**BIOREMEDIATION**

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**Introduction:**

Globally, there is a trend toward the development of quick, affordable, and effective simple solutions that may be applied in-situ to restrict the migration of pollutants into the ground or other neighbourhoods, eliminate pollutants, and restore the environment. Accelerating the biodegradation process is possible with the use of decontamination technology. The biodegradation of petroleum hydrocarbons existing in different environments, particularly in the ground, is based on one hand on the use of microorganisms indigenous existing in nature and adapted to the pollutant in question and on the other hand, the introduction of micro-organisms specific species .

The term "bioremediation" refers to the employment of biological processes to essentially eliminate pollutants or other impairments from soil and water. "Bioremediation" is a procedure that involves breaking down natural or synthetic elements by triggering specific strains of specialised microbes, producing end products that are either helpful or acceptable in terms of their influence on the environment. Detoxification, reduction, degradation or transformation of hazardous chemicals into less toxic ones is the primary mechanism underlying this procedure. The major factors that affect the bioremediation process are the nature of the contaminants, such as the kind of pesticides, agricultural chemicals, organic halogens, heavy metals, xenobiotic compounds, heavy metals, plastics, organic halogens, greenhouse gases *etc.* Nuclear waste processing also employs this technique.

**1. Benefits and drawbacks of bioremediation:**

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| **Benefits** | **Drawbacks** |
| Bioremediation is an sustainable and eco-friendly approach | Bioremediation technology is restricted to biodegradable compounds. |
| There are no hazardous chemicals used in bioremediation. It occasionally uses nutrients, such as fertilisers, to stimulate the microbial population. | Occasionally, the new substance that develops from biodegradation may be far more damaging to the environment than the original chemical. |
| Bioremediation is economical and less labor-intensive. | Bioremediation is time-consuming process. |
| It does not generate waste and self-sustaining. | Extrapolating bio remediation to the field level from the laboratory level is challenging. |
| Microorganisms involved in bioremediation are environmentally safe and are capable to decompose a variety of pollutants. | The effectiveness of the microorganisms of the soil may be restricted due to lack of oxygen and other nutrients. |
| Bioremediation can be made on the site directly without any ecosystem disruption. | Contaminants that had been stabilised could get remobilized if the hydrological and geochemical conditions change over time. |
| Bioremediation can eliminate the transportation and operating costs associated with waste management. | Ex-situ bioremediation required excavation and pumping. |
| Bioremediation can be combined with other treatment technologies. | Public awareness and education is necessary for wide acceptance of bioremediation. |

**2. Principles of bioremediation:**

The utilisation of living creatures, particularly microorganisms, in the reduction of toxic contaminants to less toxic chemicals is the fundamental principle behind bioremediation. This technology detoxifies or degrades dangerous compounds from the environment using bacteria, fungi, and/or plants. Living organisms change pollution through metabolic processes including enzyme synthesis. Several microorganisms are used in this approach to break down contaminants at the contaminated site.

**3. Techniques of Bioremediation**

**There are two major techniques of bioremediation such as ex-situ and in-situ bioremediation techniques (Figure 1). When selecting a bioremediation method, factors such as the type of pollutant, the extent and volume of the contamination, the type of environment, the location, the cost, and environmental regulations are taken into account. The efficiency of bioremediation procedures is influenced by a variety of abiotic parameters including temperature, pH, nutrient concentrations, oxygen concentrations, and others (Frutos et al. 2012; Smith et al. 2015).**

**3.1 Ex-situ bioremediation techniques**

**Ex-situ bioremediation strategies include removing pollutants out of polluted areas and treating them elsewhere once they have been moved. The amount of pollution, the type of pollutant, the severity of the pollution, the cost of treatment, and the location of the contaminated site are often taken into consideration when ex-situ bioremediation methods are being evaluated. The ex-situ bioremediation technology is influenced by performance standards as well.**

