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ORIGINAL PAPER

A sandwiched denitrifying biocathode in a microbial fuel cell for electricity generation and waste minimization

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Abstract A denitrifying biocathode in a microbial fuel cell was developed to investigate the replacement of the costly Pt-coated abiotic cathodes for electricity generation. The denitrifying biocathode was sandwiched between the dual-anode systems. The study investigated the performance for simultaneous treatment of wastewater on the anode, biological denitrification on the cathode and the potential recovery of electrical energy. Autotrophic biofilms performed denitrification on the cathode using supplied electrons by the biodegradation of organics on the anode. Graphite granules were used as electrodes for biofilm attachment, and nafion membranes were used as separators between electrodes. The system achieved a volumetric power of 7 ± 0.4 W m^{-3} net cathodic compartment (NCC) with the simultaneous removal of 229.5 ± 18 mg L^{-1} COD on anode and 88.9 g m^{-3} NCC day^{-1} nitrogen on cathode, respectively. The columbic efficiency for cathodic and anodic reactions was 98.9 ± 0.57 and 23.54 ± 0.87 %, respectively. This is a combined study for domestic wastewater treatment and biological denitrification in a compact MFC reactor. Further optimization of the system is desired to improve its performance and applicability.

Keywords Autotrophic biofilms · Biocathode · Bioelectricity production · Biological denitrification · Microbial fuel cell · Microbial electrochemistry

Introduction

Microbial fuel cells (MFCs) oxidize organic matters in an anaerobic environment to produce electrons by using bacteria as biocatalysts (Liu and Logan 2004; Rabaey and Verstraete 2005; Lovley 2006; Lefebvre et al. 2010; Nasirahmadi and Safekordi 2012). The produced electrons from the organic substrates are transmitted to the negative anode electrode and flow to the positive cathode electrode continuously through the external electric circuit, producing electricity. The electrons at the cathode will then combine with protons (diffuses through the PEM from the anode) and oxygen to form water (Chaudhuri and Lovley 2003; Min and Logan 2004; Juang et al. 2011; 2012). The flow of electrons and the potential difference between the respiratory enzyme of microbes and oxygen generate the current and voltage, respectively. Hence, a typical MFC comprises an anaerobic anode chamber and an aerobic cathode chamber separated by the proton exchange membrane (PEM) that allows the protons to pass through it.

The biological anode has already been proven a cost-effective and sustainable solution for MFC operation. Thus, only needs to optimize the operating conditions of bioanode for maximizing the electricity generation. On the other hand, the metallic and chemical cathodes used with the biological anode in a MFC system had either an unsustainable process, such as a hexacyanoferrate solution needs

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regeneration (Rabaey et al. 2005), or an expensive platinum-coated cathode for oxygen reduction (Logan et al. 2007). The second one is also affected by sulfide poisoning from the biological activity in wastewater (Bard and Faulkner 2001). Similar investigation on pyrolyzed iron (II) phthalocyanine (FePc)- and cobalt tetramethylphenylporphyrin (CoTMPP)-coated cathode has provided promising and inexpensive abiotic cathode alternatives (Zhao et al. 2005). Recent investigation on a biologically regenerated ferric iron solution combined with a bipolar membrane or Pt-free cathode system has shown an innovative way of metallic reduction on cathode (terHeijne et al. 2006; Lefebvre et al. 2009). However, the above-mentioned abiotic cathodes are questionable, as they cannot provide sustainable and long-term application of the cost-effective MFC. For these reasons, some researchers have recently started working on biocathodes that would use bacteria as biocatalyst for sustainable and economic operation of MFC. Recent studies on aerobic biocathodes in which oxygen as terminal electron acceptors have increased interest because of their sustainable performance and high power production compared to the chemical cathodes (Bergel et al. 2005; Clauwaert et al. 2007a). Gregory et al. (2004) first found that bacteria could recover electrons directly from a potentially poised graphite cathode to reduce nitrate to nitrite (Gregory et al. 2004). Park et al. (2005) observed a similar finding where nitrate is completely reduced to nitrogen gas in potentiostat-poised half-cells (Park et al. 2005).

