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Perspective

Microbial Bio-Treatment of Hydrocarbons in Petroleum Refining

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DESCRIPTION

Petroleum pollution of soils and sediments is an international concern due to the toxicity and fire resistance of aromatic components in the absence of oxygen. Gaseous oxygen can be injected into the anaerobic zone of a contaminated environment to stimulate biodegradation, which is costly and inefficient. Alternatively, other more soluble electron acceptors such as nitrates and sulphates can be used, but they oxidize slowly and are incompletely decomposed hydrocarbons. This section describes how perchlorate-reducing bacteria can use chlorate invasion as an alternative source of oxygen to break down contaminants. This conversion of chlorate to molecular oxygen and chloride is an intermediate step in the microbial reduction of perchlorate or chlorate. As part of our study on microbial perchlorate reduction, we isolated new microorganisms. From waste sludge from the Pennsylvania paper mill, distorted CKB that grows anaerobically by perchlorate or reduction of chlorate. When chlorite is inoculated into an oxygen-free petroleum-contaminated soil sample of the CKB strain, [^{14}C]-benzene is rapidly oxidized to $^{14}CO_2$ and about 40% of the original ^{14}C is recovered in this form after 2 days of incubation. If 1 mM chlorite is added to the sediment on the 3rd day, about 60% of ^{14}C will be recovered as $^{14}CO_2$ by the 6th day. No $^{14}CO_2$ is produced in samples without chlorite or CKB strains.

Similar results are obtained with anoxic soil samples that have not been previously exposed to hydrocarbons. However, there is a slight delay stage of 24 hours that is consistent with the adaptation of the microbial population to benzene. The use of low concentrations of chlorite does not significantly change the stimulating effect. 1 mM chlorite resulted in more than half of the degree of benzene degradation observed in 1 mM chlorite.

This stimulating effect also occurs in defined mixed cultures without soil. When an anaerobic washed cell suspension of the CKB strain is combined with an aerobic hydrocarbon oxide *Pseudomonas* JS150 strain and treated with chlorite under anaerobic conditions, [^{14}C] naphthalene is rapidly oxidized to $^{14}CO_2$. Unless you add O_2 to your headspace, if you omit either the creature or chlorite, $^{14}CO_2$ will not be produced. Therefore, the degradation of naphthalene is directly dependent on the presence of the combination of CKB strain and chlorite.

Since the CKB strain cannot degrade aromatic hydrocarbons in pure culture, we thought that the stimulation of hydrocarbon degradation could be the result of the CKB strain's recognition of chlorites for chloride and O_2. The resulting O_2 is used by natural aerobic hydrocarbon-oxidizing bacteria that are inhibited by the anoxic condition of the soil. This has been demonstrated to add chlorite to the anaerobic-washed whole cell suspension of the CKB strain, producing O_2 rapidly and proportionally. O_2 production does not occur if the cells are left unattended or killed by heat.

Our results show that the accreditation of chlorite by perchlorate-reducing bacteria in anaerobic environments can produce extracellular O_2. This O_2 can be used by hydrocarbon-oxidizing bacteria to degrade hydrocarbons, such as benzene, which is a particularly important environmental contaminant owing to its toxicity and relative solubility. Little is known about perchlorate-reducing bacteria, but they are ubiquitous in a variety of environments, including pristine soil and petroleum-contaminated sediments.

High concentrations of chlorite can be toxic to many microbial species, but our results are limits set by the World Health Organization (200 mg/liter) and the US Environmental Protection Agency (1 mg/liter). As a bioremediation strategy, the use of chlorite influx to stimulate hydrocarbon oxidation in contaminated environments offers a new alternative to other infusion methods.

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