**3.1.1 Solid-phase treatment**

**In solid-phase treatment or solid-phase bioremediation, the contaminated soil is removed from its natural environment and piled up. It also comprises home, industrial, and municipal trash, as well as organic wastes like leaves, animal dung, and agricultural wastes. Pipes installed all over the heaps are used to transport bacterial growth. For ventilation and microbial respiration, air must be pulled via the pipes. In comparison to slurry-phase procedures, a solid-phase system needs more area and cleanup takes longer time to carry out. Processes for solid-phase treatment include composting, land farming, windrows, bio piles *etc*.(**Kulasrestha et al. 2014**).**

 Bioremediation Techniques

 Ex-Situ

 In-situ

Biopiling

Windrow

Bio

reactor

Land farming

Natural attenuation

Enhanced

Bioslurping

Bioventing

Biosparging

Phyto

remediation

Permeable reactive barrier

Solid phase

Slurry phase

**Fig.1.** Bioremediation techniques

**3.1.1.1 Types of ex-situ bioremediation**

**3.1.1.1.1 Bio pile**

Bio pile method of bioremediation entails piling of excavated contaminated soil above ground, followed by nutrient supply and occasionally aeration, in order to increase bioremediation by successfully raising microbial activity. This method consists of a treatment bed, fertiliser and leachate collecting systems, irrigation, and aeration. Ex situ technique is being explored more frequently due to its advantageous properties such as cost effectiveness, effective biodegradation under adequate nutrient, temperature and aeration (Whelan et al. 2015).

**3.1.1.1.2 Windrows:**

Windrows technique is the regular rotation of the stacked polluted soil to improve the biodegradation activities of the native and/or transitory hydrocarbonoclastic bacteria such as Agrobacterium, Sphingomonas, *Pseudomonas*, Diatzia, Nitratireductor, Acinetobacter etc. present in the contaminated soil. Regular turning of contaminated soil and the addition of water encourage aeration, uniform distribution of pollutants, nutrients, and microbial degradative activities. It also quickens the process of bioremediation by absorbing pollutants and transforming them into other forms of life, such as minerals (Barr, 2002). Windrow treatments shown a higher rate of hydrocarbon removal in comparison to bio pile treatments; nevertheless, the soil type, which was thought to be more friable, was responsible for the windrow's superior efficacy in this area (Coulon et al. 2010). The establishment of an anaerobic zone inside heaped polluted soil is associated with CH4 (greenhouse gas) emission (Hobson et al. 2005).

**3.1.1.1.3 Land farming:**

Though occasionally seen as an in situ bioremediation technology, land farming is commonly thought of as ex situ bioremediation. The treatment site is at the centre of this controversy. The depth of the contamination has a big impact on whether land farming can be done in situ or ex situ. The common procedure in land farming is to excavate and/or till the contaminated soils. When treated on site, excavated polluted soil may be considered as in situ; otherwise, it is ex situ because it more closely resembles other ex situ bioremediation methods. According to studies, bioremediation can occur without excavation when a pollutant is less than one metre below ground level, but when it is more than one and a half metres below ground, it is necessary to transport the pollutant to the surface for effective bioremediation (Nikolopoulou et al., 2013). In order to facilitate aerobic biodegradation of pollutants by native microorganisms, excavated polluted soils are typically carefully spread on a fixed layer support above the ground surface (Philp and Atlas 2005; Paudyn et al. 2008; Volpe et al. 2012; Silva-Castro et al. 2015). It was found that in a soil with sufficient biological activity, tillage and irrigation without the addition of nutrients increased the number of heterotrophic and bacteria that break down diesel, speeding up the process of bioremediation. Dehydrogenase activity was also discovered to be an excellent bio stimulation treatment indication and may be employed as a biological metric in land farming techniques (Silva-Castro et al. 2015).

**Advantages of land farming**

* Land farming is the most simple bioremediation technique.
* Land farming is very economical and required very few numbers of equipments for operation.