Among the electron acceptors in biocathodes, nitrate is significantly important due to its relatively higher redox potential for reducing nitrate to molecular nitrogen gas, $NO_3^-/N_2 (E' + 0.74\,V)$. Due to higher redox potential of the denitrification reactions, there are few studies done to combine this biological denitrification in a complete MFC (Clauwaert et al. 2007b; Virdis et al. 2008; Lefebvre et al. 2008a, b). However, none of the previous studies has applied this technology for direct sewage treatment. That is why, an MFC operated with denitrifying biocathode would be a great potential for cost-effective energy production and waste minimization. The additional benefit of the denitrifying biocathode is that it limits the risk of oxygen leaking into the anode chamber. Thus, it will increase the efficiency of the anodic reaction as well as it will achieve simultaneous removal of carbonaceous substrates and nitrogenous compounds in anode and cathode chambers, respectively. Zhang et al. (2013) first investigated the combined removal of carbon and nitrogen form single wastewater stream using single chambered biocathode MFC (Zhang et al. 2013). Few recent studies on the biocathode denitrification for simultaneous carbon and nitrogen removal by MFC have used synthetic wastewater as electron suppliers (Virdis et al. 2008; Zhen and Largus 2006; Xie et al. 2011; Zhang and Zhen 2012). That is why, those studies do not reveal the practical significance of using this technology for raw wastewater treatment. Two recent studies have used direct raw wastewater as an electron supplier for biocathode denitrification in MFC using a double-chambered MFC configuration (Virdis et al. 2010; Li et al. 2014).

Due to the limited information available for scaling up denitrifying biocathode in complete MFC for continuous treating of municipal wastewater in anode chamber, this study was carried out (1) to check the feasibilities of applying a sandwiched denitrifying biocathode in scaled-up MFC, (2) to optimize cathodic nitrate loading rate using domestic wastewater as a sole electron donor for denitrification, (3) to determine the possible electrical power generation based on this cathodic denitrification process and (4) the operational environment for the denitrification process.

Materials and methods

Design of up-flow channeled MFC

The entire study was carried out with two separate reactors. The biocathode MFC system used in this study is illustrated in Fig. 1. The MFC reactor was constructed by three parallel acrylic chambers. The middle chamber acted as biocathode. It was sandwiched by the two chambers at the sides, which were acting as bioanode. The domestic wastewater continuously pumped through the anodic chambers, where bacteria degraded the organics by its

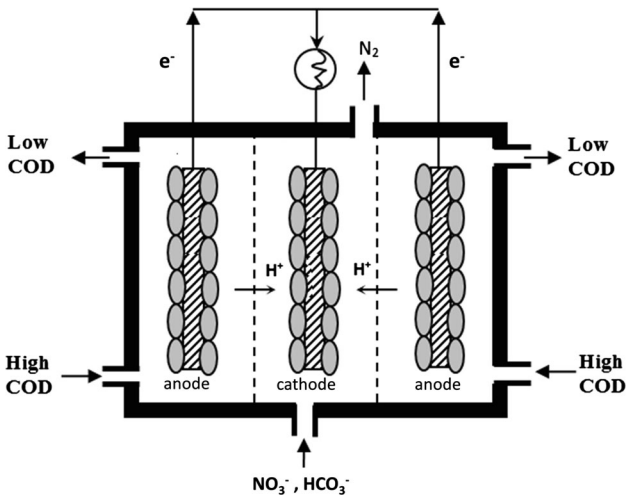

Fig. 1 MFC-based system for simultaneous carbon and nitrogen removal

metabolic pathway to produce CO_2, protons and electrons. The electrons produced through the metabolic process were collected at the anode electrode and diverted toward the cathode electrode, where they were retrieved by denitrifying bacteria to reduce nitrate to nitrogen gas. A synthetic wastewater containing nitrate was continuously pumped into the cathode chamber by a peristaltic pump (Master Flex, USA). Due to pH gradient, the produced protons in the anode chamber migrate to the cathode surface by means of diffusion within the electrolytes and throughout the cation exchange membrane.

Each chamber had an internal dimension of 18 × 18 × 4 cm with the inlet at the bottom and outlet at the top. The vertically up-flow channel facilitated the collection of possible gases (biogas in the anode chamber and N_2 gas in the cathode chamber) produced. The up-flow channel volume in each chamber was 1.08×10^{-3} m^3. Both the anode and the cathode chambers were filled with granular graphite (type 00514; diameter 1.5–5 mm, estimated projected surface between 817 and 2720 m^2 m^{-3}, Le Carbone, Belgium), and the electrical contact were provided by a graphite rod (5 mm diameter). The density and porosity of graphite bed were 1.83 kg L^{-1} and 0.55, respectively (industry specified). The volume of the total cathodic chamber (TCC) was 1.08 L, and the liquid volume between graphite granules, the net cathodic chamber (NCC), was 0.6 L. The total volume of the anodic chambers (TAC, total anode chambers) was 2.16 L, and the volume of the net anodic chamber (NAC) was 1.2 L. Both the anode and cathode chambers were separated by a proton exchange membrane (nafion-112, 51-micron thickness, GasHub Technology, Singapore). Before use, the graphite granules were washed at least five times with distilled water and then sequentially submerged in 1 N NaOH solution and 1 N HCl solution, each for 24 h. Subsequently, the granules were washed again five times with distilled water (Virdis et al. 2010) prior to being filled into the chambers. The PEM (nafion-112) was sequentially boiled at 80 °C in H_2O_2 (3 %), deionized water, 0.5 M H_2SO_4 and then deionized water, each for 1 h and then stored in deionized water prior to being used.

