

Concepts of bioremediation, bio augmentation and biosensors

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Abstract

Bioremediation uses microorganisms to destroy or immobilize waste materials. Microorganisms include archaea, bacteria, fungi and actinomycetes. Microorganisms destroy organic contaminants when they are using chemicals for their growth and reproduction. Although it is an old technique, it is still frequently used to remove environmental contaminants which have increased rapidly due to increased population, industrialization, and urbanization. There are various types of bioremediation, and different mechanisms associated with them. The process of bioremediation is enhanced by biostimulation. When bacterial culture is added to the contaminants to increase the rate of biodegradation, it is known as bio augmentation. Biosensors are devices used to detect the presence or concentration of a biological analyte, such as a biomolecule, a biological structure or a microorganism. This paper gives an idea of bioremediation, its scope, factors affecting bioremediation, types, biostimulation, bioaugmentation and biosensors.

Keywords: Mycoremediation, phytosequestration, rhizodegradation, phytodegradation and phytoextraction.

Introduction Bioremediation

In the current century, the entire world faces environmental contamination due to increased population, industrialization and urbanization. Environmental degradation is an enigma. However, science and technology have solved this problem up to a great extent by various techniques. Bioremediation is one among these and is now emerging as an innovative technology to remove environmental contaminants. Bioremediation involves living organisms, mainly microorganisms, to degrade the environmental contaminants into nontoxic forms. It works on very little investment and is applied to remove contaminants from soil, groundwater, surface water, and sediments, including air. Now, this technique is growing at an exponential rate.

Bioremediation operates through vegetation to sequester, extract, or degrade hazardous waste present in soils, sediments, groundwater, surface water and air. There are six essential mechanisms associated with bioremediation: 1. phytosequestration, 2. rhizodegradation, 3. phytodegradation, 4. phytohydraulics, 5. phytoextraction and 6. Phytovolatilization. The success of bioremediation depends upon many factors such as climate, soil conditions, suitable plant species and associated rhizosphere microbes [1,2].

The scope of environmental bioremediation extends to –

- *Inorganics* viz., Arsenic, Mercury, Chromium, Fluoride, Cyanide, abandoned mines, fly ash disposed sites, engineered Phyto treatment technologies, biological permeable barriers
- *Organics* viz., petroleum hydrocarbons, pesticides and explosives.

Different types of bioremediation are based on agents used like microbial bioremediation, which uses microorganisms to break down contaminants, using them as a food source. Mycoremediation uses fungi's digestive enzymes to break down contaminants such as pesticides, hydrocarbons and heavy metals. Moreover, when plants are used to bind, extract and clean up pollutants such as pesticides, petroleum hydrocarbons, metals and chlorinated solvents, it is known as phytoremediation.

Contaminants can be removed or reduced through in-situ bioremediation techniques or ex-situ [3]. Bioremediation techniques are also classified based on the treatment locality [4]. In-situ techniques help harmlessly in treating pollutants and is cost-effective. At the same time, ex-situ techniques require the contaminated site to be excavated, which increases the costs [5]. In both these approaches, nutrients, vitamins, minerals, and pH buffers need to be added to optimize the activity of microorganisms. Sometimes spe-

cialized microbial cultures are added (biostimulation) to enhance biodegradation further.

For the process of bioremediation, three main ingredients are essential. An electron acceptor, contaminant and microorganisms are needed. Naturally occurring compounds such as petroleum hydrocarbons allow degradation to occur quickly. Biodegradation of hydrocarbons in the soil can be limited by many factors such as nutrients, pH, temperature, moisture, oxygen, soil properties and contaminant presence [6-8].