**Limitations of land farming**

* Large operating space is required.
* Unfavorable environmental circumstances can cause a decrease in microbial activity.
* Excavation work in land farming could result in extra costs.
* Land farming is less efficient in removal of inorganic pollutant.
* The design and method of pollutant removal (volatilization) enable land farming inappropriate for treating soil contaminated with harmful volatiles, especially in hot (tropical) climate zones.
	+ 1. **Slurry-phase bioremediation**

Slurry-phase bioremediation is a little faster method of treatment than the others. For instance, in a bioreactor, polluted soil is mixed with water, nutrients, and oxygen to provide the ideal environment for the microorganisms to degrade the toxins that are present in the soil. During this procedure, stones and debris are removed from the contaminated soil. The physicochemical properties of the soil, the rate of biodegradation, and the quantity of pollutants all affect the concentration of added water. Centrifuges, vacuum filters, and pressure filters are used to extract the soil after completion of this process and then the soil is dried. The next step is to dispose of the soil and treat the resulting fluids in advance.

**3.1.2.1** **Bioreactor:**

A bioreactor is a container where certain products are produced from raw materials as a result of a series of biological reactions. There are different methods for operating the bioreactor including batch, fed-batch, sequencing batch, continuous, and multistage. The operational method is greatly influenced by the market economy and capital investment. The natural environment of the cells being examined is replicated and maintained in a bioreactor to produce the perfect growing environment. The bioreactor technique has number of advantages. One of the main benefits of bioreactor-based bioremediation is its excellent control of the bioprocess parameters (temperature, pH, agitation and aeration rates, substrate and inoculum concentrations). The ability to regulate and alter a bioreactor's process parameters suggests that the biological reactions taking place inside can be improved to significantly speed up bioremediation.

**3.1.3 Advantages of ex-situ bioremediation**

* It is effective against a variety of pollutants
* Can be evaluated very easily from site investigation data.
* It has more manageable, predictable and controlled environment for remediation.

**3.1.4 Disadvantages**

* Non-permeable soil requires further processing.
* Ex situ bioremediation is not relevant to contamination caused by heavy metals or chlorinated hydrocarbons like trichloroethylene.
* Before adding soil to a bioreactor, the pollutant might be removed physically or by washing the soil.

**3.2 In-situ bioremediation techniques**

In-situ bioremediation techniques entail handling polluted materials right where the pollution occurred. Since there is no need for excavation, the soil structure is not significantly disturbed. These methods are less expensive than ex situ bioremediation methods since there is no additional cost for excavation processes. Some in situ bioremediation methods (such as bioventing, biosparging, and phytoremediation) may be improved, while others may continue as it is (intrinsic bioremediation or natural attenuation). Techniques for in-situ bioremediation have proved effective in cleaning up sites that have been contaminated by heavy metals, hydrocarbons, dyes, and chlorinated solvents (Folch et al. 2013; Kim et al. 2014; Frascari et al. 2015; Roy et al. 2015). However, the presence of nutrients, moisture content, pH, and temperature are among the crucial environmental factors that must be satisfied for an successful in situ bioremediation process (Philp and Atlas 2005).

**3.2.1 Types of in-situ bioremediation**

There are two types of in-situ bioremediation: enhanced (engineered) bioremediation and natural (intrinsic) bioremediation.

**3.2.1.1 Natural(intrinsic) bioremediation**

Intrinsic or natural bioremediation entails passive repair of polluted environments without the use of any external force such as human intervention. Polluting compounds, especially those that are refractory, are biodegraded via both aerobic and anaerobic microbial processes. Due to the lack of an external force, this method may be less expensive than other in situ techniques. However, the process must be observed in order to prove that bioremediation is continuing and sustained; termed as "monitored natural attenuation" (MNA). MNA is frequently used to refer to a more comprehensive strategy for intrinsic bioremediation. The US National Research Council (US NRC) states that intrinsic bioremediation must meet three requirements: proof of contaminants loss from contaminated sites, laboratory-based proof that microorganisms isolated from contaminated sites have the innate potential to biodegrade or transform contaminants present at the contaminated site from which they were isolated, and proof of realising biodegradation (Philp and Atlas 2005). The majority of European nations are gradually accepting MNA because to the cold weather conditions that are likely to have a negative impact on the normal biodegradation process (Declercq et al., 2012). Since no outside force is used to speed up the cleanup process in intrinsic bioremediation, one of its significant drawbacks is that it could take longer to reach the required level of pollutant concentration. In order to ensure that the remediation duration is less than the time allotted for the pollutant to reach the exposure point in relation to the closest human and animal populations, risk assessment must be finished prior to intrinsic bioremediation. Additionally, it was shown that intrinsic bioremediation is insufficient to remove polyaromatic hydrocarbons (PAHs), which would reduce the ecotoxicity of contaminated soil (Garcia-Delgado et al. 2015).

