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BIOREMEDIATION OF TERESTRIAL FUEL SPILLS

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الإصلاح البيولوجي للتربة من الوقود المنسكب

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إن تصدع الأنابيب وإنهيار الخزانات وغير ذلك من عمليات الإنتاج وكذلك حوادث التخزين والنقل تلوث التربة بالهيدروكاربونات ، وأحياناً على نطاق واسع . يلفت الإصلاح البيولوجي حالياً الكثير من الإنتباه كتقنية إصلاحية .

تهدف الدراسة إجراء تجارب في نطاق المختبر لمعرفة أي منسكب من الوقود يمكن تنظيفه بعملية معالجة للتربة بالاصلاح البيولوجي وبسعر اقتصادي .

إن المعالجة بالإصلاح البيولوجي تتكون من تعديل الرقم الهيدروجيني للتربة ومن ثم تسميدها وفلاحتها . وقد تم تقييم فعالية الإصلاح البيولوجي ، في نطاق المختبر ، على تنظيف التربة من الملوثات كوقود الطيران ووقود الديزل والوقود الثقيل . المتغيرات العملية هي درجة حرارة الإحتضان واللامعالجة والمعالجة بالإصلاح البيولوجي ومراقبة التبخر السام .

تم تحديد المتبقى من الهيدروكربونات باستخدام كروماتوغرافيا الغاز وفي حالة الوقود الثقيل بطريقة تحديد الوزن المتبقى .

لقد تم الحصول على قيم النقاط الخمس الموجودة على المنحنيات من التغيرات العملية المذكورة آنفاً . إن نواتج التقطير الوسطي ووقود الطيران ووقود الديزل المتزايدة مع الإستمرارية تستجيب للمعالجة بالإصلاح البيولوجي تحت شروط التجربة العملية .

بفضل المعالجة بالإصلاح البيولوجي يمكن تخفيض مستوى التلوث الهيدروكاربوني للتربة إلى أدنى مستوى في فترة موسم زراعي واحد .

ABSTRACT

Pipeline rupture, tank failures and various other production, storage and transportation accidents create hydrocarbon contaminated soils, occasionally on a large scale. Bioremediation is currently attracting a lot of attention as a remedial technique. This study is designed to test, on the laboratory scale, what type of fuel spills could be cleaned up by a cost-effective bioremediation approach based on a land treatment process.

A bioremediation treatment that consisted of pH adjustment of soil, fertilisation, and tilling was evaluated on the laboratory scale for its effectiveness in cleaning up a soil contaminated by jet fuel, diesel oil, or heavy fuel oil. Experimental variables included; no treatment, incubation temperature, bioremediation treatment, and poisoned evaporation controls. Hydrocarbon residues were determined by gas chromatography or, in the case of heavy fuel oil, by residual weight determination. Five-point depletion curves were obtained for the prescribed experimental variables. The medium distillates, jet fuel and diesel oil increased in persistence in the listed order but responded well to bioremediation treatment under test conditions. With bioremediation treatment, it is possible to reduce hydrocarbons to insignificant levels in contaminated soils within one growing season.

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INTRODUCTION

Pipeline rupture, tank failures and various other production, storage and transportation accidents create hydrocarbon contaminated soils, occasionally on a large scale. Soil that is accidentally contaminated by petroleum fuel spills is classified as hazardous waste [1]. Hydrocarbon contaminated soil fails to support plant growth and is a source of ground water contamination. The currently accepted cleanup and

disposal method, by excavation and incineration or transport and burial of the soil secure chemical landfills, is prohibitively expensive when large amounts of soil are involved. This often results in cleanup delays while the contaminated soil continues to pollute scarce ground water resources [2].

Bioremediation is currently attracting a great deal of attention as a remedial technique. This is because other techniques, by themselves, are either inadequate, do not permanently solve the problem or have the potential to be very expensive. Bioremediation is being evaluated by both industry and the US–EPA as one technology for cleaning up hazardous waste sites. The process usually involves the stimulation of indigenous subsurface microorganisms to degrade chemicals on-site, although there have been cases in which microorganisms with some specialised metabolic capabilities have been added. One advantage of this process is that soil excavation is not required.

