International Biodeterioration & Biodegradation xxx (xxxx) xxx–xxx

Contents lists available at ScienceDirect

International Biodeterioration & Biodegradation

journal homepage: www.elsevier.com/locate/ibiod

Anode modification with palladium nanoparticles enhanced Evans Blue removal and power generation in microbial fuel cells

Xiangchun Quan*, Hengduo Xu, Bo Sun, Zhutian Xiao

Key Laboratory of Water and Sediment Sciences, Ministry of Education, State Key Laboratory of Water Environment Simulation, School of Environment, Beijing Normal University, Beijing 100875, PR China

ARTICLE INFO

Keywords:
Microbial fuel cell
Azo dye
Anode decoration
Power generation
Palladium nanoparticles

ABSTRACT

Anode electrode was modified using palladium nanoparticles (PdNPs) aimed to increase power generation and simultaneous pollutants removal from azo dye containing wastewater in Microbial Fuel Cells (MFCs). Results showed that the MFCs with Pd modified anode (Pd-MFC) demonstrated a higher Evans Blue decolorization rate, maximum power density and Coulombic Efficiency compared to that with control anode. The Pd1-MFC (1 mg Pd cm^{-2} anode) and Pd1-MFC (2 mg Pd cm^{-2} anode) generated a maximum power density of 499 ± 11 mWm^{-2} and 447 ± 15 mWm^{-2}, respectively, from Evans Blue containing wastewater, enhanced by 3.8 and 3.3 folds compared to the control MFC (106 ± 5 mWm^{-2}). Illumina Miseq high-throughput sequencing method was used to investigate the microbial community structure for the anode biofilm. The Pd anode enriched more exoelectrogen *Geobacter* (53% for the Pd1 anode, 81% for the Pd2 anode) than the control anode (36%). Anode decoration with Pd NPs reduced charge transfer resistance, enriched more exoelectrogen, and endowed the electrode with direct Pd catalytic ability, which might contribute to the enhanced performance of Pd-MFCs.

1. Introduction

Bioelectrochemical system (BES), as a promising technology, has drawn increasing interests recently for simultaneous pollutants removal and electricity generation (Logan, 2009). Microbes, as the biocatalyst in BES, can transform chemical energy stored in organic compounds to electrical energy. Azo dyes, characterized with a structure of aromatic ring and double bonds, widely exist in the effluent from textile industries. Azo dyes are toxic and hard to be degraded by microorganisms. BES has been explored for azo dyes degradation recently (Fernando et al., 2014; Gao et al., 2016). Although some anode exoelectrogens can transform azo dyes via co-metabolism, their microbial activity may be inhibited and power generation ability may be weakened due to the toxicity of azo dyes. Therefore, a long term pre-acclimation is generally required for the anode bacteria to adapt to azo dyes and degrade them for power generation (Wang et al., 2013).

Electro-catalysts loaded on anode electrodes could help to oxidize some organic molecules. For example, palladium nanoparticles (PdNPs) deposited on anode electrode could catalyze formate, lactate and acetate directly (Quan et al., 2015a). Pd nanoparticles, which were produced by microbial reduction, were also used for azo dye reduction in the presence of different electron donors, such as hydrogen, formate, acetate, ethanol and glucose (Quan et al., 2015b). Whether is it possible to accelerate azo dye reduction and power generation in the anode chamber of BES through anode decoration with Pd catalyst? This question has not been answered and deserves further study.

PdNPs based catalysts have attracted great attention due to large specific surface and more active sites for catalyzing (Erikson et al., 2011). PdNPs are generally prepared using chemical methods with many toxic chemical agents under harsh conditions (Moon et al., 2014; Watts et al., 2017). Recent studies have found that some microorganisms, such as *Paracoccus denitrificans* (Bunge et al., 2010) and *Shewanella oneidensis* (*S. oneidensis*) (Hennebel et al., 2009), can reduce soluble Pd (II) and form biogenic nano-palladium in periplasms or cell walls. This microbial reduction method occurred under benign and gentle conditions with much less chemical agents (Hennebel et al., 2009). Biogenic Pd also shows the advantages of high catalytic activity and good biocompatibility (Yates and Logan, 2014).

