



Enhanced biodegradation of hydrocarbon-contaminated sediments using microbial fuel cells

Jeffrey M. Morris^a, Song Jin^{b,c,*}

^a Stratus Consulting, Boulder, CO 80302, USA

^b Advanced Environmental Technologies LLC, Fort Collins, CO 80525, USA

^c Department of Civil and Architectural Engineering, University of Wyoming, Laramie, WY 82071, USA

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ABSTRACT

A sediment microbial fuel cell (MFC) was tested to determine if electron transfer from the anaerobic zone of contaminated sediments to the overlying aerobic water could facilitate an enhanced and aerobic equivalent degradation of total petroleum hydrocarbons (TPH). Results indicate that voltages as high as 190 mV (2162 mW/m³) were achieved in a sediment MFC with an anode buried in sediments containing TPH concentrations at approximately 16,000 mg kg⁻¹. Additionally, after approximately 66 days, the TPH degradation rates were 2% and 24% in the open-circuit control sediment MFC and active sediment MFC, respectively. Therefore, it appears that applying MFC technology to contaminated sediments enhances natural biodegradation by nearly 12 fold. Additionally, a novel sediment MFC was designed to provide a cost-effective method of passive oxidation or indirect aerobic degradation of contaminants in an otherwise anaerobic environment. In addition, the use of a wicking air cathode in this study maintained dissolved oxygen concentrations 1–2 mg l⁻¹ higher than submerged cathodes, demonstrating that this technology can be applied to environments with either aerobic or anaerobic overlying water and an anaerobic matrix, such as shallow lagoon, ponds, and marshes, and groundwater.

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1. Introduction

A microbial fuel cell (MFC) provides anaerobic bacteria on its anodic side with an alternative electron acceptor (AEA) in the form of a solid-state anode, which is connected through an electrical circuit to a cathode in contact with oxygen or other oxidizing compounds. This system allows anaerobic bacteria to pass electrons onto the anode while oxidizing various compounds (e.g., organic materials and reduced metals including contaminants) in the absence of terminal electron acceptors (TEAs) such as oxygen, nitrate, or sulfate (e.g., Logan et al. [1]) in the immediate vicinity. The presence of TEAs such as sulfate and nitrate are necessary for bioremediation of hydrocarbon contaminants under anaerobic conditions where the contaminants serve as substrate for capable microbial populations such as sulfate reducing and denitrifying bacteria; therefore, a lack of or deficiency in these TEAs could result in a decreased rate of bioremediation or no degradation at all (e.g., Boopathy [2]). In a previous study, we determined that the MFC system can enhance the rate of diesel biodegradation by >2.6 fold in anaerobic groundwater, providing the microbial consortium with

an inexhaustible source of AEA in the form of the solid anode and the TEA of oxygen at the cathode [3,4]. The purpose of this work was to report for the first time that MFC technology can also be utilized to enhance total petroleum hydrocarbon (TPH; compounds eluting with n-alkane markers ranging in size from C-18 to C-50) biodegradation in contaminated sediments. Additionally, a novel MFC design is described for field applications in anaerobic sediment or groundwater with anaerobic overlying water (e.g., shallow ponds and marshes).

2. Materials and methods

2.1. Passive oxidation of contaminated sediments

This experiment was to determine if an MFC system could enhance the biodegradation of hydrocarbon-contaminated sediments. Sandy sediment was collected from a contaminated beach on the Pacific Coast of the United States and shipped on ice to Western Research Institute in Laramie (Laramie, WY, USA). Following baseline characterization, 200 g of sediment was mixed with 28 ml of anoxic medium [5] under anaerobic conditions to form a sediment paste. This paste was divided evenly and added to two 100-ml glass test tubes and an additional 30 ml of the medium was added to each test tube to bring the overlying water level to the top of each tube. The sides of each tube were covered in aluminum foil

* Corresponding author at: Advanced Environmental Technologies LLC, Fort Collins, CO 80525, USA. Tel.: +1 307 461 3294; fax: +1 307 461 3294.

E-mail address: sjin@uwyo.edu (S. Jin).

(tops were left open) and placed in a water bath at 30 °C. One test tube contained a carbon cloth anode (15 cm × 6 cm; E-TEK Division, B-1/B no wet-proofing) that ran from the bottom to just over the top of the tube where it was connected to a 1 k Ω resistor that was wired to a carbon paper/platinum (Pt) cathode (E-TEK Division, A3STD5I, 0.35 mg Pt/cm²). The other tube contained only sediment and growth media with no electrodes.

