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Short Communication

Successful bioremediation of fuel-polluted seashore sand: a case study

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Plain Language Summary This study looked at how bioremediation – a process that uses microorganisms to break down pollutants – can clean up coastal sand contaminated with fuel. Over 106 days, the amount of petroleum hydrocarbons in the sand dropped dramatically, from 5,753 mg/kg to just 37 mg/kg, a reduction of about 98.21%. This final level is well below the safe limit of 100 mg/kg set by Israel's environmental authorities, showing that the cleanup was successful. Most of the pollution was removed within the first 48 days, emphasizing the importance of good conditions, like proper aeration, to support microbial activity early on. These findings suggest that bioremediation is an effective way to restore polluted coastal areas and could be used more widely to reduce oil pollution in marine environments.

Abstract This study examined the bioremediation of fuel-contaminated coastal sand, addressing petroleum hydrocarbon pollution. Over 106 days, total petroleum hydrocarbons (TPH) levels significantly dropped from 5,753 mg/kg to 37 mg/kg – a decrease of about 98.21%. This final concentration was well below the 100 mg/kg threshold set by the Israel Ministry of Environmental Protection, indicating a successful remediation process. The most significant TPH reduction occurred within the first 48 days, highlighting the importance of optimal conditions for microbial activity, like aeration, soon after contamination. These results demonstrate the potential of bioremediation to restore ecological integrity in areas affected by oil spills and suggest its broader implementation to mitigate petroleum pollution in marine ecosystems.

Keywords bioremediation; seashore sand; petrol contamination; Israel

Introduction

Oil spills frequently occur during the transportation, storage, and extraction of crude and refined petroleum products and pose significant environmental hazards by contaminating soil, groundwater, and marine ecosystems (Li et al., 2022). Global reports estimate that such incidents release millions of tons of oil annually, polluting thousands of hectares of land and water yearly (Nikiforov et al., 2016; State Report, 2017). This contamination has far-reaching consequences, as the hydrocarbons present in oil induce hydrophobicity in soils, severely compromising their ecological health and functionality (Tursi et al., 2018). This phenomenon is evident in affected areas like the Evrona Nature Reserve in Israel, which endured two major incidents of oil contamination, separated by 39 years in 1975 and 2014. These events have had both short- and long-term impacts on local ecosystems, disrupting key ecological processes such as annual vegetation recruitment, soil bacterial community structure, and predator-prey dynamics (Nothers et al., 2017; Ignat et al., 2021; Ferrante et al., 2020; Ferrante et al., 2021a). They have also affected arthropod populations (Möller, 2019), altered parasitoid community composition (Möller et al., 2020), and influenced predation rates among both invertebrates and vertebrates (Ferrante et al., 2021b). Similarly, oil spills in marine environments have been shown to cause widespread ecological damage. For example, in western Korea, they have altered the composition and abundance of intertidal benthic communities. In Alaska, spills have negatively impacted seabird populations, killer whales (*Orcinus orca*), and subtidal communities. In the Gulf of Mexico, oil spills have led to large-scale effects on deep ocean corals, disrupted oyster recruitment, degraded local wetlands, and caused population declines in dolphins, sea turtles, and seabirds (Barron et al., 2021). Collectively, these disruptions have led to a rapid and unidirectional decline in ecosystem integrity.

While traditional remediation strategies such as burning and mechanical separation have been implemented, they often fall short of environmental safety standards, proving inadequate for effective decontamination

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(Babaniyi et al., 2023). As a result, there has been a shift toward various remediation technologies, including physical, chemical, and biological methods, which aim to expedite the cleanup of oil-contaminated soils (Agarwal et al., 2016; Li et al., 2022; Fang et al., 2024). However, physical methods can produce stable oil-water emulsions that are difficult to separate, and chemical methods may lead to unintended ecological harm due to the introduction of additional pollutants, such as surfactants, heavy metals, and toxic byproducts that can persist in the environment (Ceschia et al., 2014).

