Increased power generation from cylindrical microbial fuel cell inoculated with *P. aeruginosa*

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A B S T R A C T

Herein, carbon brush cylindrical microbial fuel cell is constructed to decrease internal resistance and increase electricity production. The application of cylindrical single-chamber microbial fuel cell gives full play to the role of anode carbon brush and increases contact with surrounding oxygen by enlarging the surface area of the cathode. Both cylindrical design and *P. aeruginosa* inoculation in anode have positive effects on power output of microbial fuel cells. Changing the configuration from cubic to cylindrical resulted in a substantial reduction in internal resistance from $127.21\,\Omega$ to $49.66\,\Omega$. Meanwhile, the cylindrical microbial fuel cell inoculated with mixed anaerobic bacteria exhibits power overshoot, and the dissolved oxygen in the electrolyte is raised. Thus, we also select *P. aeruginosa* inoculation in cylindrical reactor, where the maximum power density is increased to $3322 \pm 38\,\mathrm{mW\,m^{-2}}$ and internal resistance is reduced to $34.0 \pm 1.1\,\Omega$, and then power overshoot is improved. Thus, *P. aeruginosa* showed better electrogenic performance than anaerobic mixtures. In addition, chemical oxygen demand removal efficiencies (about one cycle) of the three microbial fuel cells are similar, but the cylindrical cell handles about 0.7 times of sewage more than the cubic cell showing that cylindrical microbial fuel cell has a higher capacity of sewage treatment.

1. Introduction

As we all know, since the third scientific and technological revolutions, energy has become the lifeblood of the national economy, and the energy on earth is limited (Shaffer et al., 2009). To solve the global energy crisis and environmental pollution problems, human beings are constantly exploring and developing new types of alternative energy and trying to make it on a large scale. Microbial fuel cell (MFC) is a kind of sewage treatment technology which can treat wastewater and generate electricity at the same time (Meng et al., 2015), and single chamber air-cathode MFCs are typically considered as a promising configuration (Liu et al., 2004; Bidart et al., 2014; Ahn, 2013). It can be used in water treatment, biosensing technique, environmental pollution control and so forth (Qiao et al., 2015a). At present, low yields of energy output, however, is a key bottleneck in the practical application of MFCs (Oliveria et al., 2013).

The performance of MFC could be affected by a lot of factors, such as electrode materials, microbial species and different MFC designs. The ideal anode materials should have high conductivity, non-corrosiveness, high specific surface area, high porosity and biocompatibility to facilitate microbial colonization or electron transfer (Zhu et al., 2014).

Currently, various carbon-based materials are widely used as anode materials in MFCs because of their low charge-transfer resistance and great stability in microbiological culture media, including carbon paper (Kim et al., 2007; He et al., 2012), carbon cloth (Ishii et al., 2008), carbon brush (Aelterman et al., 2008; Wang et al., 2018) and so on. Among them, carbon brush anode has received extensive attention due to their advantages of high bioavailable area, low electrical resistance and high porosity (Xie et al., 2017). Herein, carbon fiber brushes were used as anodes in this research.

In addition, battery resistance is one of the key factors affecting MFC performance (Fan et al., 2008). In order to reduce the internal resistance of MFC, the researchers used different battery configurations, such as: plate type (Min and Logan, 2004), U type (An et al., 2009), upflow type (He et al., 2005), H type (Logan et al., 2005) and tubular type (Rabaey et al., 2005; Sun et al., 2015; You et al., 2007a). He et al. (2007) applied a rotating cathode in MFC to increase the oxygen availability to the cathode, resulting in a higher power production $(49\,\mathrm{mW\,m^{-2}})$ compared to the nonrotating cathode system $(29\,\mathrm{mW\,m^{-2}})$. This configuration, however, required additional power input to rotate the cathode. Meanwhile, standard cubic MFC is a kind of widely used reactor (Meng et al., 2015; Aelterman et al., 2008; Wang