Enrichment of electrochemically active microbes

Both anodic and cathodic liquid streams were circulated (1.25 ml min^{-1} or 0.075 L h^{-1}) in an up-flow mode during enrichment period. In the anode chambers, sieved sewage (355-μm sieve, effluents from a primary settling tank, Ulu Pandan wastewater treatment plant, Singapore) was circulated continuously as a substrate and seed simultaneously during the start-up of the MFC by a peristaltic pump (master flex, USA). The characteristics of domestic wastewater averaged as 283 ± 32 mg L^{-1} of total COD, 82 ± 13 mg L^{-1} of soluble COD, 217 ± 64 mg L^{-1} of suspended solids (SS), 197 ± 56 mg L^{-1} of volatile suspended solids (VSS), 405 ± 5 mg L^{-1} of total dissolved solids (TDS), 38 ± 4 mg L^{-1} of total nitrogen (TN), 39 ± 34 mg L^{-1} of phosphate (PO_4^{3-}). The pH was 7.2 ± 0.1 (All those data show averages and standard deviations based on a series of four samples).

During enrichment stage, the cathodic liquid stream was recirculated in up-flow mode by an external recirculation vessel. The cathodic liquid stream consisted of a modified buffer medium containing per liter: 4.4 g KH_2PO_4, 3.4 g K_2HPO_4, 2 g $NaHCO_3$, 0.5 g NaCl, 0.2 g $MgSO_4 \cdot 7H_2O$, 0.0146 g $CaCl_2$ (Virdis et al. 2010), 12.5 ml trace mineral solution and 12.5 ml vitamin solution (Lovley and phillips 1988; Balch et al. 1979; Lovley et al. 1984). Different types of aerobic and anaerobic sludge (activated sludge, digester sludge) and sediments (sediments from the primary and the secondary settling basin) were mixed in order to obtain the seeding microbes (2.5 ml of dewatered sludge in 1 L of cathodic liquid stream) for cathodic enrichment with sufficient microbial diversity. A concentrated KNO_3 solution was added in the recirculation vessel twice a day to achieve a desired volumetric loading rate of 0.1 kg NO_3^- –N m^{-3} NCC day^{-1} (33.33 mg NO_3^- –N L^{-1} of buffer solution). The liquid in the recirculation vessel was replaced after every 15 days in order to avoid potassium accumulation. By ending microbial enrichment at 30th days, the cathodic liquid circulation was switched to continuous flow mode.

Calculations

Cell potential (E_{cell}, V) was measured with a multimeter connected to a computer by a data acquisition system (PC1604, TTi, RS, Singapore) at every 30-min interval. Power (P, W) was calculated as: $P = I \cdot E_{cell}$, where current (I, A) was determined according to the ohm's law: $I = E_{cell}/R_{ext}$, and R_{ext} (Ω) was the fixed external resistance. Volumetric power (P_V, W m^{-3} NCC) was determined by $P_v = E_{cell}^2/(V \cdot R_{ext})$, where V (m^3) was the net volume of the cathodic compartment.

The open circuit voltage (OCV) measured for an MFC was the maximum cell potential generated at the system under infinite resistance (no current). Polarization and power density curves were drawn by varying the external resistance from infinity to 1 Ω using a resistor box (RS201, USA). The cell potential values were recorded only after the pseudo-steady-state conditions had been established. The establishment of this pseudo-steady-state had taken several minutes or more, depending on the cathodic nitrate concentration and the external resistance. By changing the external resistance, we obtained a new cell potential, and

hence a new current density as $I = E_{cell}/(V \cdot R_{ext})$ and power density as $P_v = E_{cell}^2/(V \cdot R_{ext})$, where V was the net volume of the cathodic compartment.