In light of these limitations, bioremediation has emerged as a viable and environmentally friendly alternative. This approach leverages the natural degradative capacities of microorganisms to break down contaminants, offering a cost-effective and sustainable solution with minimal ecological impact (Zeneli et al., 2019; Liu et al., 2011). Bioremediation techniques such as biostimulation and bioaugmentation can enhance microbial degradation, mainly when supported with essential nutrients and electron acceptors to optimize microbial growth and activity (Farber et al., 2019; Kim et al., 2018). Additionally, biosurfactants produced by oil-degrading bacteria increase the bioavailability of hydrocarbons by improving soil permeability, further enhancing the effectiveness of the remediation process (Varjani, 2017; Banet et al., 2021; Li et al., 2022). Among the employed products for bioremediation, Petroleum Remediation Product (PRP; Petrol Rem, Inc., Pittsburgh) and Oil Sponge have shown particular promise.

PRP, selected for this study, acts as a natural substrate that supports bacterial growth when applied to hydrocarbon spills (Lee et al., 1997). Its properties, such as low density, hydrophobicity, and oleophilicity, allow it to float on water surfaces, absorbing and concentrating floating oil (Chukwuma, 2019). Upon contact with hydrocarbons, PRP releases nutrients that promote microbial activity, ultimately facilitating the microbial conversion of oil into shorter, less harmful hydrocarbons, such as the gases methane (CH_4) through butane (C_4H_{10}). A key advantage of PRP in bioremediation is its lack of toxicity, making it an environmentally safe option for hydrocarbon cleanup (Chukwuma, 2019). Unlike many chemical remediation agents that can introduce secondary pollutants or harm soil and water ecosystems, PRP does not add toxic substances to the environment (Chukwuma, 2019).

Oil Sponge, also used in this study, is a 100% organic, renewable product specifically designed for effective hydrocarbon absorption (Hadji et al., 2020). It is characterized by its high absorption efficiency and its composition of fully organic materials derived from renewable sources (Ramakrishnan et al., 2021). Oil Sponge contains a blend of oil-degrading bacteria and nutrients that accelerate the breakdown of hydrocarbons into carbon dioxide and water, producing non-toxic byproducts that are safer for the environment (Hadji et al., 2020; Ramakrishnan et al., 2021). The product's formulation includes components such as cotton stalks and pecan shell membranes, which provide a supportive habitat for microbial communities. This organic matrix (consisting of a variety of bacteria – Pseudomonas, Bacillus, Chryseobacterium, Ochrobacterium) facilitates rapid and efficient hydrocarbon degradation, as the microorganisms break down the oil into harmless compounds that can be assimilated naturally, thereby enhancing the environmental safety of the remediation process (Ebadi et al., 2021).

Effective bioremediation strategies, however, require careful consideration of site-specific conditions and contaminant types (Kuppuswamy et al., 2016). For example, the carbon-to-nitrogen ratio in contaminated soils often limits microbial activity, making nutrient supplementation essential to optimize degradation rates (Zhang et al., 2020). Treatability studies, such as measuring evolved CO_2 as an indicator of biodegradation, are crucial for assessing the feasibility and effectiveness of bioremediation approaches tailored to specific environmental conditions (Castro-Aguirre et al., 2017).

In September 2007, following a maritime collision between two vessels, fuel contamination occurred along ca. 900m of the Kiryat Haim Beach. In response, the Marine and Coastal Division of the Ministry of Environmental Protection set specific treatment targets and methodologies, overseeing the remediation efforts until all fuel-related hydrocarbons were successfully removed from the beach sand. This report presents the results of our sand treatment efforts and evaluates the potential enhancement of remediation efficacy by applying Petroleum Remediation Product (PRP; Venosa, 2004) and Oil Sponge (Bayat et al., 2015).

Methods

Sampling of contaminated sand

The biological treatment of contaminated sand was conducted at Kiryat Haim Beach (32°83'87"N, 35°05'55"E). The authorities designated a 1200 m^2 area of the beach and fenced off to prevent the spread of contamination. Within this area, representatives from the Ministry of Environmental Protection layered the area with polyethylene sheets and created six piles of contaminated sand to prevent contact with the surrounding substrates (Fig. 1). All of the contaminated sand from the coastline was transferred into this enclosed area and thoroughly mixed to ensure homogeneity.