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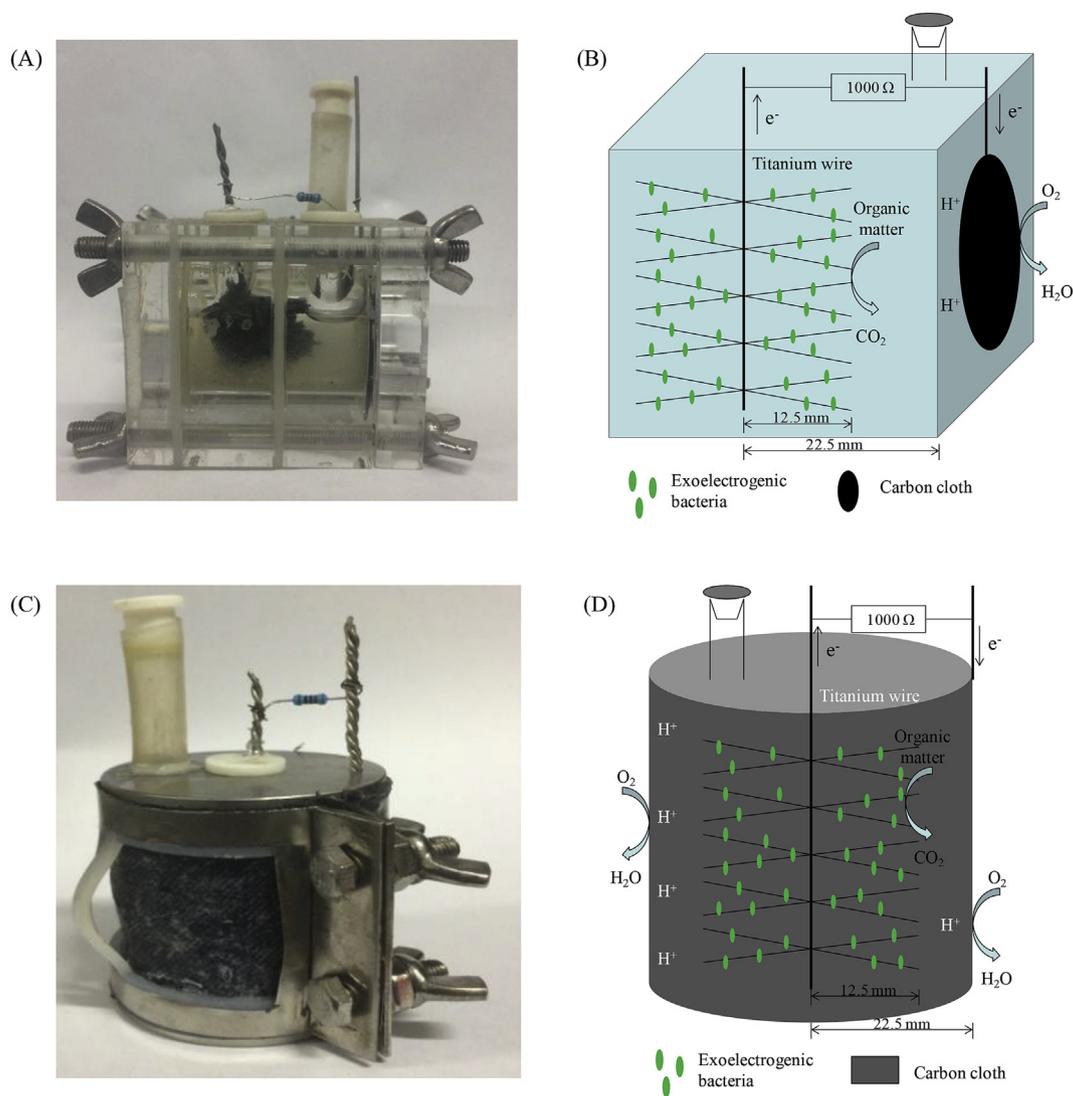 *E-mail address:* mazk@mail.buct.edu.cn (Z. Ma).