**3.2.1.2 Enhanced (engineered) in-situ bioremediation**

Enhanced (engineered) in situ bioremediation includes introducing certain microorganisms to the contaminated site. It uses genetically modified bacteria to speed up the degradation process by enhancing the physicochemical conditions that promote the growth of microorganisms.

**3.2.1.2.1 Bioventing**

This method uses a regulated airflow stimulation to promote bioremediation by increasing the activity of local bacteria by providing oxygen to the unsaturated (vadose) zone. The ultimate goal of bioventing is to encourage the microbial transformation of contaminants into a harmless state. For this generally nutrients and moisture are added to enhance bioremediation (Philp and Atlas 2005). This method has become more popular than other in situ bioremediation methods, particularly for cleaning up areas where light petroleum compounds have been spilled (Hӧhener and Ponsin 2014). Bioventing can be used for anaerobic bioremediation even though its principal objective is to improve aeration in unsaturated zones, particularly for the treatment of vadose zones contaminated with chlorinated chemicals that are resistant to aerobic treatment, where hydrogen acts as an electron donor, and to reduce chlorinated vapour, a mixture of nitrogen, small amounts of hydrogen and carbon dioxide can be introduced in place of air or pure oxygen (Mihopoulos et al. 2000; Shah et al. 2001).

**3.2.1.2.2 Bioslurping**

This technique uses vacuum-enhanced pumping, bioventing and soil vapour extraction to indirectly add oxygen and encourage pollutant biodegradation while remediating soil and groundwater (Gidarakos and Aivalioti 2007). The method is made to recover free products such light non-aqueous phase liquids (LNAPLs), which can be used to remediate saturated, unsaturated, and capillary zones.  Additionally, soils that have been contaminated with organic volatile and semi-volatile compounds can be cleaned up using it. Similar to how a straw sucks liquid from any vessel, the device uses a "slurp" that extends into the free product layer to suck out liquids (free products and soil gas) from this layer. The pumping mechanism launches LNAPLs upward to the surface, where they are cut off from water and air. The system is readily set up to operate as a typical bioventing system once all free products have been eliminated, which will complete the remediation procedure (Kim et al. 2014). In this method, too much soil moisture restricts air permeability and slows the pace at which oxygen is transferred, which in turn lowers microbial activity. The technique saves money because it produces less groundwater as a result of the operation, which reduces the expenses associated with storage, treatment, and disposal even though it is not ideal for remediating soil with low permeability (Philp and Atlas 2005). The biggest drawback of this specific in situ technology is the prospect of establishing a vacuum on a deep, highly porous location and t he fluctuating  ground water table which can result in saturated soil lenses that are difficult to aerate.

**3.2.1.2.3 Biosparging**

Biosparging, that is similar to bioventing, involves pumping air into the subsurface of the soil to encourage microbial activity and aid in the removal of pollution from polluted areas. Unlike bioventing, air is blasted at the saturated zone, perhaps transporting volatile organic molecules upward to the unsaturated zone to encourage biodegradation. The soil permeability, which affects the pollutant's bioavailability to microorganisms, and the pollutant's biodegradability are the two main parameters that influence how successful biosparging is (Philp and Atlas 2005). While biosparging encourages biodegradation, in situ air sparging (IAS), which is comparable to soil vapour extraction (SVE), uses high airflow rates to induce pollutant volatilization. Each technique's pollution removal methods do not compete with one another. Diesel and kerosene contamination of groundwater has been successfully eliminated in numerous cases via biosparging. According to Kao et al., biosparging of a benzene, toluene, ethylbenzene, and xylene (BTEX)-contaminated groundwater plume resulted in a shift from anaerobic to aerobic conditions (2008). This was demonstrated by an increase in total culturable heterotrophs and a decrease in dissolved oxygen, as well as redox potentials, nitrate, and sulphate.

