolize N-heterocyclic compounds and transfer electrons to solid extracellular substrates, the initial anaerobic enrichment cultures used were obtained from the anaerobic digested sludge of the Second Wastewater Treatment of Changsha City in China and kept in a refrigerator at 4 °C. The raw anaerobic sludge was incubated in a sealed conical flask (250 mL) and fed with artificial wastewater containing a different N-heterocyclic compound. In the beginning, the readily biodegradable glucose was provided as the supplementary carbon source. The concentration of each substrate was gradually increased to 120 mg L^{-1} according to the stability of COD and the removal efficiency of organic matter. The optimal dosing quantity of artificial wastewater was determined by substrate effluent concentration and COD removal during acclimation, which could prevent the deaths of microorganisms due to high strength organic artificial wastewater containing toxic refractory pollutants and also keep proper substrate level. Meanwhile, the sealed conical flasks were added into ample nutrients and incubated in biochemical incubator (30 °C, 180 r min^{-1}) (Zhou and Gao, 2000). The acclimation was ended when stable removal rates of both COD and substrate reached up to 90%. An example of anaerobic sludge acclimation process is shown in Fig. 1 (quinoline), while the adaption times and cultivation methods were similar for pyridine and indole. After acclimation, the mixed bacteria in the anode culture might have been changed and/or were prone to degrade substrate quickly.

2.3. Microbial medium

A medium solution (without carbon sources) was prepared by dissolving the following compounds (per liter of distilled water): $Na_2HPO_4{\cdot}12H_2O$ 2.75 g, $NaH_2PO_4{\cdot}H_2O$ 4.97 g, NH_4Cl 0.31 g, KCl 0.13 g, a vitamin stock solution 12.5 mL and trace metal solution 12.5 mL as described in a previous study (Lovely and Phillips, 1988). Three N-heterocyclic compounds medium solutions were prepared by dissolving each heterocyclic compound in the carbon-free media solution. Concentrations of each substrate were varied in the range of 20–120 mg L^{-1} to investigate their effects on power generation at a fixed resistance of 1000 Ω. The cathode medium (phosphate buffer solution) consisted of $Na_2HPO_4{\cdot}12H_2O$ (2.75 g L^{-1}) and $NaH_2PO_4{\cdot}H_2O$ (4.22 g L^{-1}). The initial pH of all the solutions was adjusted to 7.0 using NaOH solution. The composition of artificial wastewaters was prepared by dissolving different concentrations of pure substrate in the medium solution. The MFCs were inoculated with 60 mL of anaerobic sludge containing a greater diversity of electrochemically active bacteria through acclimation.

2.4. Analysis and calculations

Voltage (V) was continuously measured by a multimeter with a data acquisition system and used to calculate the power according to $p = IV (I = V/R)$. Power density P (mW m^{-2}), was obtained according to $P = IV/A$, where I (mA) is the current, V (V) is the voltage and A (m^2) is the projected surface of the anode. The Coulombic efficiency was calculated as $CE = (C_p/C_{Ti}) \times 100\%$, where C_p is the total coulombs calculated by integrating the measured current with respect to time. C_{Ti} is the theoretical amount of coulombs that can be produced from pyridine ($i = a$), quinoline ($i = b$) and indole ($i = c$), calculated as $C_{Ti} = Fb_iS_iv/M_i$, where F is Faraday's constant (96,485 C mol^{-1} e^{-1}), b_i the number of moles of electrons produced per mole of substrate, S_i the substrate concentration and M_i the molecular weight of the substrate ($M_a = 79.1$, $M_b = 129.16$, $M_c = 117.15$), v the liquid volume (L). An empirical Monod-type equation was used for modeling of power as a function of substrate concentration (S):

$$P = \frac{P_{max}S}{K_s + S}$$

where P_{max}, the maximum power and K_s (s), the half-saturation constant were determined using the Excel solver (Microsoft, version 2003). The anode and cathode potentials were measured by placing a saturated calomel electrode (SCE, +0.242 V vs. SHE) in the anode and cathode chambers for reference.

