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Degradation of *p*-nitrophenol by coupled cathodic reduction and anodic oxidation in a self-powered bioelectrochemical system and analysis of microbial community

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ABSTRACT

Cost-effective treatments of recalcitrant pollutants in wastewaters are required. The coupling degradation of *p*-nitrophenol (PNP) reduction in cathode and *p*-aminophenol (PAP, reduction product of PNP) oxidation in anode was studied in a bioelectrochemical system (BES) solely catalyzed by bacteria consortia, with no power input. In the cathode chamber, 50 mg L^{-1} PNP was reduced by 96.2 ± 2.4% within 96 h. PNP reduction efficiency was notably improved than that (63.8 ± 2.6%) in the abiotic cathode control. In the anode chamber, 20 mg L^{-1} PAP was removed by 94.0 ± 0.6% within 30 h. The reduction and oxidation peaks in cyclic voltammetry curves of the cathode and anode verified the coupled degradation process. Illumina Mi-seq sequencing revealed similar predominant bacteria with different percentages on the cathode and anode. The bacteria composition was more diverse on the anode. At the phylum level, higher prevalence of *Chlorobi, Bacteroidetes, Thermi* and *Actinobacteria* on the cathode than that on the anode were discovered. Meanwhile, *Thiobacillus*, *Methanomethylovorans*, *Sphingobium* and *Geobacter* were superior on the anode than the cathode at the genus level. Coupling treatment of PNP reduction in cathode and PAP oxidation in anode was realized in a bio-catalyzed BES. Enhanced degradation in a self-powered BES is an economical and efficient strategy for the treatment of nitroaromatic pollutants.

Keywords: Bioelectrochemical system (BES); Self-powered; *p*-nitrophenol (PNP); *p*-aminophenol (PAP); Microbial community

1. Introduction

Cost-effective treatment of toxic and refractory pollutants in wastewater is a tough problem nowadays. Conventional physical/chemical methods usually require high financial input, while biological methods often suffer from long-time operations [1]. In recent years, bioelectrochemical method is regarded as a promising strategy, because of its superiority in both higher efficiency and lower cost [2,3]. Bioelectrochemical systems (BESs) operated individually or assembly have the ability to degrade lots of pollutants [4–7], including nitroaromatics. Nitrophenol is a representative nitroaromatic pollutant which can be involved in bioelectrochemical process. Most studies on nitrophenol degradations in BESs focused on the cathodic reduction from nitrophenol (NP) to aminophenol (AP) [8]. In such a process, extra electron donors (e.g., acetate, glucose) must be provided at the anode to guarantee the reduction efficiency at the cathode [9]. Moreover, further treatments of the reduction products are still needed.

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However, if the reduction products of cathodes can be utilized as the electron donor of the anodes, a simultaneous treatment coupling cathodic reduction and anodic oxidation will be achieved [10]. BESs can be operated in modes of microbial electrolysis cell (MEC, with voltage input) and microbial fuel cell (MFC, without voltage input). Sun et al. reported a sequential reduction/oxidation of nitrobenzene with 2~5 V applied voltage [11]. Combining cathodic dechlorination and anodic mineralization of 4-chlorophenol had been studied in a BES with applied voltage of 0.5 V and photosynthetic bacteria [12]. In these studies, the pollutant degradations were mainly driven by the extra electric power input. However, in a MFC with both biocatalyzed cathode and anode, it is expected to realize the sequential pollutant degradation in a more cost-effective way. The cathodic reduction and anodic oxidation process could be accomplished in a self-powered style: solely catalyzed by bacteria on anode and cathode, with no need for extra power input. However, it is a challenge to degrade recalcitrant pollutants at the anode. Recently, several recalcitrant organic compounds were successfully oxidized in the anodes of MFCs, including pyridine, indole and aniline [13–17]. Liu et al. demonstrated the feasibility of coupling cathodic reduction of azobenzene with anodic oxidation of its reduction product, aniline, in a MFC with low output voltages (up to 8 mV) [18]. Cheng et al. reported the anodic degradation of aniline stimulated by oxygen in a MFC [19]. The AP (reduction product of NP) also holds great potential to be oxidized in the anode of MFCs, and provide electrons to the cathode reduction of NPs. Hence the simultaneous PNP reduction and PAP oxidation is realizable in a MFC. More importantly, the coupled degradation is a self-powered strategy of treating nitro-aromatic pollutants in BESs without any voltage input. The electron donor (PAP in the anode), electron acceptor (PNP in the cathode), and catalyzer during the cathode/anode reactions are all inside the system, avoiding extra power consumption. The coupled degradation process is catalyzed merely by the bacteria on the electrodes. Therefore, the microbial communities are very important for a deeper understanding of the self-powered degradation process. To the best of our knowledge, the community structure and key roles of the bacteria consortia in such systems are yet to be discovered.

