EVALUATION OF SOIL ADDITIVES AND MICROORGANISMS ON THE LEACHING POTENTIAL AND REMOVAL OF CRUDE OIL AND PENTACHLOROPHENOL FROM SOIL

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INTRODUCTION

Land treatment has been accepted as a viable soil remediation technology for many classes of organic pollutants. A significant body of both laboratory and field experimental work has demonstrated the biodegradability of petroleum wastes and chlorinated organics, as well as the environmental safety of this process (Bossert et al. 1984; Lamar and Dietrich 1990; McGinnis et al. 1991). However, the physical properties of soil can impede the biological destruction of contaminants. Successful soil remediation depends on the interaction of contaminants with microorganisms living within the soil. Low permeability soils with strong sorption characteristics usually exhibit slower degradation rates. Additives can be applied to the soil to maximize the interaction between microorganisms, thus increasing degradation rates. However, these additives may also affect contaminant migration by accelerating or hindering the mobility of target chemicals in the soil. The amount and concentration of organic pollutants that leach out of contaminated sites are receiving considerable attention. Increased awareness and concerns are primarily due to environmental mandates and regulations to reduce the impact of the contaminants on rivers, lakes, streams, and oceans. The Environmental Protection Agency (EPA) has adopted a TCLP test for estimating the leaching potential of organic and inorganic pollutants. TCLP measures a constituent's potential to contaminate groundwater. The rule defers regulation of certain petroleum-contaminated wastes, such as underground storage tank sites, and exempts polychlorinated biphenyls (PCBs) waste already regulated under the Toxic Control Substance Act.

The overall goal of this project was to evaluate the use of fungi and different soil additives on bioremoval and the leaching potential of two different crude oils and pentachlorophenol (PCP).

METHODS

The studies were carried out in three phases. In Phase 1A peat, kenaf, and an oil degrading fungus, *Cladosporium sp.*, were evaluated for the degradation and leaching of crude oil in sand. Four-liter aquaria were constructed with a perforated metal divider that retained the sand but allowed water to pass through to the collection end. These aquaria were tilted approximately 15 degrees. Sand was contaminated with 1% (vol/vol) of Saudi Arabian crude oil and thoroughly mixed. One hundred and fifty grams of contaminated sand were placed into three aquaria for controls. Three aquaria received contaminated sand plus 1% peat (wt/wt) (Exsorbet™ brand, Exsorbet Industries, Mulberry, Arkansas), and the remaining three aquaria received contaminated sand with 1% kenaf (wt/wt) core plus *Cladosporium sp.* fungus. This fungus was isolated at a super-fund site in California and shown to be a biodegrader of nonchlorinated organics (Borazjani et al. 1991). Fifty ml of fungal culture, previously grown in potato dextrose broth (Difco, Detroit, Michigan) for three weeks, were mixed with kenaf and sand. Kenaf was primarily used as a carrier for this fungus. Each aquaria for peat and kenaf treatment also received 150 grams of contaminated sand. Samples were placed in the upper ends of the aquaria and distilled water was added several times daily to simulate tide action. Leachate samples were collected daily during a one-month period and sand samples from each aquaria were collected at Day 30 for total petroleum hydrocarbons (TPH) analysis by Modified Standard Method 5520 (Standard Methods 1987).

In Phase 1B of this experiment, sand was contaminated with 2% (vol/vol) Saudi Arabian crude oil with the following treatments: controls (sand only contaminated with 2% crude oil), peat (contaminated sand amended with 3% wt/wt peat), peat plus fungus (contaminated sand amended with 3% peat wt/wt and *Cladosporium sp.* culture), and kenaf (contaminated sand amended with 3% wt/wt kenaf). Each aquaria contained 150 grams of contaminated sand excluding their amendments. Fifty ml of fungal culture was added to each aquaria for the fungal treatment. Watering and sampling procedures were the same as in the Phase 1A experiments. No sand samples were taken.

In Phase 2, clean sand was contaminated with 3% (wt/wt) Iranian crude oil and amended with the following specific additives (wt/wt):

Peat (contaminated sand with 1% peat)

Kenaf (contaminated sand with 1% kenaf)

Absorb-N-Dry (contaminated sand with 5% Absorb-N-Dry) (Balcones Minerals Corporation, Flatonia, Texas)

Safety-Absorbent (contaminated sand with 5% Safety-Absorbent) (Molten Company, Memphis, Tennessee)

Vermiculite (contaminated sand with 3% Vermiculite)

Control: (contaminated sand alone)

One hundred sixty five grams of contaminated sand excluding additives were placed in each aquaria. Sea water from the Gulf of Mexico was used to flush the treatments three times a day, and water samples were collected daily for a period of one month. A TPH analysis was performed on the water with the standard method listed in Phase 1.

