



Decarbonization and denitrification characteristics of a coupling ABR-MFC-MEC process treating black water

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ABSTRACT

Untreated black water is harmful to the environment due to its contents of large amounts of organic compounds, ammonia ($\text{NH}_4^+\text{-N}$) and pathogenic bacterium. The combined ABR-MFC-MEC (anaerobic baffle reactor-microbial fuel cell-microbial electrolysis cell) process was developed in this study to treat black water without external input of energy or oxygen. The degradation features of carbonaceous matter and nitrogen pollutants in black water were investigated using the combined ABR-MFC-MEC process in this study. Chemical oxygen demand (COD) and $\text{NH}_4^+\text{-N}$ removal rates of the study process were over 92% and 50%, respectively, when feeding an influent (black water) with COD concentration of $1,500 \pm 100 \text{ mg L}^{-1}$ and $\text{NH}_4^+\text{-N}$ concentration of $60 \pm 5 \text{ mg L}^{-1}$, while maintaining the whole process hydraulic retention time as 24 h. Proteobacteria, Firmicutes and Acidobacteria were found to be the dominant electrogenesis flora supporting decarbonization and denitrification in the combined ABR-MFC-MEC process.

Keywords: Black water; Decarbonization; Denitrification; Microbial fuel cell; Microbial electrolysis cell

1. Introduction

Black water contains feces and urine, which requires integrated treatment before reused [1,2]. The energy content of feces is around 24.3 kJ g^{-1} dry mass with a moisture content between 65% and 85% [3]; while the urine accounts for merely 1% of the waste water volume, but brings about 75% of the nitrogen load [4,5]. Averagely the urine contains 1 g L^{-1} phosphorus, 9 g L^{-1} nitrogen and 10 g L^{-1} chemical oxygen demand (COD) [6]. Incomplete treatment of black water could lead to the accumulation of nitrogen, phosphorus and other nutrients in water bodies, causing eutrophication and/or other types of secondary pollution [4].

Microbial fuel cells (MFCs) had been used to treat wastewater and produce electricity from different

compounds simultaneously [7]; microbial electrolysis cells (MECs) could achieve sustainable hydrogen production from various types of biomass and promote transport and transformation of pollutants with the stimulation of electric current on bacterial growth [8]. Single-chamber MFCs where nitrifiers are pre-enriched at the air cathodes have been previously demonstrated as a passive strategy, for which nitrogen removal could be integrated into the current-generating bio-electrochemical systems (BES) [9]. Current generation was also investigated using non-membrane single chamber MFCs treating urine [10]. The electrical energy required for the total ammonia ($\text{NH}_4^+\text{-N}$) recovery was $4.9 \pm 1.0 \text{ MJ kg-N}^{-1}$, which was insufficient to complete the electrochemical nitrogen removal/recovery processes; in this case, MFC and MEC can be coupled using the power produced from MFCs to drive MECs [11].

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External power supply was omitted in the MFC-MEC coupled system since the electric energy generated from the MFC unit was provided to the MEC unit simultaneously, which can save electricity storage demand and diminish the power loss. However, the combination strategy had no obvious effect on performances of some treatment practices due to high concentrations and/or complex ingredients of the influent wastewater (e.g., black water), which hinders wide application of the coupled process [12]. Anaerobic baffle reactor (ABR) can be considered as a series of up-flow anaerobic sludge bed reactors that still play a significant role within many wastewater treatment plants [13]. Some benefits of the ABR process are the simplicity of the reactor design, long sludge retention time with low hydraulic retention time (HRT) and tolerable to shock load [14]. Nevertheless, fermentative acidogens usually grow faster than methanogens while the methanogens are sensitive to environmental conditions such as pH and hydrogen partial pressure in ABR [15]. This may result in the accumulation of volatile fatty acid (VFA) and consequently inhibit the methanogenesis process. Thus, it is desired to develop a novel anaerobic process that could degrade organic matters more efficiently.

The coupling of MFCs and anaerobic treatment units indicated that the methane production of anaerobic treatment units could be enhanced by MFCs [9,10]; in this case $92.0\% \pm 2.5\%$ of $\text{NH}_4^+\text{-N}$ could be removed from the urine [16]. The ABR coupled with MEC has also been operated to strengthen the hydrogen production [17]. Alternatively some intermediate metabolite of the ABR unit such as VFAs can be used in the electrolysis process to produce methane [18], for example, researchers have confirmed the production of methane in single chamber MECs [17]. Therefore, the coupling of ABR and MFC-MEC units is of great value for black water treatment.

To the best of our knowledge, so far there are very few published works on the coupled ABR-MFC-MEC process treating black water with high concentration of organic wastewater. Our previous study has demonstrated that the coupled ABR-MFC-MEC process is very effective in terms of COD and $\text{NH}_4^+\text{-N}$ removal from fecal wastewater treatment; however, the mechanisms were not well understood [19]. The coupled MFC-MEC-ABR system has multiple benefits. First, the sludge residence time of the combined process was extended comparing with single units, which can reduce sludge production. Nitrification-denitrification could also be possible with the combination of MFC and MEC units when using micro-aeration to improve quality of the effluent [20]. Moreover, the balance of internal energy supply demand can be realized using the power produced from MFCs to support the operation of MECs. This study further investigated performances, characteristics and degradation mechanism of the coupling black water treatment process; dominant electrogenesis flora performing decarbonization and denitrification in the process was also studied.