In this study, coupling of cathodic PNP reduction and anodic PAP oxidation with no power input is explored in a BES. The degradation efficiencies of PNP in the cathode and PAP in the anode were studied with comparison of abiotic control. In addition, 16S rRNA gene based Illumina high-throughput sequencing was employed to investigate the consortia compositions and functions of microbes on both the cathode and anode.

2. Materials and methods

2.1. Reactor configurations

The rectangular MFC reactors were fabricated of poly-acrylic plastic. The working volumes of both the anode and cathode chambers of the MFCs were 200 mL. Plain carbon cloth (NOK H2315 T10A, Japan) was used as the electrode material of the anode and cathode. The electrodes were pretreated in 1 mol L^{-1} HCl and 1 mol L^{-1} NaOH. The anode and the cathode were connected externally through titanium wire and external resistances (240 Ω for startup, 100 Ω for operation). Proton exchange membrane (Nafion117, USA) was used to separate the anode and cathode chamber. The membranes were pretreated as described by Liu and Logan [20], and placed between the anode and cathode chambers. Surface areas of the anodes, cathodes, and proton exchange membranes were 50 cm^2.

2.2. Reactor inoculation and operation

Double-chambered MFCs operated in parallel were used in the study, and all the results were based on replicated experiment cycles. To compare the effect of cathodic reduction and anodic oxidation, three sets of treatments were conducted in the experiment. Treatment 1 was the coupled self-powered system, in which both the anode and cathode were inoculated. The schematic of coupling cathodic PNP reduction with anodic PAP oxidation is showed in Fig. 1. Treatment 2 with chemical cathode control aimed

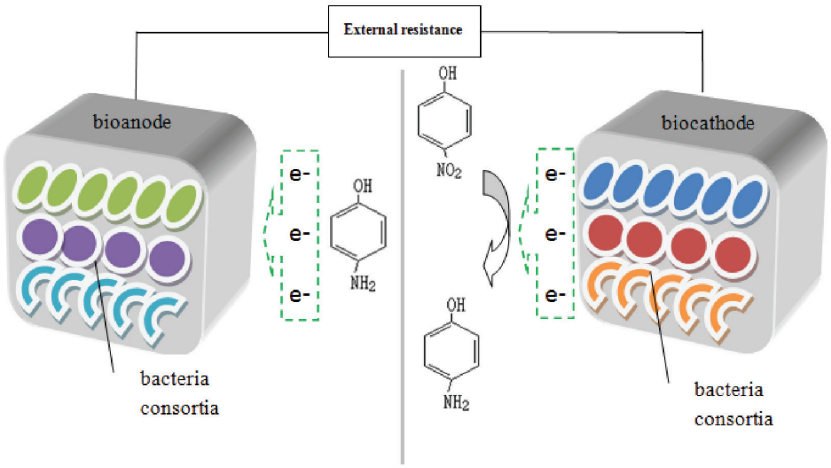

Fig. 1. Schematic of coupling cathodic PNP reduction with anodic PAP oxidation.

to compare the PNP reduction effect of biocathode and a biocathode. Treatment 3 with air-sparged cathode control was set to evaluate the anodic PAP oxidation efficiencies in the anodes. The anodes and biocathodes of the MFCs were inoculated with anaerobic sewage sludge collected from a pesticide manufacturing plant, Jiangsu, China. For each anode/biocathode, ca. 30 mL anaerobic sludge was added as the inoculums. The composition of electrolyte in the anode/cathode chamber was: NH_4Cl (310 mg L^{-1}), KCl (130 mg L^{-1}), $NaH_2PO_4{\cdot}2H_2O$ (5.6 g L^{-1}), $Na_2HPO_4{\cdot}12H_2O$ (6.07 g L^{-1}), and trace elements. The trace elements used was SL-4 (1 mL L^{-1}) solution. The composition of SL-4 was: EDTA (0.5 g L^{-1}), $FeSO_4{\cdot}7H_2O$ (0.2 g L^{-1}), and trace element stock solution SL-6 (100 mL L^{-1}). The stock solution SL-6 contained: $ZnSO_4{\cdot}7H_2O$ (0.1 g L^{-1}), $MnCl_2{\cdot}4H_2O$ (0.03 g L^{-1}), H_3BO_3 (0.3 g L^{-1}), $CoCl_2{\cdot}6H_2O$ (0.2 g L^{-1}), $CuCl_2{\cdot}2H_2O$ (0.01 g L^{-1}), $NiCl_2{\cdot}6H_2O$ (0.02 g L^{-1}), and $Na_2MoO_4{\cdot}2H_2O$ (0.03 g L^{-1}). PNP (50 mg L^{-1}) in the cathode chamber was the electron acceptor, and $NaHCO_3$ (0.84 g L^{-1}) was used as the inorganic carbon source. The pH value of the electrolytes was kept around 7.0. PAP (20 mg L^{-1}) was used as the electron donor and carbon source in the anode chambers. All MFC reactors were operated in batch mode. Electrolyte and substrate in the MFCs were replaced at the end of each treatment cycle (within 100 hr). The reactors were operated in water bath at controlled temperature of 30°C.

