Electrochimica Acta 187 (2016) 567–575

Contents lists available at ScienceDirect

Electrochimica Acta

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

Effect of polarity-reversal on electrokinetic enhanced bioremediation of Pyrene contaminated soil

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ARTICLE INFO

Article history:
Received 17 September 2015
Received in revised form 17 November 2015
Accepted 19 November 2015
Available online 22 November 2015

Keywords:
electro-bioremediation
soil
polarity-reversal
biodegradation

ABSTRACT

Polycyclic aromatic hydrocarbons (PAHs) contaminated soil was studied under electrokinetic enhanced biodegradation with polarity-reversal. The effects of polarity reversal were investigated on electric current, pH, microbial counts and the spatial distribution of pyrene biodegradation rate during the application of electro-bioremediation. Three types of treatment were conducted: PYR-degrading microbial treatment only (Bio), electro-bioremediation without polarity-reversal (EK-Bio), electro-bioremediation with polarity-reversal at an interval of 2 hours (EK-Bio-PR). After 42 days, EK-Bio-PR maintained a neutral soil pH (7.20) and relatively stable electric current. After running for 17 days, the electric current stabilized around 10 mA in EK-Bio-PR, and which was 5 times than that in KE-Bio. Polarity-reversal enhanced microbial counts, and improved the PYR degradation. The bacterial counts reached up to 4.8×10^{11} cfu/g and the best PYR degradation efficiency was 55.9% in EK-Bio-PR at the end of experiment. Thus, the application of electrokinetics with periodic polarity-reversal enabled better microbial growth and biodegradation, which further improved the overall removal of PAHs pollutant.

1. Introduction

Polycyclic aromatic hydrocarbons (PAHs) are presently considered to be mutagenic and/or known carcinogens [1,2]. PAHs have been found throughout a list of priority hazardous substances compiled by the U.S. EPA and the Agency for Toxic Substances and Disease Registry [3]. PAHs were listed as representative hydrophobic organic contaminants mainly because they are generally hazardous, ubiquitous in the environment, and were found in many contaminated sites, such as former manufactured gas plant sites and wood treatment facilities [4].

Bioremediation is a promising technology for in-situ treatment of PAHs contaminated soils because of its relatively low cost and environmental acceptability [5]. However, one major obstacle of bioremediation to PAHs contaminated soil is the limited access of microorganism to pollutants, especially in clayey soil [6]. Soil pH and nutrient supply to microorganism may also be limiting factors for such bioremediation.

Electrokinetics (EK) is an enhancing technique which is used to overcome the limitation of bioremediation, by increasing soil temperature and bioavailability of nutrients. EK can be used particularly effectively in fine grained soils of low hydraulic conductivity, which is different from other in-situ methods [7]. Several studies have shown that EK-Bio was effective for remediation of PAHs in clayey soil [8–10]. When direct current (DC) electric fields were applied to contaminated soil via electrodes placed into the ground, electrolysis occurred between the anode and the cathode. An acid front zone and a base zone were generated near the electrodes. Both zones moved towards each other and met in mid-way, causing a pH jump. The pH jump is detrimental to microorganism and thus hampers the efficiency of bioremediation. To overcome the drawback, a series of methods were used to control the pH, such as injecting solution [11–13], fixing the cathode and approaching anodes [14,15], electrolyte circulation [16,17] and non-uniform electrokinetics [5,18]. Polarity reversal was proposed for neutralizing soil pH and stimulating bioremediation of organic contaminated soil [4,9,19]. Bioremediation coupled with EK-PR could increase the temperature, bioavailability and transformation of organic contaminants without causing extreme pH conditions at the electrodes [20,21]. In addition, this technique may enhance the microbial activity for the degradation of contaminants, by producing oxygen at the anode [22]. In EK-Bio technology, the removal of organic contaminant likely depends not only on the effect of biodegradation, but also on that of electrical intensity induced stimulation [19].

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http://dx.doi.org/10.1016/j.electacta.2015.11.097
0013-4686/

However, previous studies mainly focused on the biodegradation of pollutants [13,23], and the data were not sufficient to systematically illustrate the effect of PR on electric current, pH, the microbial counts and the spatial distribution of organics biodegradation rate. Pyrene (PYR), a highly hydrophobic and persistent PAH, was used as the target pollutant in EK-Bio systems with or without PR. A mixed culture of PYR-degrading bacteria was used, which were isolated from PYR contaminated soil. The objectives of this study were to investigate the effects of electro-bioremediation on non-polar contaminant such as PYR in soil under conditions of constant versus alternating polarity electrical fields, and to identify the appropriate conditions for bacteria growth and enhanced PYR biodegradation. Furthermore, the degradation mechanism was also discussed in this study.