This study is designed to test, on the laboratory scale, what type of fuel spills could be cleaned up by a cost effective bioremediation approach based on a land treatment process and optimised for oil sludges [3]. Petroleum hydrocarbon disappearance rates were compared in contaminated but otherwise untreated soil, in bioremediation treated soil, and in soil poisoned in order to suppress biodegradation [4].

EXPERIMENTAL

Fuels

Jet fuel, diesel fuel and heavy fuel oil were selected for use in this study. All fuel products were received from a Libyan refinery. The fuel products were initially characterised and the results are shown in Tables 1 to 4.

Table 1. Analysis of Jet Fuel used in Spill Bioremediation Study

Test Method	Test Description	Result
ASTM D1298	Specific gravity at 15/15°C	0.7969
ASTM D3242	Total acidity, mg KOH/g	0.003
ASTM D1266	Total sulphur, wt.%	0.015
ASTM D1219	Mercaptan sulphur, wt.%	0.0001
ASTM D86	Distillation	
	Initial boiling point, °C	151
	10% Vol. recovered at °C	171
	20% Vol. recovered at °C	178
	50% Vol. recovered at °C	198
	90% Vol. recovered at °C	233
	Final boiling point, °C	250
ASTM D2386	Freezing point, °C	−53
ASTM D445	Viscosity at −20°C, CSt	4.3
ASTM D240	Calorific value, kcal/kg	10987
IP 57	Smoke point, mm	28

Table 2. Analysis of Diesel Fuel Used in Spill Bioremediation Study

Test Method	Test Description	Result
ASTM D1298	Specific gravity at 15/15°C	0.8323
ASTM D445	Kin. viscosity at 37.8°C, CSt	3.48
ASTM D93	Flash point, °C	63
ASTM D97	Pour point, °C	0
ASTM D129	Total sulphur, wt.%	0.27
ASTM D3242	Total acidity, mg KOH/g	0.053
ASTM D86	Distillation	
	Initial boiling point, °C	202
	10% Vol. recovered at °C	260
	20% Vol. recovered at °C	275
	30% Vol. recovered at °C	285
	40% Vol. recovered at °C	294
	50% Vol. recovered at °C	302
	60% Vol. recovered at °C	311
	70% Vol. recovered at °C	321
	80% Vol. recovered at °C	335
	90% Vol. recovered at °C	353
	Final boiling point, °C	365
ASTM D240	Gross calorific value, kcal/kg	10945

Table 3. Analysis of Heavy Fuel Oil Used in Oil Spill Bioremediation Study

Test Method	Test Description	Result
ASTM D1298	Specific gravity at 15/15°C	0.9210
ASTM D129	Total sulphur, wt.%	0.966
ASTM D93	Flash point, °C	94
ASTM D445	Kin. Viscosity at 50°C, CSt	105
ASTM D445	Kin. Viscosity at 82.8°C, CSt	28.6
ASTM D97	Pour point, °C	30
ASTM D482	Ash content, wt.%	0.017
ASTM D1796	Water content, Vol.%	0.15
ASTM D240	Gross calorific value, kcal/kg	10537

Sample Treatment

Soils were selected from the area near to the fuel storage tank form. Soils were freshly collected for each experiment. They were partially but not completely air dried to allow sieving (2 mm diameter openings) for uniform consistency, but without damaging their biological activity. Lime ($CaCO_3$) was added to semidry soil prior to column packing for adjusting [5] the pH to 7.5. The sieved soils were packed into glass columns (outer diameter, 25 mm; length, 250 mm) at the bulk density of cores collected from the field. The resulting columns were 22 mm in diameter, and 150 mm in length. The lower end of the columns were closed with a plug and a closable drain spout. After packing, water was added to the top of the column to adjust the moisture content of the soil to 50% of its holding capacity.

Nitrogen (NH_4NO_3) and phosphorous (K_2HPO_4) fertilisers were added to keep 60 μmol N and 5 μmol of P per gram of soil, respectively. They were dissolved in water that was used for adjusting, moisture

Table 4. Compositional Analysis of Fuel Products Used in the Spill Bioremediation Study

Fuel Product	Class Composition			Carbon Range
Jet Fuel	Saturates 87.7	Olefins 0.0	Aromatics 12.3 (by Vol.%)	C8-C15
Diesel Fuel	Non-Aromatics 81.8		Aromatics 18.2 (by wt.%)	C10-C25
Heavy Fuel Oil	Saturates 74.8	Aromatics 22.1	Polar 3.1 (by wt.%)	Not analysed

content. Soil columns were fed by fuel products on top of the columns and allowing them to infiltrate by gravity flow. The maximal application rate (100 mg per g of soil) was chosen so that it would not result in either fuel or water flowing out from the soil column. The evaporation of water during incubation was compensated by addition of weighed distilled water. Weekly tilling of the soil columns was performed by inserting the stainless steel wire into the soil column 15 times. This treatment was much less effective in aerating the soil than conventional tilling in the fields.