2. Materials and methods

2.1. Set-up of experiments

A four-chamber ABR, a double-chamber MFC and a double-chamber MEC were started up separately until the

steady states achieved. Afterwards the ABR-MFC-MEC system is combined. The dynamic experiments were carried out with a continuous wastewater input mode. The size of the main compartment is $640 \text{ mm} \times 180 \text{ mm} \times 250 \text{ mm}$ with an effective volume of 28 L. Effective volume of the MFC chamber equals to the MEC chamber, that is, 9.6 L. The reactor was kept at room temperature during the whole process. HRT was 24 h in the study, which could be optimized with further researches; the micro-aeration was 500 mL min^{-1} in the cathode chamber of the MFC unit forming an air rich cathode. The detailed configuration of each compartment and other operational conditions such as micro-aeration rates can refer to our previous study [19]. The diagram of the experimental set up is demonstrated in Fig. S1.

2.2. Experimental operation

The inoculated activated sludge was collected from the Dongqu sewage treatment plant, Shanghai, China, and was dosed into each cell compartment of the ABR-MFC-MEC system with the volatile suspended solids (VSS) concentration of $4,330.0 \pm 410.0 \text{ mg L}^{-1}$ [21]. Prior to inoculation, the activated sludge was acclimated for 1 month under an anaerobic condition with the feed solutions containing (per liter of distilled water): NH_4Cl 0.275 g, CH_3COONa 1.60 g, $\text{C}_6\text{H}_{12}\text{O}_6$ 0.7 g, KH_2PO_4 0.01 g, $\text{MgSO}_4 \cdot 2\text{H}_2\text{O}$ 0.01 g, NaHCO_3 0.1 g and 1 mL L^{-1} trace elements solution. The trace elements composition used in this study were (mg L^{-1}): FeCl_3 0.900, $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$ 0.120, $\text{Na}_2\text{MoSO}_4 \cdot 2\text{H}_2\text{O}$ 0.060, $\text{MnCl}_2 \cdot 4\text{H}_2\text{O}$ 0.060, KI 0.180, $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ 0.03, $\text{CaCl}_2 \cdot 6\text{H}_2\text{O}$ 0.150 and H_3BO_3 0.150. The wastewater composition was artificially made according to actual quality of the real black water during the operation period [19]; the influent concentration of COD and $\text{NH}_4^+\text{-N}$ was $1,500 \pm 100 \text{ mg L}^{-1}$ and $60 \pm 5 \text{ mg L}^{-1}$, respectively.

The ABR started with a low concentration load with the influent COD kept at $0.20 \text{ kg m}^{-3} \text{ d}^{-1}$ [15]. When the COD removal rate reached over 50%, the COD load was gradually increased until it reached to $1.6 \text{ kg m}^{-3} \text{ d}^{-1}$. When the COD removal rate could be maintained over 81.2%, the start-up of ABR process was completed. The MFC unit was started with a stable COD value as $1,200 \text{ mg L}^{-1}$. When the COD and $\text{NH}_4^+\text{-N}$ removal rates and electricity production were kept relatively constant, the MFC was considered to be started successfully. For start-up of the MEC unit, a DC electronic power supply (IT8800 series) of 0.10–0.70 V was used [22]. When the direct voltage was 0.50 V, the degradation rate of COD was 86.4%, and the increase of the voltage would affect the removal efficiency of COD; a diode was connected between two electrodes to control the direction of current from power supplier MFC to electricity user MEC. When individual units were successfully started up, all compartments were connected to constitute the ABR-MFC-MEC system, which were operated for 6 months to investigate performance of the whole process together with intermediates. Sludge samples were collected at the end of experiment to characterize the microbial community.

2.3. Analytical methodology

2.3.1. Chemical analysis

Water samples were collected from influent and effluent from each chamber daily. All the samples were collected

in triplicate. COD, $\text{NH}_4^+\text{-N}$, nitrate ($\text{NO}_3\text{-N}$), nitrite ($\text{NO}_2\text{-N}$) and total nitrogen (TN) of the samples were analyzed with standard methods [23]. In order to further analyze denitrification performance of the process, samples from sections I–IV which represent the inlet, the connection points between the ABR2 and MFCa, ABR3 and MECc, and the outlet, respectively, were analyzed by opening the connections points (Fig. S1). Intermediates were analyzed using a gas chromatograph mass spectrometer (GC–MS). A DB-5 fused silica capillary column (30 m \times 0.25 mm \times 0.25 mm) was used with helium as the carrier gas (1 mL min^{-1}). Column temperature was initially set at 40°C for 4 min and gradually increased to 230°C min^{-1} . Samples (0.2 μL) were injected automatically in a splitless mode. Injector and detector temperatures were set at 230°C and 280°C, respectively. For identification purposes, the mass spectrum of each peak was recorded in the total ion current mode of the mass spectrometer, within the m/z range of 50–500.