**3.2.1.2.4 Phytoremediation**

In order to reduce the dangerous effects of contaminants, this strategy involves plant interactions such as physical, biochemical, biological, chemical, and microbiological in polluted areas. There are a number of mechanisms involved in phytoremediation which depends on the pollutant type (elemental or organic). These mechanisms are accumulation or extraction, degradation, filtration, stabilisation, and volatilization. Extraction, transformation, and sequestering are the main methods used to eliminate elemental contaminants (toxic heavy metals and radionuclides). In contrast, organic pollutants (such as hydrocarbons and chlorinated chemicals) are primarily removed through degradation, rhizoremediation, stability, and volatilization with the possibility of mineralization when certain plants, such as willow and alfalfa, are utilised (Meagher 2000 and Kuiper et al., 2004). When selecting a plant to act as a phytoremediator, it's important to take into account a variety of factors, including the plant's root system (which, depending on the depth of the pollutant may be fibrous or tap-like), above-ground biomass (which shouldn't be fit for animal consumption), the pollutant's toxicity to plants, the plant's ability to survive and adapt to its environment, its growth rate, site monitoring and most importantly the amount of time needed to reach the required level of cleanliness are some of the key considerations. Moreover, the plant should also be immune to pests and illnesses (Lee 2013). According to Miguel et al. (2013), in some contaminated situations, contaminant removal by plants includes three steps: uptake, which is primarily a passive process (translocation from roots to shoots; and accumulation in shoot). Additionally, transpiration and the division of xylem sap between adjacent tissues are required for translocation and accumulation, respectively. Nevertheless, the procedure varies depending on other elements including the type of contamination and the plant. The majority of plants living on any polluted site are likely effective phytoremediators. In order to maximise the remediation capacity of native plants growing in contaminated areas, either through bioaugmentation with endogenous or exogenous plant rhizobacteria or through biostimulation is essential for the success of phytoremediation strategy. These are some benefits of phytoremediation:

* Cheap cost
* Environmental friendliness
* Widespread use
* Cheap installation and maintenance
* Preservation of soil structure
* Reduction of soil erosion
* Prevention of metal leaching (Van Aken, 2009 and Ali et al., 2013).
* Better soil fertility may result from phytoremediation due to the addition of organic materials (Mench et al., 2009).

Limitations of phytoremediation are:

* Long time required for remediation.
* Sluggish plant growth rates (Kuiper et al., 2004; Vangronsveld et al., 2009 and Ali et al., 2013).
* In some instances, collecting plants for biomass management after remediation may result in extra cost (Wang et al., 2012a, b).
* It is also possible that accumulated hazardous pollutants will go up the food chain because plants lack the catabolic enzymes which are necessary to completely mineralize organic contaminants to carbon dioxide and water (Lee, 2013).

There are several types of phytoremediation mechanisms as enlisted below:

1. **Rhizosphere biodegradation.** In this process, the plant releases organic compounds through its roots, feeding soil microbes with nutrition. These organisms in turn accelerate the biological decay.
2. **Phyto-stabilization:**Instead of degrading impurities throughout in this process, the plant's chemical byproducts immobilise the toxic compounds.
3. **Phyto-accumulation** (phytoextraction): During this process, the contaminants are absorbed by plant roots, along with other nutrients and water. This technique is mostly applied to metal-containing trash.
4. **Hydroponic Systems for Treating Water Streams (Rhizofiltration)**:In contrast to phytoaccumulation, rhizofiltration uses plants that are grown in greenhouses with their roots submerged in water. Ex-situ groundwater remediation can be done using this growth technique. Groundwater is pumped to the surface for irrigating these plants. An artificial soil medium (like sand blended with perlite or vermiculite) is typically used in hydroponic systems. The roots are removed and discarded as soon as they are completely saturated with pollutants.
5. **Phyto-volatilization:**In this procedure, plants absorb contaminated water that contains organic substances and then expel those substances into the atmosphere through their leaves.
6. **Phyto-degradation:**By this process, plants metabolize and destroy toxins within its tissues.
7. **Hydraulic Control:** By restricting the circulation of groundwater during this process, trees indirectly remediate. When a tree's roots descend to the water table and form a massive root mass that absorbs a lot of water, they operate as natural pumps. Example a cottonwood tree may absorb up to 350 gallons of water per day, whereas a poplar tree can extract 30 gallons of water from the ground each day.

