



## MICROBIAL BIOREMEDIATION TECHNIQUES OF SOILS CONTAMINATED WITH HEAVY METALS

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### ABSTRACT:

**Background:** Heavy metal (PM) contamination of soil is a global issue resulting from increased production activities. Unlike organic pollutants, PM cannot be chemically, physically, or biologically broken down, making remediation challenging. The objective of this study is to outline the applicability, benefits, and drawbacks of microbial bioremediation to highlight elements that may aid in its potential selection for various PM-contaminated soil remediation scenarios.

**Methods:** A descriptive and qualitative methodology built on secondary data was used to conduct this study. The analysis focused on the effects of microbial bioremediation, which involves valence transformation, biosorption, extracellular chemical precipitation, and volatilization through the actions of bacteria, actinomycetes, fungi, and algae.

**Results:** The study found that the primary determinants of successful bioremediation are:

- Climate (temperature and precipitation), Soil characteristics (pH and texture), Use of microorganisms resistant to or tolerant of contaminants, Injection of one or more species with proven remedial capability. In situ, bioremediation techniques such as bioaugmentation, biostimulation, and venting are preferred over ex-situ methods like land farming and composting due to their lower cost and reduced environmental, economic, and societal impacts.

**Conclusions:** In situ bioremediation techniques are frequently used because they are less costly and have a smaller negative impact on the environment compared to ex-situ methods. However, due to

the complex nature of bioremediation, projects and studies must adopt a multidisciplinary approach to enhance understanding of microbial ecology, physiology, evolution, biochemistry, and genetics. The diversity of microorganisms and bioremediation techniques presents opportunities to expand their application.

**KEYWORDS:** Soil Remediation, Heavy Metal Contamination, Soil Remediation, Microbial Bioremediation, Environmental Pollution, Bioaugmentation, Biostimulation, In situ Bioremediation, Ex situ Bioremediation, Biosorption.

### **INTRODUCTION:**

According to IBGE and SA (2020), soil is the earth's mineral and organic surface material that has been worn down to some extent by physical, biological, and chemical processes. It acts as a natural substrate for the growth and development of different organisms. The soil's physical, chemical, and biological properties can be impacted by soil deterioration or contamination, which can reduce the land's potential for production. Heavy metals (PM) are a global concern as one of the soil contaminants, accounting for almost 50% of the 10 million polluted sites, meaning that their concentrations exceed regulatory or regional reference values. This is because they are not biodegradable, which means that they accumulate in the food chain and are very persistent in the environment (Okpara-Elom, Onochie, Elom, Ezaka, & Elom, 2024; Saad et al., 2024; Singh & Kostova, 2024).

The production of safe food and human health is in danger because heavy metals have toxicological properties that surpass set limits. These pollutants are primarily the result of human activities such as mining (tailings ore piles and tailings dams), foundries, electronic industries, textile and petrochemical production, consumption of fossil fuels, and agriculture (use of fertilizers, pesticides, manure, fertilizers, compost, biosolids, or wastewater with heavy metals in its composition). These pollutants are introduced into the soil through steam, flue gas particles, RISES (ash and others), and RILES (Gupta, Khan, Alqahtani, Hashem, & Ahmad, 2024; Roy et al., 2024).

The study by Zhang et al., which discovered a substantial correlation between the total concentrations of Zn, Cu, Cd, As, and Ni in agricultural lands and 58 abandoned iron, copper, and lead-zinc mines in China, illustrates those above. These metals can travel through the soil through wind, sediment entrainment, leaching into surface waters, infiltration into aquifers, and accumulating in the food chain. Therefore, there are several heavy metal remediation techniques for soil, including surface coating, encapsulation, casting, soil washing, electrokinetic extraction, stabilization, solidification, vitrification, phytoremediation, and bioremediation, to reduce risks to human and environmental health (Fig 1) (Madhav et al., 2024; Reddy, Ranjit, Priyanka, Maddela, & Prasad, 2024).