Fig. 1. The photo and schematic of cubic MFC (A,B) and cylindrical MFC (C,D).

et al., 2018; Nor et al., 2015), and its anode needs to be placed on the side of the cathode. If the cylinder carbon brush is used as anode, the distance from the front and back of the brush to the cathode is not the same in such a MFC (Fig. 1A and B), which leads to greater internal resistance. Accordingly, in order to demonstrate the advantages of cylindrical MFC, we compared cylindrical MFC with the common cubic MFC. In the cylindrical MFC (Fig. 1C and D), the anode is vertically placed in the center of the battery, and the entire side of the MFC is encased in a cathode so that the distance from the outside of the anode to cathode is equal. Consequently, the internal resistance is significantly decreased. Houghton et al. (2016) constructed a predictive linear model to predict the performance of a hypothetical cylindrical MFC. Otherwise, there will be many problems when cylindrical MFC is applied in practice, such as the phenomenon of power overshoot and the high oxygen content in the electrolyte, which will affect the performance of the MFC. Thus, we conducted a more in-depth exploration of cylindrical MFC here.

The distance between the anode and cathode also has an effect on battery performance, and scientific research can only have one variable. For configuration study, we put the cylindrical MFC and the commonly cubic MFC together for comparison. Compared with frequently-used cubic MFC, we can more effectively explore the reasons for the performance improvement of cylindrical MFC. The distance between the anode and cathode of the cube MFC is 1 cm. In order to maintain

consistent, we keep that of the cylinder MFC at 1 cm, so that the size of the cell can only be fixed.

Although the cylindrical MFC configuration could significantly reduce the internal resistance (You et al., 2007b), power overshoot was found in our polarization test. Power overshoot was related to the decrease in the electroactivity of the anode biofilm at high current regime, which resulted from a lack of sufficient electron transfer components to shuttle electrons at rates required for these more positive potentials (Watson and Logan, 2011). It is rarely found in pure cultures, such as *Shewanella oneidensis* or *Geobacter sulfurreducens*, and often occurs in the mixed cultures (Hong et al., 2011). Therefore, we envisage the use of pure bacteria to improve overshoot so as to correctly evaluate the performance of the cylindrical MFC.

In addition, as the cathode area of cylindrical MFC increases, the content of dissolved oxygen in the reactor will also increase, which will affect the activity of anaerobic bacteria (Wu et al., 2017). *P. aeruginosa* is a facultative aerobic bacterium that can metabolizes phenazines, such as pyocyanin, which can transfer electron extracellularly using the redox state (Rabaey et al., 2004). *P. aeruginosa* is a common opportunistic pathogen. It is widely distributed in nature and easy to grow in moist places with low nutritional requirements. Although the power output is not satisfactory, some researches (Pang et al., 2018; Watson et al., 2013) have already used *P. aeruginosa* as anodic bacteria in MFCs. This kind of facultative aerobic bacteria would also be used in our

experiments. We found that the *P. aeruginosa* has better electrogenic performance than anaerobic mixtures. Unexpectedly, power overshoot of the cylindrical MFC inoculated with *P. aeruginosa* was effectively improved, and the internal resistance was further reduced and the electricity generation was up to 3322 mW m^{-2}.

2. Materials and methods

2.1. MFC construction and set up

The air cathode single-chamber cubic MFC and cylindrical MFC were shown as Fig. 1. The effective volume of cylinderical MFC was 48 mL and its entire profile was almost a cathode with the cathodic area of about 42.4 cm^2. The single chamber cubic MFC with effective volume of 28 mL and the cathodic projection area of 7 cm^2 each was constructed as previously described (Cheng et al., 2006a; Zhang et al., 2008). Pt/C (20 wt. %) was used as air-cathode catalyst in microbial fuel cells. The detailed preparation method of the catalyst can be found in previous studies (Li et al., 2010; Xia et al., 2013; Cheng et al., 2006b). Cathodes were made of carbon cloth (HCP330, Heseng Co., Ltd., China) which contained a Pt/C catalyst loading of 0.5 mg cm^{-2} on the water-facing side (Houghton et al., 2016; Cheng et al., 2006c). For all the experiments, the anodes were composed of acid-treated carbon fiber brushes (2.5 cm in diameter, around 2.5 cm in length and 0.5 g in weight) with a titanium wire (Feng et al., 2010) that were placed vertically in the MFCs with the edge of about 1 cm from the cathode (Fig. 1). The cathode and anode were connected externally by titanium wires with a diameter of 1 mm.