2.3.2. Sludge characterized by XRD

The sludge samples used in this study were collected in six points (Fig. S1: A1, A4, E, F, Ea and Fa). The mixed liquor suspended solids were determined by drying the sludge samples in an oven at 105°C for 24 h to a constant weight. The crystalline structures and compositions of the sludge samples were measured by powder X-ray diffraction (XRD) patterns on a Bruker D8 Advance X-ray diffractometer (Bruker AXS, Germany) using a Cu $K\alpha$ radiation source ($\lambda = 1.5406 \text{ \AA}$).

2.3.3. Electrode material characterized by SEM

The collected electrode material samples were observed using a scanning electron microscope (SEM) (ZEISS Merlin, Germany) equipped with an energy-dispersive X-ray spectrometer to identify changes on electrode material before and after the reaction.

2.3.4. Microbial community analysis by high-throughput pyrosequencing

To study the spatial distribution of bacteria in the selected reaction chamber, the suspended sludge and anode graphite felt samples were collected. The total DNA of all samples were extracted using the PowerSoil™ DNA isolation kit (Mo Bio Laboratories Inc., CA). Labels of “A1”, “A4”, “E”, “F” stand for the suspended sludge samples taken from ABR1, ABR4, MEC, MFC, respectively; while “Ea” and “Fa” stand for biofilm samples taken from the anode of double MEC and double MFC, respectively. The concentration of DNA samples was analyzed with a UV–Vis spectrophotometer (Nanodrop 2000, Thermo Scientific, USA). DNA extraction of each sample had been conducted in triplicate, and then the combined DNA solution was stored for the following analysis.

The microbial community of samples was analyzed by using the high-throughput pyrosequencing on an Illumina platform (Illumina MiSeq PE 300). Amplicon libraries were constructed for pyrosequencing using bacterial primers 338F (5'-ACTCCTACGGGAGGCAGCAG-3') and 806R (5'-GGA CTA CHV GGG TWT CTA AT-3') for the relevant region of the microbial 16S rRNA gene, which were selected as the sequencing primer set to obtain bacterial information simultaneously.

The raw pyrosequencing data were deposited to the NCBI Sequence Read Archive database (PRJNA305812).

3. Results and discussion

3.1. Performance of the combined ABR-MFC-MEC process

As shown in Figs. 1 and 2, when the influent concentration of COD and $\text{NH}_4^+\text{-N}$ was maintained at $1,500 \pm 100 \text{ mg L}^{-1}$ and $60 \pm 5 \text{ mg L}^{-1}$, respectively, and the HRT was 24 h, the removal rate of COD increased from 92% to 98% over time, with its effluent concentration kept below 75 mg L^{-1} . Besides the anaerobic process that consumes COD, the COD content in black water entering MFCs and MECs could also be used as substrate and/or electronic donor. The degradation of COD reached to a maximum limit in the long run cycle with the removal rate between 92%–98%. Performance of the combined process was superior to the individual ABR, MFC and MEC units in terms of COD removal. The $\text{NH}_4^+\text{-N}$ removal rate was only 5%–10% during the start-up period and slowly increased to over 50% in the long run (Fig. 2). The degradation of $\text{NH}_4^+\text{-N}$ mainly attributed to microbial metabolism during the start-up period; while in the stable period, biochemistry

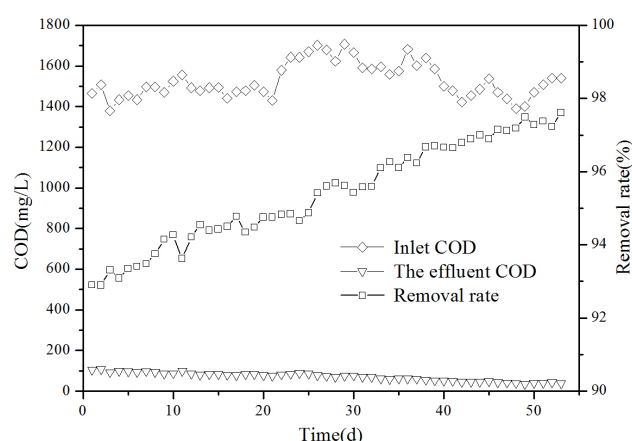


Fig. 1. COD removal profile of the combined process.

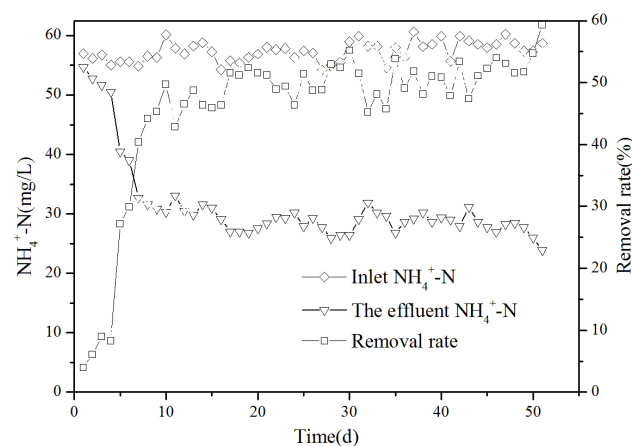


Fig. 2. $\text{NH}_4^+\text{-N}$ removal profile of the combined process.

and electrochemistry had synergistic effect on the removal of $\text{NH}_4^+\text{-N}$ with the action of microbiological weak electricity. Li et al. [24] suggested that the BES had an advantage in the low current electric field due to current electron transfer involved in the microbial metabolism that could enhance biological activity. MEC was used to remove ammonia nitrogen and MFC to consume organic matter [25]; the high potential of anode in the MEC unit could prompt nitrification rate, even with limited dissolved oxygen.

