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Mixing of an anthracene-contaminated soil: A simple but efficient remediation technique?

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ABSTRACT

Contamination of soils with polycyclic aromatic hydrocarbons (PAHs) is a serious problem in petroleum producing countries, such as México, and environment-friendly easy to apply techniques are required to accelerate the removal of the contaminants. Removal of anthracene was monitored in an arable and a pasture soil regularly mixed or amended with organic material, a non-ionic surfactant (Surfynol® 485) or earthworms (*Eisenia fetida* (Savigny, 1826)). In both soils the same results were obtained although the removal of anthracene was faster from the pasture than from the arable soil. The fastest removal of anthracene was obtained when the soil was mixed every 7 days and no contaminant was detected in both soils after 56 days. The second fastest removal of anthracene was obtained when earthworms were added to soil and no contaminant was detected in both soils after 112 days. Application of organic material that served as feed for the earthworms also accelerated the removal of the contaminant compared to the unamended soil, but application of the surfactant inhibited the dissipation of the contaminant. Only 37% of the spiked anthracene was removed from soil when surfactant was applied, while 62% was dissipated in the unamended soil after 112 days. It was found that simply mixing a soil removed anthracene faster than when earthworms were applied, while the application of the surfactant inhibited the removal of anthracene by the autochthonous soil microorganisms.

1. Introduction

Polycyclic aromatic hydrocarbons (PAHs) are a group of organic compounds with 2–13 aromatic rings. Some PAHs might be carcinogenic to humans and animals (Skupinska et al., 2004). The removal of PAHs from contaminated ecosystems is thus of great importance and different techniques have been applied to remediate hydrocarbon-contaminated soils (Khan et al., 2004). The choice of technology and remediation strategy depends on site conditions, contaminant, and the impact of the chosen technology (Khan et al., 2004). Remediation techniques are based on biological, physical, chemical, physicochemical methods or combinations of these (Van Hamme et al., 2003), but those commonly used include burying, evaporation, dispersion and washing. These technologies, however, are expensive and can lead to incomplete decomposition of contaminants (Das and Chandran, 2011), while affecting the soil microorganisms that degrade the contaminant.

Earthworms are perhaps the most important soil organisms in terms of their influence on soil properties (Udovic and Lestan, 2007). Earthworms are important processors of soil organic matter (SOM) and nutrient turnover in terrestrial ecosystems. In agroecosystems, they are often seen as beneficial organisms to crop growth and actively promoted (Fonte et al., 2010). In soil they stimulate PAHs biodegradation, and by consuming organic matter, they may reduce adsorption of the contaminant, thereby increasing its bioavailability and removal from soil (Natal-Da-Luz et al., 2012).

Although some contaminants may be soluble in water, other inherent physicochemical properties make them particularly problematic, especially, water immiscibility. The use of surfactants to enhance the removal of soil contaminants has received increased attention in recent years. Surfactants are a class of natural and synthetic chemicals that promote the wetting, solubilization, emulsification and removal of various types of organic and inorganic contaminants from soil (Das and Chandran, 2011; Wang and Mulligan, 2004). Surfactants molecules may furthermore influence the dissolution or desorption process by attaching to the PAHs-water interface. They form hemi-micelles, which may accelerate the PAHs-release and subsequently their removal from soil (Johnsen et al., 2005).

In previous research, it was found that mixing a soil accelerated the removal of anthracene (Delgado-Balbuena et al., 2013). Mixing a soil liberates organic material, such as organic contaminants, as soil aggregates are broken-up so that contaminants become available for microbial degradation. Autochthonous soil microorganisms remove PAHs from soil (Das and Chandran, 2011; Khan et al., 2004; Qasemian et al., 2012) and bioremediation techniques