2.2. Passive aeration of anaerobic environments

This experiment was to determine the feasibility of deploying an MFC system in a contaminated area with anaerobic sediments and anaerobic overlying water, which would be applicable to locations such as shallow surface water impoundment (e.g., lagoon, estuary, and marsh) or impacted groundwater with a well bore installed. Three test columns were constructed using 1.2-m long sections of 5-cm diameter PVC pipe. The bottom 20 cm was filled with organic-rich sediment from a local pond with about 100 cm of overlying water from the pond in each (Fig. 1). The first two columns were equipped with stainless steel bottle brush anodes (brush portion 2.5-cm diameter × 6.4-cm long, double spiral power brush, #50248, Gordon Brush) buried in the sediment in each column with wires leading to a 1 k Ω resistor attached to a carbon cloth cathode. The carbon cloth cathodes measured 10 cm × 4 cm and were constructed using carbon cloth (E-TEK Division, B-1/B, no wet-proofing) coated on one side with a paste made from carbon black powder with 10% Pt (E-TEK Division, C1-10 with 10% HP Pt) and Nafion[®] perfluorinated ion-exchange resin (Sigma–Aldrich, 274704). The cathode was either a wicking air cathode that was

suspended above the column with only half the cathode submerged (e.g. [6]) or a fully submerged cathode suspended about 20 cm below the water surface. The third column was a control with no electrodes (Fig. 1). Dissolved oxygen (DO) profiles were measured through the water column of each treatment at 20-cm intervals starting just below the air–water interface using a YSI DO probe and meter.

2.3. Analyses

All sediment samples were characterized for total petroleum hydrocarbons using a dichloromethane extraction, silica gel clean up and analyzed using a gas chromatograph equipped with a flame ionization detector (GC–FID), as described in [3]. Particle size analysis of the sediment was conducted using the hydrometer method described by Gee and Bauder [7]. Bulk density was determined by packing sediment into a known volume and then determining the moisture content by drying at 105 °C for 12 h. Electrical conductance, pH, and major anions were measured in the water collected from a 1:1 sediment:reverse-osmosis water paste extraction that was agitated overnight. Sediment extractions for total metals and major cations were conducted through acid digestion (USEPA Method 3050b). Total organic carbon (TOC) of the sediment was measured with a TOC solid state matrix analyzer. The voltage across the resistors on all MFCs was continuously monitored every 3–10 min using a data logger (ADC-16; Pico Technologies Limited, UK) connected to a computer.

3. Results

3.1. Passive oxidation of contaminated sediments

The initial sediment characterization indicated a high conductivity (4.5 mS/cm), consistent with the fact that this sediment is from a marine environment. Additionally, the TPH concentration was also high (18,750 mg kg⁻¹) and the TOC and TPH concentrations were within <1% of each other, indicating that most of the TOC in the sediment was in the form of petroleum hydrocarbons (Table 1). This is significant because it demonstrates that the majority of substrate available to bacteria in the sediments in the test tubes was in the form of petroleum contaminants and, therefore,