Currently, MFCs inoculated with mixed cultures show substantially better power than those with pure cultures (Qiao et al., 2015a). Thus, this paper gives priority to use anaerobic sludge as inoculum. The MFCs were inoculated with a mixture of sludge suspension and medium by a volume ratio of 1:1, and they were named as AMB-Cylindrical-MFC and AMB-Cubic-MFC, respectively. Inoculation sludge was from Beijing Gaobeidian Wastewater Treatment Plant. Each reactor was fed a medium solution containing 1 g L^{-1} glucose, 50 mM PBS electrolyte (pH 7.0), vitamins and minerals (Li et al., 2010). The external resistance was fixed at 1000 Ω and all reactors were operated at 30 ± 2 °C.

The cylindrical MFC inoculated with *P. aeruginosa* was the same as above, except that the type of bacteria was different, and named as PA-Cylindrical-MFC. *P. aeruginosa* was purchased from Shanghai Luwei Microbial Sci. & Tech. Co. Ltd. and had already been treated as zero or very low toxicity by the company before purchase. At first, the nutrient agarwass was insulated for 15 min at 120 °C (Choi et al., 2015). Then, poured it into a Petri dish to cool down room temperature. Finally, some of the bacteria were put into the Petri dish and incubated for 24 h at 25 °C.

2.2. Characterization

The output voltage across the resister was recorded. The feed solution was replaced once the voltage dropped below 150 mV to obtain one complete cycle of cell operation. Polarization curves were measured by steady discharging method. The external resistance was gradually changed from 10 kΩ to 69 Ω. For each external resistance, the potential differences between anode and cathode, anode and Ag/AgCl, cathode and Ag/AgCl were measured under pseudo-steady state condition (10 min stabilization time) using the multimeter. The polarization curve is a common way to calculate the apparent internal resistance (Rint) of MFC, and the slope of the linear part of the polarization curve corresponds to the total internal resistance of MFC (Qiao et al., 2015b). Therefore, the internal resistance of the reactor can be obtained as long as the polarization curve is linear. Since only the anode carbon brushes are the same in the three MFCs, current density and power density were normalized to the side projected area of the anodic carbon fiber brush (6.25 cm^2).

Ohmic resistance (R$_\Omega$) was determined by current interrupt method. This experiment used the data acquisition system (NEWARE, Shenzhen, China) at a time interval of 0.1 s to measure the potential because of the need for rapid and accurate measurement of potential after the interruption of current. The ohmic resistance could be calculated by ohm's law, R (Ω) = ΔU/I, where I stands for the steady state current before interrupt.

The content of dissolved oxygen (DO) in cubic MFC and cylindrical MFC was measured by portable dissolved oxygen meter (HACH, HQ 30d). We measured the dissolved oxygen with hydraulic residence time of 8 h to evaluate the changes in dissolved oxygen content of the two different battery configurations.

Chemical oxygen demand (COD) was determined for the influent and effluent electrolyte of MFC using COD rapid titrator (5B–6C V7, Shanghai Lian-hua Tech. Co., Ltd.) by digestion spectrophotometry method. Each time the nutrient solution in MFC was replaced, COD was periodically measured when the hydraulic retention time was equal to 24 h (about one cycle), and COD removal rate was calculated according to eq. (1).

$$\eta_{COD} = (COD_{in}/COD_{ef} - 1) \times 100\% \tag{1}$$

Where COD$_{in}$ is the COD content of the influent of MFC, and COD$_{ef}$ is the content of the effluent of MFC.