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that increase the availability of a contaminant, such as mixing, will increase its dissipation. As part of a study into the techniques that remove PAHs from soil, four different methods were compared for their capacity to remove anthracene from soil. Anthracene was used as a model for PAH degradation in soil (Moody et al., 2001; Prasanna et al., 2008; Qasemian et al., 2012). However, removal of a three-ring PAHs is faster from soil than the dissipation of a five-ring PAHs such as benzo(a)pyrene (Álvarez-Bernal et al., 2006). Two soils (an arable and a pasture soil) were spiked with anthracene and amended with organic material (carrot, *Daucus carota* L.) that served as food for earthworms, the organic material plus the earthworm *Eisenia fetida* (Savigny, 1826), a non-ionic surfactant (Surfynol® 485) or mixed regularly while the removal of the contaminant was monitored in an aerobic incubation experiment. The objective of this research was to compare different strategies to remediate anthracene-contaminated soil and determine which technique might be the best to remove PAHs from soil.

2. Materials and methods

2.1. Chemicals used

Anthracene with purity >98% was obtained from Sigma-Aldrich (USA) and acetone with purity >99.7% from J.T Baker (USA). The non-ionic surfactant Surfynol® 485 was obtained from Air Products and Chemicals de México S.A. de C.V. (México). It is an ethoxylated molecule of 2,4,7,9-tetramethyl-5-decyne-4,7-diol with 30 mol of ethylene oxide (EO) per molecule ($C_{14}H_{14}(OH)_2EO_{30}$) and a molecular weight of 1546 and a critical micelle concentration (CMC) of 11.2 mmol/L (Musselman and Chander, 2002).

2.2. Sampling site, collection and characterization of soil

Two soils were used in this study. One arable soil was collected in Otumba (State of México, México) (N.L. 19° 42′, W.L. 98° 49′). Its average altitude is 2349 masl. and characterized by a sub-humid temperate climate with a mean annual temperature of 14.8 °C and average annual precipitation of 577 mm mainly from June to August (http://www.inegi.gob.mx). Details of the sampling site can be found in Méndez-Bautista et al. (2010). The sandy loam soil had a pH 7.6 and EC 1.15 dS/m and an organic C content of 7.2 g C/kg with a particle size distribution of 870 g/kg sand, 90 g/kg clay and 40 g/kg silt. The water holding capacity (WHC) was 650 g/kg soil.

The second was a pasture soil collected in Juchique de Ferrer (State of Veracruz, México) (N.L. 19° 50′, W.L. 96° 42′). The weather at the sampling site is warm with an average annual temperature of 25 °C. It is characterized by abundant precipitation in summer and early autumn averaging 1000 mm/y. The loamy sand soil with pH 5.9 and EC 1.0 dS/m had an organic C content 13.2 g C/kg and a particle size distribution of 721 g/kg sand, 42 g/kg clay and 237 g/kg silt. The WHC was 1130 g/kg soil.

Soil was sampled at random by augering 30 times the 0–15 cm top-layer of five plots of approximately 0.5 ha. The soil from each plot was pooled and as such a total of ten soil samples were obtained (five replicates of two soils). This field based replication was maintained in the laboratory study.

2.3. Experimental set-up

A total of 100 kg of each soil was contaminated with 500 mg anthracene/kg dry soil. Initially, 10 kg soil was spiked with 50 g anthracene dissolved in 4.7 L acetone and placed under vacuum in a desiccator for 30 min so that the acetone was removed from the soil. The 10 kg contaminated soil was then mixed with the remaining 90 kg soil. Five different treatments were applied to the anthracene-contaminated soil. In a first treatment, soil was amended with two adult *E. fetida* earthworms of 0.35 g obtained from INECOL (Xalapa, Veracruz, México) and with a developed clitellum. The earthworms were fed 60 g carrot every two weeks. In a second treatment, soil was amended with 60 g organic material (carrot) every two weeks. As such, the effect of the earthworms on the removal of anthracene could be differentiated from that of the organic material applied. In a third treatment, soil was mixed every 7 days for 10 min. In a fourth treatment, soil was amended with 24.9 g/kg soil surfactant Surfynol® 485 and mixed (Salomón-Hernández, 2012, unpublished M.Sc. thesis). In a fifth treatment, soil was left unamended and served as control so that remediation capacity of the autochthonous microorganisms could be determined.