2% mercuric chloride was used as biologically inactive poisoned control to differentiate losses from biodegradative losses [4]. The poisoned controls showed the maximum evaporative loss that may occur under the incubation conditions. However, biodegradation and evaporation compete in the removal of petroleum hydrocarbon. The difference between the loss of hydrocarbons from the poisoned controls and the loss observed in active soil samples strongly underestimates the true contribution of biodegradation.

ANALYTICAL PROCEDURES

For each point of analysis, the fuel in the soil of an entire column was extracted. Jet fuel, diesel oil and heavy fuel oil were soxhelet .extracted by methylene chloride for 6 h. Anhydrous sodium sulphate was added to the extraction of heavy fuel oil which has no highly volatile components, the solvent was evaporated in a preweighed dish, and the residual was determined gravimetrically. Extracts of jet fuel and diesel oil were brought to volume and the extracts were analysed by gas chromatography using the Chrompack Packard Gas Chromatograph model 439 equipped with flame ionisation detector. The column used was 25 m long capillary column packed with Cp-Cil 5CB WCOT. Nitrogen was used as carrier gas. The flow rate of carrier gas was adjusted at 1 ml/min. The temperature of the detector and injection port was maintained at 300°C.

For class separation, jet fuel was separated into saturates, olefins and aromatics using the relevent standard procedure [6]. A jet fuel sample (0.75 ml) was introduced into a glass adsorption column packed with silica gel. The upper small layer of the silica gel contains a mixture of fluorescent dyes. When all the sample has been adsorbed on the gel, isopropyl alcohol was added to desorb the sample and force it down to column. The hydrocarbons were separated according to their adsorption affinities into aromatics, olefins and saturates. The fluorescent dyes were also separated selectively, with the hydrocarbon types and made the boundaries of the aromatics, olefins and saturate zones visible under ultraviolet light.

Diesel fuel was separated into aromatics and non-aromatics by using the standard procedure described in ASTM D2549 method [7]. A weighed amount (10 g) of sample was charged to the top of a glass chromatographic column packed with activated bauxite and silica gel. Normal pentane (150 ml) was added to the column to elute the non aromatics. When all of the non-aromatics were eluted, the aromatic fraction was first eluted by additions of diethyl ether (100 ml), then chloroform (100 ml) and by ethyl alcohol (175 ml). The solvents were completely removed from the extracts and the residues were weighed and calculated as aromatics and non-aromatics fractions of the sample.

For class separation of heavy fuel oil, it was fractionated on a silica gel column. The silica gel (100–200 mesh) was activated at 105°C for 12 h. The glass column (2 by 28 cm) was packed with silica gel suspended in hexane. The 0.5 g hydrocarbon samples were adsorbed on 3 g of silica gel and placed on the column. A 3 g layer of anhydrous sodium sulphate was placed over the sample to absorb any water and to prevent the disturbance of the sample with the solvents. The class fractions of heavy fuel oil was accomplished by successive elution in a discontinuous solvent gradient of increasing polarity. The saturated, aromatic, and asphaltic classes were eluted with 120 ml of hexane, benzene, and chloroform-methanol (1:1; vol./vol.), respectively.

RESULTS AND DISCUSSION

The studies generated a five-point depletion curve for each fuel type under a variety of incubation conditions (untreated soil, bioremediation treated soil, poisoned soil). The disappearance pattern of fuel hydrocarbons are shown in Figs. 1 to 3. Although the classical pattern of material depletion shows exponential behaviour but not exactly first order kinetics. Because of diffusion limitations and increase in the degrading microbial populations, the depletion of a homogeneous substrate in soil is rarely a first order but, rather it is an intermediate between first order (exponential) and zero order (linear)