In this study, Pd nanoparticles were prepared through microbial reduction by a strain *S. oneidensis*. Anode electrode was modified using Pd catalyst and applied in microbial fuel cells (MFCs) aimed to improve pollutant removal and power generation from azo dye containing wastewater. Evans Blue (EB) was selected as the studied azo dye, because it is widely used as chemical and biochemical analysis agent, and the removal of Evans Blue in BES has seldom been reported (Antonin et al., 2015). The electrochemical behavior and morphological property

* Corresponding author.
E-mail address: xchquan@bnu.edu.cn (X. Quan).

https://doi.org/10.1016/j.ibiod.2018.01.001
Received 31 October 2017; Received in revised form 4 January 2018; Accepted 4 January 2018

of the Pd electrode was characterized using Cyclic Voltammograms (CVs) and Scanning Electron Microscopy (SEM), respectively. The performance of the MFCs with Pd anodes was evaluated from pollutant removal rate and power generation capacity. Effects of Pd decoration on microbial community changes of anode biofilm were investigated through high-throughput sequencing based on Illumina Miseq platform.

2. Materials and methods

2.1. Microbial culture and Pd NPs preparation

S. oneidensis MR-1was used as the bacteria to form PdNPs. This strain was from a lab from Tsinghua University (Beijing, China). The strain *S. oneidensis* MR-1 was capable of forming Pd NPs in cells via reducing Pd (II) ion (Windt et al., 2005). *S. oneidensis* MR-1 was first cultured through incubation in Luria-Bertani (LB) medium for 24 h at 28 °C. The microbial culture was then collected and production of Pd proceeded in M9 medium according to the method described previously (Windt et al., 2005). The microbial cells after Pd formation was collected through centrifugation (6000 rpm for 15 min), cleaned with distilled water, and then dried in an oven controlled at 105 °C. The Pd-associated biomass was burned further at 600 °C under nitrogen atmosphere for 20 min to obtain Pd power.

2.2. Electrodes preparation

With carbon cloth as the supporting material, Pd modified electrodes were prepared according to the following procedure. First, the above prepared Pd powder was mixed with carbon black powder and added by 5% Nafion (DuPont Co., Delaware) to make paste mixture. The paste was then spread to each side of carbon cloth and dried in an oven at 121 °C. Through adjusting the amounts of Pd in the carbon black powder, the Pd modified electrodes with different Pd loading amounts, i.e. 1 mg Pd cm^{-2} carbon cloth (named Pd1 electrode) and 2 mg Pd cm^{-2} carbon cloth (named Pd2 electrode) were obtained. Another electrode was coated with carbon black only (15 mg cm^{-2}) according to the above procedure and used as the control electrode. Each type of electrode was prepared in triplicate for use in the following experiments.

2.3. MFC design and operation

Dual-chamber MFCs were constructed and loaded with different anode electrodes to evaluate their performance in treating Evans Blue containing wastewater. The MFCs had a cubic anode chamber (working volume 100 mL) and a cubic cathode chamber (working volume 100 mL) which were separated by a membrane (Nafion 117, DuPont). The above prepared electrodes with different Pd loading amounts (Pd1 and Pd2) and control electrode were fixed to the MFCs. The cathode was Pt coated carbon paper (4 cm × 2.5 cm, 1 mg Pt cm^{-2}). The MFCs were first started using the same method described by Quan et al. (2015a). Briefly, the anode chamber of MFCs was inoculated with anaerobic sludge from a wastewater treatment plant (Beijing, China) to enrich electro-active bacteria on anode electrode. The medium fed to the anode chamber contained (g L^{-1}): 1.64 CH_3COONa, 0.31 NH_4Cl, 0.13 of KCl, 3.32 of $NaH_2PO_4{\cdot}2H_2O$, 10.32 $Na_2HPO_4{\cdot}12H_2O$, 12.5 mL of trace mineral solution and 5 mL of vitamin solution. The catholyte was Phosphate Buffer solution (PBS). Air was supplied to the cathode chamber through aeration by an air-diffuser. Voltage output data across a 1000 Ω external resistor was monitored using a data acquisition system. When voltage output reached a steady level, the feeding medium to the anode chamber turned to the medium containing 100 mg L^{-1} Evans Blue and 500 mg L^{-1} glucose with other components the same as the above described. The MFCs ran in a fed batch mode at a constant temperature (30 ± 1 °C). Effluent samples were withdrawn from anode chamber at certain time intervals. Evans Blue concentration was determined using an Ultra-Violet (UV) device (HACH, DR6000) at a wave length of 481 nm. Chemical Oxygen Demand (COD) was measured according to standard methods (Walter, 1998).