3. Results and discussion

3.1. Bioelectricity generation of MFCs

Fe–N/CB was smeared on the air-cathode of MFCs. All MFCs were operated for five cycles at least after start-up and the output voltage profiles are shown in Fig. 2A. Repeatable and stable output voltage of 554 ± 7 mV was obtained in AMB-Cubic-MFC with the 1000 Ω external resistors, voltage of 625 ± 5 mV in AMB-Cylindrical-MFC, voltage of 638 ± 9 mV in PA-Cylindrical-MFC. Based on the power density curve, the maximum power density of AMB-Cylindrical-MFC was 1364 ± 88 mW m^{-2} which is much higher than that of AMB-Cubic-MFC (917 ± 26 mW m^{-2}) (Fig. 2B and C), and the current density was nearly identical (about 2.75 A m^{-2}). The high energy output showed that the electrogenic microorganism community was formed on the anode surface. Obviously, their performances were mainly affected by the MFC configuration because at this time the microbial species, electrolyte, the electrode material and even the concentration of the cathodic catalyst were the same.

However, there is a defect in the cylindrical MFC when inoculated with anaerobic mixed bacteria. As shown in Fig. 2D, power overshoot occurred in the cylinderical MFC at high current regime (300-22 Ω R$_{ext}$) which could cause lower power than it actually was, and the reason for the rapid decrease of power density was mainly caused by the anode mentioned in the literature (Liu et al., 2012).

To further improve the performance of cylindrical MFC, additional experiments were conducted using MFC reactors inoculated with *P. aeruginosa*. According to previous reports (Yong et al., 2017; Baranitharan et al., 2013), the power density of *P. aeruginosa* remains low for practical applications. In this paper, the performance of cubic MFC inoculated with *P. aeruginosa* (1033.7 mW m^{-2}) is much better than that of anaerobic mixed bacteria (Fig. S1). To date, it does not appear that pure bacteria produce as much power density as mixed bacteria. Moreover, researchers have recognized that anaerobic bacteria are suitable for MFC electrogenic bacteria. This may open a new door for future research on MFC. In addition, when applied to cylindrical MFC, the inflection phenomenon of the power density was improved, and the maximum power density and current density further rose to 3322 ± 38 mW m^{-2} and 8.81 A m^{-2} (Fig. 2B) respectively. It should be noted that, in addition to the different bacteria in AMB-Cylindrical-MFC and PA-Cylindrical-MFC here, all the other conditions
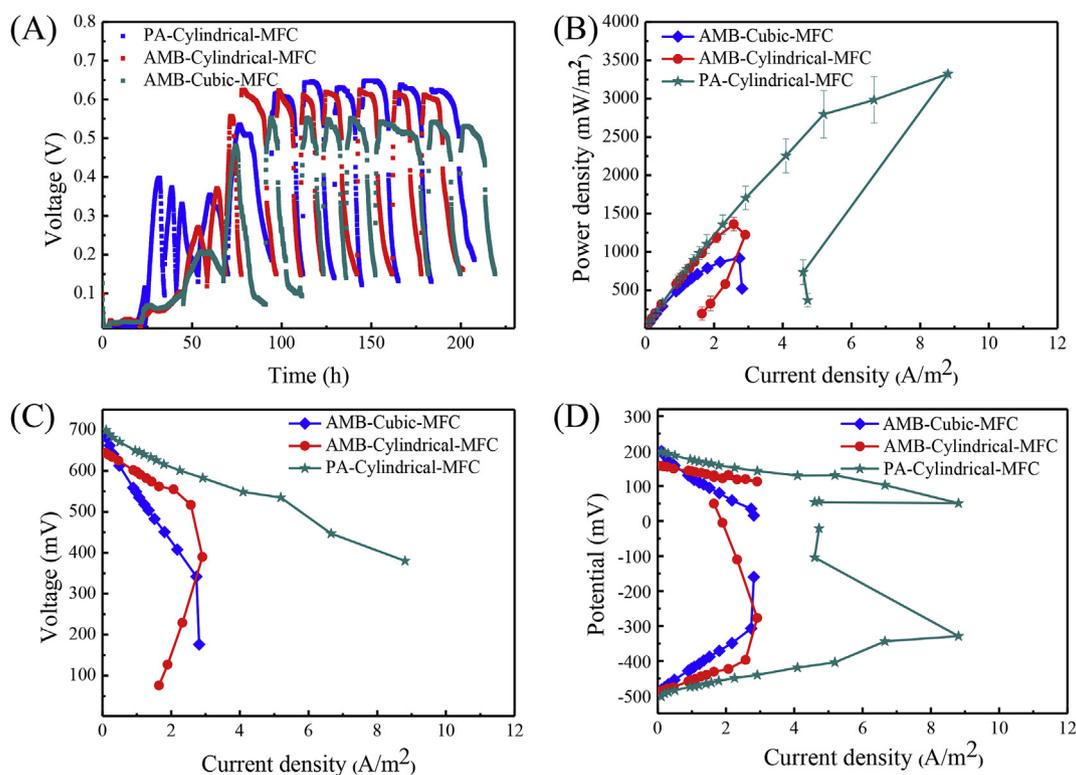
Fig. 2. Bioelectricity generation of MFCs. (A) is voltage output variation trend of AMB- Cylindrical-MFC, AMB-Cubic-MFC and PA-Cylindrical-MFC. (B) and (C) are power density and polarization curves of the MFCs, respectively. (D) is the anode and cathode electrode potentials.