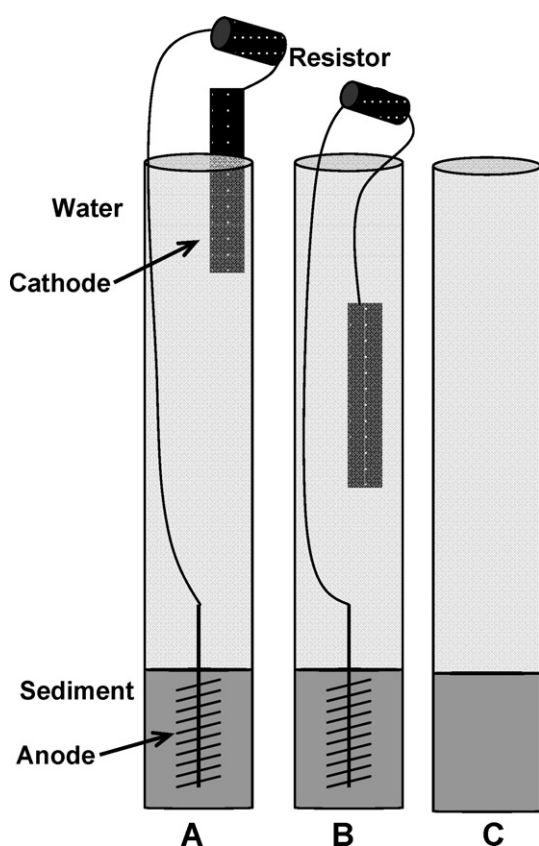


Fig. 1. Sediment column design for passive aeration experiments. Overall length of the columns is 120 cm with approximately 20 cm of sediment and 100 cm of overlying water. Column descriptions: (A) the column with the anode buried in the sediment and a wicking cathode, (B) the column with the anode buried in the sediment and a fully submerged cathode, and (C) this column was a control with no electrodes.

Table 1
Baseline analysis of contaminated sediment.

Parameter	Value
pH	7.2
Conductivity (mS/cm)	4.54
Bulk density (g/m ³)	1.56
% sand	52.3
% silt	30.5
% clay	17.2
USDA textural classification	Sandy loam
	mg kg ⁻¹
Total organic carbon	18,590
Total petroleum hydrocarbons	18,750
Chloride	228.1
Nitrate	2.5
Sulfate	385.3
Calcium	9550
Magnesium	1120
Potassium	935
Sodium	585
Aluminum	3450
Copper	22.5
Iron	4455.0
Manganese	76.8
Nickel	1.1
Zinc	23.3

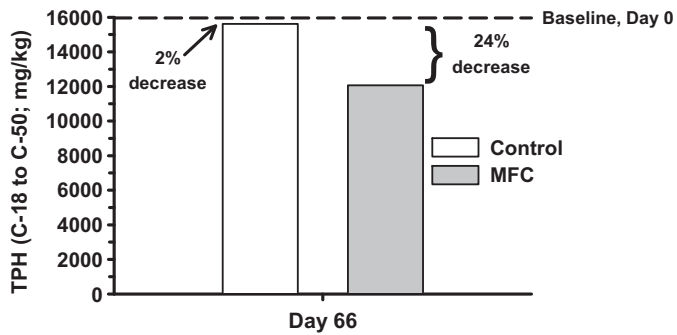


Fig. 2. Decreases in total petroleum hydrocarbons (TPH) over 66 days in the control (no electrodes) and MFC (anode and cathode) treatments.

any power produced from the MFC was likely due to contaminant oxidation by the bacteria.

The initial TPH concentration in the sediment (after being mixed with growth media) was $15,958 \text{ mg kg}^{-1}$ on day 0. After 66 days the concentrations in the control and MFC decreased to $15,621$ and $12,066 \text{ mg kg}^{-1}$ or a decrease of 2.1% and 24.4%, respectively (Fig. 2). The average voltage generated from the MFC during TPH degradation over 66 days was 25 mV with spikes ranging from about 60 to 190 mV (Fig. 3). Given the sediment volume of about 70 ml in each test tube, the average current density was calculated to be 37 mW/m^3 and the current density ranged from about 216 to 2162 mW/m^3 throughout the experiment based on continuous voltage measurements.

3.2. Passive aeration of anaerobic environments

The average voltages in the wicking cathode column and the submerged cathode column were 124 and 34 mV, respectively, over 20 days (Fig. 4). The voltage in the submerged cathode column increased briefly to about 140 mV during aeration but aeration did not affect the voltage in the wicking cathode column. The DO profiles measured 12 days after startup, 24 h after aeration, and 6 days after aeration demonstrate that the control and submerged cathode columns tended towards anoxic conditions with DO concentrations $< 1.0 \text{ mg l}^{-1}$ while the wicking cathode column maintained DO concentrations from 1 to 2 mg l^{-1} higher than the control or submerged cathode columns (Fig. 5). The optimal external resistance for the wicking and submerged cathode columns was about $4 \text{ k}\Omega$

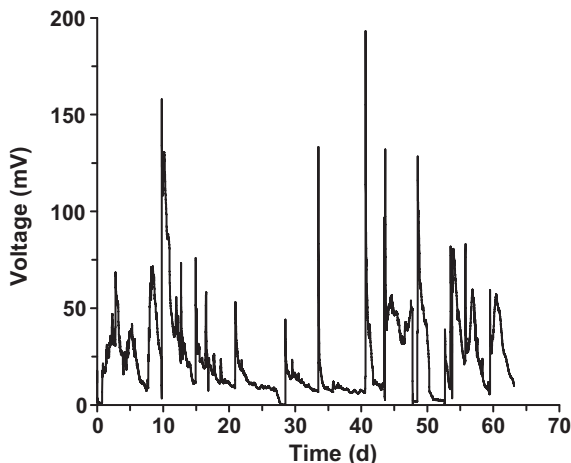


Fig. 3. Voltage measured across a $1 \text{ k}\Omega$ resistor from the MFC treatment over 66 days.