2.3. Chemical and electrochemical analysis

Both PNP and PAP were quantified using high performance liquid chromatography (HPLC) (LC-20AT, SHIMADZU, Japan) equipped with RP18 column (5 μm, 4.6 × 250 mm). The mobile phase was 40% methanol and 60% H_2O pumped at 1 mL min^{-1}. The detection was performed at 254 nm. Voltage (V) across the external resistance (RE) was measured using a multimeter (Victor, VC9804A+, China). Analysis of cyclic voltammetry (CV) was performed using electrochemical workstation (VMP3, BIO-LOGIC, France), equipped with three-electrode system. When CV curves of the cathode were plotting, the cathode and anode of the MFCs were used as the working and counter electrode, respectively. The saturated calomel electrode inserted in the cathode chamber was used as reference electrode. Reversely, anode was the working electrode, cathode was the counter electrode, and reference electrode was in the anode chamber, when CV curves of the anode were plotting. The potential range was –0.8 V ~ +0.5 V, and the scan rate was 10 mV s^{-1}.

2.4. Microbial community analysis

Pieces of bio-cathode and bio-anode were cut off with sterile knife to analyze the microbial community. After mixing with sterile saline solution, the treated samples were placed in sterile containers and vortexed for 1 min. The suspended biofilms were collected by centrifugation at 13,000 r min^{-1} for 10 min. Biofilms for 16S rRNA gene sequencing amplicon were processed as follows: DNA extraction with FastDNA™ Spin Kit for soil (MP Biomedicals, USA), 16S rRNA gene PCR amplification and PCR products purification. To amplify and sequence the V1~V2 hypervariable region of the 16S rRNA gene, forward primer (5′-AGAGTTTGATYMTGGCTCAG-3′) and reverse primer (5′-TGCTGCCTCCCGTAGGAGT-3′) were selected and different 8-bases barcodes and a Guanine were linked to the 5′ end of each primer. Then the purified products were sent for sequencing using Illumina Miseq sequencing platform (Illumina Inc., USA) at Beijing Genome Institution (BGI Shenzhen, China). The acquired data was processed by Sickle and Mothur program to remove the low quality sequences and reduce noises. The 16S rRNA gene sequences were classified into operational taxonomic units (OTUs) at 3% cutoff (or 97% similarity). The filtered sequences were assigned to a taxon by the stand-alone RDP classifier [21,22]. Sequences of the samples were submitted to NCBI SRA (Accession Numbers: PRJNA395124, SRX3051282, SRX3051285).

3. Results and discussion

3.1. PNP reduction and PAP formation in the cathodes

PNP served as the cathodic electron acceptor in the self-powered BES. It was also the initial reactant in the whole coupled reduction-oxidation degradation process. Fig. 2 shows an almost complete degradation (96.2 ± 2.4%) of the 50 mg L^{-1} PNP within 96 h in the biocatalyzed cathode (Treatment 1) during one operating cycle. In comparison, the degradation efficiency of PNP in the abiotic cathode control was 63.8 ± 2.6% (Treatment 2), which was much lower than that of the biocathode. The bio-catalyzed cathode also raised the PNP reduction rate from 7.9 ± 0.7 mg L^{-1} d^{-1} in the abiotic control to 12.0 ± 0.6 mg L^{-1} d^{-1}. The significantly promoted PNP reduction rate in the biocathode than in the abiotic control should be ascribed to the catalysis of the cathodic microbial consortia. Bacteria on the cathode could improve the PNP degradation by accelerating electron transfer and reducing the overpotential of electrode reactions [9,23].