were the same.

The apparent internal resistance of MFC can be divided into ohmic internal resistance and non-ohmic resistance. Ohmic resistance arises from electrolytes and proton exchange membrane (if present) that inhibit electron and ion conduction. The non-ohmic resistance is produced by the two-phase interface between the electrode and electrolyte, due to a low activation rate of the electrode surface and the low diffusion rate of the reactants or reaction products onto the electrode surface or into the solution. The internal resistance of AMB-Cubic-, AMB-Cylindrical- and PA-Cylindrical-MFCs were calculated to be $127.2 \pm 3.0\,\Omega$, $49.7 \pm 1.5\,\Omega$ and $34.0 \pm 1.1\,\Omega$, respectively (Table 1). To further understand the specific distribution of the internal resistance, we also used the current inperrupt method to measure the ohmic resistance of the MFCs (Fig. 3). The ohmic resistance of AMB-Cubic-, AMB-Cylindrical- and PA-Cylindrical-MFC was determined to be 61.4 ± 0.1, $28.4 \pm 0.9\,\Omega$, $22.1 \pm 0.4\,\Omega$, respectively (Fig. 3A–C, and Table 1). When anaerobic mixed bacteria were used as electrobacteria, changing the battery configuration from the cubic MFC to the cylindrical MFC resulted in a significant reduction in MFC internal resistance, especially non-ohmic resistance, leading to a significant increase in the maximum power density. Therefore, we should consider reducing both ohmic resistance and non-ohmic resistance when designing new MFC configuration. For the cylindrical MFC configuration, the internal resistance (primarily non-ohmic internal resistance) decreased after inoculation with *P. aeruginosa,* and the current density and maximum power density was further increased. The probable cause is

that *P. aeruginosa*, in addition to direct electron transfer, can also produce redox mediators such as phenazine compounds that promote the electron transfer extracellularly to the anode (He et al., 2007).

Compared with AMB-Cubic-MFC, the output voltage of AMB-Cylindrical-MFC increased by 69 mV (Fig. 2A) and internal resistance decreased by 61% (Table 1), while the maximum power density increased by only 51% (Fig. 2B). Otherwise, compared with AMB-Cylindrical-MFC, the output voltage of PA-Cylindrical-MFC was similar and the internal resistance was only slightly reduced, but the maximum power density was 1.44 times higher. Why only change the electrogenic bacteria to *P. aeruginosa*, the performance of cylindrical MFC is improved so much? The power density is calculated as $P = U^2/(R \cdot A)$, the areas (A) are the same and the external resistance (R) of each point is fixed, so it's the cell voltage (U) that causes the above difference. As can be seen from Fig. 2D, the cathode potential of PA-Cylindrical-MFC was similar to that of AMB-Cylindrical-MFC, while the potential of PA-Cylindrical-MFC declines significantly than that of AMB-Cylindrical-MFC.

