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# Uneven biofilm and current distribution in three-dimensional macroporous anodes of bio-electrochemical systems composed of graphite electrode arrays



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## HIGHLIGHTS

- A 3-D macroporous anode composed of graphite rod arrays was developed.
- Current promotion was limited after the electrode array reached a high number.
- A significantly uneven current distribution was found in the macroporous anode.
- Proton transfer influenced the pH and biofilm distribution in the macroporous anode.
- The closer the segment was to the center, the lower the contribution to the total current.

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# G R A P H I C A L A B S T R A C T



### ABSTRACT

A 3-D macroporous anode was constructed using different numbers of graphite rod arrays in fixedvolume bio-electrochemical systems (BESs), and the current and biofilm distribution were investigated by dividing the 3-D anode into several subunits. In the fixed-volume chamber, current production was not significantly improved after the electrode number increased to 36. In the case of 100 electrodes, a significant uneven current distribution was found in the macroporous anode. This was attributed to a differential pH distribution, which resulted from proton accumulation inside the macroporous anode. The pH distribution influenced the biofilm development and led to an uneven biofilm distribution. With respect to current generation, the uneven distribution of both the pH and biofilm contributed to the uneven current distribution. The center had a low pH, which led to less biofilm and a lower contribution to the total current, limiting the performance of the BESs.

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# 1. Introduction

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Bio-electrochemical systems (BESs) are green electrochemical technologies that use applied biological techniques (Kim et al., 2015; Mook et al., 2013; Mohan et al., 2014). In a typical BES, microorganisms on the electrode are used to catalyze the electrochemical reactions, and then, several types of energy or chemicals are harvested. Recently, many typical BESs, such as microbial fuel



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cells (MFCs), microbial electrolysis cells (MECs) and microbial electrosynthesis cells (MESs), have been developed to produce electricity, hydrogen and valuable compound chemicals, respectively (Kim et al., 2015; Mohan et al., 2014). During the past decade, BES research has been a rapidly evolving field, and many works have contributed to construction materials (Liu et al., 2012; Song et al., 2015; Yousefi et al., 2016; Yang et al., 2016), biofilms (Jerome et al., 2012; Zhang et al., 2011; Zheng et al., 2016; Yang et al., 2015), electron transfer mechanisms (Sun et al., 2017; Cai et al., 2013; Qiao et al., 2015), key influencing factors (Liu et al., 2014; Lorenzo et al., 2010) and reactor designs (Yang et al., 2015; Logan, 2010; Zou et al., 2014). Up to now, BESs researchers reported several aspects of practical application such as the treatment of actual wastewater, small power sources, biosensor and saline water desalination. However, many inherent challenges still occur when BESs are applied in practice.

Scale-up is an important consideration for future practical application of BESs (Logan, 2010; Zou et al., 2014; Liu et al., 2008). With respect to scale-up, an 3-D electrode with a compact porous structure is the expected choice for the enlargement of specific surface areas, showing the attractive and potential application of large BESs in the future (Lorenzo et al., 2010; Cheng and Logan, 2011). Currently, several types of 3-D electrodes have been adopted to increase the performance of BESs, including graphite rods, graphite granules, graphite felt, graphite wool and graphite fiber brushes, among others. (Wei et al., 2011; Zhang et al., 2010a; Clauwaert et al., 2009). Among these 3-D electrodes, tenuous graphite rods would be more suitable to the scale-up of BESs, not only because they are relatively inexpensive and have a large, defined specific surface area but also because they can be easily arranged as designed (Zhu et al., 2013). In a specific chamber of BESs, the more tenuous electrodes are used, the more electrodes can be integrated. Thus, a larger electrode surface area can be obtained, demonstrating an attractive application (Zhu et al., 2013). A previous MFC study reported that a further improvement in the volumetric power density can be expected by inserting more graphite rods with a smaller diameter into the anode chamber (Zhang et al., 2013b).