Two hundred sub-samples of 500 g of both soils were added to polyvinyl chloride (PVC) tubes (diameter 10.5 cm, length 20 cm) containing a 5 cm layer of tezontle or porous volcanic rock. The amount of soil added to the PVC tubes was such that a 10 cm layer was obtained. The PVC columns were covered with perforated aluminium foil so that aeration was possible, but evaporation limited.

Each other day in the evening around 6:00 p.m., 2 g was taken from each treatment, weighted and dried overnight at 100 °C. The next morning, the dried soil was weighted, the water content calculated and the soil column adjusted to 60% WHC when necessary. The soil columns were placed in a greenhouse. After 0, 1, 3, 7, 14, 28, 56 and 112 days, a 20 g soil sample was taken from each column and extracted for anthracene with an exhaustive extraction method (Song et al., 1995).

2.4. Soil chemical analysis

The concentration of anthracene in the soil was determined using a modified exhaustive ultrasonic extraction method described by Song et al. (1995). Briefly, a 1.5 g sub-sample of soil was mixed with 3 g anhydrous sodium sulphate to form a fine powder, placed in a Pyrex tube and 10 mL acetone was added. The mixture was mechanically shaken on a vortex for 1 min, and the tubes were placed in a sonicating bath at 30–40 °C for 20 min. The extracts were separated from the soil by centrifugation at 2000 *g* for 7 min. The whole process was repeated three times. The extracts were evaporated and dissolved in 1 mL acetone. Each sample was analyzed for anthracene on a Agilent Technology 4890-D (Pennsylvania, USA) fitted with a flame ionization detector.

A HP-5 column from Hewlett-Packard with length 15 m, inner diameter 0.53 mm, and film thickness 1.5 μm was used to separate the anthracene with carrier gas He flowing at a rate of 7 ml/min. The oven temperature at 140 °C was increased to 170 °C at a rate of 2 °C/min maintained at 170 °C for 5 min. The temperature of the injector was 280 °C and that of the detector was 300 °C. The detection limit of our GC analysis was 0.3 mg of anthracene per kg of dry soil. The amount of anthracene recovered with the exhaustive technique was 98%. Although the amount of anthracene lost during the procedure was <2%, data were adjusted for these small losses.

2.5. Statistical analysis

Concentrations of anthracene were subjected to an analysis of variance using PROC GLM (SAS, 1989) to test for significant differences between the remediation techniques used. Repeated measurements, i.e. the sampling was none destructive, was considered in the statistical analysis.

3. Results and discussion

The concentration of anthracene decreased in both the arable and pasture soil and approximately 190 mg anthracene/kg dry soil was still detected in the control treatment after 112 days (Fig. 1a, b). It is well known that autochthonous microorganisms remove PAHs from soil (Hosokawa et al., 2009). The decrease in anthracene over time also followed a well known pattern with most of the dissipation occurring in the first weeks of the incubation (Contreras-Ramos et al., 2008).

Application of surfactant is known to accelerate the removal of PAHs from soil, but not always (Paria, 2008; Singh et al., 2007). It is assumed that the surfactant increases the bioavailability of the hydrocarbon by a parallel action of desorption and solubilization of the contaminant, thereby favouring the removal of the contaminant (Christofi and Ivshina, 2002). In the study reported here, the surfactant inhibited the removal of the contaminant (Fig. 1a, b). After 112 days, more than half of the applied anthracene (approximately 300 mg anthracene/kg dry soil) was not removed from the soil amended with the surfactant. Several studies reported that biodegradation of PAHs in the presence of synthetic surfactants was inhibited (Paria, 2008; Singh et al., 2007). Different processes might explain a decrease in the removal of anthracene from the surfactant-amended soil. First, the surfactant might inhibit the microbial activity in soil or the surfactant might reduce the bioavailability of anthracene (Chen et al., 2000). Second, if the surfactant is highly biodegradable, then there might be competition between carbon sources, so that the degradation of the contaminant is inhibited (Makkar and Rockne, 2003). Third, the surfactant might increase the availability of the contaminant and