Environmental Pollution 236 (2018) 343–354

Contents lists available at ScienceDirect

Environmental Pollution

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

Time-dependent bacterial community and electrochemical characterizations of cathodic biofilms in the surfactant-amended sediment-based bioelectrochemical reactor with enhanced 2,3,4,5-tetrachlorobiphenyl dechlorination☆

Hui Wan [a], Xiaoyun Yi [a, b], Xiaoping Liu [a], Chunhua Feng [a, b, *], Zhi Dang [a, b], Chaohai Wei [a, b]

[a] The Key Lab of Pollution Control and Ecosystem Restoration in Industry Clusters, Ministry of Education, School of Environment and Energy, South China University of Technology, Guangzhou 510006, PR China
[b] Guangdong Provincial Engineering and Technology Research Center for Environmental Risk Prevention and Emergency Disposal, South China University of Technology, Guangzhou 510006, PR China

ARTICLE INFO

Article history:
Received 30 August 2017
Received in revised form
13 January 2018
Accepted 16 January 2018

Keywords:
PCB dechlorination
Electrical stimulation
PCB contamination
Surfactant
Electrochemical impedance spectra
High-throughput sequencing

ABSTRACT

Applying an electric field to stimulate the microbial reductive dechlorination of polychlorinated biphenyls (PCBs) represents a promising approach for bioremediation of PCB-contaminated sites. This study aimed to demonstrate the biocathodic film-facilitated reduction of PCB 61 in a sediment-based bioelectrochemical reactor (BER) and, more importantly, the characterizations of electrode-microbe interaction from microbial and electrochemical perspectives particularly in a time-dependent manner. The application of a cathodic potential (−0.45 V *vs.* SHE) significantly improved the rate and extent of PCB 61 dechlorination compared to the open-circuit scenario (without electrical stimulation), and the addition of an external surfactant further increased the dechlorination, with Tween 80 exerting more pronounced effects than rhamnolipid. The bacterial composition of the biofilms and the bioelectrochemical kinetics of the BERs were found to be time-dependent and to vary considerably with the incubation time and slightly with the coexistence of an external surfactant. Excellent correlations were observed between the dechlorination rate and the relative abundance of *Dehalogenimonas*, *Dechloromonas*, and *Geobacter*, the dechlorination rate and the cathodic current density recorded from the chronoamperometry tests, and the dechlorination rate and the charge transfer resistance derived from the electrochemical impedance tests, with respect to the 120 day-operation. After day 120, PCB 61 was resistant to further appreciable reduction, but substantial hydrogen production was detected, and the bacterial community and electrochemical parameters observed on day 180 were not distinctly different from those on day 120.

1. Introduction

The use of electrodes as continuous electron acceptors or electron donors to stimulate the growth of specific microorganisms for in situ bioremediation of contaminated sites represents a highly promising, efficient, and cost-effective approach that has attracted considerable attention (Thrash and Coates, 2008; Rabaey et al., 2007). This innovative technology minimizes the need for external chemical amendments and requires a small amount of electrical energy, offering the possibility to manipulate and control a wide range of microbial processes in remediation of anoxic environments, such as sediment and groundwater. Existing examples include the degradation/transformation of petroleum hydrocarbons (Venkidusamy et al., 2016), food wastes (Du and Li, 2016), heavy metals (Gregory and Lovley, 2005), nitrate (Puig et al., 2012), and chlorinated compounds (Aulenta et al., 2009). In recent years, a process named bioelectrochemically assisted reductive

☆ This paper has been recommended for acceptance by Charles Wong.

* Corresponding author. The Key Lab of Pollution Control and Ecosystem Restoration in Industry Clusters, Ministry of Education, School of Environment and Energy, South China University of Technology, Guangzhou 510006, PR China.
E-mail address: chfeng@scut.edu.cn (C. Feng).

https://doi.org/10.1016/j.envpol.2018.01.048
0269-7491/

dechlorination (BARD) has been proposed and verified to be an effective way to promote dechlorination of trichloroethene (TCE) (Aulenta et al., 2007), tetrachloroethene (PCE) (Strycharz et al., 2008), pentachlorophenol (PCP) (Zhang et al., 2014), and 2-chlorophenol (Strycharz et al., 2010). We have also previously demonstrated that electrical stimulation shows substantial promise as a strategy for bioremediation of sediment contaminated with PCBs (Yu et al., 2016, 2017; Liu et al., 2017a). Taking 2,3,4,5-tetrachlorobiphenyl (PCB 61) as an example, we found that the utilization of a sediment-based bioelectrochemical reactor (BER) with low-voltage electrical fields significantly accelerated PCB 61 dechlorination, as evidenced by an apparent increase in its rate and efficiency obtained under the closed-circuit conditions compared to an anaerobic digestion system without electrical stimulation.