Throughout the treatment process, which involved the application of PRP and Oil Sponge, three 100g sea sand samples were collected randomly from each of the six piles every four weeks between September 17, 2007, and January 1, 2008. Ecotech Laboratories analyzed all samples to assess the effectiveness of the remediation efforts. We chose PRP because it is marketed as a dry powder in barrels and various absorbent packaging sizes. It is a concentration of nutrients created through a natural enzymatic process derived from bees' wax (Venosa, 2004). The substance is non-toxic and serves as a nutrient for the naturally decomposing organisms at the contamination source, participating naturally in the decomposition process. The shelf life of the material is unlimited.

On October 12, 2007, 50 sacks of Oil Sponge weighing 675 kg were introduced into the contaminated sand. The contaminated sand was mixed and irrigated with

Figure 1. Initiation of the project on the Kiryat Haim beach. Note the polyethylene sheets underneath the contaminated sand piles.

seawater to achieve the required moisture level of up to 20% (González-Alcaraz et al., 2015). Freshwater was deemed unsuitable due to its chlorine content, which can harm microbial life and inhibit natural biological degradation processes, including influencing the bioremediation bacteria applied to the soils (Singh et al., 2021).

On October 15, 2007, samples were taken from the sand at six predetermined locations within the treatment area.

The primary material in the biological treatment, PRP, was introduced on October 18, 2007. In total, about 295 kg of PRP in 10 barrels was used. About 100 kg of the material was a mixture of PRP and ground corn to maintain moisture levels at 15% in the sandy soil.

On October 22, 2007, seawater was pumped from the nearby beach to the biological treatment surface, totaling about 100 cubic meters. At the end of the irrigation, the moisture level measured on the sand surface was 23%.

On November 4, 2007, an excavator was used to mix and till the contaminated sand surface. On the same date, samples were taken from the contaminated sand at six points according to the attached sampling plan. The samples were sent to Aminolab laboratories in central Israel for independent analysis of total recoverable petroleum hydrocarbons (TPH) using infrared spectroscopy.

Further mixing, tilling, and wetting of the contaminated surface were carried out on 12 and 22 November 2007.

Each sample was placed in a glass vial with a Teflon-coated cap and preserved in a cooler with ice until delivery to the laboratory. Personnel collecting the samples utilized clean disposable gloves, and each vial was labeled with a sticker indicating the date and sampling location.

Statistical analyses

To analyze differences in TPH concentration between sampling days, we applied a Kruskal-Wallis test. Following this, Dunn's post hoc test with Bonferroni correction was used to identify specific pairs of sampling periods with significant differences in TPH concentrations. To assess the decrease in TPH concentration over time during the bioremediation process (from September 17, 2007 to January 1, 2008), we applied a log-log linear regression model to capture the exponential decay of TPH levels. The model takes the following form:

$$ln_{TPH} = a + b \times ln_{days}$$

Where a represents the initial concentration level (intercept), and b represents the decay rate (slope). A negative slope indicates a reduction in TPH levels over time, as expected in bioremediation. The model fitting was conducted using ordinary least squares, minimizing the sum of squared residuals between observed and predicted log-transformed TPH values. Coefficient estimates, along with their standard errors, t-values, and p-values, were calculated to assess the significance of each parameter.

For each sampling period, standard deviation (SD), standard error (SE), median, and median absolute deviation (MAD) were used as measures of central tendency and variability in TPH concentration. All statistical analyses were performed in R (R Core Team, 2023), using the dplyr package for data manipulation (Wickham et al., 2023), ggplot2 for data visualization (Wickham, 2016), dunn.test for Dunn's post hoc test (Dinno, 2017), and stats for regression modeling and the Kruskal-Wallis test (R Core Team, 2023).

Results

Over the 106 days of the experiment, TPH concentration decreased by 98.21% (Table 1).

Statistical analysis revealed significant differences in TPH concentrations between the sampling periods

Table 1. Summary statistics (mean, median, MAD, and SD) of TPH concentration across five sampling dates illustrate a progressive decline in contamination levels over the bioremediation period.