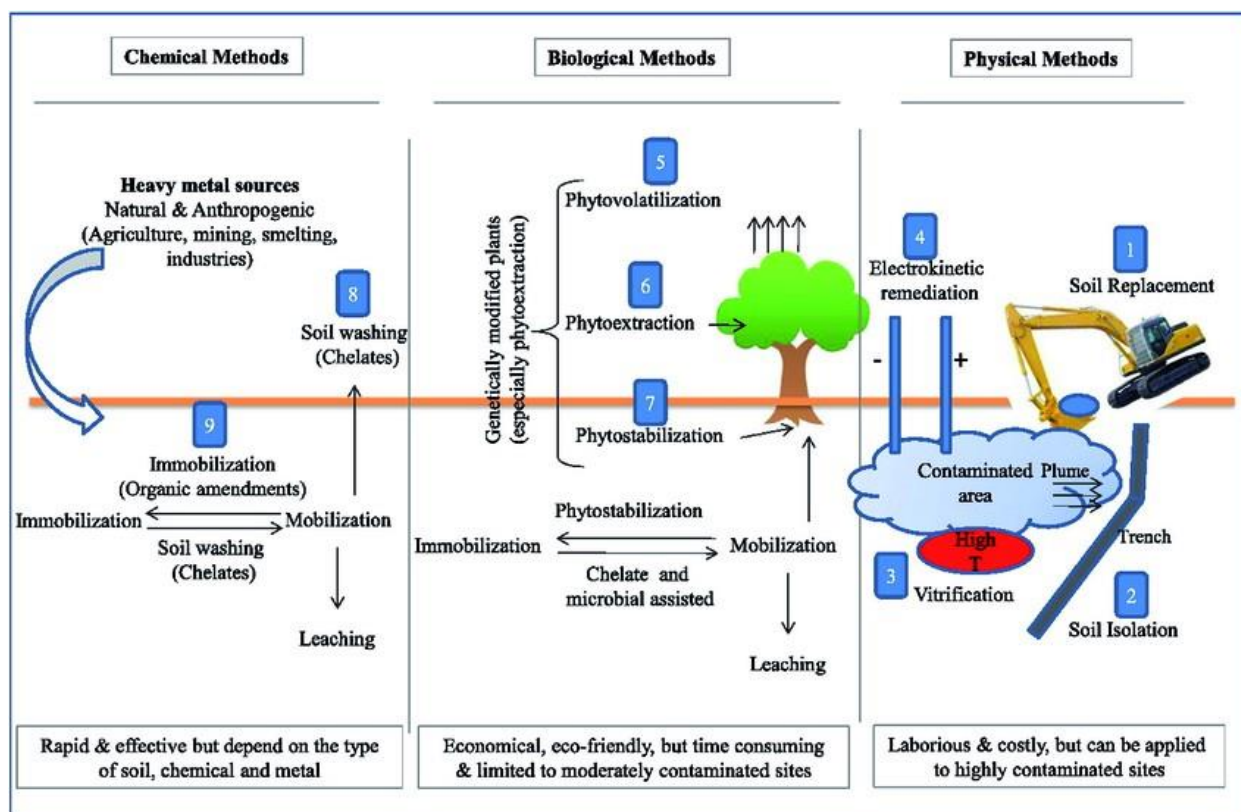


Fig. 1. Techniques for remediation of soil contaminated by heavy metals.

The application location (in situ or ex-situ), process type (physical, chemical, and biological), degree of development (traditional or innovative), and goal (containment, which lowers exposure, isolation from the environment without having to act on it, extraction/removal, which lowers or eliminates its concentration, and immobilization, which prevents its dispersion/mobility) are all factors that can be used to characterize these techniques. However, contaminant removal/removal strategies are preferable over contamination immobilization and containment techniques due to the long-term efficacy of treatment (Khatun, Kobir, Miah, Sarkar, & Alam, 2024; Yaashikaa, Palanivelu, & Hemavathy, 2024).