2. Experimental

2.1. Materials

The soil sample was taken from ground surface soil up to 20 cm deep at a random location in the city of Shenyang (Liaoning Province, China). The soil was brown earth and the main characteristics of the soil were as follows: sand, silt and clay proportions were 23.2%, 46.2% and 30.6%, respectively; soil organic matter comprised 2.4%; the total nitrogen is 0.084% and the total phosphorus is 0.042%; soil pH was 7.32. Its cation exchange capacity (CEC) was 18.63 cmol/kg. The soil was air-dried and sieved through a 2-mm sieve. The dried soil was spiked with PYR [4]. The target concentration of PYR was 150 mg/kg. Following that, the soil was left in open air at room temperature for seven days in order for sufficient adsorption to occur. Then, it was stored in non-transparent bag at 4 °C until it was needed for the experiment.

A mixed culture of PYR-degrading bacteria was used for the experiment. The bacteria were isolated from soil near a manufactured gas plant. The culture media contained PYR as the sole carbon source. The bacteria were incubated in Luria–Bertani (LB) media [19]. In this study, the bacteria were introduced into contaminated soil and included *Acinetobacter baumannii, Bacillus pumilus, Pseudomonas putida, Pseudomonas nitroreducens and Bordetella sp*. All of the bacteria are specific aerobic and strict aerobic [24].

2.2. Electrokinetic setup

Fig. 1 is a schematic representation of the EK setup and sampling sites. The apparatus system included a soil chamber of 22 cm in length and 22 cm in width and 20 cm in height, two pairs of cylindrical graphite electrodes (22 cm in length and 1 cm in diameter). A constant electical potential was applied at 1.0 V/cm during the experiment using a DC power supply and an controller capable of reversing the polarity every 5 minutes.

2.3. Testing method

The contaminated soil was inoculated with PYR-degrading bacteria at an initial concentration of 1.43×10^{10} cfu (colony forming units)/g dry soil, before being added to the soil chamber. The soil was then rehydrated to a moisture content of about 30% (w/w) with distilled water and maintained at this moisture level during the 42-day period. The wet soil was spread into the soil chamber in layers with a pressure of 0.1 kg/cm. Three sets of bench-scale experiments were conducted. Only bacteria were applied for the control experiment. Two additional soil chambers were set up, with EK-Bio-PR and EK-Bio, respectively. The soil samples were collected from ten positions in all experiments every 7 days. Each position was 5 cm apart between the anode and cathode, as shown in Fig. 1. All experiments were carried out in triplicate.

2.4. Analysis

Cultivable soil bacteria were counted using the viable cell counting method [25]. Soil pH was measured based on a 1:2.5 ratio of soil to water using a pH meter (Model PHS-3C, Shanghai, China) during the treatment process.

The PYR concentration was determined using a UPLC (Waters, USA) equipped with a Waters XSELECT CSH™ C18 column

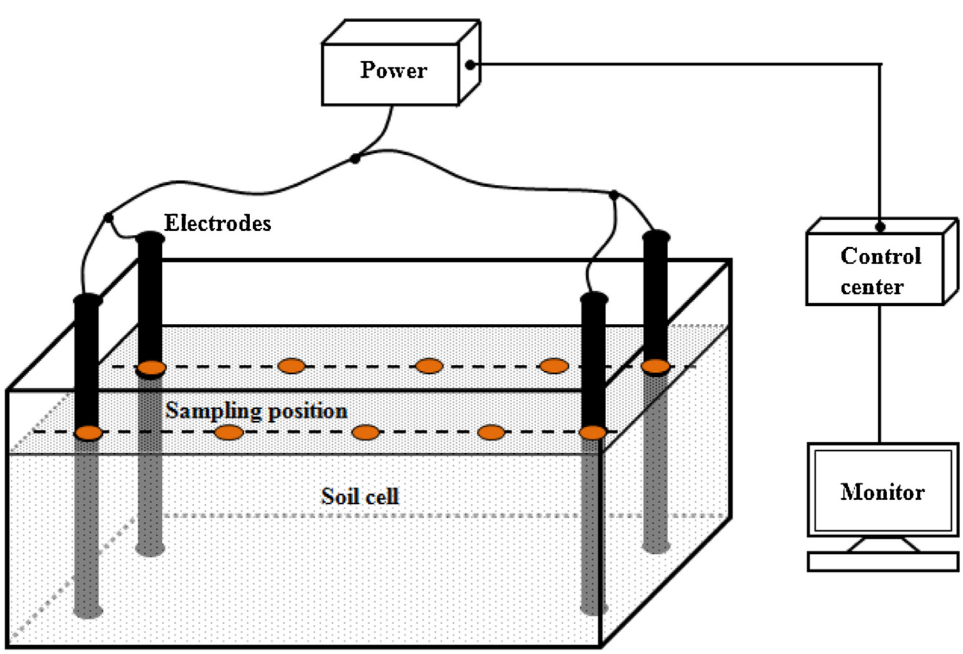

Fig. 1. Schematic diagram of Electrokinetic setup (○ sampling position).