1. Nutrients- Nitrogen and phosphorus can increase the rate of hydrocarbon degradation. There is an increase in cell growth rate, reducing the microbial lag phase enabling the microbial population to achieve high energy levels. However, a limitation is there as excessive content of nitrogen and phosphorus in soil cause microbial inhibition [9]
2. Moisture- Water content affects the diffusion of water and soluble nutrients entering and exiting the microorganism. However, excess moisture reduces the amount of oxygen during the aerobic process [9]
3. PH- The pH level of the soil affects the amount of nutrients present. For biodegradation of petroleum hydrocarbons, pH 7 is the ideal level for microbes to survive [9]

4. Temperature- Temperature controls the enzyme reactions of microorganisms. Mesophiles (25°C-45°C) is the ideal temperature for microbes to biodegrade [9]

Many types of researches are being carried out for bioremediation using various microorganisms Saranya *et al* [10]. carried out experiments of bioremediation of Mercury present in industrial effluents using *Vibrio fluvialis*. The results demonstrated that *V. fluvialis* has a solid ability to detoxify mercury from mobile solutions Saha *et al* [11]. Bharti *et al.* (2018) worked on measuring the bio remedial efficacy of *Nostoc Carneum* Agardh in industrial effluents treatment. Different physicochemical parameters were taken into considerations, and it was observed that the cyanobacterium *Nostoc* reduced the concentration of different wastes in industrial effluents Sharan *et al* [12]. reported Cyanobacterium adsorption on Cadmium which depends on Contact time. Many researchers like Kshirsagar & Sengar *et al.*[13] have reported a very high reduction in BOD using different algal species such as *Chlorella* and *Gloeocapsa*. They confirmed that microalgae are the best candidates for wastewater purification and improvement in their physicochemical parameters. According to Kalaivani *et al.* [14], the microalgae was very efficient in reducing BOD when sewage water was diluted in different concentrations.

Table1: Advantages and limitations of bioremediation

Advantages	Limitations
In situ	Limited to shallow soils, streams, and groundwater
Passive	High concentrations of hazardous materials can be toxic to plants
Solar driven	Mass transfer limitations associated with other biotreatments
Costs 10% to 20% of mechanical treatments	Slower than mechanical treatments
Transfer is faster than natural attenuation	Only effective for moderately hydrophobic contaminants
High public acceptance	Toxicity and bioavailability of degradation products is not known
Fewer air and water emissions	Contaminants may be mobilized into the groundwater
Generate less secondary wastes	Potential for contaminants to enter food chain through animal consumption
Soils remain in place and are usable following treatment	Unfamiliar to many regulators
Phytovolatilized contaminants could be transformed to less toxic forms (e.g. elemental mercury and dimethyl selenite gas) ii) phytovolatilization accelerates degradation processes	The contaminant or a hazardous metabolite might accumulate in vegetation and be passed on in later products such as fruit or lumber. Low levels of metabolites have been found in plant tissue.
Phytostabilization: i) circumvents the removal from soil, ii) It has a lower cost and is less disruptive than other more-vigorous soil remedial technologies, iii) Revegetation enhances ecosystem restoration.	i) The contaminants remain in place. ii) The vegetation and soil may require long-term maintenance to prevent re-release of the contaminants and future leaching. iii) Require extensive fertilization or application of soil amendments. iv) Plant uptake of metals and translocation to the aboveground portion must be avoided. v) The root zone must be monitored to prevent metal leaching.

<p>In phytoextraction, the plant biomass containing the extracted contaminant can be a resource (phytoextraction). For example, biomass that contains selenium (Se), an essential nutrient, has been transported to areas that are deficient in Se and used for animal feed. In green house experiments, gold was harvested from plants.</p>	<p>i) Metal hyper accumulators are generally slow-growing with a small biomass and shallow root systems. ii) Plants harvested must be properly disposed. iii) Phytoextraction studies conducted using hydroponically grown plants, with the contaminant added in solution, may not reflect actual conditions and results occurring in soil. v) Phytoextraction coefficients measured under field conditions are likely to be less than those determined in the laboratory</p>
<p>Rhizofiltration using terrestrial plants removes contaminants more efficiently than aquatic plants. ii) This system can be either in- situ (floating rafts on ponds) or ex- situ (an engineered tank system). iii) An ex- situ system can be placed anywhere because the treatment does not have to be at the original location of contamination.</p>	<p>i) The pH of the influent solution may have to be continually adjusted to obtain optimum metals uptake. ii) The chemical speciation and interaction of all species in the influent have to be understood and accounted for. iii) A well-engineered system is required to control influent concentration and flow rate. iv) The plants (especially terrestrial plants) may have to be grown in a greenhouse or nursery and then placed in the rhizofiltration system. v) Periodic harvesting and plant disposal are required. vi) Metal immobilization and uptake results from laboratory and greenhouse studies might not be achievable in the field.</p>