Date	Mean (±SD) TPH	Median± (MAD) TPH
17.09.2007	2917 ± 1690	2847 ± 1993
15.10.2007	621 ± 563	582 ± 1993
04.11.2007	309 ± 327	242 ± 282
04.12.2007	178 ± 150	120 ± 81.5
01.01.2008	30 ± 30.2	23.5 ± 34.8

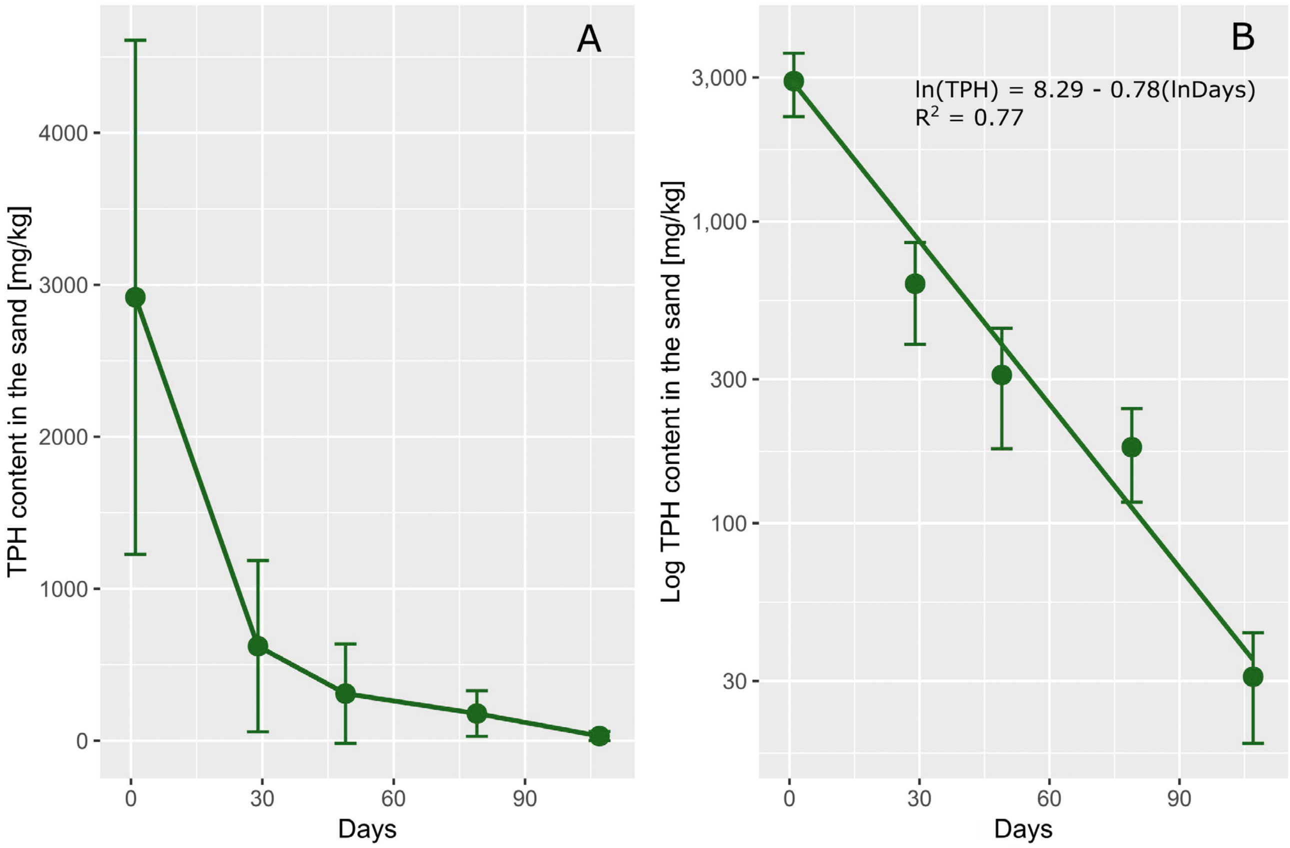

Figure 2. Changes in TPH concentration over time, showing statistically significant reductions across sampling periods (A) and a log-log model illustrating an exponential decay TPH trend with time (B). Points indicate mean values and error bars represent standard errors.

(Kruskal-Wallis test: $H = 23.077$, $p < 0.001$; Fig. 2A). However, Dunn's post-hoc test indicated that these differences were limited to specific pairs of sampling periods: specifically, the first vs. fourth (difference = 2.14, $p = 0.006$) and first vs. fifth (difference = 4.29, $p < 0.001$) periods, as well as the second vs. fifth period (difference = 3.22, $p = 0.006$).