We hypothesized that because the cylindrical MFC was inoculated with anaerobic bacteria, and the activity of anaerobic bacteria was low due to the increase of dissolved oxygen content. Accordingly, the DO test was done. The dissolved oxygen content (Fig. 3D) of AMB-Cubic-MFC, AMB-Cylindrical-MFC and PA-Cylindrical-MFC is $3.07 \pm 0.36\,\mathrm{mg\,L^{-1}}$, $6.19 \pm 0.13\,\mathrm{mg\,L^{-1}}$ and $6.22 \pm 0.23\,\mathrm{mg\,L^{-1}}$, respectively. Because the cathode area of cylindrical MFC was enlarged, the DO content doubled or so. This indicates that the poor electrical performance of AMB-Cylindrical-MFC may be due to the high content of

Table 1
The calculated internal and ohmic resistance in MFCs.

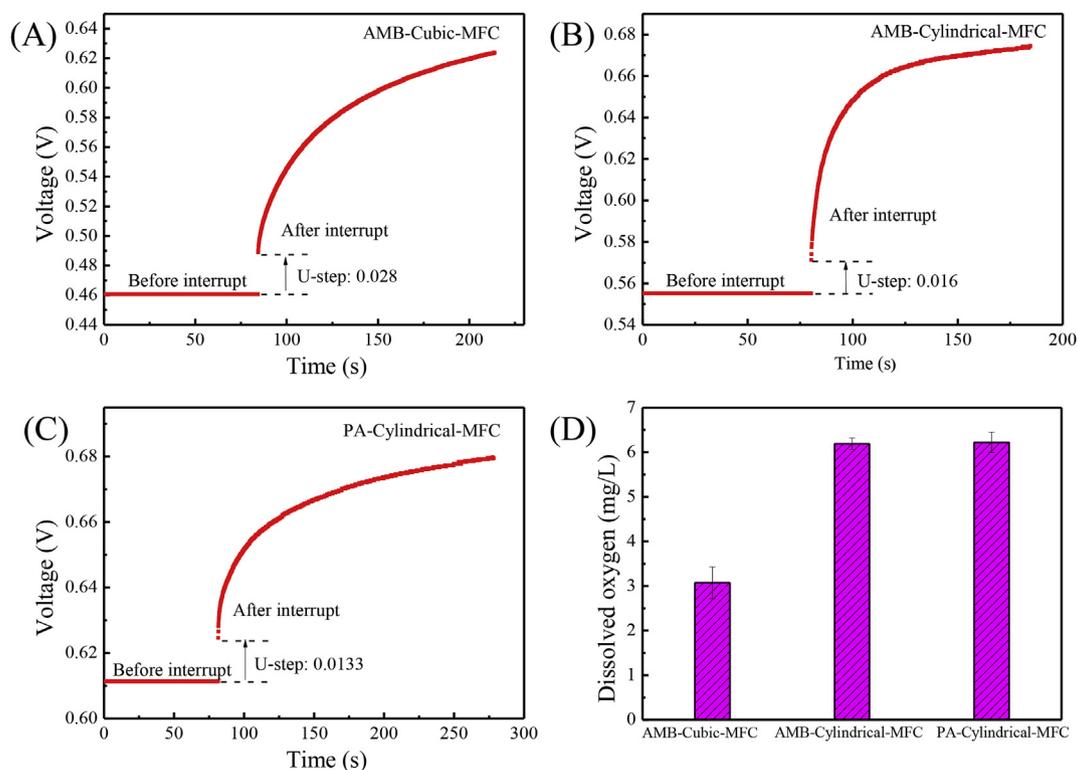| MFC name	R_Ω Ohmic resistance (Ω)	Non-ohmic resistance (Ω)	R_{int} Internal resistance (Ω)
AMB-Cubic-MFC	61.4 ± 0.1	65.8	127.2 ± 3.0
AMB-Cylindrical-MFC	28.4 ± 0.9	21.3	49.7 ± 1.5
PA-Cylindrical-MFC	22.1 ± 0.4	11.9	34.0 ± 1.1

Fig. 3. (A), (B) and (C) are the U-T profiles of AMB-Cubic-MFC, AMB-Cylindrical-MFC and PA-Cylindrical-MFC when using current interrupt method in MFCs (external resistance 1000 Ω), respectively. (D) Dissolved oxygen content of MFCs.

