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Bioremediation: Prospects and limitations

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Abstract

Many microorganisms possess the inherent ability to transform hazardous compounds. However, the long-term persistence of many of these contaminants in the environment is a testament to the fact that these naturally occurring processes often do not occur at rates that are fast enough to protect ecosystem and human health. Frequently, the microorganisms are limited by the availability of the pollutant or another key substrate or are not present in sufficient numbers. In many cases, bioremediation can overcome these limitations through careful engineering of the contaminated environment, thereby enhancing the rates of key microbial processes. Thus, successful bioremediation involves the integration of environmental microbiology and engineering techniques with other disciplines, such as geochemistry and hydrology.

Keywords: Bioremediation, environment, hazardous compound, microorganisms.

Introduction

The popularity of bioremediation is increasing because it often consumes less energy and fewer resources and thus is less expensive and more sustainable than physicochemical treatment approaches, such as land filling or incineration. Further, many alternative remediation techniques simply transfer organic contaminants to another medium without detoxifying the compounds. Nevertheless, the implementation of bioremediation is not without its challenges. Difficulties caused by the inaccessibility and heterogeneity of many contaminated environments are encountered in all remediation endeavors. However, additional complexities are faced in bioremediation because of the need to understand how (1) microorganisms alter contaminant structures, (2) microbial activities are affected by environmental conditions and interactions with other populations (microbial ecology) and (3) beneficial microbial processes can be enhanced through engineering approaches.

Bioremediation

Bioremediation is a waste management technique that involves the use of organisms to remove or neutralize pollutants from a contaminated site. According to the EPA, bioremediation is a 'treatment that uses naturally occurring organisms to break down hazardous substances into less toxic or non toxic substances'. Technologies can be generally classified as *in situ* or *ex situ*. *In situ* bioremediation involves treating the contaminated material at the site, while *ex situ* involves the removal of the contaminated material to be treated elsewhere. Some examples of bioremediation related technologies are phytoremediation, bioventing, bioleaching, land farming, bioreactor, composting, bioaugmentation, rhizofiltration and biostimulation.

Basis of bioremediation

Bioremediation is based on the idea that all organisms remove substances from the environment to carry out growth and metabolism. Bacteria, Protista and Fungi are found to be very good at degrading complex molecules and incorporating the breakdown products into their metabolism (Bouwer et al., 1993). The resultant metabolic wastes that they produce are generally safe and somehow recycled into other organisms. Fungi are especially good at digesting complex organic compounds that are normally not degraded by other organisms. Bioremediation does not involve only the degradation of pollutants but also, at times it is sufficient to remove the pollutant from the environment without degrading it (Broda, 1992). Bacteria in particular take up large amounts of metals and minerals to ensure adequate resources for binary fission. Algae and plants are very good at absorbing nitrogen, phosphorus, sulfur and many minerals and metals from the environment. For example, plants like locoweed remove large amounts of the toxic element selenium (Caplan, 1993). The selenium is stored in plant tissues where it poses no harm until the plant is eaten. Many algae and bacteria produce secretions that attract metals that are toxic in high levels. The metals are in effect removed from the food chain by being bound to the secretions.

Factors required for bioremediation

The control and optimization of bioremediation processes is a complex system of many factors (Day, 1992). These factors include**:**

i) The existence of a microbial population capable of degrading the pollutants.

ii) The availability of contaminants to the microbial population.

iii) The environment factors (type of soil, temperature, pH, the presence of oxygen or other electron acceptors and nutrients) (Vidali, 2001).

Types of Compounds for bioremediation

Pollutants found in soils present a variety of different human health risks including. Pollutants that are being studied for bioremediation potential are listed below.

Petroleum byproducts

BTEX - benzene, toluene, ethylbenzene and xylene - are byproducts of petroleum products. The biodegradability of these compounds is relatively well known and remediation can be achieved by creating favorable conditions for BTEX degrader's growth.

Methyl tert-butyl ether

MTBE is a gasoline additive introduced to replace lead. MTBE raises the oxygen content of fuel, allowing for more complete combustion and less emissions. MTBE, however, is highly soluble, does not adsorb well in soil and can therefore move quickly through soil and into groundwater.

Polychlorinated biphenols

PCBs are used in industrial applications, are very recalcitrant, and many are known carcinogens.