It was worth noting that smaller improvements in the reduction efficiencies were showed between the biocathode and abiotic control in the first half than that in the second half of the whole reduction process (47.6 ± 3.2% vs. 32.8 ± 4.4% at 48 h, and 89.0 ± 2.8% vs. 56.4 ± 3.4% at 72 h). The results implied the important role of adsorption

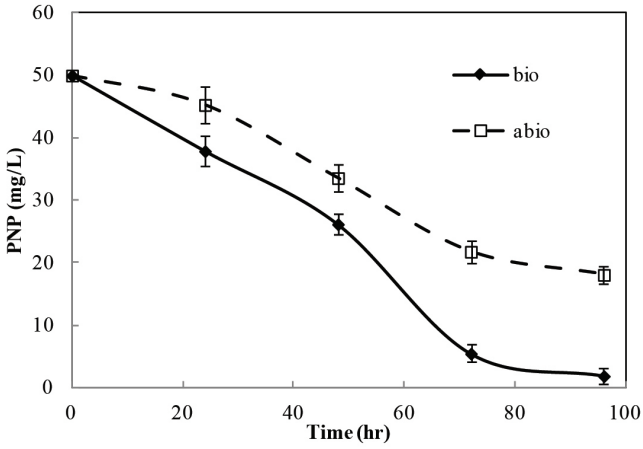

Fig. 2. PNP reduction in the cathodes.

effect at the beginning of the degradation process [11], no matter in the biocathode or the abiocathode. During the PNP degradation, the electrons from the cathode were more efficiently transferred to the absorbed PNP, with the catabolism of microbes on the biocathode. However, the biocatalysis was absent in the abiotic cathode control. Thus more efficient removal of PNP was achieved in the biocathode. So far, the first step in the coupled reduction-oxidation degradation process was realized in the system.

PNP in the cathodes was reduced to PAP, which is less toxic and easier to be mineralized than PNP. The conversion efficiencies of PNP to PAP were 38.5 ± 2.2% in the biotic cathode (Treatment 1) and 22.2 ± 1.4% in the abiotic control (Treatment 2). The higher PAP formation in the biocathode also suggested more efficient electron transfer facilitated by cathode bacteria. Accordingly, excessive intermediate products were avoided in the biocathode. The PAP formation efficiency in self-powered system (Treatment 1) was lower than that (49.4 ± 2.4%) in PNP reduction system with applied voltage [24], owing to the absence of extra electron provision from electric power input.

To study the PAP formation process in the cathodes, a prolonged (as to PNP reduction) cycle was tested (Fig. 3). There were two peaks of PAP concentrations at 48 h and 144 h within the test of 216 h in both the biotic cathode (Treatment 1) and abiotic control (Treatment 2). However, the PNPs in the cathodes were almost completely degraded in 96 h. More than one peak of PAP concentration could be attributed to the following reasons. Firstly, PAPs produced from PNP degradation are able to form different dimers, thus a decline of PAP concentration appeared after the first peak. Meanwhile, the dimers which are not stable could be reduced in the reducing environment at the cathode [25]. The further transformation of the dimers may result in the subsequent increase of PAP concentration after the valley in 96 hr. Secondly, there were usually intermediate products except for the end product during nitroaromatics reduction, especially in the abiotic control [26]. In a prolonged cycle showed in Fig. 4, it is probable that the intermediates were further transformed to PAP. Therefore, PAP concentration (8.7 ± 0.7 mg L^{-1}) at the second peak was higher than that (6.5 ± 0.7 mg L^{-1}) at the first peak in the abiotic cathode.

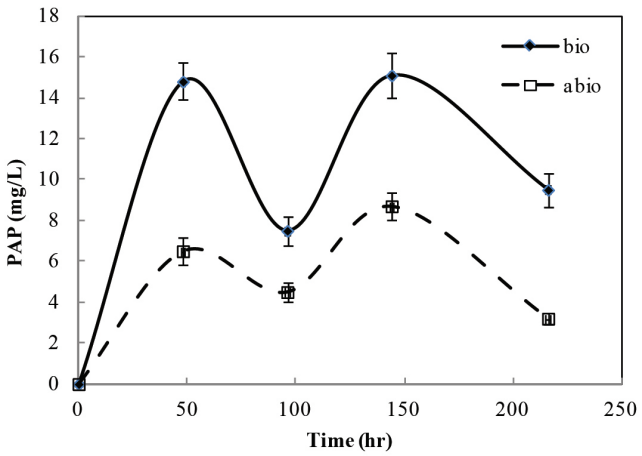

Fig. 3. PAP formation in the cathodes.

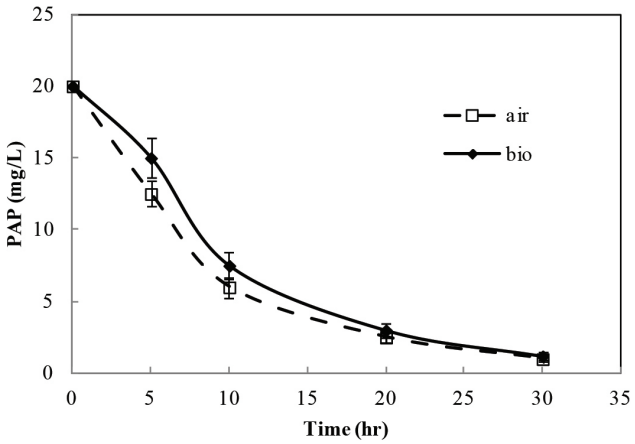

Fig. 4. PAP degradation in the anodes.