At regular intervals, samples were withdrawn from each of the anode solution for measurement of substrate concentration and COD removal efficiency, respectively. GC/MS analyses were conducted on a Finnigam-TRACE Gas Chromatography coupled with a TRACE Mass Spectrometry (EI 70 eV) to identify the possible metabolites of each N-heterocyclic compound. A TR-5MS capillary column with inner diameter of 0.25 mm and length 30 m was adopted in the separation system. Samples of 1 μL were injected and analyzed. The temperature of the GC/MS column was maintained as follows: 40 °C for 3 min; then increased to 280 °C at the rate of 5 °C min^{-1} and kept the temperature for 2 min. All the samples were centrifuged for 5 min at 12,000 r min^{-1} (TGL-16G, Anke, China) and then filtered through a 0.45 μm pore diameter needle-type filter dish. Each substrate concentration was analyzed using a UV-2550 Phramasper UV–vis spectrophotometer. COD (TCOD and SCOD) was determined according to Standard Methods. The pH values of the culture media in each MFC were measured in the experiments.

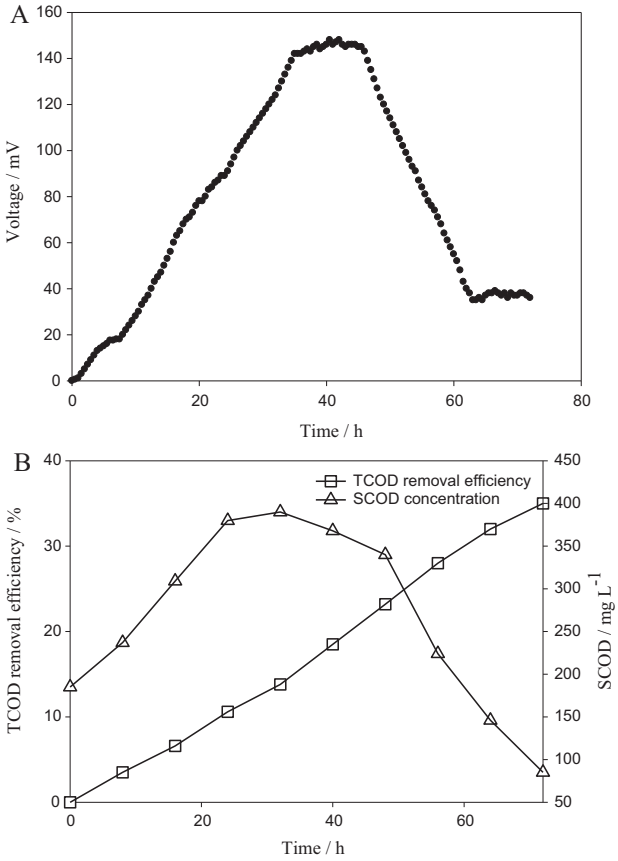

Fig. 2 – Electricity voltage output over testing time with 1000 Ω load (A) and TCOD removal efficiency as well as corresponding SCOD of sludge supernatant within MFC (B) under steady operation.

3. Results

3.1. Electricity production from anaerobic sludge-MFC

The MFC system generated constant electrical power from anaerobic sludge after a successful acclimation period, yielding a maximum voltage output of 145 ± 5 mV with a 1000 Ω resistor (Fig. 2A). As Fig. 2B shown, an increase in TCOD removal efficiency was noted and reached about 35% over testing time. Conversely, the SCOD of sludge supernatant was sharply increased and was then kept stable, gradually decreased at the end of periodicity. The experimental results are similar to the published studies (Liu et al., 2009; Jiang et al., 2009). The MFC had sludge hydrolysis function which was validated by the increase of sludge SCOD after MFC operation. Apparently, the particulate COD of sludge was efficiently hydrolyzed into soluble organic matters by the microbes in the MFC and the soluble organics was readily utilized for power generation. The control experiment without N-heterocyclic compounds as carbon sources clearly indicated that: (1) electricity generation at relatively lower power density could be sustained using anaerobic sludge; (2) the sludge supplied biodegraded matters to methanogenic