The internal resistance of the MFC was calculated from its open circuit voltage (no load) OCV, loaded voltage E_{cell} and the load resistance R_{ext} as $R_{int} = (OCV/E_{cell} - 1)\cdot R_{ext}$, R_{int} being the MFC internal resistance. R_{int} could also be determined simply as the slope of the linear part of the polarization curve (i.e., $R_{int} = -\Delta E/\Delta I = 16.5\ \Omega$, Fig. 3, voltage polarization curve for the cathodic nitrate loading of 0.12 kg N m^{-3} NCC day^{-1}). From the fuel cell electrochemistry, it is known that the power output of a fuel cell would be maximum when $R_{int} = R_{ext}$ during the voltage polarization period. That is why, R_{int} values were the direct values of R_{ext}, when the maximum power density achieved during polarization.

The cathode chamber columbic efficiency (ϵc, %) was calculated as: $\epsilon c = 100\ MI/(Fbq\Delta NO_3)$, where $M = 62$ g mol^{-1} was the molecular weight of nitrate, $F = 96{,}485.3$ C mol^{-1} of electron was Faraday's constant, $b = 5$ was the number of electrons exchanged per mole of nitrate reduction, q (L s^{-1}) was the flow rate of the cathodic liquid stream and ΔNO_3 (g L^{-1}) was the change in nitrate concentration in the cathodic liquid stream. The nitrate reduction at the cathode chamber follows the equation as $NO_3^- + 5e^- + 6H^+ \rightarrow 0.5N_2 + 3H_2 - O$.

The anode chamber columbic efficiency (ϵc, %) was calculated as: $\epsilon c = 100MI/(Fbq\Delta COD)$, where $M = 32$ g mol^{-1} was the molecular weight of oxygen, $F = 96{,}485.3$ C mol^{-1} of electron was Faraday's constant, $b = 4$ was the number of electrons exchanged per mole of oxygen reduction, q (L s^{-1}) was the flow rate of anodic liquid stream and ΔCOD (g L^{-1}) was the change in total organic concentration in the anodic liquid stream. The electrons produced from the organic oxidation at the anode were used by the oxygen reduction at cathode chamber following the equation as $O_2 + 4H^+ + 4e^- \rightarrow 2H_2O$.

Analytical methods

COD, TS, TVS, SS, VSS, TDS were measured according to standard methods. At the enrichment stage, an optical method (ultraviolet absorption spectrometry at a single wavelength of 220 nm) was used for rapid measurements of nitrate concentration in the cathodic liquid stream (Collos et al. 1999). Later on, nitrate, nitrite, phosphate and NH_4^+ and SO_4^{-2} in the anodic and the cathodic liquid stream were determined by an Ion Chromatograph (DIONEX-500 fitted with a GP50 gradient pump and a CD20 conductivity detector) with IonPac CS12A cation and IonPac AS9-HC anion column. In those measurements, samples were first filtered through a 0.2-μm pore diameter membrane before analysis. Produced N_2 gas analyses were performed using a gas chromatograph (GC-17A, Shimadzu) with a charlston 80/100 porapak column. Total nitrogen was measured using a Shimadzu TNM-1 unit coupled with a TOC-V analyzer. In both case, samples were pre-filtered through a 0.2-μm pore-sized membrane. pH of the anodic and the cathodic liquid was measured by a standard pH probe with a pH meter.

Results and discussion

Cell potential generation during enrichment stages

During the enrichment period, both the reactors were operated at an external resistance of 12.5 Ω. Domestic wastewater (Sieved effluents through a 355-μm size from the primary settling basin) was the sole electron donor for the anodic microbial consortium, and the synthetic nitrate solution with a loading rate of 0.1 kg NO_3^- −N m^{-3} NCC day^{-1} (33.33 mg NO_3^- −N L^{-1} of solution) was the sole electron acceptor for cathodic microbial consortium. Figure 2 shows the cell potential generation at the enrichment period. Two distinct zones of cell potentials increase had been found. The first zone (Fig. 2a) was the exponential increasing zone, and the second zone (Fig. 2b) was the gradual increasing zone of cell potential. The first exponential increase in cell potential during the first 6 days indicated the growth of anodic microbial consortium, which was the electron donor for cathodic denitrification.

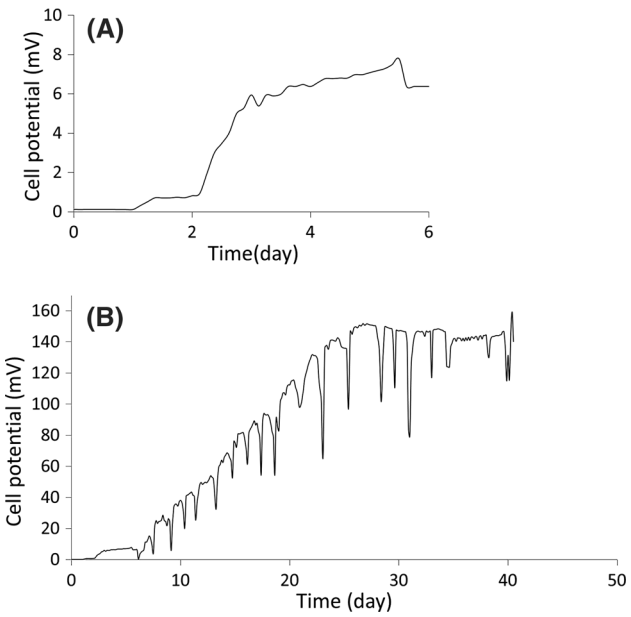

Fig. 2 **a** Cell potential generation during first 6 days. **b** Cell potential generation during entire enrichment period

