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Review

Bioremediation: A tool for environmental cleaning

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Sites contaminated by heavy metals and other pollutants are common through out the world. Researchers developed bioremediation as one feasible way to accelerate or encourage the degradation of pollutants from such affected sites. The basis of bioremediation is that all organisms remove substances from the environment to carry out their growth and metabolism. Bioremediation is not effective only for the degradation of pollutants but it can also be used to clean unwanted substances from air, soil, water and raw materials form industrial waste. With this in view, though many engineered processes for applying bioremediation have been developed but the inexpensive treatment of such sites has remained an elusive goal. Unlike organic contaminants, which often can be metabolized inexpensively into harmless substances such as carbon dioxide and water, metals and their salts that typically inhibit rather than support biological processes. However, in recent years there has been a flurry of interest developed in the implementation of biological approaches for bioremediation of at least some forms of inorganic contamination and paved the way for some other promising technologies to emerge.

Key words: Bioremediation, growth and metabolism, pollutants, environment.

INTRODUCTION

All substances in nature ultimately succumb to decay. Much of this phenomenon is a natural consequence of the laws of thermodynamics. Many molecules degrade by the action of oxygen, halogens and radicals naturally found in the environment. While a large proportion of materials degrade because their components are subject to the action of enzymes. Unfortunately, for humans, many of the wastes do not decay as fast as other substances. They end up polluting the air, land and water. 2 major factors prevent material wastes from decaying rapidly. One such is that waste we produce so much at one time that the rate of natural decay is insignificant compared to the amount present. Another factor is that most waste end up in areas not conducive to rapid degradation. Contaminated lands have generally resulted from past industrial activities at which time the awareness of health and environmental effects connected with the production, use and disposal of hazardous substances was less well recognized than today. This problem is worldwide and the estimated numbers of contaminated sites have grown significantly to a large number (Cairney, 1993).

The conventional techniques used for remediation have generally been to dig up the contaminated soil and remove it to a landfill, or to cap and contain the contaminated areas of a site. These methods had some drawbacks. The first method simply moving the contamination elsewhere was found to involve significant risks in the excavation, handling and transportation of hazardous material. Additionally, it was very difficult and increasingly expensive to find new landfill sites for the final disposal of the material. The cap and contain method was only an interim solution since the contamination remains on site, requiring monitoring and maintenance of the isolation barriers long into the future, with all the associated costs and Potential liabilities (Vidali, 2001).

Biotechnology emerged an essential tool in this endeavor because it can provide new approaches such

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such as bioremediation for understanding, managing, preserving and restoring the environment. It can be used to assess the well being of ecosystems, transform pollutants into benign substances, generate biodegradable materials from renewable sources and develop environentally safe manufacturing and disposal processes. Bioreediation has also provided problem solving opporunities in this field by detoxifying industrial wastes/effluents; where in bacteria can be altered to produce certain enzymes that metabolize industrial waste components and also new pathways can be designed for the biodegradation of various wastes. Due to its compaatively low cost and generally benign environmental impact, bioremediation offers an attractive and/or suppleent to more conventional clean-up technologies, which generally have a high public acceptance and can often be easily carried out on sites. It will however not always be suitable, as the range of contaminants on which it is effective is limited, the time scales involved are relatively long and the residual contaminant levels achievable may some times be in appropriate. Although the methoologies employed are not technically complex, even still considerable experience and expertise may be required to design and implement a successful bioremediation program, in accordance to need to thoroughly assess a site for suitability and to optimize conditions to achieve a satisfactory result.

Since bioremediation seems to be a good alternative to conventional clean-up technologies substantial research in this field is rapidly increasing. Bioremediation has been used at a number of sites worldwide (King et al., 1997; National Research Council, 1993), including Europe, which resulted in varying degrees of success. Techniques are improving in as much as greater knowledge and experience are gained. There is no doubt for the fact that bioremediation has great potential for dealing with certain types of site contamination (Norris et al., 1993; Hinchee et al., 1995) shown in the form of table (Table 1). Unfortunately, the principles, techniques, advantages and disadvantages of bioremediation (Table 2) have not been widely known or understood, especially of those who will have to deal directly with bioremediation proposals, such as site owners and regulators. Some tests make an exhaustive examination of the literature of bioremediation of organic (King et al., 1997; National Research Council, 1993; Norris et al., 1993) and inorganic pollutants (Hinchee et al., 1995) and another test takes a look at pertinent field application case histories (Flathman et al., 1993).

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

Microbial populations and bioremediation processes

Certain enzymes produced by microbes attack hydrocarbons molecules, causing degradation. The degradation of oil relies on having sufficient microbes to degrade the oil through the microbes' metabolic pathways (series of steps by which degradation occurs). Fortunately, nature has evolved many microbes to do this job. Throughout the world there are over 70 genera of microbes that are known to degrade hydrocarbons (Glazer et al., 1995). These microbes usually account for less than 1% of natural population of microbes, but can account for more than 10% of the population in polluted ecosystems (Vidali, 2001).