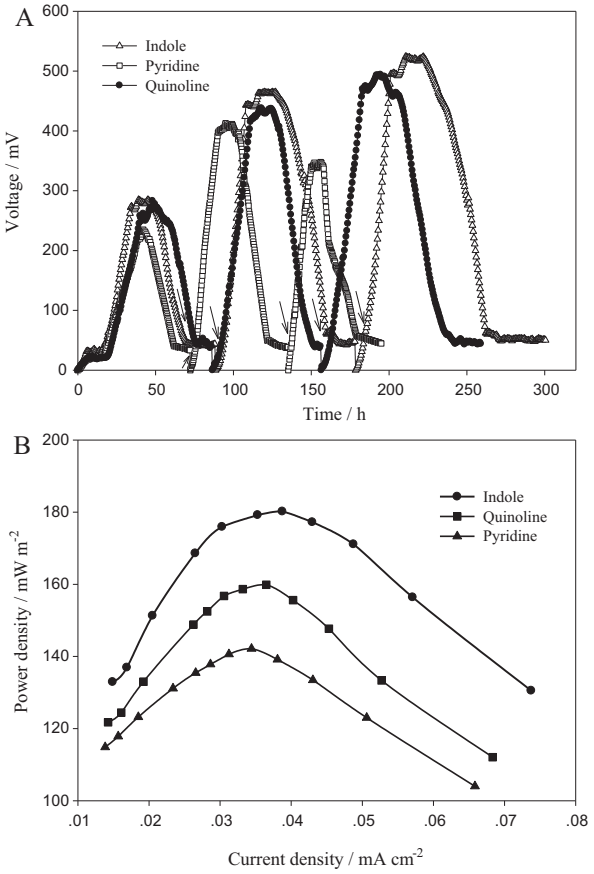

Fig. 3 – (A) Electricity generation from N-heterocyclic compounds (pyridine, quinoline and indole) using a mixed bacterial culture at a fixed resistance of 1000 Ω. (B) Power generation as a function of current density for MFCs using external resistors of 50 Ω to 5000 Ω. The arrows show the time of anode solution replacement with a different concentration (20 mg L^{-1}, 60 mg L^{-1}, 120 mg L^{-1}) of fresh medium solution.

strains and electricity-generation microbes under electric load, which evidenced by the variation of SCOD in sludge supernatant.

3.2. *Power generation from different substrates using two-chambered MFCs*

The experiments demonstrated the feasibility of power generation using N-heterocyclic compounds in MFCs and the bacteria needed to perform this process were already present in the anode chamber after inoculation. The cycles of power generation for reactors fed the same initial and final substrate concentrations (20–120 mg L^{-1}) are shown in Fig. 3A. Reproducible electricity cycles were obtained from all the tested compounds. A circuit voltage of 25 ± 5 mV was quickly generated within only a few hours when the medium solution containing different substrates was added into the anode chamber. The initial voltage might have been due to both chemical and biological factors based on the difference of the potential between the two chambers. Subsequently, the voltage immediately increased due to biological activity and reached a stable value. Voltage generation was recovered by subsequent replacement of artificial wastewaters with fresh ones. It is observed that the maximum voltage was decreased when pyridine concentrations were larger than 60 mg L^{-1}. The results suggest that the pyridine concentration above a particular level produced an inhibition on electrochemical active bacteria.