The second gradual increase in cell potential during 6th–29th days indicated the growth of cathodic microbial consortium, which was the electron acceptor for biocathode denitrification. After approximately 6 days enrichment of the cathode chamber, there was a gradual increase in cell potential over the next 23 days, reached a saturated value of 150.8 ± 4.2 mV (20.11 ± 0.56 A m^{-3} NCC). After approximately 30 days, both the MFC reactors achieved a complete denitrification without nitrite accumulation, whereas nitrate and nitrite were partially removed in the start-up period due to less electron retrieval at cathode.

Steady-state electricity and power production with fixed resistor at different cathodic nitrate loading

The MFCs were operated with domestic wastewater as anodic influent and synthetic nitrate solutions as cathodic influent at 12.5-Ω fixed external resistance. The initial nitrate loading rates in the cathode chamber were varied from 0.025 to 0.14 kg NO_3^- −N m^{-3} NCC day^{-1}. Figure 3 and Table 1 summarize the results of the 8 months of continuous feeding operations, in terms of electrical performances well as nitrogen and COD removal in the cathode and anode chamber, respectively. The results showed that with the gradual increase in initial nitrate loading in the cathodic stream, specific current and power productions were increased, while cathodic denitrifying efficiency was decreased. The maximum specific current achieved was 28.9 ± 0.4 A m^{-3} NCC at the initial cathodic nitrate loading of 0.12 kg NO_3^- −N m^{-3} NCC day^{-1}, leading to the cathodic nitrate removal efficiency of 71.1 ± 3.7 % and the anodic COD removal efficiency of 32.7 ± 2.6 %. The nitrate loading rate above 0.120 kg NO_3^- −N m^{-3} NCC day^{-1} showed a slight decrease in currents and power productions which indicated the anodic depletion (the anode suffers limitations to supply electrons) of this MFC. The optimum cathodic nitrate loading indicated that domestic wastewater had a limit to act as a sole electron donor for cathodic denitrification. The Table 1 also demonstrated that the current and power productions were proportional to the denitrification rate. The optimum nitrate loading corresponding to the electron supplied from domestic wastewater for complete denitrification was 0.120 kg NO_3^- −N m^{-3} NCC day^{-1}. However, a significant amount of biogas was produced in the anode chamber, while the produced gas in the cathode chamber was mostly nitrogen with a trace amount of methane (data not shown).

The COD removal obtained here was quite low, e.g., 32.7 ± 2.6 %, which should to be further increased. Since

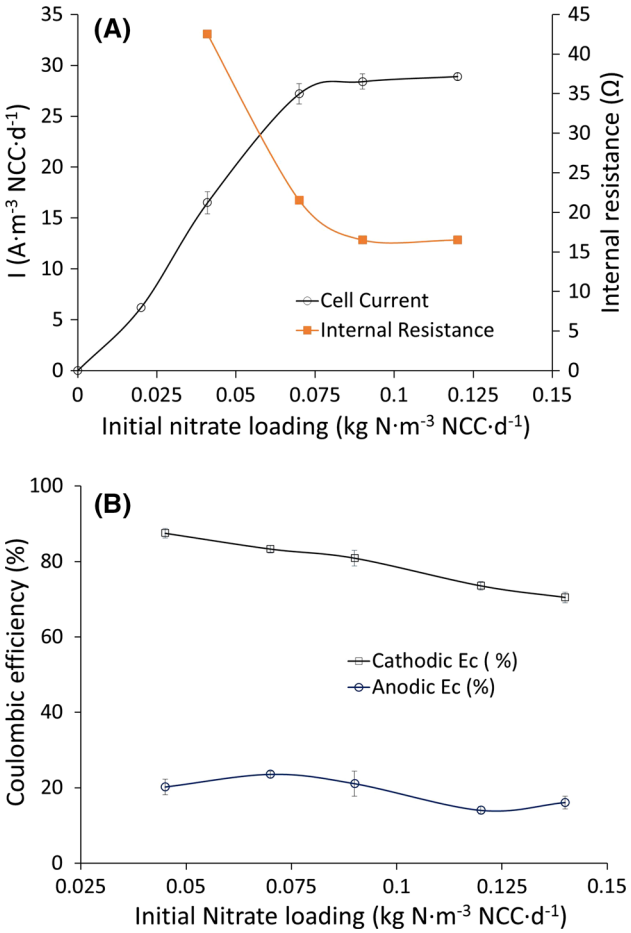

Fig. 3 **a** Current production and internal resistance of MFC. **b** Columbic efficiency for cathodic and anodic reaction (results are average and standard deviation of four samples)