To expand on these findings, a log-log linear model was developed to describe the decreasing TPH concentration in the bioremediation area. The estimated $\beta \pm SE$ for log-transformed time (Log_Days) was −0.778 (± 0.241), with $t = -3.232$ and $p = 0.0481$, demonstrating a statistically significant decay in log-transformed TPH concentration (Fig. 2B). Additionally, the intercept ± SE was estimated at 8.290 ± 0.884, with $t = 9.379$ and $p = 0.0026$, indicating a statistically significant starting concentration level. This model explains a substantial portion of the variance in TPH concentration over time ($R^2 = 0.78$), further confirming the effectiveness of the applied method.

Discussion

The data collected during the bioremediation treatment period reveals a significant reduction in total petroleum hydrocarbons (TPH) levels, decreasing from an initial concentration of 2,917 mg/kg to 30 mg/kg over 106 days. This reduction of approximately 98.21% demonstrates the effectiveness of the bioremediation techniques employed, successfully restoring sand quality to TPH levels below the 100 mg/kg threshold established by the Israel Ministry of Environmental Protection for residential areas.

The most substantial reduction in TPH concentrations occurred during the initial 48 days of treatment. This early phase of bioremediation appears crucial, likely due to the rapid growth of microbial populations stimulated by the application of organic amendments such as Oil Sponge and coarse sorghum. These amendments provided essential nutrients and moisture, creating favorable conditions for microbial degradation of hydrocarbons. The subsequent stabilization of TPH levels suggests that, beyond a certain threshold, the degradation rate may reach an asymptote, emphasizing the need for continuous monitoring and potential supplementary interventions to sustain microbial activity. This observation aligns with the findings of Li et al. (2022), who reported a 99.2% reduction in soil hydrophobicity within seven days in a desert habitat, highlighting the impact of early interventions in similar contexts.

Our findings also suggest the importance of environmental factors, including moisture content and aeration, in enhancing bioremediation efficiency. The application of seawater for irrigation not only maintained optimal moisture levels but also introduced natural nutrients, further supporting microbial growth. This emphasizes the importance of developing bioremediation strategies tailored to local environmental conditions and specific contaminant characteristics. This perspective is supported by Banet et al. (2021), who found that while biostimulation accelerated biodegradation in a hyper-arid environment, bioaugmentation with sediments from older oil spills proved ineffective, suggesting the critical role of timely interventions.

The effectiveness of bioremediation can vary based on the contamination level and the choice of amendments. For example, Slyusarevsky et al. (2017) demonstrated that the combination of activated carbon and diatomite (ACD) significantly improved TPH degradation in highly contaminated soils by reducing toxicity and enhancing microbial activity. Other studies also support that adsorptive materials like ACD can limit the mobility of toxic petroleum metabolites, thereby reducing the risk of leaching and protecting groundwater quality (Aivalioti et al., 2010; Mao et al., 2009). These results highlight the potential of integrating adsorbents with biostimulation or bioaugmentation to extend the capability of *in situ* bioremediation, especially in highly contaminated soils.

Moreover, our findings align with studies showing that bioaugmentation alone may have limited effectiveness in highly toxic soils, as introduced microbes often struggle to compete with indigenous microorganisms or adapt to high toxicity levels (Macaulay and Rees, 2014). In these cases, adsorptive bioremediation, as demonstrated in other research, can help mitigate toxicity by immobilizing contaminants and supporting indigenous microbial communities (Bakhaeva et al., 2001).

In conclusion, our study provides compelling evidence for the effectiveness of bioremediation in addressing petroleum contamination in coastal environments. Combining organic amendments and environmental management techniques significantly reduced TPH levels, underscoring the potential for similar approaches in other contaminated areas. These findings contribute to the broader goal of restoring ecological balance and protecting marine and coastal ecosystems from oil pollution. Additionally, as highlighted by previous studies, the integration of adsorptive materials may represent a promising strategy for further improving bioremediation outcomes in highly contaminated or sensitive environments.

Ethical guidelines

According to the guidelines supplied by, and supervision of the Coastal and Marine Division of the Ministry of Environmental Protection of the State of Israel.

Conflict of interest

The authors declare no conflicts of interest.

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