However, in the biocathode, the PAP concentrations of the two peaks (14.8 ± 0.9 mg L^{-1} vs. 15.1 ± 1.1 mg L^{-1}) were close to each other, owing to less intermediate products. Thirdly, PAP degradation is the source of electrons for PNP reduction in the system. In this study, there was a time lag between PNP reduction in the cathode (96 h) and PAP degradation in the anode (30 h) (section "PAP degradation in the anodes"). The time lag implied the further transformation of PAP and its degradation products [25]. The transformation probably continued the electron provision for PNP reduction after 30 h. The CV test in section "Electrochemical analysis of the electrodes" also suggested the multi-step transformation of PNP & PAP. Besides, adsorption to the electrodes contributed to the variations of PAP concentration as well. The difference in PAP formation between the biotic and abiotic cathode demonstrated the higher PNP reduction efficiency in the biocathode.

3.2. PAP degradation in the anodes

PAP was the objective reduction product of the PNP in the cathodes. PAP holds great potential to be oxidized for the two groups (–NH_2 and –OH) which could be oxidized in the molecular structure [27]. Thus, PAPs were utilized as the electron donor in the anodes of the BES. The electrons released from the PAP oxidation could be transferred to the cathode and consumed during the reduction of PNPs. Fig. 4 shows the PAP oxidation process in the anodes during an operating cycle. In the biocathode reactors (Treatment 1), 20 mg L^{-1} PAP was removed by 94.0 ± 0.6% within 30 h, showing almost equal removal efficiency with that in the air-cathode control (95 ± 0.4%) (Treatment 3). The oxidation rate of PAP in the anode reached 15.0 ± 0.5 mg L^{-1} d^{-1}, which is also comparable to that of the air-cathode control (15.2 ± 0.3 mg L^{-1} d^{-1}). The oxidation pathway of aminophenol in phosphate buffer solution was reported as follows: aminophenol could be firstly oxidized to quinonimine, and then *p*-benzoquinone [28]. PAP (reduced from PNP) in Fe^0–PM–PS and Fe/Cu bimetal systems were supposed to be oxidized to hydroquinone and *p*-benzoquinone, and then ring opening compounds (fumaric acid & maleic

acid), finally degraded into CO_2 and H_2O [29,30]. Formation of benzoquinone was detected during PAP degradation process. Meanwhile, TOC in the anode was reduced by 77.6 ± 0.9%, with increasing concentration of ammonia nitrogen during one cycle in this study. Thus the PAP oxidation process in this system is proposed to follow the similar pathway. The aim of this study is to demonstrate the feasibility of simultaneous anodic PAP oxidation with cathodic PNP reduction. The specific PAP oxidation process should be investigated in further studies.

Oxygen in the air-cathode control is an ideal electron acceptor in the cathode of BES [14]. Anodic substrate oxidation with electron release can be efficiently driven by oxygen reduction in the cathode [31]. As a result, electrons in PAPs are prone to be drawn to the anode via microbial metabolism, when powered by air-cathode. It is noteworthy that the comparable PAP degradation efficiencies were obtained in the biocathode and air-control in this study. The result demonstrated the likewise effective electron delivery from PAP to PNP in the bio-cathode. It revealed the high feasibility of coupled cathodic PNP reduction and anodic PAP oxidation in a BES. More importantly, reduction in biocathode is a low-cost way, in which the extra power consumption for air-sparge is not required. In addition, the relatively higher degradation rate of PAP in the anode (within 30 h) than PNP in the cathode (within 96 h) should be partly ascribed to the much lower toxicity of the aromatic amines than that of the corresponding nitroaromatics [1]. For a clearer understanding of the degradation process, further work could be focused on the degradation mechanisms, such as formation and transition of the intermediates.

3.3. Electrochemical analysis of the electrodes

Very low voltage outputs at only several mV (maximum 10 mV) were obtained during the coupling degradation process in this study. The result was comparable to the output voltages (maximum 8 mV) reported in the study of coupling cathodic reduction and anodic oxidation of azobenzene in an MFC [18]. The advantage of such a self-powered system is no electric energy consumption. Therefore, avoiding extra power inputs, but not obtaining high voltage output, is the pursuit of self-powered system.