Table 1: Definitions and Impacts of Soil and Heavy Metal Contamination

Source	Definition and Impacts
IBGE and SA (2020)	Soil is the earth's mineral and organic surface material worn down by physical, biological, and chemical processes. It acts as a natural substrate for the growth and development of organisms. Soil deterioration or contamination impacts its physical, chemical, and biological properties, reducing land's production potential.
Okpara-Elom, Onochie, Elom, Ezaka, & Elom (2024); Saad et al. (2024); Singh & Kostova (2024)	Heavy metals are a global concern, accounting for almost 50% of the 10 million polluted sites. They are non-biodegradable, accumulate in the food chain, and are persistent in the environment.
Gupta, Khan, Alqahtani, Hashem, & Ahmad (2024); Roy et al. (2024)	Heavy metals from human activities such as mining, foundries, electronic industries, and agriculture pose a risk to food production and human health due to their toxicological properties. These pollutants are introduced into the soil through steam, flue gas particles, ash, and other residues.
Zhang et al.	There is a substantial correlation between the total concentrations of Zn, Cu, Cd, As, and Ni in agricultural lands and abandoned mines. These metals travel through the soil via wind, sediment entrainment, leaching, infiltration into aquifers, and accumulation in the food chain.

**Table 2: Heavy Metal Remediation Techniques**

Source	Techniques
Madhav et al. (2024); Reddy, Ranjit, Priyanka, Maddela, & Prasad (2024)	Remediation techniques include surface coating, encapsulation, casting, soil washing, electrokinetic extraction, stabilization, solidification, vitrification, phytoremediation, and bioremediation.
Khatun, Kobir, Miah, Sarkar, & Alam (2024); Yaashikaa, Palanivelu, & Hemavathy (2024)	Techniques can be characterized by application location (in situ or ex-situ), process type (physical, chemical, and biological), degree of development (traditional or innovative), and goal (containment, isolation, extraction/removal, and immobilization). Contaminant removal strategies are preferable due to long-term efficacy.
Agrawal, Ruhil, Gupta, & Verma (2024)	Microbial bioremediation is practical and environmentally sound for treating heavy metal-contaminated soils compared to costly and harmful physicochemical procedures.

**Table 3: Sources and Pathways of Heavy Metal Pollution**

Source	Description
Gupta, Khan, Alqahtani, Hashem, & Ahmad (2024); Roy et al. (2024)	Heavy metals enter the soil primarily through human activities such as mining (tailings ore piles and dams), foundries, electronic industries, textile and petrochemical production, consumption of fossil fuels, and agriculture (use of fertilizers, pesticides, manure, compost, biosolids, or wastewater containing heavy metals).
Zhang et al.	Heavy metals travel through the soil via wind, sediment entrainment, leaching into surface waters, infiltration into aquifers, and accumulation in the food chain.

**Table 4: Factors Influencing the Choice of Remediation Techniques**

Source	Factors
Khatun, Kobir, Miah, Sarkar, & Alam (2024); Yaashikaa, Palanivelu, & Hemavathy (2024)	Factors include soil and pollutant characteristics, anticipated efficacy, technological and financial viability, and implementation time.
Agrawal, Ruhil, Gupta, & Verma (2024)	Microbial bioremediation is favoured due to its practicality and environmental benefits compared to costly and environmentally harmful physicochemical methods.

**Table 5: Toxicological Properties and Risks of Heavy Metals**

Source	Risks and Properties
Okpara-Elom, Onochie, Elom, Ezaka, & Elom (2024); Saad et al. (2024); Singh & Kostova (2024)	Heavy metals are non-biodegradable, accumulate in the food chain, and persist in the environment. Their toxicological properties exceed set limits, posing risks to food production and human health.
Gupta, Khan, Alqahtani, Hashem, & Ahmad (2024); Roy et al. (2024)	Heavy metals from human activities lead to environmental contamination, impacting food safety and human health due to their toxicological properties.