the domestic wastewater is a mixture of easily biodegradable and complex organics. The most possible way of achieving higher organics removal from the domestic wastewater stream is to add another anaerobic pre-treatment chamber prior to the MFC. The pre-treatment chamber will enhance fermentative microbial degradation (hydrolysis, acidogenesis and acetogenesis) of complex polymers such as starch, cellulose and lignin and convert them into simple volatile fatty acids (Donoso-Bravo et al. 2009). At the same time, the MFC reactor design and the selection of materials should be optimized to achieve sufficient microbial density within the vicinity of the MFC electrode to harvest maximum electrons.

The highest denitrification rate obtained here was 88.93 g NO_3^- −N m^{-3} NCC day^{-1}, which was 39 % lower than that found by Clauwaert et al. (2007b) in which they used acetate instead of domestic wastewater as electron donors (Clauwaert et al. 2007b; Virdis et al. 2010). The

Table 1 Parameters describing the system performance using 12.5-Ω external resistance (results showing as averages and standard deviations of four samples for two separate reactors)

Initial nitrate loading (kg NO_3^- –N m^{-3} NCC day^{-1})	Specific I production (A m^{-3} NCC)	Specific P production (W m^{-3} NCC)	Cathodic denitrification (g NO_3^- –N m^{-3} NCC day^{-1})	Cathodic denitrification (%)	Anodic COD removal (mg/L)	Anodic COD removal (%)	NH_4^+ lost in anodic flow (mg NH_4^+ h^{-1})	NH_4^+ gained to cathodic flow (mg NH_4^+ h^{-1})	OCV (V)	Max. I* (A m^{-3} NCC)	Max. P* (W m^{-3} NCC)
0.041 (12.5 mg N/L)	16.2 ± 1.1	1.77 ± 0.1	33.3 ± 2.4	82.1 ± 2.9	84.6 ± 7	13.6 ± 1.2	1.29	0.95	0.524	16.6	2.03
0.07 (18.9 mg N/L)	27.2 ± 0.5	5.07 ± .3	53 ± 2.4	**90.5 ± 1.4**[a]	115.5 ± 12	18 ± 0.6	1.09	1.02	0.578	29.73	5.83
0.09 (28.2 mg N/L)	28.4 ± 0.8	5.6 ± 0.3	69.42 ± 1	76.4 ± 1.5	148.4 ± 28	21.7 ± 4.3	1.62	1.31	0.579	34.54	6.41
0.12 (37 mg N/L)	**28.9 ± 0.4**[a]	**5.74 ± 0.1**[a]	83.73 ± 3.7	71.1 ± 3.7	**229.5 ± 18**[a]	**32.7 ± 2.6**[a]	1.52	1.38	0.593	42.98	**6.6**[a]
0.14 (45 mg N/L)	27.4 ± 0.3	5.2 ± 0.1	**88.93 ± 2.2**[a]	64.2 ± 2.5	194.4 ± 18	27.9 ± 2.6	0	1.37	0.584	41.63	6.5

I current, *P* power, *OCV* open circuit voltage

* Refers to the results in case of voltage and power polarization

[a] Refers to the maximum values in the series investigation

denitrification rate found in this MFC-based system was significantly lower than the heterotrophic denitrification in conventional system, but the second one did not recover electrical energy from the organic degradation. Similarly, the autotrophic denitrification by the anoxic and anaerobic NH_4^+ oxidizing bacteria (anammox) could also obtain higher nitrogen removal with a longer start-up period, but it could not remove organic carbon (Pynaert et al. 2004). That is why, the MFC-based denitrification is a cost-effective and sustainable process.