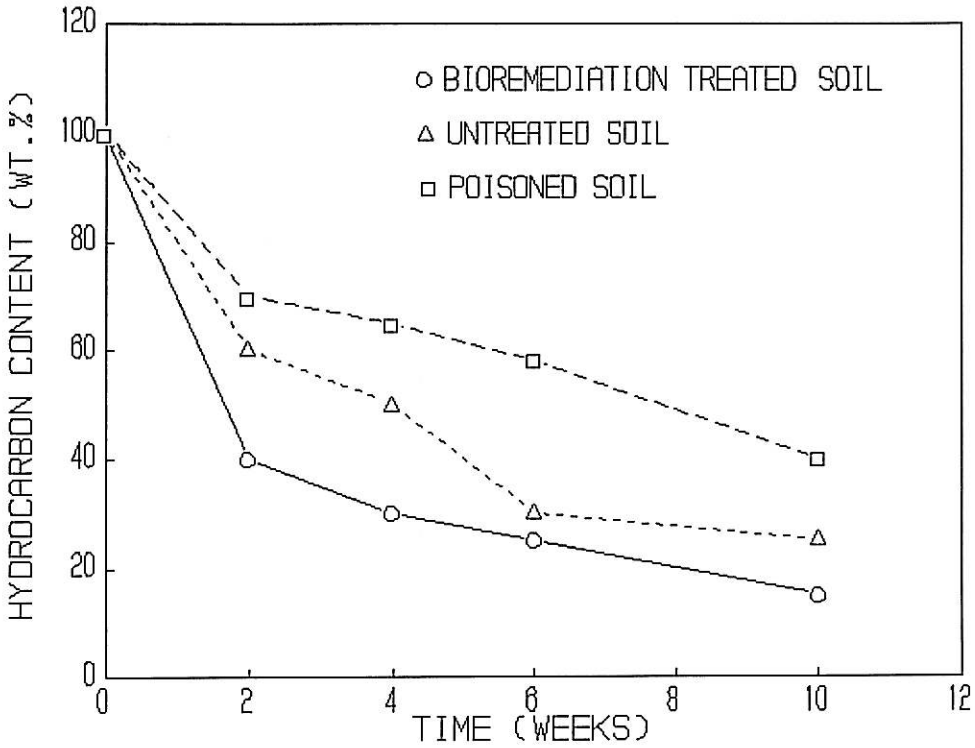

FIG. 1. Disappearance of jet fuel hydrocarbons.

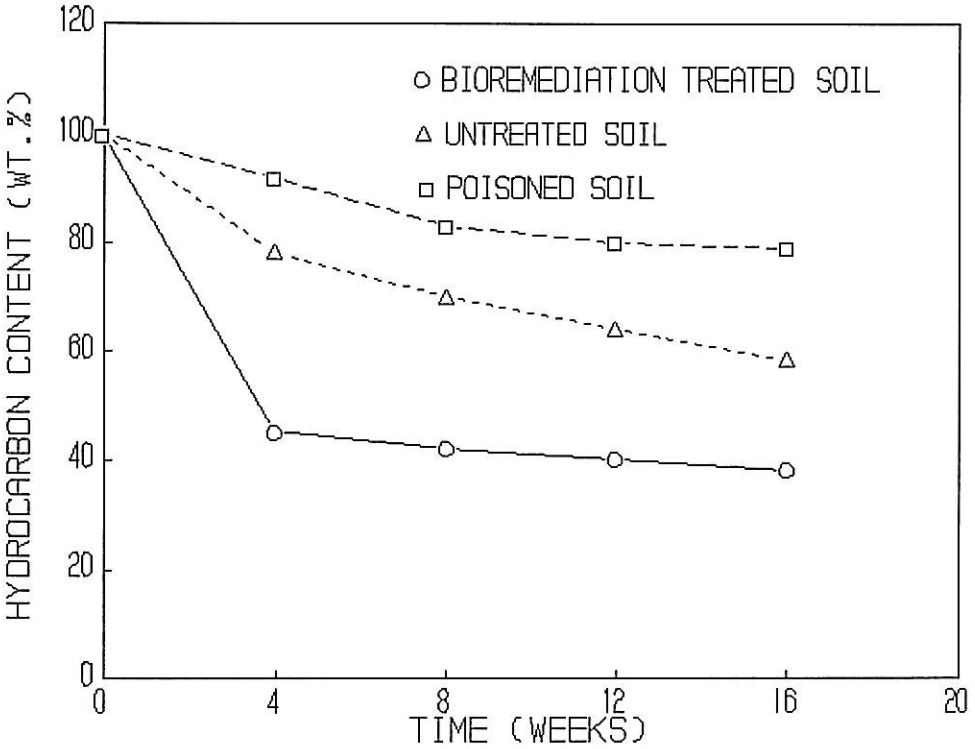

FIG. 2. Disappearance of diesel fuel hydrocarbons.