Chlorinated solvents

Chlorinated solvents are used extensively as cleaning agents. Plumes have been found to contaminate groundwater below dry cleaners in many places, including Davis, Ca. Many chlorinated solvents are carcinogenic. Trichloroethylene (TCE) can be degraded to vinyl chloride under anaerobic conditions. Vinyl chloride, in turn, needs different conditions to transform, and this should be seriously considered due to its high toxicity.

Polycyclic aromatic compounds

PAHs are found in high concentrations at industrial sites especially sites that use or process petroleum products. They are considered carcinogens and mutagens, and are very recalcitrant, pervading for many years in the natural environment. Other contaminants include residuals from flares (per chlorate) and explosives (TNT, RDX), metals (chromium, lead), plutonium and uranium; potassium and nitrogen. Much of the high levels of these contaminants found in nature are a result of human activity.

Examples of Microorganisms involved in bioremediation

Pseudomonas putida

Pseudomonas putida is a gram-negative soil bacterium that is involved in the bioremediation of toluene, a component of paint thinner. It is also capable of degrading naphthalene, a product of petroleum refining, in contaminated soils.

Dechloromonas aromatica

A soil bacteria genus which are capable of degrading per chlorate and aromatic compounds.

Nitrosomonas europaea, Nitrobacter hamburgensis and Paracoccus denitrificans

Industrial bioremediation is used to clean wastewater. Most treatment systems rely on microbial activity to remove unwanted mineral nitrogen compounds (i.e., ammonia, nitrite and nitrate). The removal of nitrogen is a two stage process that involves nitrification and denitrification. During nitrification, ammonium is oxidized to nitrite by organisms like *Nitrosomonas europaea*. Then, nitrite is further oxidized by microbes like *Nitrobacter hamburgensis*. In anaerobic conditions, nitrate produced during ammonium oxidation is used as a terminal electron acceptor by microbes like *Paracoccus denitrificans*. The result is dinitrogen gas. Through this process, ammonium and nitrate, two pollutants responsible for eutrophication in natural waters are remediated.

Phanerochaete chrysosporium

The lignin-degrading white rot fungus, *Phanerochaete chrysosporium*, exhibits strong potential for bioremediation of: pesticides, polyaromatic hydrocarbons, PCBs, dioxins, dyes, TNT and other nitro explosives, cyanides, azide, carbon tetrachloride, and pentachlorophenol. White rot fungi degrade lignin with nonselective extracellular peroxidases, which can also facilitate the degradation of other compounds containing similar structure to lignin within the proximity of the enzymes released.

Deinococcus radiodurans

Deinococcus radiodurans is a radiation-resistant extremophile bacterium that is genetically engineered for the bioremediation of solvents and heavy metals. An engineered stain of *Deinococcus radiodurans* has been shown to degrade ionic mercury and toluene in radioactive mixed waste environments.

Methylibium petroleiphilum

Methylibium petroleiphilum (formally known as PM1 strain) is a bacterium is capable of methyl tert-butyl ether (MTBE) bioremediation. PM1 degrades MTBE by using the contaminant as the sole carbon and energy source.

Biodegradation mechanisms

Many environmental contaminants are subject to chemical or photochemical reactions. However, biological organisms—particularly microorganisms—play a more important role in the removal of many hazardous organics from the environment. Thermodynamically feasible contaminant transformations often do not occur in the absence of a biological catalyst, due to kinetic limitations, but are facilitated by microorganisms via enzymes, which lower the activation energy that must be overcome for a reaction to proceed, and the investment of biochemical energy to convert oxygen and other key co-reactantants to more reactive forms.

In situ and ex situ bioremediation processes

Different techniques are employed depending on the de degree of saturation and aeration of an area. In situ techniques are defined as those that are applied to soil and groundwater at the site with minimal disturbance. Ex situ techniques are those that are applied to soil and groundwater at the site which have been removed from the site via excavation (soil) or pumping (water). In situ bioremediation by indigenous microbial population is an increasing popular, eco-friendly option for cleanup of contaminated sites and currently considerable effort is being spent to design cheap and feasible strategies using this technology, which shows promise as a relatively good alternative (Atlas, 1981). Mercury resistant bacteria have been considered as a potential approach to biological remediation. The bacterial mer operon encodes a cluster of genes involved in detection, mobilization and enzymatic detoxification of mercury. The mer genes are inducible with regulatory control being exerted at the transcriptional level both positively and negatively. Ionic mercury (Hg^{++}) is transported into the cytoplasm by a set of transport genes, where the merA gene, which encodes mercuric ion reductase, reduces this highly toxic ionic mercury (Hg^{++}) to the much less toxic volatile Hg^0 released from contaminated sites is far too slow to be effective for large scale field applications and therefore, naturally occurring bacteria are not suitable for remediation of mercury pollution. The mercury systems are of interest both biochemically and biologically because of their specificity to mercuric ions (Atlas, 1984; Atlas and Bartha, 1981). No other metal ion is known to be transported or reduced by the genes of the mer operon. An increasing awareness of the contribution of mercury resistant bacteria to the environment management process and possibility of intentionally introducing genetically modified organisms into the environment has forced microbial ecologists and scientists to explore these prokaryotic systems as potential means of bioremediation and use molecular intervention in the abatement of mercury pollution (Wistreich and Lechtman, 1988; Pritchard, 1991).