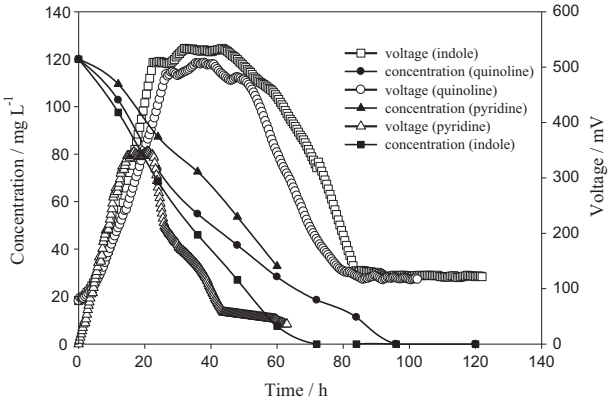

Fig. 4 – Electricity voltage output and degradation of pyridine, quinoline and indole as the sole fuel in the MFCs at the same initial concentration of 120 mg L^{-1}.

Power density was obtained by measuring stabilized voltages at external resistances ranging from 50 to 5000 Ω. By varying the circuit resistance, it is determined from a polarization curve that the maximum volumetric power density was 180.2 mW m^{-2}, 159.9 mW m^{-2}, and 142.1 mW m^{-2} when using indole, quinoline and pyridine as the pure fuel at the concentration of 60 mg L^{-1}, respectively (Fig. 3B). The maximum power output is similar to that obtained using the same two-chambered system in several other studies.

3.3. *Power generation as a function of degradation efficiency*

To compare studies on electricity generation and removal rates using different organic materials, we monitored power output as a function of degradation efficiency (Table 1 and Fig. 4). The operation period was up to 63 h with pyridine and the maximum output voltage reached 346 mV (the external resistance $R = 1000\ \Omega$), while at the same time the concentration of pyridine was 32.8 mg L^{-1} in the anode solution. It took longer time for the bacteria to produce electricity from quinoline (102 h) and indole (122 h), and to achieve the maximum voltage (494 mV, 524 mV). At the end of each power cycle, the detected concentrations obtained from the anode chamber were 11.3 mg L^{-1} and 7.6 mg L^{-1}, respectively. The time of power generation still lasted about 60 h and output voltage exceeded 50 mV with indole when the removal rate reached more than 93.7%. Similar results were observed for quinoline and pyridine. The experimental data indicated that: (1) degradation efficiencies and power generation were different in the circumstances of the same initial concentration, which might be related to chemical structures, mass fraction of benzene ring in compound, biological activities; (2) there was a disparity between the biodegradation rates and electricity generation of organics in the MFC, the electricigens were able to continuously produce electricity using intermediate metabolites of refractory organic compounds until completely consumption of all the substances. The amount of Coulombs recovered by the MFCs was calculated based on the electrical cycles. The CEs measured at each substrate concentration were less than 8.0% in the MFCs and decreased with an increase in substrate concentration. The inverse relationship between CE and substrate concentration is similar to that observed by Liu et al. (2005). The CEs were quite low,

Table 1 – MFC performance using different substances as carbon sources.

Compound	Molecular formula	Chemical structures	Mass fraction of benzene ring in compound (%)	V_{max} (V)	Maximum removal efficiency (%)	Maximum COD removal efficiency (%)	Power density (mW m^{-2})	Current density (mA m^{-2})
Pyridine	C_5H_5N			0.413	72.7	86	142.1	344
Indole	C_8H_7N		67	0.494	90.6	95	228.8	437
Quinoline	C_9H_7N		60	0.524	93.7	93	203.4	411

indicating a substantial loss of electrons in the systems. The significant electron losses may be due to the electron transfer from the substrates to other electron acceptors in solution and/or assimilation for bacterial growth (Liu et al., 2005).

