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ISBN: 978-93-94570-62-7

11. Bioremediation

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Key Concepts:

After completing this chapter, you will be able to:

- Understand the concept of bioremediation.
- Discuss how selection of microorganisms is done and how biodegradation is done using natural and modified microbes.
- Details about techniques used in bioremediations.
- List of different pollutants which can be brake down using bioremediation.

11.1 Introduction:

Absorption and degradation of the xenobiotic present in the environment with the help of microorganisms is called biodegradation. The focused pollutants are metals and radionuclides which make the site unsuitable for the living organism to flourish. The microorganisms used for bioremediation are either genetically modified or selected from nature. Bioremediation can either be carried out in situ, which is at the site of contamination itself, or ex-situ, which is located away from the site. The selection of technic depends on the range of factors like type, concentration, or characteristics of pollutants. Microbus utilizes contaminants as a source of food and energy and converts them into simple elements which are harmless to the environment. The optimum condition for the growth of microorganisms should be fulfilled for proper growth, which is provided by adding amendments like molasses, vegetable oil, or gases. Which thereby accelerates the bioremediation process. The various factors involved in bioremediation are an energy source, nutrition, pH, temperature, oxygen content, characteristics of contaminant, etc. The advantage of bioremediation includes its being cost-effective and fewer harmful by-products.

Some disadvantage like the growth of microbes is not controlled resulting in toxic byproducts, providing optimal condition for the growth of microbes In situ is of major concern, also the process take long time as compare to other remediation technique. Bioremediation can be classification into three phytoremediation, bacteria remediation and mycoremediation. Examples of bioremediation are contaminated soil, oil spill, ground water, crime scene clean up.

11.1.1 Definition:

Bioremediation is a branch of biotechnology that employs the use of living organism like microbes and bacteria in the removal of contaminants, pollutants and toxins from soil, water and other environments. (Cory Mitchell).

Bioremediation is a waste management technique that includes the Living Organism living organisms to eradicate or neutralise pollutants from a contaminated site. Bioremediation is a treatment technique that uses naturally occurring organisms to break down harmful material into less toxic or non-toxic material.

11.2 Selection of Microbes:

Microorganisms can grow at temperature below 0°C to extreme heat. The microbes are adaptive to new environment and their biological system made them suitable for remediation process. Carbon is the main requirement for microbial activity. The native microorganisms can stimulate by providing resources to grow and proliferate the process is called as bio-stimulation. It also increases the rate of degradation.

Microbes break down pollutants via there inherent metabolic processes with or without slight pathway modification. By using genetic engineering and transgenic technics specially designed microbes can be used at the site which can degrade the pollutants more efficiently. Naturally occurring microbes can degrade hydrocarbons, polychlorinated compounds, polyromantic hydrocarbons, radionuclides and metals. Aerobic microorganisms used for bioremediation are *Pseudomonas*, *Acinetobacter*, *Sphingomonas*, *and Mycobacterium* etc. They degrade pesticides, hydrocarbons, alkanes and polyromantic compounds. Anaerobic bacteria are not used frequently as aerobic bacteria.

11.2.1 Biodegradable Pollutants:

Pollutants which can be degrade by bioremediation are -

- Pesticides
- Agrochemicals
- Non- chlorinated pesticides and herbicides
- Organic halogens
- Inorganic metals (lead, chromium, mercury)
- Gases (ozone, nitrogen dioxides, sulphur dioxide)
- Petroleum hydrocarbons
- Radionuclides
- Dyes
- Plastics explosive and sludge.

Soil gets polluted by various industrial and agricultural activities due to deposition of heavy metals, chemical spillers or pesticide usage. It can be clean by bio stimulation or addition of microbes to the site of contamination. Air gets polluted by industrial emission carrying volatile compounds, dust particles, toxic gases etc. Bio filtration is used for cleaning industrial gases by passing polluted air over a microbial culture medium.

That degrades contaminants into co2, water or salts. Bio filtration is the only biodegradable technique currently available to remediate air born pollutants. Water is treated by aerobic and aerobic strains of bacteria. That degrades pollutants and organic matter present in waste water.

11.2.2 Factor Responsible for Bioremediation Process:

- Temperature.
- Moisture.
- Electron acceptors.
- Cost.
- Nutrients.

- pH.
- Soil density, permeability, texture etc.
- Size of contaminated area.
- Concentration of contaminants.
- Properties of pollutants.

11.2.3 Bioremediation Techniques:

Bioremediation can be generally classified as in-situ, intrinsic and ex-situ.

Table showing classification of bioremediation technique-



Figure 11.1: Classification of Bioremediation Technique

11.2.4 In-situ bioremediation:

In in-situ techniques contaminants are treated at the site only it does not require excavation from the site. It is less costly than ex-situ as no equipment is required on site.

The major concern is to increases efficiency of microbes by providing optimum conditions (nutrient, pH, moisture content, temperature) for growth of microorganism. Soil porosity and texture play important role in in-situ technique.