Nitrogen (N) removal is more challenging than COD by the combined process and thus should be put in the priority. The difference of COD and $\text{NH}_4^+\text{-N}$ removal rates was determined by the types and content of microorganisms in the system [26]. In the later stages, the removal rate of $\text{NH}_4^+\text{-N}$ was obviously improved, which may relate to the enrichment of electricity producing and denitrifying bacteria on electrodes through the analysis of microbial community. To conduct denitrification analysis further, it is necessary to carry out nitrogen distribution study of the whole process. The direction of wastewater flow can be changed by opening or closing the valve in the middle, where the water quality at cross-sections of compartments could indicate the tendency of N transformation. Sections I–IV represent the inlet, the connection point of ABR2 and MFCa, the connection point of ABR3 and MECc, and the outlet, respectively, in Fig. 3.

TN and $\text{NH}_4^+\text{-N}$ decreased to different extent in section II. The occurrence of nitrate and nitrite was observed in sections III and IV together with removal of TN and $\text{NH}_4^+\text{-N}$, whereas the contents of nitrate and nitrite were almost close to zero in section II. Nitrate and nitrite in section IV mainly come from the oxidation and transformation of $\text{NH}_4^+\text{-N}$ in the join point of ABR3 and MECc. COD degradation mainly occurred near the MFC anode area of section II while denitrification mainly occurred near the MEC cathode of section III, which led to the accumulation of intermediates, that is, nitrate and nitrite during the nitrification-denitrification process. The degradation rates of COD and $\text{NH}_4^+\text{-N}$ were improved by the coupling of anaerobic degradation, MFCs, MECs and the stimulation with low voltage electricity [8,27]. The BES had high

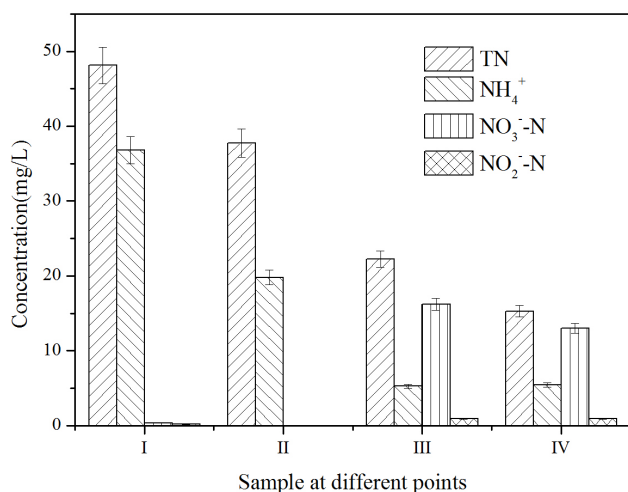


Fig. 3. Distribution of N species among different treatment chambers.

removal efficiencies of organic matters with a low-current electric field; while a high-current electric field destroyed the microbes [24]. Therefore, it will be necessary to apply appropriate weak current to the system for improving the removal of $\text{NH}_4^+\text{-N}$ in future studies. The coupled ABR-MFC-MEC system could be combined with the anaerobic ammonium oxidation (ANAMMOX) technology due to its low cost and high efficiency in nitrogen-contained wastewater treatment.

3.2. Characteristics of bio-sludge, electrodes and effluent

The sludge from different points of the electrodes and the compartments was sampled for characteristic analysis. The XRD graphics of the dried sludge taken from different compartments were shown in Fig. 4(A). As seen from the picture, silicon and silicon dioxide were found in the sludge samples of six different location points, which were the common composition among sludge samples [28]. Electrode materials played important roles as supporting body of microorganisms and electron conductor. The structures of original carbon felt electrodes had been determined by SEM in Fig. 4(B) while Fig. 4(C) showed the images of electrodes in different compartments after operation. Fig. 4(B) demonstrated that the electrode material was smooth and uniform, whereas relatively rough particles were accumulated on the electrode at different locations, the original stratified structure is interrupted and fragmented in Fig. 4(C). The presence of large quantities of particulate matter would block the mass transfer process gradually and have a significant impact on the power production. This is in consistent with the observations of gradually decreased performance of ABR-MFC-MEC, in terms of pollutants' removal efficiencies and power production.

Intermediate product of the coupled process is presented in Fig. 5. The common intermediate products could be classified into oleamide and palmitoylethanolamide. These intermediate products were very low toxic or even non-toxic to the environment and can be further degraded by microorganisms effectively [29]. Rhamnolipid produced by *Pseudomonas aeruginosa* ATCC 9027 enhanced the bio-availability of hexadecane and thus benefited the bacterium growth with hydrophobic substrates [30]. The appearance of intermediates at the beginning of the anaerobic reactors was more complex; similarly, the intermediate in MFC was far more complex than in the single chamber MEC system. The MEC unit could be stimulated with low voltage electric field and transformed to intermediates. The anaerobic degradation of organic matter and electro-chemical process caused the accumulation of intermediate product to some extent. In addition, GC-MS analysis implied that intermediate products changed from complexity to simplicity from ABR to MFCc. These compounds transportation across microbial cell barriers was enhanced by the hydrocarbons entering a more aqueous phase and acting as the carbon source for microorganisms [30,31].