3.4. *Power as a function of substance concentration*

The effect of power generation for reactors fed different initial substrate concentrations (120 mg L^{-1}) at 1000 Ω external resistance was examined (Fig. 5A and Table 1). Maximum power density ranged from 142.1 mW m^{-2} to 228.8 mW m^{-2}. Indole produced the highest maximum power density (228.8 mW m^{-2}) with K_s = 33.5 mg L^{-1}, and pyridine generated the lowest power density (142.1 mW m^{-2}) with K_s = 28 mg L^{-1} among the compounds examined. A plot of the maximum power output at each initial substrate concentration showed saturation kinetics at four different circuit loads (Fig. 5B). A maximum power density of P_{max} = 196.4 mW m^{-2} and a half-saturation constant of K_s = 29.8 mg L^{-1} (R^2 = 0.9959) were obtained using a 600 Ω resistance, while those using 2000 and 5000 Ω resistance were P_{max} = 181.5 mW m^{-2} and K_s = 28.1 mg L^{-1} (R^2 = 0.9903), and P_{max} = 142.1 mW m^{-2} and K_s = 29.1 mg L^{-1} (R^2 = 0.9799). Our experimental results demonstrate that the half-saturation constant was not only dependent on substrate type but also external resistance. Results showing how external resistance corresponds to maximum power would enable us to better evaluate the effect of substrate concentration on MFC operation because MFCs are preferentially operated at an external resistance that correlated to a maximum power output for bioenergy applications.

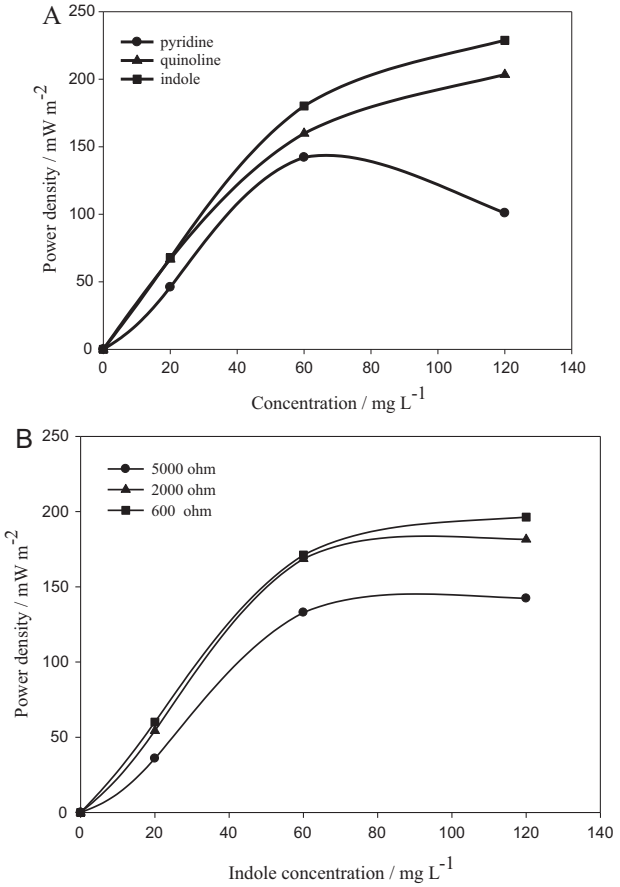

Fig. 5 – Maximum power density as a function of (A) initial various substrate concentration using a 1000 Ω resistance and (B) initial indole concentration using different resistors.

3.5. *Influence of COD removal on voltage generation*

Consumptions of three selected compounds and COD corresponded with the increase of output voltage are shown in Fig. 6. It was observed that the COD biodegradation rates exceeded 86% in all the treatments (Table 1). As shown in the figure, voltage production and COD removal rates of indole were higher than other substances under the same experimental conditions. MFC performance was affected significantly by the different types of fuel.

3.6. *The variation of anode and cathode potentials*

Indole-containing (120 mg L^{-1}) medium solution was fed into the MFC, yielding a stable cathode potential of 380 ± 20 mV vs. SHE with a 1000 Ω resistor (Fig. 7). The variation in cathode potential was due to electrochemical losses. Evidence from experimental data showed that the potentials of anode tend to decrease from 380 mV to −208 mV vs. SHE and then steadily increased to 380 mV as the time passing. The redox potential of the anode indicated that indole was oxidized in the MFC. The exact potential at which indole are oxidized in a MFC cannot be extrapolated from the data set because anaerobic sludge contains biological organic matters. Such phenomenon was also observed in other MFCs with N-heterocyclic compound as the fuel.

