

Ryegrass enhancement of biodegradation in diesel-contaminated soil

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Abstract

A time-course pot experiment was conducted with ryegrass grown in soil experimentally contaminated with diesel oil. Relationships among plant growth variables, microbial activity and the dissipation rate of diesel oil over time were analyzed.

Results indicate that ryegrass growth can lower the dissipation threshold. The residual rate of diesel oil in the rhizosphere was 55% lower than in the corresponding root-free soil, and the threshold reduction occurred after the development of plant roots. In the rhizosphere, the number of aerobic bacteria and the amount of soil dehydrogenase activity were higher than in the root-free soil and also showed a correlation with the growth of roots.

The dissipation rate of diesel oil showed a correlation with soil dehydrogenase activity in both the rhizosphere and the root-free soil. A positive correlation was observed between the growth rate of roots and soil dehydrogenase activity in the rhizosphere. Moreover, the dissipation rate per dehydrogenase activity of the rhizosphere was higher than in the root-free soil. Ryegrass roots were determined, therefore, to be effective at enhancing the biodegradation of diesel-contaminated soil.

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1. Introduction

As a result of human economic activity, large quantities of soil have been contaminated with petroleum products. Several technologies satisfactorily treat soils with petroleum products, including physical/chemical

techniques such as thermal treatment, soil washing, the extraction of gases or liquid matter, solidification and stabilization or combinations of the above (Langbehn and Steinhart, 1995). However, such techniques are not only expensive, but also require the use of heavy equipment and a substantial amount of energy.

Bioremediation is a cost-effective and environmentally-friendly technology, and there are several successful bioremediation applications for petroleum-contaminated soils (Lindstrom et al., 1991; USEPA,

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2004). One variation is phytoremediation, the use of higher plants to further enhance the degradation of soil contaminants (Banks et al., 2003a). Phytoremediation, a low-cost alternative to traditional contaminant removal and disposal techniques, has been proposed for the restoration of petroleum-contaminated soil. Phytoremediation has the added benefit of being potentially very popular with the public. The rhizosphere offers increased potential for the degradation of petroleum hydrocarbon (Günther et al., 1996); by focusing there, we can develop an effective method of remediating petroleum-contaminated soil. To fully exploit this technology, however, we must understand the complex rhizosphere dynamics that result in the reduction of petroleum contaminants.

Several studies have examined the relationship between plants and rhizosphere-based microbial communities. The presence of mycorrhiza generally enhanced plant growth and PAH dissipation (Joner and Leyval, 2003), and phytoremediation systems were observed to increase the catabolic potential of the rhizosphere soil by altering the functional composition of the microbial community (Siciliano et al., 2003). However, the specifics of how the rhizosphere enhances biodegradation are still unclear.

We hypothesize that roots enhance the degradation of contaminants by stimulating microbial activity. Plant exudates and sloughed tissue, which serve as sources of energy, carbon, nitrogen, or growth factors have been shown to increase microbial activity (Alexander, 1977; Banks et al., 2003a). Root growth also opens deeper soil to better water infiltration and oxygen diffusion (Singer et al., 2003). Root surfaces provide adhering space for soil microorganisms, and roots can disrupt soil aggregates and enhance biodegradation of entrapped hydrophobic contaminants (Banks et al., 2003a). These studies demonstrate how root growth increases microbial activity.

Little research has been conducted, however, on the relationships among root growth, microbial activity and degradation of contaminants. Günther et al. (1996) showed that enhanced hydrocarbon disappearance is caused by an increased rhizosphere microbial community in comparison with rooftop soil, but root growth was not studied. Hou et al. (2001) found an increase in biodegradation subsequent to the formation of higher ryegrass root intensity, but did not evaluate time-course change. Banks et al. (2003b) demonstrated

that the greatest decrease in total petroleum hydrocarbon (TPH) concentrations occurred in the period with the greatest root growth, but did not evaluate concurrent microbial activity.

The purpose of our study was to examine the periods of greatest TPH dissipation in order to contribute to the practical use of phytoremediation in field operations. A time-course pot experiment was conducted to measure the TPH decrease, plant growth variables and microbial activity in the rhizosphere of ryegrass grown in soil experimentally contaminated with diesel oil. We analyzed the relationships among plant growth variables, microbial activity and the rate of diesel-oil dissipation at 1 month intervals from the time of seeding to maturity. The data collected on root growth, microbial activity and TPH dissipation were then analyzed to evaluate the relationships among these factors. Results from this research may allow for increased effectiveness in phytoremediation field operations.

Ryegrass was selected as the model plant for the experiments due to its hardiness and ability to grow in diesel-contaminated soil. Ryegrass also demonstrated effective dissipation of diesel oil in preliminary experiments.

2. Materials and methods

2.1. Preparation of the experimental soil

The experimental soil was prepared by adding diesel oil, leaf mold and perlite to masado, a decomposed granite soil. Because preliminary experiments indicated that the use of 2% (w/w) diesel-contaminated soils inhibited the growth of ryegrass from seeds, the initial concentration of diesel-oil was set to 1.8%. Allowing for loss by volatilization during the soil contamination process, the weight of diesel oil added to the masado was set to 3.0%.

Prior to the addition of the diesel oil, the masado was sun-dried in a green-house and mixed regularly to ensure consistent evaporation until the water content was less than 1%. In order to obtain a homogeneous soil/pollutant mixture, the diesel-oil was gradually sprayed into the masado in a mechanical mixer. After the requisite amount of diesel oil was added, mixing was continued for an additional 5 min. The masado–diesel mixture was further stabilized by 3

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