2.4. Electrochemical characterization

The electrochemical property of MFCs was characterized by measuring polarization curves and power density-current when adjusting external resistors over the range 50–10 000 Ω. The impedance of anode electrodes was investigated through conducting electrochemical impedance spectroscopy (EIS) using an electrochemical working station (ChenHua CH660, China) under the conditions of 100 kHz–0.01 Hz frequency and a potential amplitude of 5 mV. CVs were performed for the different anode electrodes in a three-electrode system, with Pt rod and Ag/AgCl as the counter and reference electrode, respectively. During conducting EIS and CVs experiments, a nitrogen environment was maintained through purging high-purity nitrogen. All the above tests were repeated in triplicate.

2.5. DNA extraction, PCR and high-throughput sequencing

At the end of MFC operation, anode electrodes were taken out and microbial community for anode biofilm was analyzed according to the following procedure. First, genomic DNA was extracted from the biofilm sample using DNA Isolation Kit (MoBio) according to the manufacturer instruction. The V3—V4 region of the 16S rRNA gene was amplified using the bacteria primers 338F (5′-ACTCCTACGGGAGGCA GCA-3′) and 806R (5′-GGACTACHVGGGTWTCTAAT-3′). The PCR was run on GeneAmp® 9700 (ABI). A total PCR reaction mixture volume was 20 μL, containing 0.4 μL (5 μM) forward and reverse primers, 10 ng template DNA, 4 μL dNTPs (2.5 mM), 4 μL 5 × FastPfu Buffer and 0.4 μLFastPfu Polymerase. The PCR was conducted under the following conditions: an initial denaturation at 95 °C for 2 min, 25 cycles denaturation at 95 °C for 30 s, 55 °C annealing for 30 s, and 72 °C for 45 s extension, followed by a final extension at 72 °C for 10 min. The PCR products were purified using the AxyPrep™ DNA Gel Extraction Kit (AXYGEN). After purification and quantification, samples were sent to a biotechnology company (Shanghai Majorbio Bio-pharm Biotechnology Co., China) for 16S rRNA gene sequencing basing on Illumina Miseq platform.

3. Results and discussion

3.1. Electrochemical characteristics of Pd modified electrode

Cyclic voltammetry was performed to evaluate the electrochemical activity of different electrodes in 5.0 mM $Fe(CN)_6^{3-/4-}$ and 0.2 M Na_2SO_4 solutions (Fig. 1). As shown in Fig. 1(a), the CV graph of the Pd electrode displayed apparent peaks of current density at the potential of 293 mV (reductive peak) and 490 mV (oxidative peak), resulting from redox reaction of potassium hexacyanoferrate. The bare electrode showed much smaller redox peaks compared to the Pd electrode, suggesting higher electrochemical activity of the Pd electrode. The Pd electrode displayed a larger CV graph area and higher redox peaks than the control electrode, indicating decoration with Pd improved electron transfer and faradic charge capacity of electrodes, possibly because of the increased specific surface area and conductivity (Qiao et al., 2007; Sun et al., 2010). Similar results were also found for a study with gold particles modified electrode conducted by Sun et al. (2010).

EIS was further conducted for the above electrodes in a three-electrode system in order to know resistance and electron transfer properties (Janek et al., 1998). The complex impedance (Z) results are presented in Nyquist plot (Fig. 1(b)) which comprises a straight line at low frequencies and single semicircle at the high frequencies. EIS results were fitted to an equivalent electrical circuit with a combination of resistance and capacitance. The charge transfer resistance (R_{ct}) was