The principle of the BARD process lies in the fact that the application of electrical fields (referring to the applied potential exerted at an electrode surface) can stimulate the growth and enrichment of naturally existing dechlorinating microorganisms, which are ultimately the core "workers" driving the relevant microbial reactions. These bacteria are also named as organohalide-respiring bacteria (OHRB) that are capable of colonizing the electrode surface and establishing redox interactions with the electrode (Aulenta et al., 2016; Field and Sierra-Alvarez, 2008). Thus far, however, there is limited information available in the literature, revealing specific OHRB that can directly or indirectly interact with the electrode and that are responsible for dechlorination. The only OHRB in pure culture, reported previously to enable reductive dechlorination with a cathodically applied potential, are *Geobacter lovleyi* dechlorinating tetrachloroethene (PCE) to *cis*-dichloroethene (*cis*-DCE) (Strycharz et al., 2008) and *Anaeromyxobacter dehalogenans* reducing chlorophenol to phenol (Strycharz et al., 2010) with the cathode as the electron donor. There are also some OHRB detected from the mixed consortia including *Dehalobacter* and *Desulfovibrio*, which could contribute to the enhanced reduction of pentachlorophenol (PCP) at a biocathode with the cathodic potential (Zhang et al., 2014), and *Dehalococcoides*, which was possibly involved in the dechlorination of TCE to *cis*-DCE directly utilizing electrons from a negatively polarized electrode (Aulenta et al., 2007). In fact, any shift in the environmental factors, operation conditions, and inoculation time will directly influence bacterial communities of OHRB and other related communities. However, bacterial diversity in the BARD system is still largely unexplored; in particular, none of the previous studies have investigated the time-dependent variations in bacterial composition.

The commonly used method for electrical stimulation is the application of a constant potential polarized to the working electrode, which is known as the chronoamperometry method. The resulting current as a function of time provides important information of faradaic processes occurring at the electrode, and the integration of currents over long time intervals is helpful for understanding the efficiency of electron utilization for the BARD process. The rate of extracellular electron transfer (EET) is a predominant indicator with respect to the extent and efficiency of the bioelectrochemically dechlorination driven by OHRB. In general, the electrochemical impedance spectra (EIS) measurements, known as a non-destructive characterization tool, are frequently adopted for acquiring the data of the change transfer resistance that is inversely proportional to the ease of EET. Over the past years, most of the literature articles (Dominguez-Benetton et al., 2012) have used EIS to obtain information of ohmic and charge transfer resistances with respect to the anodic electron transfer that is involved in the bioelectrochemical process using the anode as the electron acceptor. Only one report published by (Aulenta et al., 2010) used EIS to characterize the charge transfer resistance of a dechlorinating biocathode that serves as the electron donor. No information is available in the literature on the time-dependent impedance variations of cathodic biofilm, which is indeed of crucial importance in understanding the electron transfer rate and thereby the dechlorination performance.

In this study, we make the first attempt to evaluate time-dependent variations in the bacterial community and the electrochemical property of the cathodic biofilms that were enriched from a surfactant-amended sediment-based bioelectrochemical reactor (BER). A BER with addition of surfactant was previously demonstrated to achieve enhanced reductive dechlorination of 2,3,4,5-tetrachlorobiphenyl (PCB 61). Here, a more reducing cathodic potential (−0.45 V *vs.* SHE) compared to that previously applied (Yu et al., 2017) was used to drive the BARD process with greater energy forces. In addition, the effect of two surfactants (Tween 80 and rhamnolipid) on PCB 61 reduction in the BERs was examined. For comparisons relating to the extent and the rate of PCB 61 dechlorination, the reactors were also run without electrical stimulation of the cathode.

2. Materials and methods

2.1. Chemicals

All PCB standards were of >99% purity and were obtained from AccuStandard (New Haven, CT, USA). Further, *n*-Hexane, dichloromethane, and acetone were of HPLC-grade and were obtained from ANPLE (Shanghai, China). The reagent-grade surfactant, polyoxyethylene sorbitan monooleate, known as Tween 80, was purchased from Sigma-Aldrich. The biosurfactant rhamnolipid was obtained from Huzhou Zijin Biological Technology Company (Zhejiang, China).

2.2. Configuration and operation of sediment-based BER

The sediment-based BER was constructed according to a method previously reported in the literature (Yu et al., 2016, 2017). The BER has two chambers (each with a void volume of 300 mL) separated by a cation exchange membrane (Qianqiu Corporation, Zhejiang, China). Two pieces of graphite felt (6.0 cm × 5.5 cm × 0.5 cm) were placed into the two chambers and connected to an external circuit using a titanium wire (0.6 mm): one was horizontally positioned on the bottom of the cathode chamber and used as the cathode, and the other was vertically placed in the anode chamber and used as the anode. Conductive graphite granules with diameters between 3 and 5 mm (Beijing Sanye Co. Ltd, China) were added to the cathode chamber in order to enlarge the accessible surface area of the cathode. This reduced the effective volume of the cathode chamber to 230 mL. The graphite granules were cleaned by soaking in 37% HCl solution for 24 h, followed by thorough rinsing with deionized water and drying overnight at 100 °C. A CHI1000C potentiostat (Chenhua Corporation, Shanghai, China) in the three-electrode mode was then applied to control the cathodic potential, with the cathode as the working electrode, the anode as the counter electrode, and a saturated calomel electrode (SCE, 0.24 V *vs.* standard hydrogen electrode, SHE) placed in the cathode chamber as the reference electrode. Unless stated otherwise, all potentials reported in this paper were in reference to SHE. The BER was sterilized in an autoclave before inoculation.

Seed sediments were collected from 40 cm below the water interface in a small river (23°36′ 12.30″ N, 113°04′ 38.84″ E) near Longtang town (Qingyuan, Guangdong, China), where an electronic waste recycling site has been in operation for forty years. Live and sterilized sediments were thoroughly homogenized in an anoxic