4. Discussion

4.1. *Power generation*

Experimental results of the control experiment with carbon paper but without bacteria showed N-heterocyclic compounds absorption by electrode were 1.8–4.2%, which seems to be

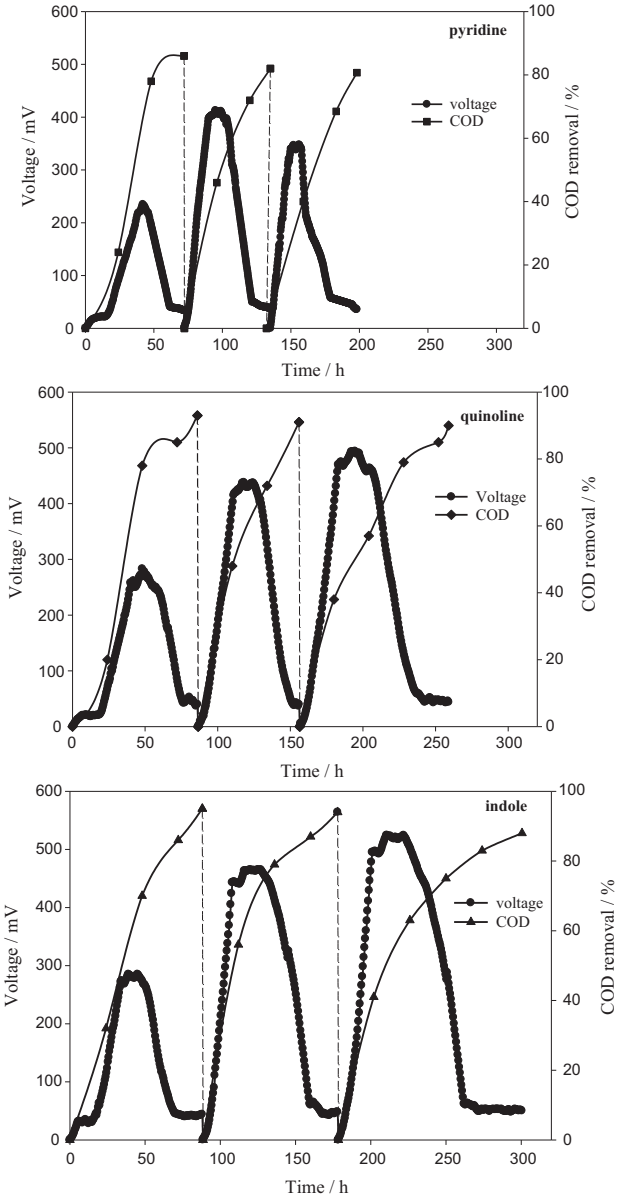

Fig. 6 – Electricity production and COD removal rates using pyridine, quinoline, and indole as the sole fuel.

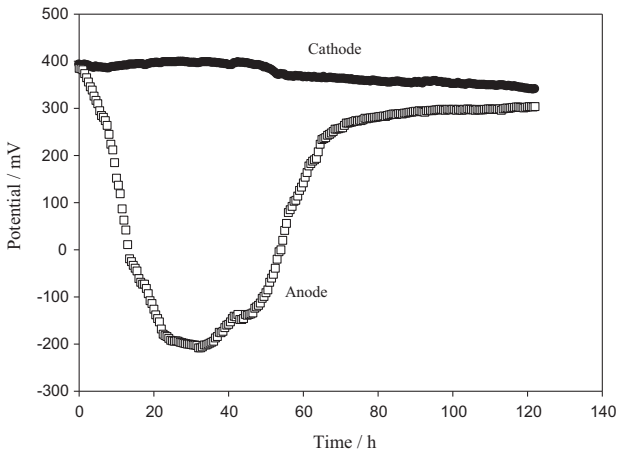

Fig. 7 – Typical evolution of redox potential the anode and the cathode for an indole-fed MFC (potentials calculated vs. SHE).