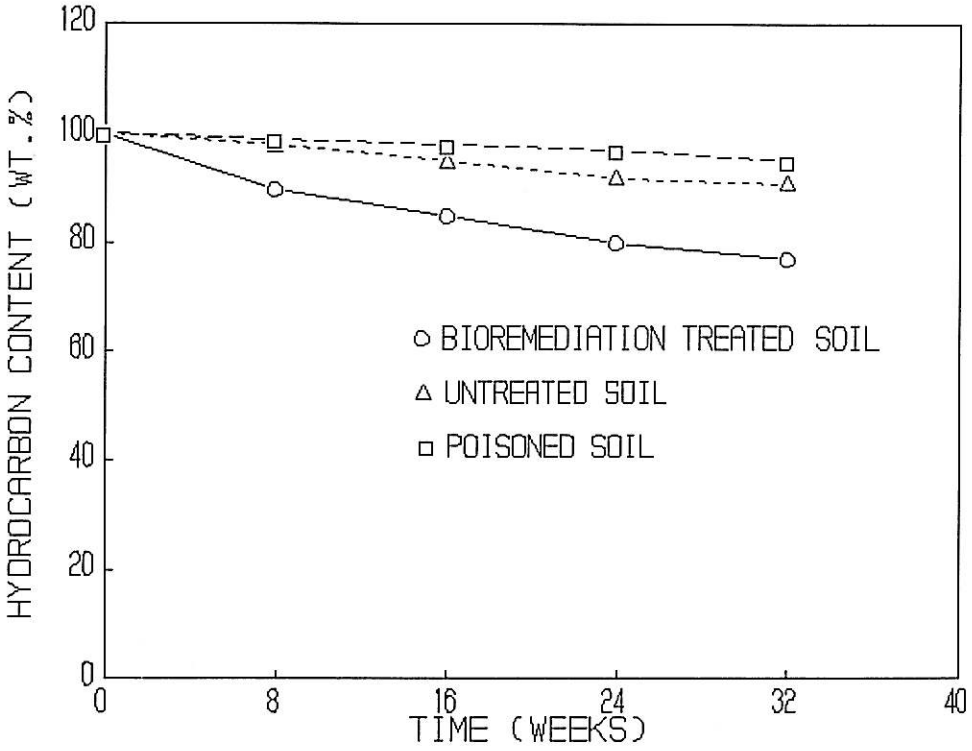

FIG. 3. Disappearance of fuel oil hydrocarbons.

kinetics. The kinetics are further complicated in the case of fuels by the fact that there consist of numerous individual hydrocarbons, each of which degrade at a different rate. The slowing tendency of utilisation is caused not only by substrate depletion but also by the fact that the remaining hydrocarbons are structurally less degradable than the ones already disappeared [8, 9]. To some extent this is compensated by the increasing numbers (enrichment) of the hydrocarbon-degrading microorganisms in the soil with time. For the reasons described above, there is no precise way to convert the curves obtained in this study to constants.

Table 5. Half-Lives of Fuel Disappearance in Soil under Incubation Conditions

Fuels and incubation conditions	Half-life
Jet fuel	
Poisoned soil	7 weeks
Untreated soil	4 weeks
Bioremediation treated soil	1.7 weeks
Diesel Fuel	
Poisoned soil	>16 weeks
Untreated soil	16 weeks
Bioremediation treated soil	3.8 weeks
Heavy Fuel Oil	
Poisoned soil	>32 weeks
Untreated soil	>32 weeks
Bioremediation treated soil	>32 weeks

Table-5 represents the half-lives of fuels under various incubation conditions. The half-life is simply the time needed to reduce the total fuel concentration in soil to 50% of the initial amount. In case of a 50% reduction is not achieved within the time period of the experiment, Table-5 indicates this fact as half-life > 16 weeks. The half-lives used in this table give a useful comparison on the relative biodegradability of the fuels and the environmental conditions that favour or restrict the process.

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