**3.2.2 Permeable reactive barrier (PRB)**

Permeable reactive barrier (PRB) is an in-situ method for cleaning up underground water that has been contaminated with a variety of contaminants (such as heavy metals and chlorinated substances,natural pyrite (FeS2), zero-valent iron (ZVI) powder, sodium citrate, etc). In this method, the path of contaminated groundwater is covered with a permanent or semi-permanent reactive barrier (medium), which is primarily composed of zero-valent iron (Garcia et al., 2014 and Zhou et al., 2014). Contaminants become trapped and undergo a series of reactions. It passes through the barrier under its natural gradient and produces clean water (Thiruvenkatachari et al., 2008 and Obiri-Nyarko). These barriers trap pollutants and it is permeable to allow the flow of water but not pollutants as well as passive with little energy input. It is also inexpensive, readily available and accessible (De Pourcq et al., 2015). The effectiveness of this technology is largely dependent on the choice of medium, which is influenced by the type of pollutant, biogeochemical and hydrogeological conditions, environmental and health consequences, mechanical stability, and cost. (Obiri-Nyarko et al., 2014 and Liu et al., 2015). Scientistshave recently concentrated on combining PRB with other techniques, such electrokinetics, for treating various classes of pollutants (Garcia et al., 2014; Mena et al., 2015; Ramirez et al., 2015). Although sustaining barrier reactivity is important for PRB technique performance, retaining barrier permeability is equally important for PRB success and may be accomplished by preserving the proper particle size distribution (Mumford et al. 2014). The issues with the PRB approach are the decline in long-term performance brought on by the barrier's decreased reactivity, zero-valent iron (ZVI), loss of porosity, and inability to apply the technique to sites contaminated with chlorinated hydrocarbons and recalcitrant compounds.

**3.2.3 Microorganisms used in bioremediation**

In nutritional chains, which are a crucial component of the biological balance in life, microorganisms play a significant role. With the aid of bacteria, fungi, algae, and yeast, polluted materials are removed during bioremediation. Under the presence of hazardous substances, microbes are capable of growing in wide range of temperatures (below zero as well as in severe heat). The biological system and adaptability of microorganisms make them suited for the cleanup process. The fundamental ingredient needed for microbial action is carbon. Microbial consortiums worked in many situations to do bioremediation. *Achromobacter*, *Arthrobacter*, *Alcaligenes*, *Bacillus*, *Corynebacterium*, *Pseudomonas*, *Flavobacterium*, *Mycobacterium*, *Nitrosomonas*, *Xanthobacter*, etc. are some of these microbes.The following microorganisms are employed in bioremediation: *Acinetobacter*, *Sphingomonas*, *Nocardia*, *Flavobacterium*, *Rhodococcus*, and *Mycobacterium*. According to reports, these bacteria can break down polyaromatic chemicals, hydrocarbons, alkanes, and pesticides. These bacteria utilise the pollutants as a source of carbon and energy. Aerobic bacteria are utilised in bioremediation to break down and transform contaminants into less harmful forms.

**3.2.3.1 Factors affecting microbial bioremediation**

The bioremediation technique involves using bacteria, fungi, algae, and plants to break down, remove, change, immobilise, or detoxify various chemicals as well as the physical pollutants from the environment. Microorganism’s enzymatic metabolic pathways speed up the biochemical processes that helps in contaminant breakdown. Only when microorganisms come in touch with substances that aid in their ability to produce energy and nutrients for cell division do they begin to react to contaminants. The chemical composition and quantity of contaminants, the physicochemical properties of the environment, and their accessibility to already-existing microorganisms are only a few of the variables that affect how effective bioremediation is. The key contributing components include the microbial population's capacity to degrade pollutants, the ease with which contaminants may be accessed by the microbial population, and environmental conditions such soil type, pH, temperature, and oxygen level and nutrients.