DO. And because *P. aeruginosa* has good tolerance to oxygen, it can adapt to higher oxygen content and release more electricity.

3.2. COD analysis

In addition, we measured the COD removal efficiency after running for 24 h (one cycle) to compare the capacity of the MFCs to treat sewage. COD removals (Fig. 4) of the three different MFCs were 90.0 ± 1.9%, 92.0 ± 3.1% and 85.3 ± 5.2%, respectively. Although the COD removal rate of the AMB-Cylindrical-MFC was only slightly better than that of the cubic MFC, it handled about 0.7 times of sewage, which means that the cylindrical MFC has a higher ability of handling sewage and is helpful for the scale-up of MFCs. When using cylindrical MFC as reactor, the COD removal efficiencies obtained with *P. aeruginosa* were slightly less than that of anaerobic mixed bacteria. This is presumably because the inoculated sludge contains a wide variety of microorganisms that develop into a community and consume more organic substances from the electrolyte. Although using *P. aeruginosa*
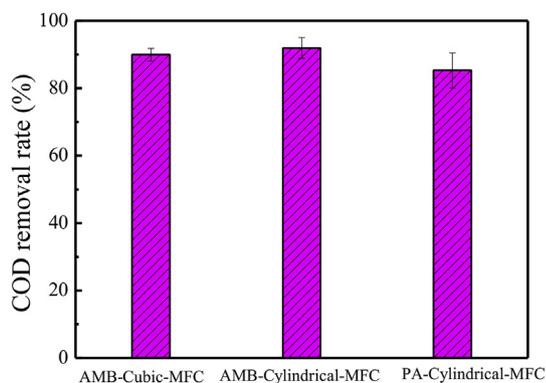
achieves the minimum COD reduction, the power generation is quite high. Similar situations have previously occurred, in which COD removal is inversely proportional to electricity production (Logan et al., 2005; Estrada-Arriaga et al., 2018).

4. Conclusion

In this research, we have demonstrated the superiority of carbon brush cylindrical MFC with increased cathode area and reduced internal resistance. Compared with the cubic MFC, the maximum power density of AMB-Cylindrical-MFC increased by 52% while the internal resistance reduced by 60.9%. This shows that it is of great significance to combine cylindrical MFC with carbon brush anode. We also found that the electrogenic characteristics of *P. aeruginosa* were different from those reported previously. *P. aeruginosa* shows better electrogenic performance than anaerobic mixtures. After inoculation with *P. aeruginosa*, overshoot of cylindrical cell has been greatly alleviated, and the current density and maximum power density are further improved reaching to $8.81 \, A \, m^{-2}$ and $3322 \pm 38 \, mW \, m^{-2}$, respectively. The possible reason is that *P. aeruginosa*, in addition to direct electron transfer, can also produce redox mediators that accelerate the electron transfer extracellularly to the anode. Due to the oxygen resistance and the ability of extracellular electron transfer of *P. aeruginosa*, we speculated that this might be the reason for the excellent performance of cylindrical batteries. The main advantages of this system are high power generation, small internal resistance and good oxygen resistance. Overall, our findings show that cylindrical MFC equipped with carbon brush anode and *P. aeruginosa* should be further studied in MFC.

Declaration of interests

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

The authors declare the following financial interests/personal

Fig. 4. COD removal rates (HRT = 24 h) of MFCs.

relationships which may be considered as potential competing interests.

CRediT authorship contribution statement

Man Zhang: Investigation, Writing - original draft. **Zhaokun Ma:** Resources. **Na Zhao:** Formal analysis. **Kaixuan Zhang:** Formal analysis. **Huaihe Song:** Supervision.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.bios.2019.111394.

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