3.3. Microbial structural distribution

High-throughput pyrosequencing was used to characterize the biomass from different chambers and electrodes of the combined process when the stable performance was reached. Detailed information as about the sampling

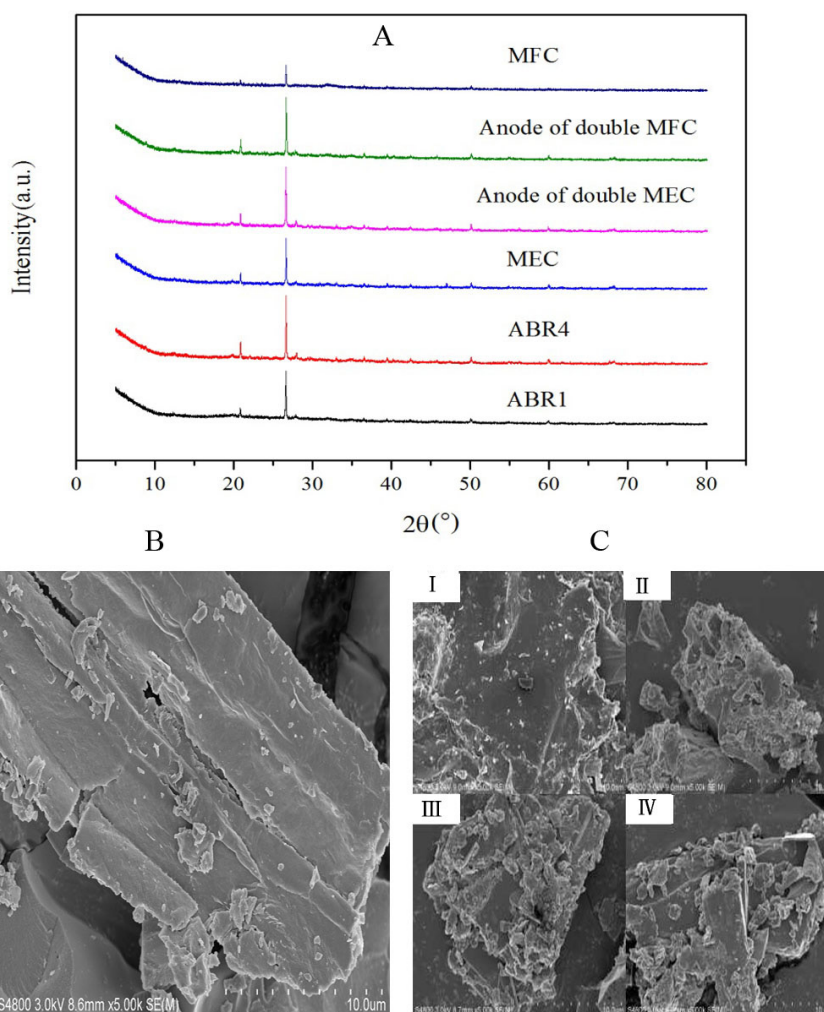


Fig. 4. Characteristics of the carbon felt material. (A) XRD graphics of the dried sludge taken from different compartments; (B) SEM of the original carbon felt material and (C) SEM figure of the carbon felt material after cultivation (I: MFC; II: MEC; III: MECa; IV: MFCa).

positions and sequences were shown in Table S1. Points of longer total genomic DNA fragment had been selected to indicate whether there was certain tailing phenomenon in the selected target, and to determine if the sample DNA had a small amount of pollution of genome in the selected microorganism. The microbial community structures of samples at the phylum, family and genus levels are shown in Fig. 6.

The bacterial community abundance was identified at the phylum level in Fig. 6(a). Proteobacteria and Firmicutes and Acidobacteria were found to be the high efficient electrogenesis flora obtained in the reactor [32]. Proteobacteria, Firmicutes, Chloroflexi, Bacteroidetes, Chlorobi, Acidobacteria and Fusobacteria were the most common observed stereotypes of microorganism in the system, accounting for more than 80% of the total microbial population. The most abundant classes within the Proteobacteria phylum are β -Proteobacteria, similar results have been reported elsewhere [33,34]. Proteobacteria, Chloroflexi, Bacteroidetes and Firmicutes were found to be the dominant bacteria in ABR1, accounting for 23.4%, 19.5%, 12.1% and 9.6% of the compartmental population, respectively. Other types of non-dominant bacteria appeared in the cultivation

and domestication period but decreased with the operation time. The content of Proteobacteria and Firmicutes has increased to 34.3% and 30.4%, respectively, in ABR4, indicating that the ABR configuration had an optimal screening effect on the target bacteria. Gulhane et al. [35] found that anaerobic baffled reactor provided a selective environment for the microbial community and their respective metabolic activities.