However, increasing the number of graphite electrodes resulted in the decreased pore size of the 3-D electrode. With respect to 3-D macroporous electrode, an optimal pore and electrode size are expected for the porous anode considering the surface for bio-electrochemical reaction, the channel for mass transfer and the framework for electron transfer. In the fixedvolume BESs porous electrode, a small pore size would be beneficial for the reaction area and electron transfer but would result in a high mass transfer resistance. It is reported that mass transfer would significantly influence biofilm development and BESs performance (Torres et al., 2008a,b; Zhang et al., 2011; Yang et al., 2015). The previous study showed that the microchamber limited biofilm formation and, subsequently, current generation in a micro-sized microbial fuel cell (Choi and Chae, 2013). The mesh pore size influenced hydrogen production in MECs using stainless steel mesh cathodes (Zhang et al., 2010b). The performance of the porous bio-anode in MECs was improved by enhancing the mass transport of substrates and protons inside the porous anode (Sleutels et al., 2009). Many studies indicated that proton transfer is one of the key factors for the performance of BESs (Zhang et al., 2011, 2015; Torres et al., 2008a), and proton accumulation limits the bio-electrochemical reaction rate (Torres et al., 2008b; Nevin et al., 2009; Franks et al., 2009). It is expected that mass transfer, especially proton transfer, is the bottleneck for BESs using a 3-D macroporous electrode. Thus, it's necessary to investigate the structure-induced mass transfer effects in 3-D macroporous electrode for better understanding of the electrode design.

In addition, it was expected that, in 3-D electrodes with small pore size, mass transfer limitation would occur in the electrode center, resulting in uneven distributions of biofilm and current. The uneven current distribution led to poor utilization of the substrate and electrode and then low current (Arjan et al., 2009). A nonuniform current distribution was reported in a liter-scale multiple-electrode microbial fuel cell with a relatively large electrode spacing of the electrode arrays; this resulted from the different electrode spacing between the anode and cathode electrode (Zhang et al., 2013b). However, there was noted that a large electrode distance (2.0 mm) indicated that the mass transfer limitation would not occur inside the array in above study. Thus, it is still necessary to investigate the current distribution in 3-D macroporous electrodes of BESs.

In this study, graphite electrode arrays with different electrode numbers were constructed for the 3-D macroporous anode in BESs. The pore size of porous electrode was adjusted by arranging different numbers of graphite rods in the fixed-volume BESs. Current generation as function of the graphite rod numbers and the current distribution in the 3-D macroporous electrode was investigated. Also, the possible reasons for the uneven current distribution and the suggestions for its improvement were discussed.

#### 2. Materials and methods

#### 2.1. BESs construction

As shown in Fig. S1c and d, a graphite rod-based half-cell system was constructed using a single polymethyl methacrylate (PMMA) chamber with a total volume of 135 mL  $(150 \times 30 \times 30 \text{ mm})$ . The chamber was evenly filled with 100 graphite rods ( $\Phi$  = 0.9 mm, L = 40.0 mm) in an aligned arrangement  $(10 \times 10 \text{ array})$  with an electrode distance of 0.5 mm, which served as the electrode (Fig. S1b). The electrode array was divided into 25 segments, and each segment had four graphite rods (Fig. S1e). The 25 segments were labeled sequentially as A, B, C, D and E rows based on the vertical direction and 1, 2, 3, 4 and 5 lines based on the horizontal direction. For the measurement of the current distribution, each segment was connected with a small fixed resistance  $(R = 1.0 \Omega)$  and then connected to the electrochemical workstation (Fig. S1a). For the independent measurement of the current generated from each segment, the wire from each segment was electrically isolated from other wires. The 0.1 mm diameter stainless steel wire (254SMO, TongLai Stainless Steel Wire (Xiamen) Co., Ltd., China) was used to link the graphite rod as a collector. The Ag/AgCl reference electrode and pH microelectrode were installed in this chamber. In the half-cell system, the graphite rod array, carbon cloth and Ag/AgCl reference electrode served as the working electrode, counter electrode and reference electrode, respectively.

## 2.2. BESs operation

In the experiment, the half-cell system was inoculated with the effluent from a MFC that was successfully operated for over two years. An anolyte including 0.68 g/L sodium acetate and a 8.3 mM phosphate buffer solution (6.0 g/L Na<sub>2</sub>HPO<sub>4</sub>, 3.0 g/L KH<sub>2</sub>-PO<sub>4</sub>, 0.1 g/L NH<sub>4</sub>Cl, 0.5 g/L NaCl, 0.1 g/L MgSO<sub>4</sub>·7H<sub>2</sub>O, 15.0 mg/L CaCl<sub>2</sub>·2H<sub>2</sub>O and 1.0 mL/L trace elements solution) (pH 7.2–7.4) was fed to the microorganisms. The system was started by connection to an electrochemical workstation (PRASTAT MC, USA) using chronoamperometry under a potentiostatic control of 0.05 V. The solution was de-aerated by passing through high purity nitrogen continuously during the startup process. All experiments were performed at room temperature:  $30 \pm 1$  °C.

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