For better understanding of the anodic/cathodic reactions and electron transfer process, cyclic voltammetry tests were employed to evaluate the electrochemical performance of the self-powered system. Cyclic voltammetry (CV) curves of the cathode and anode are shown in Fig. 5. There were two peaks of reduction current at –0.80 V and +0.08 V, respectively in the CV curve of the cathode (Fig. 5a), demonstrating the PNP reduction reactions in the cathode. The positions of the reduction peaks in this study were close to the result during nitroaromatics reduction [22,32]. In the positive sweep of bioanode, current increased with a minor peak at +0.04 V, and finally reached the plateau after +0.24 V (Fig. 5b). The anodic CV result was similar to the study on aniline oxidation in bioanode stimulated by oxygen [19]. It was the evidence for PAP oxidation in the anode chamber of the system. Higher currents in the cathode implied easier cathode reduction than anodic oxidation in this coupled

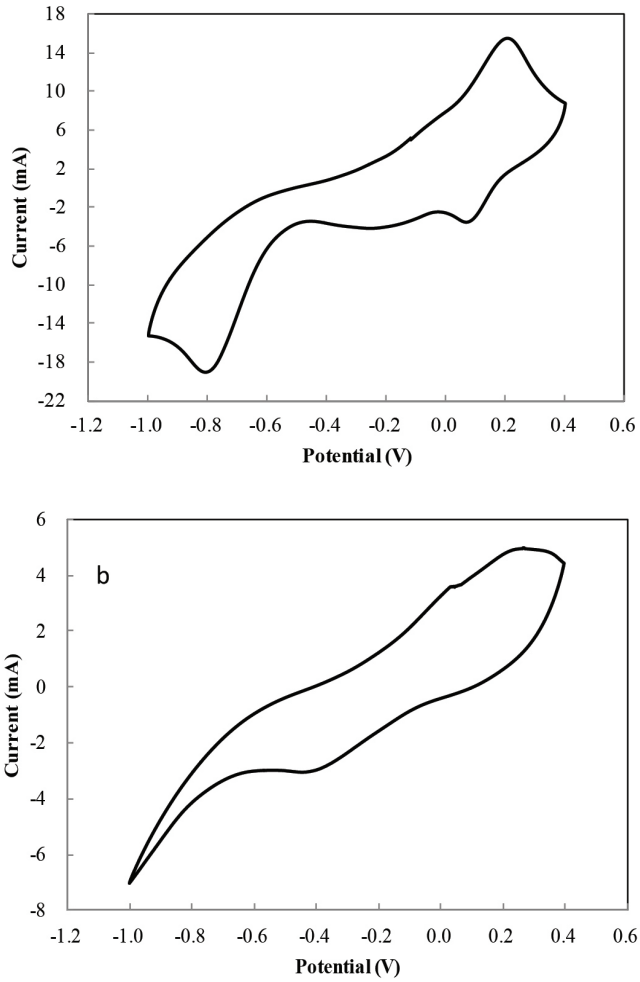

Fig. 5. Cyclic voltammetry analysis of cathode (a) and anode (b).

system [32]. As a result, the electrochemical analysis proved the realization of synergistic degradation of PNP in the cathode and PAP in the anode. However, it is a challenge to precisely interpret the voltammetric results because of the complexity of microbial and bioelectrochemical process [33]. Minor peaks showed on the curves might be the implication of further degradation of intermediates during the degradation process, which should be explored deeply in future work. Besides, optimization of operating parameters and electrode materials of the system could be further studied to improve the system efficiency [34].

3.4. Microbial community on the cathode and anode

The microbes on the electrodes play an important role in catalyzing the degradation process in the BES. Illumina Mi-seq sequencing was employed to investigate the microbial communities on the cathode/anode. For samples from the cathode and anode, 41724 and 41755 effective sequences respectively were selected and utilized to perform the bacterial diversity and abundance analysis. In total, 545 and 534 operational taxonomic units (OTUs) were obtained at 3% distance cutoff from the cathodic and

anodic samples, respectively. The species diversity indices (3% cutoff) of bacteria sample from the cathode and anode are listed in Table 1. The higher Shannon index (4.0 vs. 3.8), higher Chao index (563.3 vs. 557.6) and lower Simpson index (0.06 vs. 0.07) of the cathodic bacteria were showed. The observed species (Sobs) and species richness abundance-based coverage eastimator (Ace) were also consistent with this trend. The indices revealed higher bacterial population diversity on the cathode than that on the anode.