If microbes are not present in a system they can be added to help promote bioremediation. The added microbes can be cultures grown from other contaminated areas or they can be microbes genetically engineered to degrade oil. However, even when these microbes are present, degradation of hydrocarbons can take place only if all other basic requirements of the microbes are met. Microorganisms can be isolated from almost any environ-

Table 1. Some contaminants potentially suitable for bioremediation (Vidali, 2001).

Class of contaminants	Specific examples	More potential sources
Chlorinated solvents	Trichloroethylene Perchloroethylene	Drycleaners Chemical manufacturing
Polychlorinated biphenyls	4-Chlorobiphenyl 4,4-Dichlorobiphenyl	Electrical manufacturing Power station Railway yards
Chlorinated phenol	Pentachlorophenol	Timber treatment Landfills
BTEX	Benzene Toluene Ethylbenzene Xylene	Oil production and storage Gas work sites Airports Paint manufacturing Port facilities Railway yards Chemical manufacture
Polyaromatic hydrocarbons (PAHs)	Napthalene Antracene Fluorene Pyrene	Oil production and storage Gas work sites Coke plants Engine works
Pesticides	Atrazine Carbaryl Carbofuran Coumphos Diazinon	Agriculture Timber treatment plants Pesticide manufacture Recreational areas Landfills

Table 2. Advantages and disadvantages of bioremediation (Vidali, 2001).

Technology	Examples	Benefits	Limitations
In situ	In situ bioremediation Biosparging Bioventing Bioaugmentation	Most cost efficient Noninvasive Relatively passive Natural attenuation Process	Environmental Constraints Extended treatment time Monitoring difficulties
Ex situ	Landfarming Composting Biopiles	Cost efficient Low cost Can be done on site	Space requirements Extended treatment time Need to control abiotic loss, mass transfer problem.
Bioreactors	Slurry reactors Aqueous reactors	Rapid degradation Kinetic Optimized environmental parameters	Soil requires excavation Relatively high cost capital

mental conditions. Microbes will adapt and grow at subzero temperatures, as well as extreme heat, desert conditions, in water, with an excess of oxygen and in anaerobic conditions, with the presence of hazardous compounds or on any waste stream. The main requirements are an energy source and a carbon source. These microbes because of their adaptability and other biological systems can be used to degrade or remediate environmental hazards (Ruldolph et al., 1996).

Environmental factors

Nutrients

Although the microorganisms are present in contaminated soil, they cannot necessarily be there in the strength required for bioremediation of the site. Their growth and activity must be stimulated. Biostimulation usually involves the addition of nutrients and oxygen to help indigenous microorganisms. These nutrients are the basic building blocks of life and allow microbes to create the necessary enzymes to break down the contaminants. Carbon is the most basic element of living forms and is needed in greater quantities than other elements. In addition to hydrogen, oxygen and nitrogen it constitutes about 95% of the weight of cells. Phosphorous and sulfur contribute with 70% of the remainders. The nutritional requirement of carbon to nitrogen ratio is 10:1 and carbon to phosphorous is 30:1 (Vidali M., 2001).

Environmental requirements

Microbial growth and activity are readily affected by pH, temperature and moisture. Although microorganisms have been also isolated in extreme conditions, most of them grow optimally over a narrow range, so that it is important to achieve optimal conditions. If the soil has too much acid it is possible to rinse the pH by adding lime. Temperature affects biochemical reactions rates and the rates of many of them become double for every 10°C rise in temperature. Above a certain temperature, however, the cells die (Pritchard et al., 1992). Plastic covering can be used to enhance solar warming in late spring, summer, and autumn. Available water is essential for all the living organisms and irrigation is needed to achieve the optimal moisture level. The amount of available oxygen will determine whether the system is aerobic or anaerobic.

Hydrocarbons are readily degraded under aerobic conditions, whereas chlorinated compounds are degraded only in anaerobic ones. To increase the oxygen amount in the soil it is needed to till the land for sparge of air. In some cases, hydrogen peroxide or magnesium peroxide can be introduced in the environment. Soil structure controls the effective delivery of air, water and nutrients. To improve soil structure, materials such as gypsum or organic matter can be applied. Low soil permeability can impede movement of water, nutrients and oxygen. Hence, soils with low permeability may not be appropriate for in situ clean-up techniques.

In situ and ex situ bioremediation processes

Different techniques are employed depending on the 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, ecofriendly option for clean up 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 et al., 1981, 1993). 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 intervenetion in the abatement of mercury pollution (Wistreich et al., 1988; Pritchard, 1991).

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 (Vidali M., 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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