Types of In situ bioremediation

Different types of In situ bioremediation are as follows:

1] **Intrinsic In Situ Bioremediation and Natural Attenuation**

Intrinsic in situ bioremediation uses the natural bio-degradative activities of native microorganisms to contain contaminants and prevent their migration away from the source (NRC, 1993; Rittmann and McCarty, 2001). This is done without engineering the site to enhance the process. However, the use of field tests and site-derived samples to document thoroughly the role played by native microorganisms in removing the contaminants is required.

2] Engineered In Situ Bioremediation

Engineered in situ bioremediation strategies are designed to enhance the intrinsic biodegradation of contaminants via the input of stimulatory materials into the contaminated region (Rittmann et al., 1994). By supplying limiting substrates in such a way that no contaminant escapes biodegradation, a biologically active zone (BAZ) can be established.

This process further subdivided in to following types:

Water Circulation Systems

When contaminants are located partially or totally in the saturated zone, the stimulatory materials can be applied via water circulation systems (NRC, 1993; Rittmann et al., 1994; Rittmann and McCarty, 2001). A typical water circulation system involves the injection of dissolved limiting substrates and hydraulic control of the plume migration to force the flow of the contaminated water through the BAZ.

Bioventing

The goal of bioventing is to provide air (or oxygen) using configurations and flow rates that ensure adequate oxygenation of the BAZ for aerobic biodegradation while minimizing production of off gases (NRC, 1993; Leeson and Hinchee, 1997; Rittmann and McCarty, 2001). It is a particularly cost-effective and efficient for bioremediation of hydrocarbons in the unsaturated zone. Air may be injected or drawn through unsaturated media via a vacuum (also known as soil gas extraction), but injection is typically used. Additional stimulatory materials may be provided in dissolved form or as gases.

In Situ Bioreactive Barriers

In situ biobarrier systems are used to prevent further transport of a contaminant plume in the saturated zone (Rittmann and McCarty, 2001). Specifically, a BAZ is created in the path of the plume via the input of stimulatory materials. In some cases, hydraulic or physical controls (e.g., a funnel-and-gate system) may be needed to ensure that the plume passes through the BAZ. The stimulatory materials used to create the biobarrier can be added either in dissolved form via aqueous injection wells, infiltration galleries, or re-circulating wells, or by placing slow-release materials (e.g., proprietary sources of electron donors or acceptors, mulch, wood chips, iron filings) in wells or trenches.

Types of Ex situ bioremediation

The three main approaches of Ex situ bioremediation are— land treatment, composting/ biopiles, and slurry-phase treatment, vary primarily in how the contaminated material is aerated and the degree of water saturation.

Land Treatment

In land treatment, the contaminated materials are spread out in relatively thin layers, typically in a specially constructed above-grade treatment system or land treatment unit (LTU) (Cookson, 1995; Eweis et al., 1998). LTUs generally have controls for containing the contaminated materials, including a liner and leachate collection system and a cover to reduce volatile emissions. The contaminated material is tilled to aerate the soil and reduce mass-transfer limitations by mixing. Soil nutrient and moisture levels are monitored along with pH and adjusted if necessary to further enhance biodegradation. The soil may also be amended with a bulking agent (e.g., wood chips, shredded bark) or organic material to improve porosity and permeability.

Composting and Biopiles

In composting, the contaminated material is piled up rather than spread out in thin layers, which affects the degree of heat entrapment that occurs and the mode of aeration (Cookson, 1995; Eweis et al., 1998). In conventional composting, the contaminated material is typically mixed with an organic bulking agent to improve airflow in the pile by increasing the porosity, and possibly a thermal source whose degradation results in an increase in pile temperature. Some materials, such as manure, may serve as both a bulking agent and a thermal source. Water is added as needed to control the moisture content. Three basic composting process designs are used: (1) windrows on an impervious surface, which are aerated by mechanical mixing at regular intervals; (2) static piles on an impervious surface, which are either aerated passively or by a forced-air system; and (3) in-vessel reactors, which are mechanically mixed and sometimes include forced aeration. In low-temperature composting, also known as biopiles or soil heaping, the contaminated soil, possibly mixed with a poorly degraded bulking agent, is placed in a static pile, aerated passively or by forced air, and irrigated to control moisture, pH, and nutrients (Cookson, 1995; Fahnestock et al., 1998).