negligible compared with the removal efficiency of each N-heterocyclic compound in the MFCs (Table 1). The current experiments demonstrate that all of the N-heterocyclic compounds tested could be used as the fuels and successfully produce electricity in two-chambered MFCs. The duration of electricity generation, the maximum voltage, and the maximum power density were comparatively studied using different substrates in MFCs. The maximum voltage produced was 145 ± 5 mV and the corresponding TCOD removal efficiency was about 35% using anaerobic sludge. MFC technology could be recognized as one of a promising sludge pretreatment process. The amount of power generated through indole oxidation (maximum of 228.8 mW m^{-2} for typical operating conditions) is comparable to power reported in many other MFCs studied using readily biodegradable compounds, such as lactate of 0.6–15 mW m^{-2} (Kim et al., 2002; Park and Zeikus, 2003; Rabaey et al., 2004), and also recalcitrant organics, such as 38 mW m^{-2} from cysteine in similar types of two-chambered MFCs (Logan et al., 2005). The results suggest that the type of organic matter, and perhaps the bacterial community that developed during acclimation, affected microbial kinetics and/or the maximum power density with N-heterocyclic compounds. The maximum voltage outputs obtained from indole or quinoline were higher than pyridine, meanwhile, the voltage gradually increased and the duration of power generation extended when the substrate concentrations varied from 20 to 120 mg L^{-1} at a fixed external resistance of 1000 Ω. But pyridine was inconsistent with the above regularity. There were several possible reasons to explain these results: (1) the aerobic biodegradation of pyridine and the inadequate population of capable microorganisms in the anode chamber might have led to the lower power density and the shorter time of electricity generation; (2) less electrons could be generated in the unit time by the same or different consortia of bacterium; (3) the activities of various electricity-generating bacteria were inhibited in the presence of refractory compound and electrochemically active microbes were not the dominant species, thus the majority of organics were degraded by other strains. The characteristics of fuels had a great impact on the effect of MFC productivity.

In our experiments, all of the refractory compounds could be degraded by the mixed bacteria present in anaerobic sludge, which was cultured and acclimated over several months. However, only specific substrates can be utilized as carbon sources by most of electricity-generating bacteria. For example, strain *Geobacter metallireducens* could only incompletely oxidize a limited number of aromatic compounds such as phenol, toluene, and benzoic acid (Kazumi et al., 1995). The *Pseudomonas* species isolated from a MFC with glucose as a carbon source could not further utilize the fermentation products for power generation (Rabaey et al., 2004). Further researches are necessary to analyze the microbial community from the different substrates and understand the microbial reactions occurring at the anode and their roles for electricity generation. It is clear from the experimental results that the removal rates of pyridine were obviously lower than quinoline and indole, although the tested substrates were simple N-heterocyclic compounds containing 5–9 carbons and their molecular structures are different (Table 1). The differences in structure might influence the selection of bacterial species at the electrode and/or the different metabolic pathways for the same bacterial species.

4.2. *Metabolic pathway of substrate degradation*

A published report discussing the degradation of pyridine under aerobic conditions indicated that pyridine ring was cleaved between the N and C-2 atoms and subsequently deaminated to glutaric diadehyde, followed by successive oxidation to glutarate semialdehyde (Rhee et al., 1997). In contrast, the metabolic fate and pathway of pyridine by anaerobic microbial populations has largely been ignored. In our study, the value of pH was decreased during the pyridine degradation, indicating that some acid intermediates were produced. Additionally, some long-chain fatty acid intermediates and heterocyclic intermediates in the anode chamber were detected by GC/MS, These components suggest that pyridine in the MFC establishment is possible to be biodegraded via the metabolic pathway suggested by Zefirov et al. (1994), which is different from another published studies (Rhee et al., 1997; Zhang et al., 2009). The reports proposed that pyridine ring was cleaved between the N and C-2 atoms and subsequently deaminated to glutaric diadehyde, followed by successive oxidation to glutarate semialdehyde.