**3.2.3.1.1 Biotic or biological factors**

**Biotic factors are beneficial for the breakdown of organic compounds. Limited carbon sources show antagonistic interactions between microorganisms, the protozoa and bacteriophages. The concentration of the contaminant and the amount of catalyst present in the biochemical process are typically correlated with the rate of contaminant breakdown. Enzymatic activity, interactions (competition, succession, and predation), mutation, horizontal gene transfer, its growth for biomass production, population size, and composition are among the main biological factors ( Madhavi et al., 2012 and Boopathy et al., 2000).**

**3.2.3.1.2 Abiotic or environmental factors**

Metabolic activity and physicochemical characteristics of the microorganisms targeted during the process in relation to environmental pollutants. The environmental conditions affect how successfully bacteria and pollutants interact. Microbial activity and their growth are influenced by a number of variables, including temperature, pH, moisture, soil structure, water solubility, nutrients, site conditions, oxygen content and redox potential, resource depletion and physico-chemical bioavailability of pollutants, concentration, chemical structure, type, solubility, and toxicity. These variables regulate the kinetics of deterioration.

In most aquatic and terrestrial environments, the pH range of (6.5-8.5) is generally ideal for contaminant biodegradation. Moisture level influences metabolism of contaminantsas it is influenced by the kind and availability of soluble components, pH, osmotic pressure of terrestrial and aquatic systems (Cases et al., 2005).

**4. Prospects of bioremediation**

There are several different bioremediation strategies, and many have proven successful in repairing polluted environments. The variety, profusion, and community structure of microorganisms in polluted settings provide insight into the likelihood that any bioremediation strategy will provide other environmental elements that can restrict microbial activity. Microorganisms has a vital role in bioremediation. Advanced molecular approaches, such as "Omics," which comprises genomics, proteomics, metabolomics and transcriptomics have aided in the identification of microorganisms as well as their functions and metabolic and catabolic processes. The availability of nutrients, lack of population of microorganisms with the ability to degrade materials and the bioavailability of the pollutants can cause a delay in the completion of bioremediation.

Biostimulation and bioaugmentation techniques speed up microbial activities in contaminated sites since bioremediation rely on microbial processes. Nutrients are added to a polluted sample to biostimulate it as well as to increase microbial activity. It is noteworthy that pollutant-degrading microbes are naturally found in polluted and contaminated sites; the type and concentration of pollutants may affect their growth and metabolic activities. Later, we can use agro-industrial wastes, which contain nitrogen, phosphorus, and potassium as a nutrient source for the majority of polluted sites.Contaminants are degraded more effectively by microbial consortiums than by isolated isolates (Silva-Castro et al., 2012). When these isolates are combined, this activity could result in the complete and quick degradation of pollutants due to the metabolic diversities of individual isolates(Bhattacharya et al.2015). Additionally, compared to a non-adjusted setup (control), both bioaugmentation and biostimulations were successful in eliminating pollutants such polyaromatic hydrocarbons (PAHs) from a substantially polluted sample (Sun et al. 2012). Bioaugmentation has been proven to be an efficient strategy, but it has also been demonstrated to speed up the breakdown of several chemicals. If the proper biodegrading microorganisms are absent in the soil or if microbial populations declined due to pollutant toxicity, specific microorganisms can be reintroduced as "introduced organ-isms" to improve the current populations.However, this method is very uncertain because there is a chance that the inoculated microorganisms may not survive in the new environment. The practise is referred to as bioaugmentation. Using the bioremediation process, sewage or contaminated water or soil is treated using naturally occurring or genetically modified microorganisms with specific metabolic profiles. Some of the issues with bioaugmentation can be resolved by using carrier materials including alginate, agar, agarose, gelatin, gellan gum, and polyurethane (Tyagi et al., 2011).