Slurry-Phase Treatment

Slurry-phase treatment is performed under water-saturated conditions (Cookson, 1995; Eweis et al., 1998). The contaminated material is pretreated (often to remove large materials) and mixed with water to form a slurry that can be mixed and aerated, thereby improving contaminant mass transfer, oxygen availability, and biodegradation rates. Other amendments (e.g., nutrients, neutralizing agents, microbial enrichment cultures, surfactants, and dispersants) are added as required. Reactors are typically operated in a batch or semi batch mode and may be open or closed. Dewatering is used to separate the treated water and soil, which are treated further, reused or disposed of.

Advantages of Bioremediation

i. Can be highly specific.

ii. Less expensive than excavation or incineration processes.

iii. If mineralization occurs get complete degradation and clean up.

iv. Does not transfer contaminants from one environment to another.

v. Uses a natural process.

vi. Good public acceptance.

vii. Process is simple.

viii. Using ISB, it will treat the groundwater and soil at the same time.

Disadvantages to Bioremediation

i. Not instantaneous.

ii. Often need to develop a system.

iii. Always need to test and optimize conditions empirically – not with computer models.
iv. May have inhibitors present.
v. Compounds may not be in a biodegradable form – polymers, plastics.

Future Prospects

The application of microorganisms to enhance the fertility of soil conditions and removing the soil contaminations through bioremediation technology is extensively used in Europe and USA. In India, progress has been made in applying microorganisms to the restoration of polluted soil through bioremediation processes. However, the application of bioremediation technology in the restoration of ecosystem and soil management is used less compared to Europe and USA. Hence, we need extensive research programs to increase the capabilities of bioremediation to deep, extensive, subsurface contamination due to chlorinated hydrocarbons and complex mixed wastes, including soils and groundwater. Besides that, The American Academy of Microbiology (AAM) has concluded that enough knowledge is now available for field trials of bioremediation technology for organic compounds and further they emphasized that research is needed for the following classes of environmental pollutants: metals, metalloids, radionuclide's and complex polycyclic hydrocarbons. The on-going microbial genomics studies will deliver more robust technologies for the bioremediation of metal – contaminated waters and land. Exciting developments in the use of microorganisms for the recycling of metal waste, with the formation of novel biomaterials with unique properties are also predicted in the near future.

Limitations for bioremediation

There are several limitations to bioremediation. One major limitation has to do with the nature of the organisms. The removal of pollutants by organisms is not a benevolent gesture. Rather, it is a strategy for survival. Most bioremediation organisms do their job under environmental conditions that suit their needs. Consequently, some type of environmental modification is needed to encourage the organisms to degrade or take up the pollutant at an acceptable rate. In many instances the organism must be presented with low levels of the pollutant over a period of time. This induces the organism to produce the metabolic pathways needed to digest the pollutant. When using bacteria and fungi, it is usually necessary to add fertilizer or oxygen to the material containing the pollutant. This can be disruptive to other organisms when done in situ. In situations where simple compounds and metals are being taken up it is likely that these pollutants are at toxic levels for the organisms. These techniques (U.S. EPA, EPA/625/K-96/001; U.S. EPA, EPA/540/2-90/002) are generally the most desirable options due to lower cost and fewer disturbances since they provide the treatment in place avoiding excavation and transport of contaminants. In situ treatment is limited by the depth of the soil that can be effectively treated. In many soils effective oxygen diffusion for desirable rates of bioremediation extend to a range of only a few centimeters to about 30 cm into the soil, although depths of 60 cm and greater have been effectively treated in some cases (Validi, 2001).

Conclusion

Bioremediation is a powerful tool available to clean up contaminated sites and it occurs when there are availability of microorganisms that can biodegrade the given contaminant and the necessary nutrients. Regardless of which aspect of bioremediation that is used, this technology offers an efficient and cost effective way to treat contaminated ground water and soil. Its advantages generally outweigh the disadvantages, which is evident by the number of sites that choose to use this technology and its increasing popularity.

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