In single chamber MEC and MFC systems, Proteobacteria was still the most abundant species accounting for 39.8% and 27.1% of the compartmental population, respectively. Similarly, Proteobacteria accounted for 47.4% of the compartmental population in the double chamber MFC. The results were in good agreement with a study that showed the highest abundance of Proteobacteria in both the active sludge community and the electrode community [36]. Bacteroidetes and Chloroflexi were the second abundant microorganisms found in the single chamber MEC process that accounted for 16.5% and 15.6% of the total population, respectively; while in the single chamber MFC system, Chloroflexi and Bacteroidetes accounted for 21.7% and 10.4% of the compartmental population, respectively. Chloroflexi was a recently

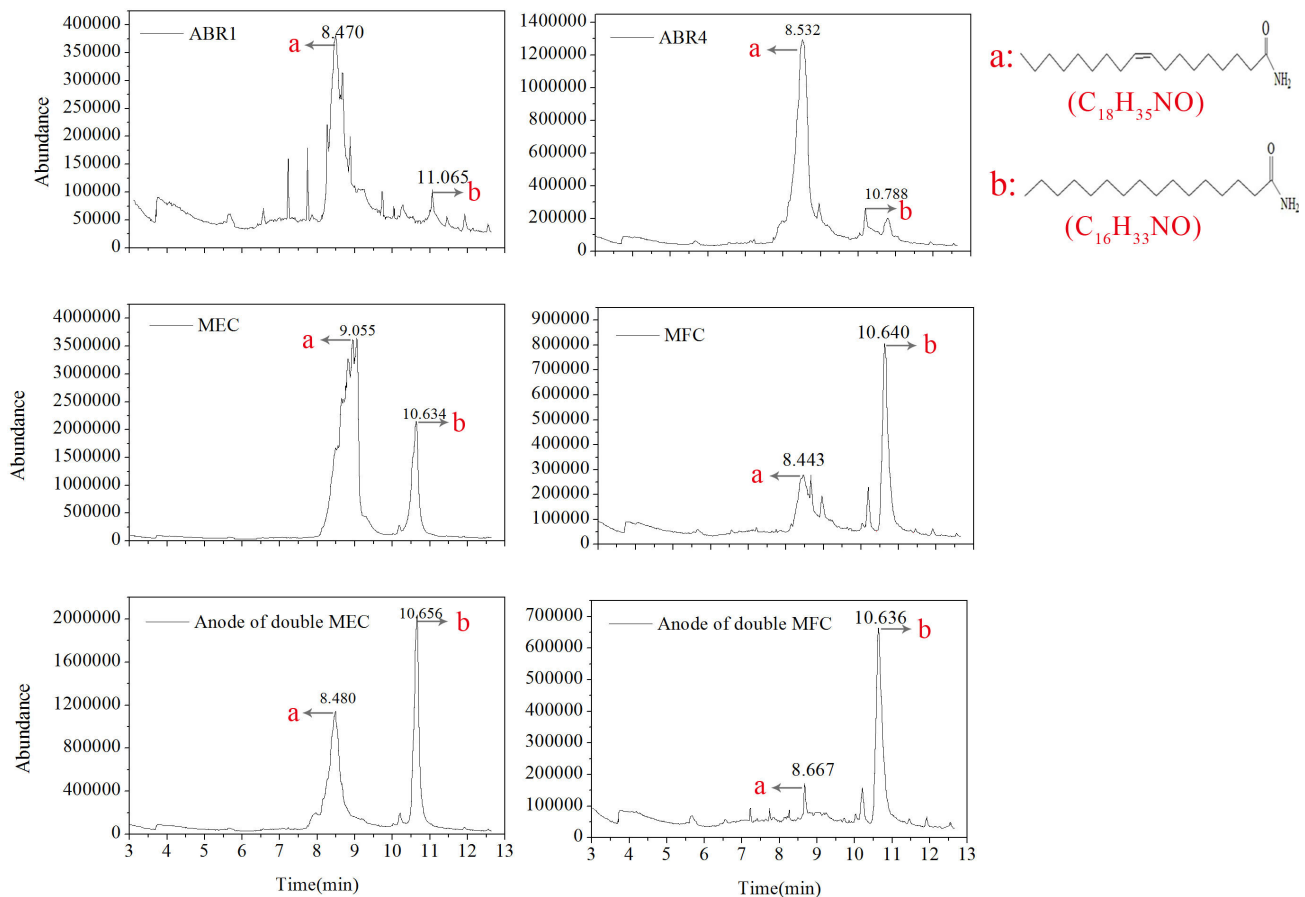


Fig. 5. Intermediate products during the coupled process.

classified phylum that was enriched in MFC reactors using cellulose as the substrate [37]. The abundance of Chloroflexi was lower than that of Proteobacteria in this study.

The bacterial community abundance was identified at the family level as shown in Fig. 6(b). The microbial community included nitrifiers and denitrifiers. The nitrifiers consisted of Nitrosomonadaceae, Bradyrhizobiaceae, Microbacteriaceae and Mycobacteriaceae [34]. Although the quantities of these microorganisms were low, the effect of decarbonization and denitrification in the coupling ABR-MFC-MEC process was obvious. It was reported that ammonium removal was limited to 7%–12% under anaerobic conditions in a MEC system, while micro-aerobic conditions increased the removal efficiency to 31% [12]. However, the micro-aerobic environment was unfavorable for the denitrifying bacteria, which explains why these special bacteria were low in content. Dissolved oxygen levels need to be strictly controlled in order to create micro-aerobic conditions that enhance the nitrification process without impeding the anodophilic and denitrification activities [12]. There were more nitrifying and denitrifying bacteria in the MFC and MEC units than in ABR units (Fig. 6(b)).