At the phylum level, the most abundant bacteria on the cathodes were: *Proteobacteria* (38.76%), *Chlorobi* (16.90%), *Bacteroidetes* (11.85%), *Chloroflexi* (6.62%), *Euryarchaeota* (6.48%), *Thermi* (5.74%), *Thermotogae* (2.30%), *Actinobacteria* (1.52%), *Gemmatimonadetes* (1.46%), *Planctomycetes* (1.40%), *Acidobacteria* (1.18%), *Firmicutes* (1.12%), Hyd24–12 (1.10%), and *Armatimonadetes* (1.04%). Meanwhile, the dominant bacteria communities on the anodes at the phylum level were: *Proteobacteria* (47.03%), *Chlorobi* (15.44%), *Bacteroidetes* (7.61%), *Chloroflexi* (7.61%), *Euryarchaeota* (7.22%), *Thermi* (3.55%), *Thermotogae* (2.35%), Hyd24-12 (1.31%), *Firmicutes* (1.29%), and *Actinobacteria* (1.22%) (Fig. 6a).

The phylum analysis indicated similar predominant bacterial phyla with different percentages on the cathode and anode. The similarity may be due to two reasons. On the one hand, both the anode and cathode were operated under anaerobic conditions. On the other hand, PAP was present at both electrodes, acting as the PNP reduction product on the cathode and the reactant on the anode. The dominant microbial phyla discovered in this system are also popular in other BESs. Most of the main bacterial phyla in this study were also reported by Cheng et al. in MFCs treating aniline [19]. Cui et al. found the enrichment of *Proteobacteria, Bacteroidetes* and *Firmicutes* in an up-flow BES treating azo dyes [35]. In the self-powered system, the absolute majority of the bacterial phyla belonged to *Proteobacteria*, which accounted for 1/3~1/2 of the total bacterial community on both the anode and cathode. *Proteobacteria* was reported as the dominant phylum in nitrifying microbial community in activated sludge [36] and municipal wastewater treatment plant [37]. It was also dominated in an UASB & BES coupling system for nitro reduction [7]. Other major bacterial phyla included *Chlorobi*, *Bacteroidetes*, and *Chloroflexi*. Wan et al. found *Proteobacteria*, *Chloroflexi* and *Chorobi* presented as primary phyla in their study of simultaneous bio-autotrophic reduction of nitrate and perchlorate [38]. Dominance of *Chlorobi*, *Bacteroidetes*, and *Chloroflexi* were also reported in anaerobic reactors [16]. *Bacteroidetes,* which showed abundance in both anode and cathode of this system, were often reported in BESs [35,39].

At the genus level, the dominant populations on the cathodes were: *Thiobacillus* (12.89%), *Methanomethylovorans* (6.41%), B-42 (5.74%), *Sphingobium* (3.30%), *Kosmotoga* (2.28%), *Rhodanobacter* (1.43%), *Fluviicola* (1.35%), *Geobacter* (1.09%), and *Paracoccus* (1.05%). At the same time, the major communities on the anodes at the genus level were: *Thiobacillus* (18.38%), *Methanomethylovorans* (7.12%), *Sphingobium* (7.04%), B-42 (3.55%), *Kosmotoga* (2.35%), *Geobacter* (1.54%), and *Pandoraea* (0.98%) (Fig. 6b).

Table 1
Species diversity indices (3% cutoff) of bacteria on cathode and anode

Sample	Sobs	Chao	Ace	Shannon	Simpson
Cathode	545	563.3	576.8	4.0	0.06
Anode	534	557.6	570.6	3.8	0.07

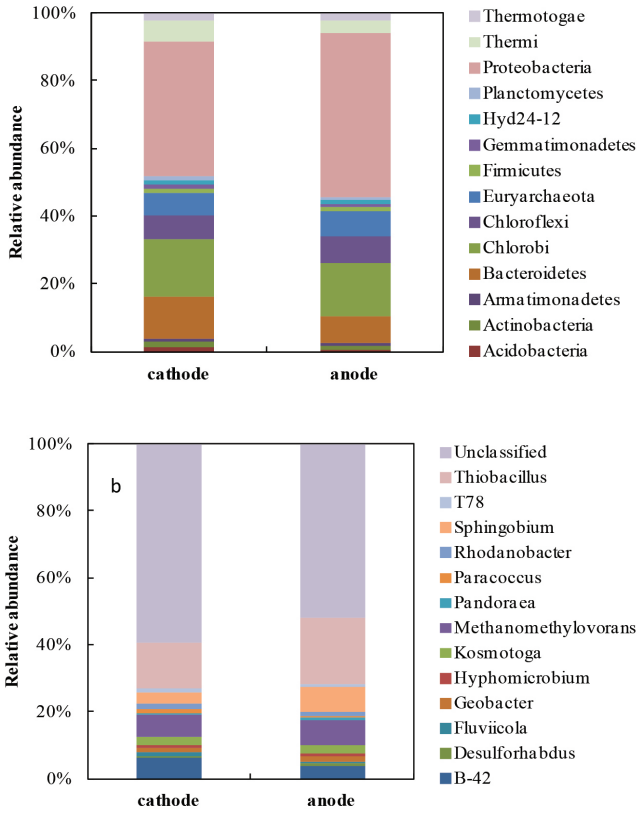

Fig. 6. Composition of bacteria consortia on cathode and anode: a) Phylum level, b) Genus level.