A. Different technique classified under in-situ technique are-

- **a. Bioventing-** In this technique airflow of oxygen is pass to unsaturated zone to stimulate the activity of indigenous microorganism. Nutrients and water is also added to soil to enhance bioremediation. It is used in diesel polluted soil, hydrocarbons polluted site, reduction of chlorinated compounds under aerobic condition. Permeability of soil enhances the biodegradation process.
- **b. Biosparging** here air is injected to soil subsurface to stimulate microbial growth. Which help in movement of pollutant from saturated zone to unsaturated zone in turn promote biodegradation? It depends upon soil permeability and pollutant biodegradability. It is used in remediation of benzene, petroleum products, toluene, xylene, etc.
- **c. Bio slurping** Here a light non-aqueous phase liquids (LNAPLs) is pumped to soil surface by upward movement. Which in turn help in increasing the permeability of oxygen and hence increase the microbial activity. It is used for volatile and semi volatile compounds remediation at saturated and unsaturated zone. Here main concern is soil moisture which decreases air permeability in turn oxygen transfer rate.
- **d. Phytoremediation-** In this technique plants are used for biodegradation of pollutants. Plants can breakdown, remove, accumulate or convert pollutants into non-toxic compounds present at the site. Hundreds of plant species are identified as accumulators of pollutants e.g., Hemp, alfalfa, Indian mustard, water hyacinth, corn, sunflower, tobacco etc. used for remediation of metals (As, Cu, Pb, Zn, Cd) gasoline, ethers etc. recombinant DNA technology is used to produce transgenic plants which increase metabolism and degradation of heavy metal.

11.2.5 Type of Phytoremediation Are:

• **Phytostabilization**- secretion from roots of the plant precipitates the toxins and made them less available.

- **Phytotransformation** plant uptake the pollutants and transform it into another product.
- **Phyrovolatilization** plant evaporate the volatile pollutants by transpiration process.
- **Phytoextraction** direct uptake of pollutants and concentration of pollutants in plant tissue followed by removal of plant from the site.
- **Phytostimulation** enhancing the process of degradation in rhizosphere.

11.2.6 Advantages of Phytoremediation:

- It's easy to observe plants growth and changes.
- It's cost effective.
- Doesn't cause any harm to environment.
- It is helpful in extraction of metals by phytomining.
- Helpful at the site where excavation is not possible.
- Prevention of erosion and leaching of metal.

11.2.7 Limitations of Phytoremediation:

- Long time is required as compare to other remediation techniques.
- Pollutant concentration, toxicity tolerance.
- Accumulated toxins can be transferred to food chain.

Permeable reactive barrier (PRB): It include precipitation, degradation and sorption of pollutants. It is in-situ technique used for remediation of heavy metal and chlorinated compounds. It is physical method where a permanent or semi-permanent reactive barrier (medium) is submerged in polluted groundwater where pollutants are trapped and undergo series of reactions in turn clean the water.

11.2.8 Advantages of In-Situ Technique:

- It is done at the site so no extra affords required for excavation and transportation.
- Less costly.

- Native microorganisms are only used most of the time so no laboratory expense is included.
- No disturbance to soil.
- No sophisticated equipment is required.

11.2.9 Limitations of In-Situ Technique:

- Providing optimal condition on site is a difficult task.
- Soil texture, porosity, permeability and environmental conditions are limiting factors in in-situ technique.

Intrinsic bioremediation-

it is a type of natural in-situ technique where no extra affords is applied for bioremediation. It totally depends on aerobic and anaerobic microorganisms to biodegrade the pollutants. It is observed in biodegradation of saturated and unsaturated hydrocarbon. It is passive form of remediation and is less expensive as compare to other bioremediation process. Intrinsic approach is limited as it takes a long time to eradicate pollutants and it is limited to some xenobiotic compounds only.

A. Different naturally occurring bioremediation processes are-

- **Bio stimulation-** In this process indigenous microorganism is provided with additional nutrients which enhances biodegradation.
- **Bio augmentation**-In this process exogamic microorganism are added which capable of detoxifying the contaminants, mostly genetically modified microbes are used.
- **Natural attenuation**-Here biodegradation is totally depended upon the indigenous microbes without any human intervention other than monitoring.

11.3 Ex-Situ Bioremediation:

Ex-situ means off site i.e., taking the contaminants to other site or treating it elsewhere. The contaminated soil can be treating in bioreactors where microorganisms can grow in control condition like pH, temperature and nutrient supply.

Bio pile, land farming or windrowing is used other than bioreactor in ex-situ bioremediation. Exogamic microbes, nutrients, water, air supply is provider to contaminated soil which enhances biodegradation process.

A. Different technique classified under ex-situ are:

a. Bio pile:

It's an ex-situ technique where contaminated soil is mixed with soil containing microbes. Nutrients, moisture, heat, nutrients, oxygen and pH can be controlled to increase biodegradation process.