Geobacter has been widely studied due to their electroactive ability for current generation in MFCs [38]. There were more *Geobacter* in the MFC unit than in other chambers at the genus level (Fig. 6(c)), which was related to electricity production capacity of the single chamber. MFC exoelectrogenic

bacteria such as *Geobacter*, *Shewanella* and *Pseudomonas* have been identified by Geochip-based functional gene analysis in the BESs [39]. Our study also implied that the enrichment of current producing bacteria in the single chamber MFC process might trigger better performance compared to the double chamber MFC.

The dominant bacterial community of ABR compartments was more similar to that of the MEC compartments, which implied that the start-up of MECs would be faster than MFCs. Since it is very important to keep steady operation at a very fast speed for the combined ABR-MFC-MEC system, the seed sludge from a running MEC unit or an ABR unit would bring faster start-up of a new combined ABR-MFC-MEC process than from the MFC unit. Compared with microbial strains from the anode of MFCs, more diversity and abundant electrode functional flora were found from the compartment of MECs. According to the changes of microbial structure, the species and abundance of Chlorobi tended to increase with operation of the process; this kind of microorganism can use sulfide as electron donor and produce elemental sulfur with deposition in the extracellular compounds [40]; the oxidized product sulfate could then be used as electron acceptor. The above finding explained why the denitrification and decarbonization efficiency improved when the MFC unit was exposed to very limited aeration [41,42]. The abundance of Spirochaetae increases with the aeration intensity, which implied that the micro-aeration

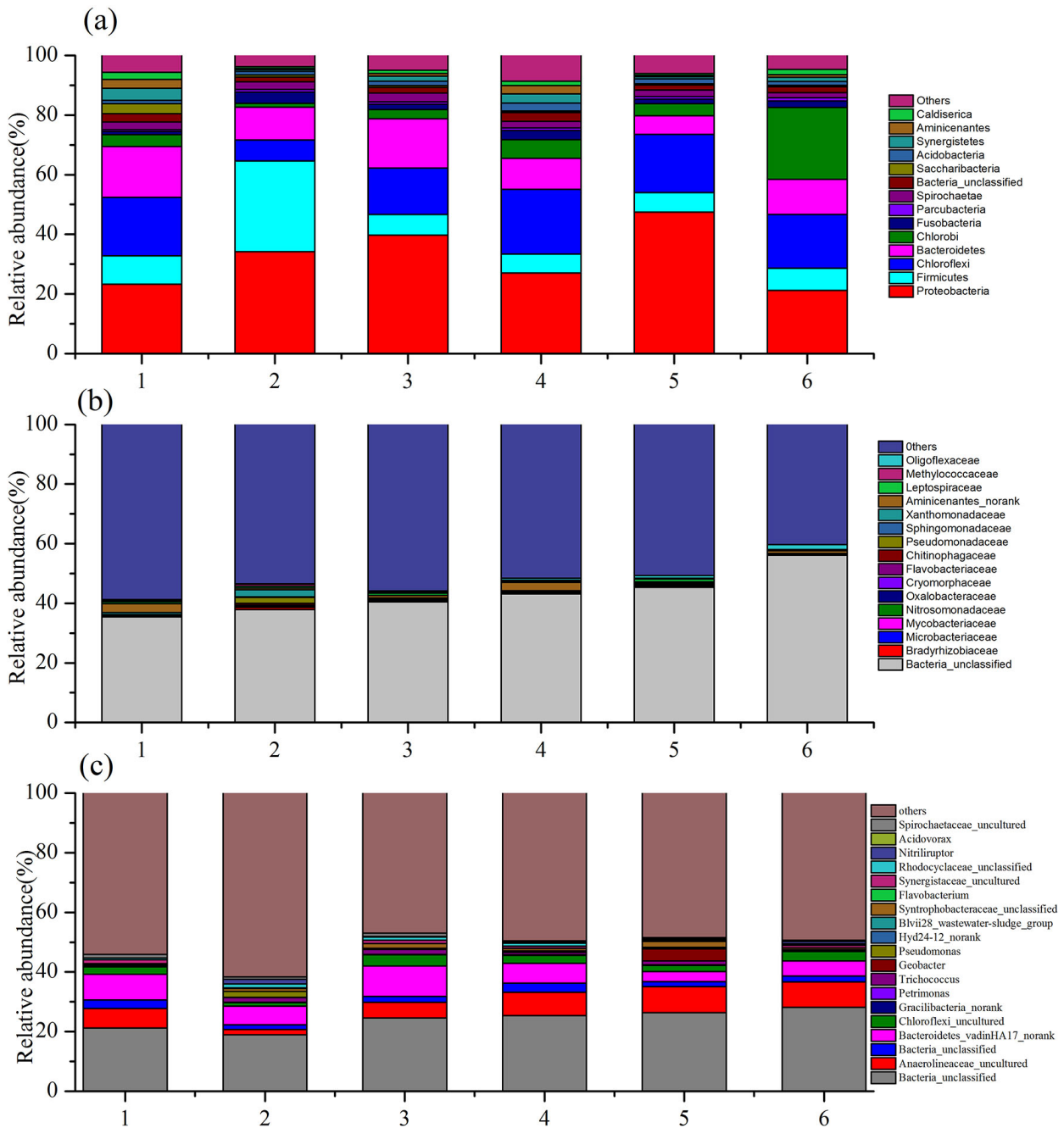


Fig. 6. Microbial community structures of samples at the (a) phylum, (b) family and (c) genus levels.