**Table 6: Remediation Techniques by Characteristics**

Source	Technique Characteristics
Madhav et al. (2024); Reddy, Ranjit, Priyanka, Maddela, & Prasad (2024)	Techniques include surface coating, encapsulation, casting, soil washing, electrokinetic extraction, stabilization, solidification, vitrification, phytoremediation, and bioremediation.
Khatun, Kobir, Miah, Sarkar, & Alam (2024); Yaashikaa, Palanivelu, & Hemavathy (2024)	Techniques can be categorized by application location (in situ or ex-situ), process type (physical, chemical, and biological), degree of development (traditional or innovative), and goal (containment, isolation, extraction/removal, and immobilization).

The features of the soil and pollutants, the anticipated efficacy, the technological and financial viability, and the implementation time all play a role in selecting the technique. However, microbial bioremediation is a practical and environmentally good method for treating heavy metal-contaminated soils because physicochemical procedures are costly and harmful to the environment (Agrawal, Ruhil, Gupta, & Verma, 2024).

### OBJECTIVE:

This study intends to emphasize elements that may aid in the potential selection of soil remediation in various circumstances by describing microbial bioremediation's uses, benefits, and drawbacks (Alabssawy & Hashem, 2024).

### METHODOLOGY:

The current work aims to provide descriptive information by characterizing microbial bioremediation methods for heavy metal-contaminated soils. The research base is bibliographic since secondary data from the natural and environmental sciences are mostly gathered from databases like Scielo, Scopus, ScienceDirect, and Web of Science. Works released since 2015 were given precedence in the search, which was conducted using the terms "soil remediation," "heavy metals in soil," and "soil decontamination." According to Lakatos and Marconi's definition, the information is processed using a qualitative approach that creates a comparative overview of the key features of bioremediation in various contexts by combining data from theoretical and empirical bibliographies, as well as by incorporating definitions of concepts, theorist examinations, and methodological problem analysis (Ashkanani et al., 2024; X. Li et al., 2024; Tennakoon, Wasana, Bellanthudawa, Sandamal, & Ratnasekera, 2024).

### RESULTS:

#### MAJOR FACTORS AFFECTING BIOREMEDIATION:

The availability of heavy metals is significantly influenced by soil and climate variables (Table 1), particularly pH, structure, temperature, and precipitation. These authors clarify that the availability of MPs in the soil increases at low pH, except for the metalloid As. This condition reduces the adsorption of metal ions on soil particles, increases competition between free metal ions and other cations in the soil solution, and disrupts microorganism metabolism in the case of as it is different since it normally resides as an anion, arsenate or arsenite, rather than as cations in the soil solution (Y.-Q. Liu et al., 2024; Maqbool et al., 2024).

Another important component is the structure of the soil particles; loamy and fine-textured clay soils have the maximum PM availability, while sandy soils have the lowest availability. Conversely, in areas with high average temperatures and precipitation, metal solubility and bioavailability will likewise be comparatively higher than in other areas. To maintain low PM availability in the soil, future remediation techniques should concentrate on controlling pH (slightly over 7.0), particularly in locations with high temperatures and precipitation (F. Liu et al., 2024; Xiang et al., 2024).

FACTORS	MICROBIOLOGICAL REMEDIATION
<b>Temperature</b>	The bioavailability of pollutants, their volatilization, the effectiveness of heavy metal cleanup, and biological physiology and enzymatic activity
<b>Type of Terrain</b>	Nutrient and oxygen transport and distribution, water and pollutant dispersion, organic content, rate of biodegradation, and interactions between coexisting contaminants in soils.
<b>Soil Fertility</b>	Metal bioavailability, microbial activity, and biodegradation rate.
<b>Metal Concentration</b>	Three strategies to modify biological activity: The synthesis of cellular polymers, respiration, and cell division of microorganisms are all impacted by MP ions, which can reduce bioremediation. 2) Low

