WATER QUALITY

Bioremediation of Sediments

Final Report

Bioremediation provides an environmentally sound method of removing polyaromatic hydrocarbons (PAHs) from contaminated sediments. A combination of surfactant treatment with application of selected strains of PAH-degrading bacteria in a slurry treatment system brought about increased levels of PAH removal in comparison to the any of the treatments alone. However, removal rates equal to those achieved by single PAHs were not achieved in the mixed PAH systems, indicating that biodegradation rates obtained using single PAHs will overestimate the time to achieve acceptable removal rates.

Objectives

- Surfactant when present at nontoxic concentrations will increase the rate and extent of utilization of high three-and four-ringed PAHs.
- Components of mixtures of PAHs may inhibit the degradation of PAHs which are otherwise utilized by bacterial strains.
- A sediment slurry system will be favorable for bioremediation of contaminated sediments.

Summary of Progress

Tests with four strains of PAH-degrading bacteria indicate that there are significant effects on both the rate and extent of biodegradation when PAHs are given in mixtures in comparison to results when PAHs are given singly. In binary mixtures, a PAH utilized for growth can enhance the rate and extent of utilization of a PAH which is utilized but which does not support growth.

On the other hand, the presence of three or four PAHs tend to inhibit the utilization of all of the component PAHs, regardless of whether they are used for growth or not. This finding has significance for bioremediation of contaminated sediments.

A surfactant has been selected for soil slurry assays. Tween 80 is nontoxic to the selected bacterial strains and is not utilized by them. Its use results in as much as a doubling of the rate of utilization in the presence of the surfactant compared with its absence. Conditions for soil slurry experiments were selected based on range-finding experiments. A sediment loading of 5% was chosen as an optimum concentration for conducting the assays. Extraction efficiencies were determined to be satisfactory using liquid-liquid extraction (as compared to Soxhlet extraction or resin extraction).

In soil slurry experiments, bacterial strains show the capacity to degrade complex mixtures of PAHs, although at rates lower than those achieved using pure cultures and pure PAHs. Thus laboratory work with single PAHs may not adequately predict biodegradation rates occurring in the environment.

Accomplishments

PAHs rarely occur alone in sediments; they invariably are components of mixtures of PAHs and may be present along with other toxicants such as phenols and heavy metals. The results reported above may explain why PAHs which can support growth of microorganisms are nevertheless persistent in contaminated sediments. These results may therefore indicate where attention must be focused in order to achieve bioremediation of environments contaminated by PAHs.

Publications

D. Dean-Ross. Submitted. Utilization of mixtures of polycyclic aromatic hydrocarbons by bacteria isolated from contaminated sediment. Microbial Ecology.

Undergraduate/graduate students

Paul Reust, undergraduate; Kym Crandell, undergraduate

Narrative Report

An additional bacterial strain was tested for utilization of mixtures of PAHs. This strain is a variant of PHEN USX4 and was obtained from it by repeated subculturing using fluoranthene as growth substrate. In contrast to the parent strain which only utilizes fluoranthene when present along with phenanthrene, this strain utilizes fluoranthene alone. As occurred with the parent strain, however, the presence of all three PAHs was inhibitory to the utilization of all of the PAHs.

Screening studies conducted last year indicated that Tween 80 was the surfactant of choice to use in combination with the bacterial strains, as it had little or no toxicity to any of the strains. Full-scale utilization studies with two of the bacterial strains confirm the desirable properties of this surfactant in producing significant increases in biodegradation rates of PAHs.

In preparation for sediment slurry experiments, experiments were conducted to determine the optimum loading rate for sediments in the slurries and to determine extraction efficiencies using several extraction procedures. Three extraction procedures were tested: liquid-to-liquid extraction (the procedure used for the utilization studies), Soxhlet extraction of filtered and dried sediment followed by liquid-liquid extraction of the filtrate, and resin extraction of the slurry. All three methods gave comparable extraction efficiencies in the 90-95% range. The Soxhlet method involved considerable manipulation of the slurry, which increased extraction time. The resin method involved additional expenses in terms of resin column and solvents. The method was also time consuming, as each sample must be filtered and washed several times. The time requirements could have been reduced with the purchase of a vacuum manifold; however, this expense was not considered worthwhile without an increase in extraction efficiency. Consequently, liquid-liquid extraction was selected as the method of choice.

Slurry experiments using pure cultures in combination with Tween 80 confirm the desirable properties of this nonionic surfactant in enhancing biodegradation rates. Rates of PAH biodegradation achieved with the strains tested are substantially lower in mixtures than alone. However, rates achieved in the slurry system are elevated in comparison to rates in undisturbed sediment, indicating that the sediment slurry provides the necessary conditions to maximize biodegradation rates.

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