strategy actually promoted current-producing bacteria growth and improved electricity production. The decreased abundance of Saccharibacteria implied that it was replaced with the high efficient electrogenesis flora or other microorganisms that had synergies with the production of electrical bacteria.

4. Conclusions

A coupled ABR-MFC-MEC process treating black water was investigated in present study. When the influent COD concentration was $1,500.0 \pm 100.0 \text{ mg L}^{-1}$, the

$\text{NH}_4^+\text{-N}$ was $60.0 \pm 5.0 \text{ mg L}^{-1}$ and HRT was 24 h under a condition of continuous wastewater input, the COD and $\text{NH}_4^+\text{-N}$ removal rates were maintained at 92% and 50%, respectively, without any external energy input. In addition, the coupled system had a short start-up time and stable operation. Microbial structure of the combined ABR-MFC-MEC process in the stabilization stage showed that Proteobacteria, Firmicutes and Acidobacteria were the dominant electrogenesis flora obtained in the system. The different functions of microbial flora with competition and cooperation keep a balance of commensalism in the black water treatment system.

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Conflict of interest

The authors have declared no conflict of interest.

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Supplementary material

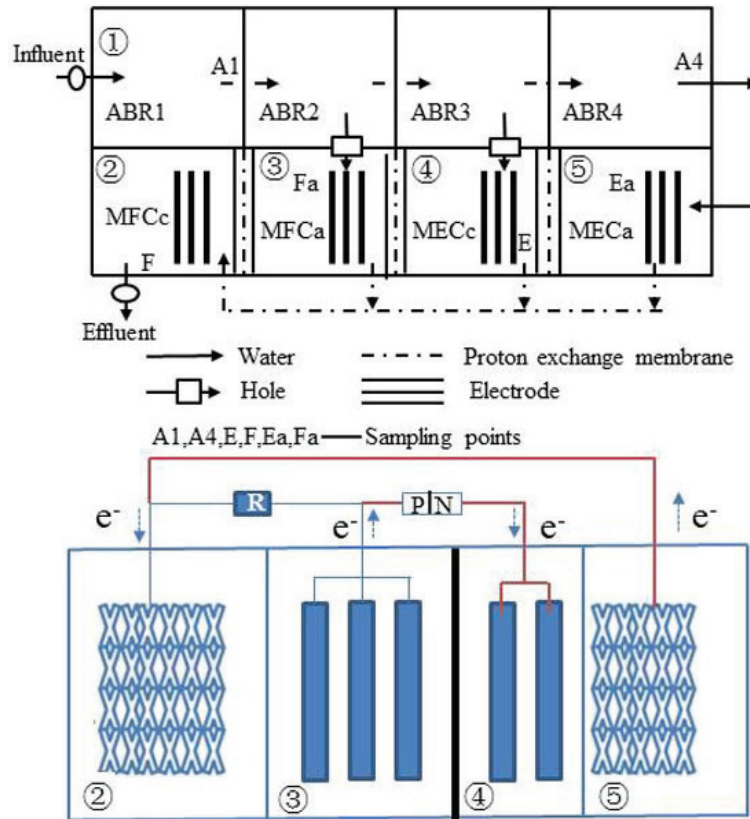


Fig. S1. Diagram of the experimental setup.

Table S1
Sampling locations and conditions for microbial structural analysis

Sampling time	Operational period					
Operational conditions	COD $\leq 3,000 \text{ mg L}^{-1}$; HRT = 12 h; $Q_{\text{air}} = 0 \text{ mL min}^{-1}$; $C_{\text{sulfate}} \leq 10 \text{ mg L}^{-1}$;					
Sampling point	ABR1	ABR4	MEC	MFC	MECa	MFCa
Sample ID	Sample I	Sample II	Sample III	Sample IV	Sample V	Sample VI
Sequences	60,116	33,138	62,899	69,005	65,199	71,199
Bases (bp)	26,370,211	14,638,885	27,787,896	30,339,643	28,819,923	31,310,751
Average (bp)	438.66	441.76	441.79	439.67	442.03	439.76
Serial no.	Sample ID	Sample type	Concentration (ng/ μL)	OD260/280	OD260/230	
A1	Sample I	Bulk sludge	54.8	1.85	0.77	
A4	Sample II	Bulk sludge	2.8	1.42	0.15	
E	Sample III	Electrode	55.8	1.87	1.15	
F	Sample IV	Electrode	14.1	2.00	0.10	
Ea	Sample V	Electrode	8.1	1.59	0.15	
Fa	Sample VI	Electrode	31.3	1.84	1.58	