Ammonium diffusion toward the cathode stream

Ammonium diffused from the anode stream toward the cathode stream during the operation period. Table 1 shows that the diffusion rate of ammonium from the anode stream toward the cathode stream was increased with the increase in current production. Higher current production was associated with the higher amount of electron flow through the outer circuit. That is why, the higher electron flow needs more cations to be transported toward cathode to counter balance more negative charges. Similar findings were presented by Rozendal et al. (2006) in which the ammonium fluxes through the cation exchange membrane toward the cathode increased with the increasing current production (Rozendal et al. 2006). The nitrogen losses through this ammonium diffusion can be reduced by the proper selection of the proton exchange membrane to enhance only H^+ diffusion as well as the establishment of in situ nitrification and denitrification at the cathode surfaces. Further investigations and optimization of this aspect are certainly needed to have a better understanding of this microbial process.

Effect of cathodic NO_3^- loading on steady-state current production and internal resistance of the MFC

Figure 3a shows the steady-state current production and internal resistance of the MFC as a function of cathodic nitrate rate. With the gradual increase in nitrate concentration (substrate) in the cathodic stream, the current production increased following a steady-state current production pattern at higher substrate loading in the cathodic stream. The current generation curve looked like the empirical Monod-type equation as-

$$I = \frac{I_{\max} xNC}{K_{nc} + NC} \quad (1)$$

where I was the specific current production, $I_{\max}$ was the maximum steady-state current production, NC was the concentration of cathodic nitrate as substrate and K_{nc} was the Monod or half-saturation constant, indicating the

concentration of cathodic nitrate that produced a current one half of the maximum current generation. The internal resistance of the MFC showed a gradual decrease with the increase in cathodic nitrate loading following a steady-state value. This data demonstrated that limited fluxes (mass transfer) of electron acceptors to the cathode could result in high mass transfer losses in the lower cathodic nitrate loading (Freguia et al. 2008; Rozendal et al. 2008).

Coulombic efficiencies

In biocathode MFCs, the electricity production was due to organic waste (COD) oxidation at anode chamber and nitrate reduction at cathode chamber. Therefore, the columbic efficiencies in both the anode and the cathode chambers need to measure for the same electricity generation. Figure 3b shows the columbic efficiencies for both the anode and cathode chamber as a function of initial cathodic nitrate loading. A maximum cathodic columbic efficiency of 87.5 ± 1.2 % was achieved at an initial cathodic nitrate loading of 0.041 kg N m^{-3} NCC day^{-1}. With the increase in nitrate loading rate, the cathodic columbic efficiency was decreased due to an abundant of unreduced nitrate in the cathode chamber at high cathodic nitrate loading. Anodic wastewater could supply enough electron flow from organic oxidation to reduce the entire nitrate in the cathode chamber at low cathodic nitrate loading. However, at high cathodic nitrate loading, the supplied electrons from the wastewater oxidation were not enough to reduce the entire nitrate in the cathode chamber, resulting in reduced columbic efficiency. A maximum anodic columbic efficiency of 23.5 ± 0.9 % was achieved at an initial cathodic nitrate loading of 0.07 kg N m^{-3} NCC day^{-1}. The lower anodic columbic efficiency demonstrated that the COD consumption occurred not only by the current generation process, but also some other processes such as microbial growth, fermentation and methanogenesis. Freguia et al. (2007) showed that a major portion of COD was consumed by the growth of electrogenic microbes in MFC (Freguia et al. 2007). That is why, there had been speculated that acetoclastic methanogenesis and bacterial growth in MFC were a significant form of COD consumption which caused the lower columbic efficiency in the anode chamber. The fermentation and methanogenesis had confirmed by the production of biogas in anode chamber. Further investigation need to be done for better understanding of these competitive microbial processes in anode chamber and minimizing their effects.

Polarization and power density curves

The polarization and power density curves were obtained by varying the external resistance from infinity to 1 Ω using a resistor box. This was done by using the domestic wastewater as anodic and the nitrate solution as cathodic influent. The values of current and voltage were recoded after the establishment of pseudo-steady-state at every point. The establishment of this pseudo-steady-state took several minutes or more, depending on the cathodic nitrate loading and the external resistance. Figure 4 shows the polarization and power density curves at six individual nitrate loadings. The results showed that with the gradual increase in cathodic nitrate loading, the OCV, maximum current and maximum power productions were increased gradually (Table 1, last three columns). The MFC produced a maximum OCV of 0.593 V at the cathodic nitrate loading of 0.12 kg $NO_3 - N$ m^{-3} NCC day^{-1}. This was significantly lower than the maximum value of 1.08 V as determined by NADH (−0.32 V) and nitrate (+0.82 V) redox potentials. However, it was in the same range as conventional MFCs. For instance, the OCV of 0.8 V was reported in a single-chamber air cathode MFC (Liu et al. 2005).