Chemically speaking, biosurfactants are comparable substances with green and biodegradable characteristics. However, using bio-surfactants to a polluted site would be economically unviable due to their high building costs and limited scalability. Combining agricultural and industrial wastes provides nutrients for the growth of biosurfactant producers throughout the fermentation process. Utilizing a variety of bioremediation strategies will help improve remediation effectiveness (Cassidy et al., 2015). It is a good idea to use genetically engineered microorganisms (GEM) strategically to improve bioremediation capacity. This is because it is possible to create a designer biocatalyst that can break down pollutants including resistant substances by merging new, effective metabolic pathways, expanding the range of substrates for existing pathways, and enhancing the stability of catabolic activity (Paul et al.2005). Parallel gene transfer and GEM expansion in an environmental application, however, are encouraging strategies. Systems for containing bacteria that allow any GEM to escape and recreate a dirty environment. Additionally, a biological strategy for genetically engineering microorganisms with a particular pollutant component could improve the effectiveness of bioremediation. Because of their increased surface area and lower activation energy, nanomaterials decrease the toxicity of pollutants to microorganisms, which speeds up and lowers the cost of bioremediation (Rizwan et al.2014).

**5. Bioremediation applications**

To be effective, bioremediation must be applied to all environmental states of matter, including solids (soils, silt, and sludge), liquids (groundwater, surface water, and industrial waste water), gases (industrial air emissions), and subsurface environments (saturated and vadose zones).

The three main methods of bioremediation are: intrinsic (natural) bioremediation, biosimulation (environmental changes made through fertiliser application and aeration), and bioaugmentation (addition of the microbes).

The natural soil microflora typically makes up the biological community used in bioremediation. However, it is also possible to manage higher plants to improve toxicant elimination (phytoremediation), particularly for cleaning up soils that have been poisoned with metal.

**6. Limitations of bioremediation**

The use of bioremediation is restricted to biodegradable substances. This process is prone to quick and total deterioration. Products of biodegradation may be more harmful or persistent in the environment than the parent chemical.

1. Specificity: The biological processes that take place are quite particular. The availability of metabolically competent microbial populations, optimal environmental growth conditions, and appropriate quantities of nutrients and pollutants are crucial site elements that are essential for success.

2. From batch and pilot scale investigations to large-scale field operations, the bioremediation technology is difficult to scale up.

3. Development of new engineering bioremediation solutions that are appropriate for sites with composite combinations of pollutants that are not evenly dispersed in the environment is still needed. It might exist in the form of solids, liquids, or gases.

4. The bioremediation procedure is time-consuming compared to alternative treatment options including excavation and soil removal from contaminated sites.

5. Lack of regulatory consensus: Since there is no universally agreed-upon definition of clean, we cannot state with certainty that remediation is 100% accomplished. There is no acceptable endpoint for bioremediation treatments as a result, making it impossible to evaluate their effectiveness.

**Conclusion**

When it comes to remediating, cleaning, maintaining, and recovering methods for resolving a polluted environment through microbial activity, biodegradation is a very profitable and alluring alternative. The rivalry between biological agents like fungi, bacteria, and algae as well as unfavourable external abiotic factors (aeration, moisture, pH, and temperature) and limited bioavailability dictate how quickly undesired waste materials degrade. The effectiveness of bioremediation depends on a number of variables, including but not limited to cost, site features, and the kind and quantity of pollutants.Site description is the main step for the successful bioremediation since it aids in the creation of the most effective and promising bioremediation technique (ex-situ or in-situ). Due to the digging and removal from the archaeological site, ex-situ bioremediation methods are typically more expensive. They can, however, be utilised to cure a variety of contaminants. Contrarily, in-situ techniques do not incur additional costs for excavation; yet, some inefficient in-situ bioremediation approaches can be reduced by the on-site installation cost of equipment, attached with successfully, and controlling the subsurface of a polluted site. Geological characteristics of the polluted site, such as soil, pollutant sort and depth, human habitation site, and performance of each bioremediation strategy, should be taken into consideration while selecting the most effective bioremediation method to successfully treat the polluted areas.

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