Gu et al. (2002) determined that indole may be degraded in the metabolic pathways through two-step hydroxylation pathway and yielding oxindole and isatin subsequent to cleavage between the C-2 and C-3 atoms on the pyrrole ring of indole. However, isatin was not observed in the culture medium of this study and the intermediates of indole were not reported in the past. When authentic oxindole and isatin standards were used for identifying the intermediates which appeared in the culture, the unidentified peak did not match those of either oxindole or isatin. On the basis of products analysis, oxidation was proposed to occur at the heterocyclic double bond, followed by deprotonation at nitrogen.

Many pure bacteria were investigated for the biodegradation of quinoline in previous studies, but few researchers used mixed microbes in the anaerobic sludge. It has been reported that the metabolic pathway of quinoline under an anaerobic environment is initiated by oxidation to 2(1H)-quinolone, and followed by oxidation to 3,4-dihydro-2(1H)-quinolone (Johansen et al., 1997). Quinolone was recognized as the main degradation product in numerous studies either under aerobic or anaerobic condition. Similarly, we also identified 2(1H)-quinolinone in the samples taken 12 h, but no degradation products were observed in the samples taken near the end of the experiment, which indicated that 2(1H)-quinolinone was being further transformed.

In the current study, it should be pointed out that anaerobic biodegradation pathways of pyridine, indole and quinoline could not be determined only by the detection of complex intermediates. Few reports were published to support the analysis results presented here, which may be related to sampling time and experimental conditions, microbial species etc. The comparison of the data suggested that the order of degradation rate of three N-heterocyclic compounds under anaerobic condition was indole > quinoline > pyridine. A similar result was found by Li et al. (2003). N-heterocyclic compounds in the MFC might produce many chemical reactions such as oxidation, reduction, polymerization and hydrolyzation, which enhanced the removal efficiency. Research into current literature about the degradation pathways and intermediate products of N-heterocyclic compounds indicated that many disparities and gaps exist, and further studies are needed.

5. Conclusions

Results from this study demonstrated the feasibility of using two-compartment MFC reactors to produce electricity and simultaneously degrade N-heterocyclic compounds. The maximum power density in the range of 142.1–228.8 mW m^{-2} was achieved using a mixed microbial communities enriched from anaerobic sludge, which were acclimated over several months so as to increase the number of electricity-generating bacteria in the anode chamber and reduce the anaerobic fermentation process of other microorganisms. Electricity generation was accompanied by $90 \pm 4\%$ removal efficiencies of COD and $88 \pm 5\%$ degradation rates of each substrate used. Effect of substrate concentrations on power generation followed an empirical Monod-type equation, with a maximum half-saturation concentration constant of $K_s = 29.8$ mg L^{-1} (indole). The variation of anode and cathode potentials suggested that MFC performance is markedly influenced by the anode solution. The sample analysis with GC/MS technique showed that the intermediate products were formed in the degradation process. The MFC technology offers a new biotechnology for energy generation to enhance the biodegradation of recalcitrant contaminants such as N-heterocyclic compounds in the anaerobic environment and a novel method to offset operating costs, making advanced remediation measures for difficult to degrade materials more affordable for practical applications.

Acknowledgements

This work was financially supported by the National Natural Science Foundation of China (20977026, 50978088), the National 863 High Technology Research Foundation of China (2006AA06Z407), the Research Fund for the Doctoral Program of Higher Education of China (20090161110009), the Hunan Provincial Natural Science Foundation of China (06JJ20062), the Hunan Key Scientific Research Project (2009FJ1010), and the Equipment Foundation of Hunan University.

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