Biostimulation

Biostimulation is when the environment is modified to stimulate the bacteria capable of carrying out bioremediation. It is being done by the addition of various nutrients and electron acceptors, such as phosphorus, nitrogen, oxygen, or carbon (e.g. in the form of molasses), which are otherwise available in quantities low enough to constrain microbial activity [15-17]. Perfumo *et al* [18], explained it as the addition of nutrients, oxygen or other electron donors and acceptors to the coordinated site to increase the population or activity of naturally occurring microorganisms available for bioremediation. According to Margesin and Schinner biostimulation is a type of natural remediation that can improve pollutant degradation by optimizing conditions such as nutrients, aeration, pH and temperature control [19]. They suggested that biostimulation can be considered as an appropriate remediation technique for petroleum pollutant removal from soil. It requires evaluating both the intrinsic degradation capacities of the autochthonous microflora and the environmental parameters involved in the kinetics of the in-situ process. The primary advantage of biostimulation is that bioremediation can be undertaken by already present microorganisms that are well-suited to the subsurface and are well distributed spatially within the subsurface. The primary challenge is that the delivery of additives must be in a manner that allows them to be readily available to subsurface microorganisms based on the subsurface's local geology. Tight, impermeable subsurface lithology (tight clays or other fine-grained material) make it challenging to spread additives throughout the affected area. Fractures in the subsurface create preferential pathways in the subsurface, which additives preferentially follow, preventing even distributing additives. The addition of nutrients might also promote the growth of heterotrophic microorganisms, which are not innate degraders of total petroleum hydrocarbon, thereby creating a competition between the resident microflora [20].

Bioaugmentation

Bioaugmentation is the manual addition of an organic culture of bacteria to an environment such as a bioreactor to speed up the rate of degradation of sewage or a contaminant. Organisms originating from contaminated areas may already break down waste, but perhaps inefficiently and slowly, which is being used in bioremediation. Bioaugmentation is to enrich the bio remedial population

to make it more efficient in removing contaminants. The addition of pre-grown microbial cultures to bioreactors and other treatment equipment make them ready to hit optimal levels of efficiency. Main objective behind the addition of archaea or bacterial cultures is to increase the efficacy of the processes within the bioreactor or another industrial application so that the contaminants can be removed quickly and effectively and at a lower cost. It makes it easier for organizations to maintain a balance, protecting profits while also striving for ecological sustainability [17].

Biosensor

A biosensor is a device that helps in measuring biological or chemical reactions through generating signals equivalent to the concentration of the analyte in the reaction. Biosensors are employed in drug discovery, disease monitoring, detection of pollutants, disease-causing microorganisms and indicators of disease in bodily fluids (blood, urine, saliva, sweat). A typical biosensor consists of the following components.

- **Analyte:** It is a substance that needs to be detected. For example, glucose is an 'analyte' and can be detected by a biosensor designed to detect glucose.
- **Bioreceptor:** A molecule that can recognize the analyte is known as a biosensor. Enzymes, cells, aptamers, deoxyribonucleic acid (DNA) and antibodies are used as receptors. The process of signal generation (in the form of light, heat, pH, charge or mass change, etc.) and interaction of the bioreceptor with the analyte is called bio-recognition.
- **Transducer:** It is an element that transforms one form of energy into another. In a biosensor, the role of the transducer is to convert bio-recognition into an appropriate signal. This process of energy transformation is called signalization. Most transducers produce either optical or electrical signals proportional to the concentration of analyte-bioreceptor interactions.
- **Electronics:** This part of the biosensor processes and transduces signal for its proper display. It consists of complex electronic circuits that perform signal conditioning involving amplification and conversion of signals from analogue into digital form. The display unit of the biosensor quantifies the processed signals.
- **Display:** The display consists of an interpretation system consisting of a liquid crystal display or a direct printer generating

numbers or curves that the user can understand. It consists of both hardware and software that would generate results of the biosensor in a user-friendly manner. The output signal on the display unit can be numeric, graphic, tabular or an image, depending on the end user's requirements.

Conclusion

Researches show that bioremediation is significantly less expensive than other technologies that are often used to clean up hazardous waste. There are several cost or efficiency advantages of bioremediation that can be employed in inaccessible areas without excavation.

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