In Phase 3, PCP and TPH contaminated soil was obtained from a wood-treating facility and sieved with a 2 mm mesh screen. After thorough mixing, 200 g of soil was placed into each of nine amber dishes. Three dishes were amended with 1% peat (wt/wt) and three with 1% peat inoculated with a 50 ml culture of *Phanerochaete chrysosporium*. This fungus was purchased from the American Type Culture Collection (ATCC 24725, Rockville, Maryland). The remaining three dishes contained only PCP contaminated soil. The dishes were incubated at 24°C with a moisture content of 15% (dry wt. basis). The dishes were monitored weekly for aeration and moisture content. Samples were taken at 45-day intervals and analyzed for PCP [EPA Method 3540 and 8151 (U.S. EPA 1986)]. Soil samples from Day 90 were also tested for PCP by the toxic characteristic leachate procedure (TCLP). Leachate samples were extracted by EPA Method 3520 and analyzed by Method 8151 (U.S. EPA 1986). Soil TPH was extracted by EPA Method 3540 and analyzed by Modified Standard Method 5520.

RESULTS AND DISCUSSION

In both Phase 1A and 1B experiments, contaminated sand amended with peat exhibited lower TPH concentrations in the collected leachates (Tables 1 and 2). In the Phase 1B experiment, peat showed a 75% lower TPH concentration than the kenaf and control treatments. The addition of peat plus fungus was not as effective due to wetting by the fungal solution. The peat treatment alone also performed slightly better than the kenaf plus fungus treatment in the Phase 1B experiment. The performance of peat is probably due to its superior petroleum absorption properties.

Leachate results from Phase 2 are summarized in Figure 1. Peat and Safety-Absorbent (SA) amended sands released the lowest concentration of TPH, while kenaf and Absorb-N-Dry (Ab) treatments were virtually identical. The vermiculite treatment released the highest TPH concentration into the leachate among all additives.

Phase III demonstrated that peat treatments were also highly effective in enhancing the biodegradation of PCP (Table 3). The addition of the fungus doubled the degradation rate of PCP in comparison to peat treatment alone. There were no significant differences among treatments for TPH degradation (Table 4). TCLP analysis of day 90 samples showed almost twice as mich of PCP in fungus-treated leachate than the control treatments. Fungal metabolite may have altered the absorption of PCP to the soil particles, making the PCP more bioavailable for degradation as well as more leachable.

CONCLUSION

The addition of peat to sand contaminated with crude oil can be a safe treatment to reduce the levels of TPH in leachate. However, more studies are needed to determine the effectiveness of peat for other soil types. Kenaf additions did not reduce the TPH concentrations in leachate as anticipated. Peat can also be used along with known degraders, such as the white rot fungus, for the bioremoval of chlorinated compounds. More studies are needed to determine the limiting factors responsible for the slow biodegradation rate of TPH and PCP during the first several weeks of application.

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Table 1. Concentration of TPH in soil and leachate for Phase 1A*.

Treatment	Soil TPH (ppm)	Leachate TPH (mg/ml)
Control	7600	43
Peat	7600	13
Kenaf plus Fungus	7400	18

* each figure represents an average of three replicates.

Table 2. Concentration of TPH in leachate for Phase 1B*.

Treatment	Leachate TPH (mg/ml)
Control	100
Peat	25
Peat plus Fungus	53
Kenaf	101

* each figure represents an average of three replicates.

Figure 1. Concentrations of oil residue (TPH) in leachate samples (mg/L) for Phase 2. Each bar is an average of three replicates.

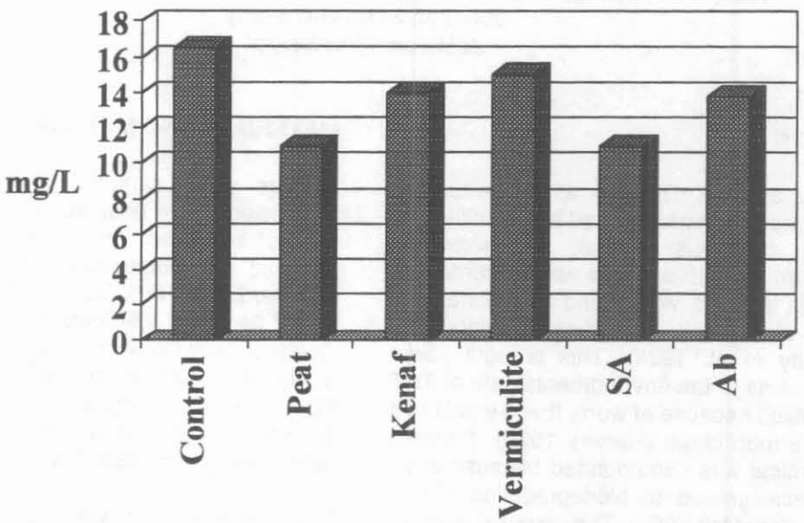

Table 3. PCP concentration (ppm) in soil and leachate for Phase 3*.

Sampling Day	Control	Peat	Peat plus Fungus
0	806	822	720
45	745	647	797
90	778	565	245
% degradation	3	31	66
TCLP	1.6	2.1	3.1

* each figure represents an average of three replicates.

Table 4. TPH concentration (ppm) in soil for Phase 3*.

Sampling Day	Control	Peat	Peat plus Fungus
0	4583	3950	3317
45	1792	2042	2983
90	2369	1795	1777
% degradation	48	54	46

* each figure represents an average of three replicates.