The genus analysis provides a deeper understanding of microbial community structure on the electrodes. The predominant five genera were *Thiobacillus*, *Methanomethylovorans*, B-42, *Sphingobium*, and *Kosmotoga*. They account for 28.43% of the cathodic bacteria and 40.63% of the anodic bacteria, respectively. The result also indicated that the bacteria consortia on the cathode were more diverse than the anode. It is accordance with the result of species diversity indices. *Thiobacillus* (belonging to β-*proteobacteria*) is a well-known autotrophic denitrifying bacterium with metabolism type of obligate autotrophy and facultative anaerobic [40]. It usually appeared in different denitrifying processes, using nitrate as the electron acceptor to achieve denitrification or accelerating nitrate reduction [41,41]. Its dominating existence in this study could be related to the nitro degradation process. The genus *Methanomethylovorans* could utilize methylated amines for methanogenesis [43]. It was also reported to be dominant in the anaerobic reactors such as UASB and anaerobic tank [44,45]. *Methanomethylovorans* presented on the electrodes in this study could be mainly involved in the degradation of PAP. *Sphingobium* is a well-known hydrocarbon degrader [46]. It was reported to double the removal rate

of bisphenol A during bioaugmented treatment of trace organic contaminants in wastewater treatment plants [47]. *Sphingobium* on the electrodes probably improved the degradation of the phenolic compounds at the cathode and anode in this system. *Kosmotoga* belonging to *Thermotogae* phylum was a recently proposed genus which was first isolated from oil production fluid [48]. Ye et al. found the extremely high abundance of *Kosmotoga* in different sections of a municipal wastewater treatment plant [37]. Enrichment of *Kosmotoga* in the anode of a microbial electrochemical cell was reported, and its abilities of degrading complex substances and maintaining high current density were demonstrated [49]. In this study, the percentage of *Kosmotoga* was also higher in the anode than that in the cathode. Thus the roles of *Kosmotoga* in this system should mainly focus on facilitating the electron delivery in PAP oxidation on the anode.

Apart from the common genera, there were also exclusive genera of the cathode or anode. The genera *Paracoccus* and *Fluviicola* represented more than 1% of the anodic genera but rarely appeared on the cathode in this system. Meanwhile, percentages of *Rhodanobacter* and *Geobacter* on the anode were 50% higher than that on the cathode. Reversely, *Pandoraea* was more than 2 times abundant on the cathode than the anode. *Paracoccus* and *Rhodanobacter* are co-existing denitrifying bacteria which were also reported in other studies [50,51]. *Rhodanobacter* also possesses the ability to degrade the metabolic products of aromatic hydrocarbons [52]. Its existence on the anode implied the occurrence of mineralization and denitrification during PAP degradation in the anode of this system. *Geobacter* is a typical anode-respiring bacteria facilitating electricity generation in microbial fuel cells [53]. The higher abundance of *Geobacter* on the anode demonstrated its potential role in promoting electron delivery of in this self-powered system. *Pandoraea* was reported to be capable of degrading *p*-xylene and products of 2,4,5-trichlorophenol [54,55]. It was also found in sediment microbial fuel cells [56]. Its presence on the cathode could be involved in the enhancement of PNP reduction efficiency. The bacteria consortia colonized in the self-powered system could cooperate with each other and finally facilitate the coupled PNP degradation process. In future work, the precise microbial function and function genes could be deeply investigated with combined metagenomic and metatranscriptomic method.

4. Conclusions

Enhanced degradation of *p*-nitrophenol (PNP) was realized by coupling the PNP reduction at the cathode and *p*-aminophenol (PAP, the reduction product of PNP) oxidation at the anode, in a self-powered BES. The cyclic voltammetry analysis of the cathode and anode testified the coupled reduction and oxidation process. The bacteria consortia on the cathode and anode could catalyze and improve the degradation process. This study demonstrated high feasibility for self-powered degradations of nitro aromatics through sequential cathode reduction and anode oxidation in BESs in the future. Solely biocatalyzed degradation of organic pollutant in a BES is a low-cost and efficient way for treatment of wastewater containing nitroaromatics.

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