The polarization curves at lower initial cathodic nitrate loading (see curves at nitrate loading of 0.02 and 0.041 kg N m^{-3} NCC day^{-1}) showed a rapid decrease in cell potential with the gradual increase in current production at the outer circuit (the current production will increase by the gradual decrease in external resistance at the outer circuit) indicating that the concentration or mass transfer loss was dominating. The possible causes of high mass transfer loss at the lower cathodic nitrate loading is the insufficient flux of nitrate to the biocathode surfaces and therefore limit the rate of reaction (Logan et al. 2006). The polarization curves at the higher cathodic nitrate loading showed three distinguished zones of voltage decrease (1) a rapid voltage drop at very low current densities indicates activation losses; (2) a nearly linear voltage decrease due to the ohmic losses dominating the maximum voltage losses due to bacterial metabolism and (3) a second rapid voltage decrease at higher current densities is possibly due to the concentration polarization losses (Logan et al. 2006).

The maximum volumetric power obtained was 6.6 (R_{ext} = 16.5 Ω) W m^{-3} NCC at a cathodic nitrate loading of 0.12 kg $NO_3 - N$ m^{-3} NCC day^{-1} indicating a good opportunity of using this denitrifying biocathode MFC for

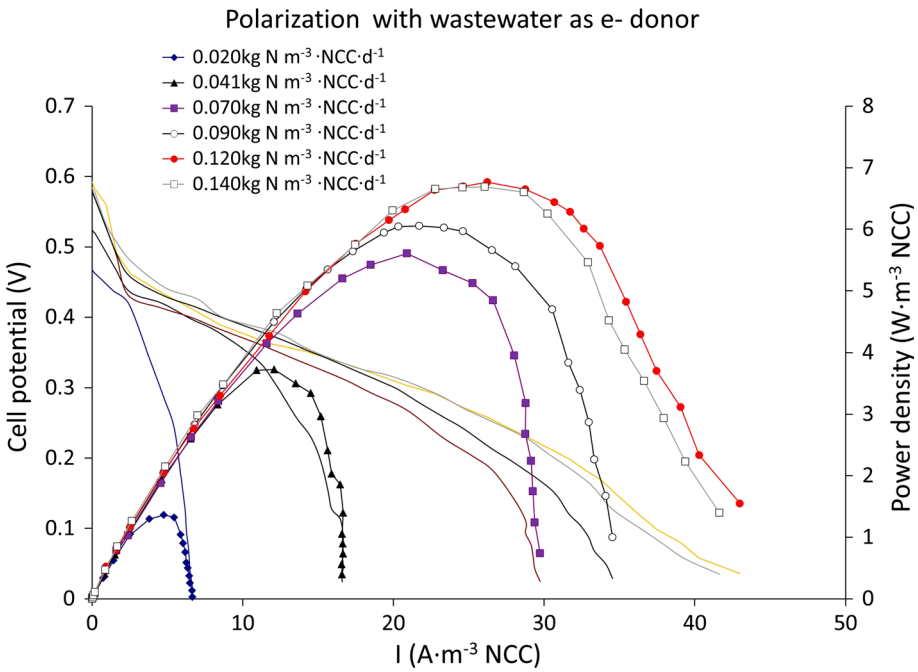

Fig. 4 Polarization and power density curves at different nitrate loading rates (*marker lines* are power density and *plain lines* are polarization curves)

wastewater treatments. The obtained maximum power was 17.5 % lower than that obtained (8 W m^{-3} NCC) by Clauwaert et al. (2007b) in which they used acetate instead of wastewater as anodic influent (Clauwaert et al. 2007b). The nitrate loading in the cathode chamber beyond 0.12 kg NO_3 − N m^{-3} NCC day^{-1} could not produce more power, which indicated the potential limitations of wastewater as anodic electron suppliers.

Conclusion

This is a combined process of achieving complete biological denitrification on cathode by using electrons supplied from oxidation of the organics present in wastewater. This is a cost-effective system, which uses no external energy for COD and nitrogen removal. Instead, the combined process produces electrical energy spontaneously. The use of sandwiched denitrifying biocathode into an MFC showed a great potential for simultaneous production of green energy and minimization of waste. The system already has achieved electrical power production of 6.6 W m^{-3} NCC, COD removal of 33 % and nitrogen removal of 88.93 g NO_3^- −N m^{-3} NCC day^{-1}. The COD removal obtained here needs to be further increased by adding a fermentative pre-treatment of domestic wastewater. However, this study shows the practical relevance of a novel denitrification process even though it has not been fully optimized at this stage. Further improvements of the denitrification process could be achieved by investigating the redox activity of denitrifying enzymes in the different microorganisms and the electron transfer mechanisms from electrode to microbes.

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