It is useful in cold environment and to treat low molecular weight volatile pollutants like petroleum pollutants, halogenated volatile organic compounds and pesticides. In this technique aeration is provided thru proper air supply system which increases the oxygen supply and hence microbial activity.

b. Land farming:

In this technique contaminated soil, sludge are incorporate into soil surface and tiled or turn over which provide aeration and thus increases the biodegradation process. Land farming may be in-situ or ex-situ depends up on depth of pollutant in the soil.

If the pollutant is present at depth of <1m below the ground then it is treated with in-situ techniques. If pollutants are present below >1.7 m then ex-situ techniques are used.it is useful in oil sludge and petrol refinery waste treatment. Inorganic contaminants can't be treated with this technique.

c. Windrows:

Here polluted soil is rotated periodically which in turn increases the aeration and nutritional supply for microbes and accelerate the bioremediation process. This is most cost-effective technique for bioremediation.

Contaminated soil is piled up into small hill which is periodically turned upside down which increases oxygen concentration, nutrient and other amendments are also added during turning process.

d. Bioreactor:

Bioreactors are large vessels in which raw material are treated to form different products. Polluted soil or slurry can be treated using bioreactor.

Bioreactors are designed according to the mode of operation like batch, fed batch or continuous.

Bioreactor provide controlled environment for microbial growth hence increases biodegradation process.

11.3.1 Advantages of Ex-Situ Technique:

- Less laborious
- Less expensive
- Result is faster as compare to in-situ.
- It's easy to control the growth condition for microorganisms.
- Preliminary assessment of site is not required.
- Use for wide range of contaminants.
- Large volume can be handled easily.

11.3.2 Disadvantages of Ex-Situ Technique:

- It is not possible to perform ex-situ under the building, in city, and crowded area.
- It disrupts soil texture and structure.
- Not applicable for heavy metals.
- Required large area for treatment thus site size is main constrain.
- Only aerobic biodegradable contaminants can be treated.
- Hard to provide optimum conditions for microbes to grow.



Figure 11.2: Steps for Bioremediation using Microbes

Advantages and Limitations of Bioremediation:

11.3.3 Advantages of Bioremediation:

- It is often be carried out on site.
- It does not require too much of sophisticated equipment.
- Bioremediation is less expensive.

• End product is safe to the environment.

11.3.4 Limitations of Bioremediation:

- Bioremediation is limited to only biodegradable compounds.
- Bioremediation take longer time than other treatment options.
- Research and development are requiring engineering microbes that are not naturally present in environment.

11.3.5 Applications of Bioremediation:

- Waste water and industrial effluent treatment-bioremediation is sustainable method accepted worldwide over expensive chemical method in sewage treatment plant and effluent treatment plant. Microorganisms are seeming to be effective in biodegradation of pollutants to less toxic substance which make waste water suitable for disposal or for further use.
- Control of air pollution- air filters were developed using microbes immobilized on inert material when polluted air come in contact with microbes undergoes biodegradation.it is useful for absorption of toxins released from industries. It is effective detoxification method and also helps in volume reduction. Phylloremidiation is also known for reduction of air pollutants like particulate matters, ozone, nitrogen dioxides, sulphur dioxide and volatile organic compounds. Phylloremidiation is a bioremediation method where plant leaves and microbes associated with leaves adsorbs air pollutants and degrades it.
- Soil and land treatment:-Bio slurping, bio venting, bio sparing, phytoremediation are various methods used for efficient bioremediation of toxic pollutants from soil. Bio remediation is effective for biodegradation of petroleum hydrocarbons, heavy metals and agricultural pesticides which is a major concern for health and environment.
- Solid waste management- bioremediation of solid waste can be done by windrow composting; here microorganisms were sprayed on the windrows of solid waste which is then converted into manure. This manure there by used in agriculture practices.

11.4 Conclusion:

Bioremediation is a natural remediation process for removal of toxic pollutants from the site. It is mainly classified in to in-situ and ex-situ, as compare to ex-situ technique in-situ is less costly as it does not required excavation of pollutants, installation of big equipment or laboratory cost. Process of bioremediation involves analysis of pollutants, finding right microbes for biodegradation, standardising the process and its pilot scale implementation. Selection of technique depends up on pollutant concentration, depth, type, location of site, performance and human habitation.

11.4.1 Questions for Review:

- Define bioremediation.
- Describe different type of bioremediations techniques. What are the criteria for selection of technique?
- Discuss the strategy to be followed for efficient bioremediation.
- What are the limitations and advantages of bioremediation?
- Discuss selection of microbes for effective biodegradation.
- What are the different applications of bioremediation?
- Discuss the factors responsible for bioremediation.

11.4.2 Questions for Discussion:

- Make a list of different pollutants can be effectively treated by using bioremediation.
- Discuss how efficiency of microbes can be increase for effective biodegradation. Using naturally occurring microbes and increasing its efficiency or using genetically modified microbes is preferred?
- Give your review on statement-"phytoremediation is the future of bioremediation".

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