	concentrations of MP can stimulate bioremediation, while high concentrations inhibit it. 3) Low concentrations of MP inhibit bioremediation, while high concentrations inhibit less.
<b>Species</b>	The capacity of living things to take in, move, gather, and process contaminants.
<b>pH</b>	pollutants' bioavailability, nutrients' accessibility, enzymatic activity and biological physiology, and interactions between contaminants.
<b>Humidity</b>	Bioavailability of pollutants and nutrients, oxygen content, microbial cell development and function, and biomass of plants.
<b>Organic Content</b>	Pollutant bioavailability, nutritional availability, and interpollutant interactions.
<b>Nature of the Contaminants</b>	The harmful impact of various metal valences and forms on organisms varies, influencing how effective metal remediation is.
<b>Interaction Between Contaminants</b>	Bioremediation processes are influenced by enzymatic activity, biological physiology, and synergistic or antagonistic effects on the bioavailability of contaminants.

*Table 1. Main factors that influence bioremediation in contaminated soil.*

The microorganisms are still alive in the contaminated site, so the components form the cornerstone of bioremediation's effectiveness. The body of research indicates that following heavy metal contamination, soil microbe diversity and structure underwent substantial changes, with the degree of exposure and the microorganisms' ability to adapt to the toxins determining these changes. For this reason, a soil remediation technique involves the employment of indigenous microorganisms resistant/tolerant to contaminants or the inoculation of one or more species with recognized bioremediation capability, which are noted below (Dhanapal et al., 2024; Wang et al., 2025).

#### **FUNGUS:**

Fungi A variety of mushrooms, including *Pleurotus eryngii*, *Coprinus comatus*, *Aspergillus sydowii*, *Penicillium griseofulvum*, *Trichoderma koningii*, *Paecilomyces marquandii*, and *Clitocybe maxima*, have the potential to be used in the bioaccumulation and biosorption of vanadium (V), lead (Pb), Cu, Cd, Cd (II), and Zn in areas where herbicides and pesticides are co-contaminated. These authors clarify that fungi can enhance and accelerate the biodegradation of agrochemicals in addition to PM remediation and that co-remediation the use of two or more fungi generally results in higher treatment efficacy (Y. Li et al., 2024; Wen et al., 2024).

#### **BACTERIA:**

According to Diels et al. and Vaxevanidou et al., MPs like As, closely linked to iron oxide, can be released more readily when iron-reducing bacteria are present. This is a less harsh pH method than utilizing acids in chemical reactions. To dissolve metals in soil, other bacteria (such as *Bacillus subtilis* and *Torulopsis bombicola*) can also create biosurfactants such as saponin, rhamnolipids, sophorolipids, escin, and surfactin. Additionally, they can enhance plant development in polluted soils and encourage plant tolerance to heavy metals. Additionally, there is a history of studies on the application of marine and transgenic bacteria for the remediation of mercury (Hg)-contaminated waste. In these cases, the bacteria take up or convert methylmercury into Hg(II), which is then reduced to Hg(0), a less volatile and dangerous form of mercury (Dou et al., 2024; Gani, Hussain, Pathak, & Banerjee, 2024; He et al., 2024).

#### **MICROALGAE:**

Through the two processes of bioassimilation and biosorption, microalgae can effectively carry out bioremediation, according to Sreekumar et al. They can grow as "algal blooms" in contaminated water and absorb many pollutants. After harvesting and lipid/protein extraction, algal biomass can

be effectively used as a biosorbent. A low-cost microalgae-based biomass with a strong bioremediation capacity to neutralize and adsorb heavy metal ions can be developed by carefully evaluating factors including growth conditions, cell shape, biomass preparation, etc. (Nag et al., 2024; Song et al., 2024).

### BIOREMEDIATION TECHNIQUES:

Soil washing and phytoextraction are employed in conjunction with bioremediation of PM-contaminated soils to encourage the solubilization of heavy metals before extraction. Each working mechanism has unique benefits, drawbacks, and applications (Table 2) (Ningombam, Mana, Apum, Ningthoujam, & Singh, 2024).