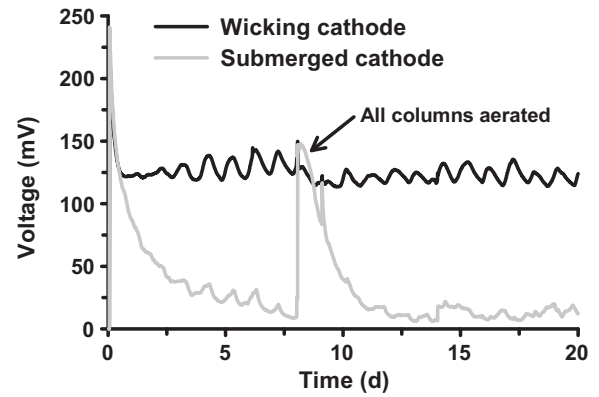


Fig. 4. Voltage measured across a $1 \text{ k}\Omega$ resistor from the two MFC sediment columns.

and the internal resistance was estimated from $3.9 \text{ k}\Omega$ to $4.2 \text{ k}\Omega$ (Fig. 6).

4. Discussion

Over the last several years, researchers studying the field of MFC systems have accomplished many significant advances in the technology and application of MFCs in aqueous environments (e.g. [1,8,9]) and sediments (e.g. [10–12]). These researches have

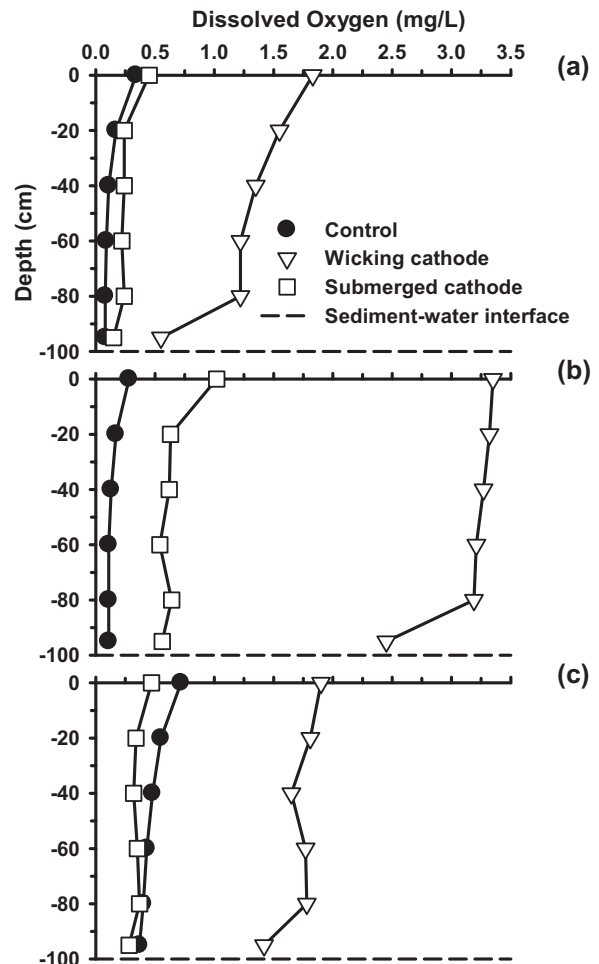


Fig. 5. Dissolved oxygen (DO) profiles in each sediment column 12 days after start-up (a), 24 h after each column was aerated to $>5 \text{ mg DO/l}$ (b), and 6 days post aeration (c).