<b>THE PROCESS OF AERATION</b>	
<b>Application</b>	Passively initiates the biodegradation of contaminants. Organic soils that are non-volatile or semi-volatile are the principal uses.
<b>Advantages</b>	Since their degradation rate is higher than that of volatilization, they are beneficial in addressing contamination with low vapour pressure chemicals (less than one mmHg). Bioventilation can be an economically viable substitute for excavation when costs are high. It doesn't call for more space or the usage of large machines.
<b>Disadvantages</b>	Low levels of soil moisture and airflow problems in the affected area. Specific soil properties, such as porosity, hydraulic conductivity, and humidity, are needed—periods of cleaning range from months to years.
<b>FARMING ON LAND:</b>	
<b>Application</b>	Less successful when treating heavy oil (big tanks), this method treats oily wastes and sludge from refineries. 2,500 ppm of heavy metals and 50,000 ppm of hydrocarbons
<b>Advantages</b>	In comparison to other bioremediation methods, it is less expensive. This method is regarded as low-tech as it doesn't necessitate complex engineering considerations and makes it simple to manipulate and regulate design and operating factors.
<b>Disadvantages</b>	If there isn't enough land, it won't work and needs big tracts for land disposal. Deep pollution might result in expensive excavation and earthmoving expenses.
<b>BIOSTIMULATION:</b>	
<b>Application</b>	Suitable for soils with sufficient porosity and air permeability values (> 10-10 cm <sup>2</sup> ) and as homogenous as feasible. The ideal pH (6 and 8), humidity (12–30% by weight), temperature (between 0 and 40 °C), and nutrition ratio (between 10:1) must all be met.
<b>Advantages</b>	It is extremely helpful for treating large, contaminated regions of industrial centres where it is not practical to pause operations to complete the treatment.
<b>Disadvantages</b>	It is not advised for very heterogeneous, highly stratified, or clayey soils as these may hinder O <sub>2</sub> transport.
<b>COMPOSTING</b>	
<b>Application</b>	It works on most organic molecules, albeit better on rapidly decomposing ones.
<b>Advantages</b>	It is effective for garbage with little hydrocarbon content. Because it

	is a closed system, process variables can be more tightly controlled.
<b>Disadvantages</b>	The soil pile must be enclosed with membranes or, akin to greenhouses, if volatile hydrocarbon gases or vapours are produced or if unfavourable weather conditions impact the process. Before being discharged into the atmosphere, the process-generated vapours must be collected and cleaned.
<b>BIOAUGMENTATION</b>	
<b>Application</b>	It is used when the amount of microflora bacteria required to break down pollutants is insufficient and when the contaminated location must be treated immediately. Used to remove residues, including high quantities of metals and pollution from herbicides and insecticides
<b>Advantages</b>	doesn't require the usage of large apparatus or extra space to perform the therapy

**Table 2. Applicability, advantages and disadvantages of bioremediation techniques.**

Treatability typically starts with methods that encourage the growth of microorganisms that may be used to bioremediate PM through infiltration tunnels or spray irrigation selectively. If toxins are deep inside the earth, injection wells could be a viable solution. To increase microbial activity and enhance bioremediation, nutrients, oxygen, and other additives are frequently combined (Janaki et al., 2024; Kou et al., 2024).

### CONCLUSION:

The variety of microorganisms and methods available for bioremediation of heavy metal-contaminated soils can pave the way for their increased use, particularly for in situ applications where minimal disturbance to the soil and cost savings are desired. To increase our knowledge of the ecology, physiology, evolution, biochemistry, and genetics of microorganisms, projects and studies of this kind must have a multidisciplinary planning process, given the variety of elements that impede bioremediation. Before bioremediation, alternative treatment methods may be used on soils where the environment is not conducive to microbial growth. As a result, it's critical to assess cooperative remediation strategies, particularly for heavily contaminated soils that need more time to heal.

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