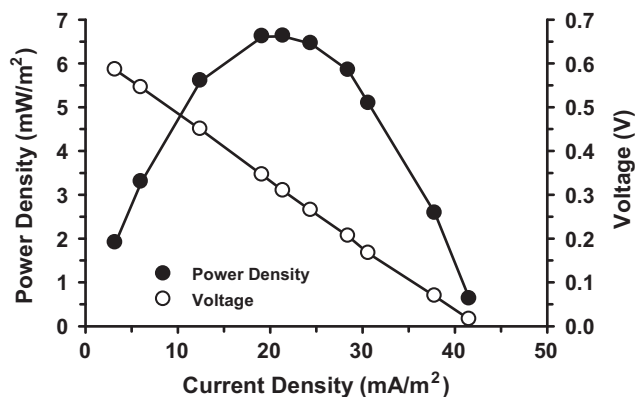


Fig. 6. Power density and polarization curves measured in the wicking cathode sediment column at external resistances ranging from 50 k Ω to 100 Ω . Based on the power density and polarization curves the optimal external resistance and internal resistance were \sim 4 k Ω and 4163 Ω , respectively.

been focusing on power generation while utilizing substrates or to reduce chemical oxygen demand (COD) loads in waste streams. Our research investigates the MFC system as a means for passive oxidation and enhancement of remediation of contaminated groundwater and sediments. The goal of passive oxidation is to enhance natural biodegradation by introducing an inexhaustible supply of electron acceptors into a contaminated system using MFC technology. This is accomplished by linking the contaminant-oxidizing bacteria in the contaminated anaerobic environment to oxygen via the electrical connection between the MFC anode in the anaerobic environment and the cathode in an aerobic environment (i.e., water/air interface). In this work we describe and quantify, for the first time, the use of an MFC system to enhance the natural degradation of petroleum hydrocarbons in contaminated sediments. The rate of TPH degradation in the MFC setup increased by >11.5 fold over background rates during the course of this experiment. Therefore, the presence of the anode increased the microbial activity in the sediment resulting in more contaminant degradation. Additionally, the lack of degradation in the control suggests that the limiting factor to biodegradation was likely the lack of TEAs or favorable TEAs. Our previous work [13] has also demonstrated that the realized thermodynamic favorability of the MFC anode may be closer to nitrate than conventional measurements indicate (e.g. [14]).

The focus of this work was to use MFC technology to enhance biodegradation in sediments. Power generation was more a concurrent process than an objective to pursue. The power production was low in our sediment MFC treatment (average of about 37 mW/m³), due to the high internal resistance (4163 Ω) in the sediments. In addition, the seemingly low efficiency in power generation of such a sediment MFC did not impact its enhancement of biodegradation. This indicates that the power efficiency loss was probably limited to the electron transfer from sediment to the anode, rather than during the electron release from organic compounds into the matrix (bio-oxidation). Therefore, the material requirements for implementing a sediment MFC for the purpose of enhanced biodegradation is less stringent in MFCs for power generation, as previously reported.

The lack of efficient electron acceptors is common in many contaminated sediment environments, which suggests that utilizing sediment MFC technology to supply an unlimited source of electron acceptors to indigenous microbial populations could significantly enhance natural attenuation rates by enhancing biodegradation. The implications of this study are wide reaching as many impaired benthic environments suffer from excessive organic contaminant and (or) nutrient loading resulting in a depletion of terminal electron acceptors in the anaerobic zone of the sediment. This anaerobic

zone could lie just millimeters below the water–sediment interface and a lack of terminal electron acceptors would effectively halt biodegradation (i.e., natural attenuation). The application of MFC technology for sediment remediation may be used as an alternative or augmentation to dredging, which is not feasible in sensitive areas that would be adversely affected by the complete removal of the upper layers of benthos (i.e., wetlands, marshes, and most aquatic environments) and potential release of metals and other environmental hazards. Furthermore, by enhancing the breakdown of excessive organic carbon sources (hydrocarbon contaminants, municipal or industrial waste, etc.) and (or) elevated nutrient loads, sediment MFCs will also be useful for remediating anoxic dead zones in impacted benthic environments (e.g. [15]). These column experiments also demonstrate that MFC technology may also be utilized to passively aerate pelagic environments through a reduction in the diffusion rate of oxygen out of these zones and into sediments by providing a direct link from the anaerobic zone to an oxygen rich environment such as overlying water or the air–water interface that by-passes the oxygen-depleted zone and allows more oxygen to accumulate. This would hold significant ecological benefits including minimizing eutrophication of sensitive water bodies and maintaining healthier, less organic-rich sediments.

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