Recent advances in the bioremediation of soils contaminated with heavy metals

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Abstract

Due to the increase in the human population, environmental pollution has increased drastically. Heavy metal pollution of the soil is one of these factors. Because of their industrial importance and advantages, heavy metals are often used in various chemical, paint, and battery industries. Bioremediation is a potential approach for addressing heavy metal soil contamination. This review covers methods for removing heavy metals from polluted soils, which include both traditional and cutting-edge techniques. Bioremediation techniques involving the use of microbes and plants and their mechanisms of action are also discussed. The bioremediation process is influenced by several factors, including energy sources, microbial and environmental factors, bioavailability, and economic resources. Although novel and recent, bioremediation is closely related to microbial biotechnology. Biostimulation, bioaugmentation, bioaccumulation, biosorption, phytoremediation, and nanotechnology are just a few of the bioremediation mechanisms used by microbes and plants. The most recent advances in the field of bioremediation, including biomineralization, phytostabilization, hyperaccumulation, dendroremediation, rhizoremediation, mycoremediation, cyanoremediation, genoremediation, and bioinformatics tools, are also discussed here. The benefits and drawbacks of bioremediation of heavy metal-contaminated soils are also emphasized.

Keywords: heavy metal, soil contamination, bioremediation, plant microbial interaction

1. INTRODUCTION

Anthropogenic activities, such as mining, the discharge of industrial waste, melting, and the creation of fossil fuels, have been the primary causes of increasing environmental contamination during the last several decades (Hou *et al.,*2023). As there is an increase in the number of industries that result in contaminated land, pesticides, heavy metals, pesticide runoff, and petroleum hydrocarbons are just a few of the contaminants that harm our water and land.(Bech, 2022; Saraswat *et al.,*2023) Contaminated lands pose potential threats to human health and animals, and therefore, it is important to make an effort to overcome this threat (Jannetto & Cowl, 2023). The primary drawbacks of conventional cleanup approaches are the high price and the significant dangers associated with the excavation, handling, and transportation of hazardous materials. (Sharma, *et al.,*2022). Hence, bioremediation is a new term for an environmentally friendly, cost-effective, and highly efficient method for cleaning polluted areas by reducing or eliminating pollutants in soil, wastewater, or industrial sludge through the use of living organisms (Sarker *et al.,* 2023).

Currently, bioremediation can be used as an emerging technology to remove or detoxify heavy metals. Bacteria, fungi, and yeast can be used to detoxify contaminants effectively (Hlihor *et al.,*2017).

Bacillus bacteria are one kind of bioremediation agent. *Bacillus subtilis*, *Bacillus cereus*, and *Bacillus thuringiensis* are among the most promising Bacillus species for bioremediation (Wrobel et al. 2023). Several methods, including biosorption, bioaccumulation, bioprecipitation, and extracellular polymeric substance-mediated biosorption, are available for this bacterial species. Additionally, certain Bacillus strains may promote plant growth, which helps with phytoremediation and the bioaccumulation of heavy metals in soil (Wrobel et al., 2023). In regard to cleaning contaminated areas, bioremediation is an option since it is both sustainable and environmentally beneficial. Microbes use contaminants, which are found at polluted sites, as their carbon source (Maqsood *et al.,*2023). Microbes have a variety of mechanisms, and contaminants are incorporated into cometabolism or mineralization pathways. Biotechnological advancements have made it feasible to alter the genetic material of microbes, which has improved the efficacy of bioremediation.(Maqsood *et al.,* 2023; Sarker *et al.,*2023)

1.1. ORIGIN OF HEAVY METALS IN THE SOIL

High-density metals and semimetals that are considered heavy may be harmful to human health (Gustin, et al.,2021). Elements with an atomic number greater than 20, metal characteristics, and an atomic density greater than 5 g cm^3 are considered heavy metals (HMs). Some examples of HMs include lead, arsenate, mercury, cadmium, zinc, aluminum, copper, iron, chromium, nickel, lead, and platinum (Briffa, *et al.*,2020; Gustin *et al.*,2021).

There are two kinds of sources through which heavy metals enter the environment (**Fig. 1**).

Natural Sources*:* The Earth's crust contains heavy metals according to design. Heavy metals may be found in rocks and other natural materials (Rajkumar, et al., 2023). Rocks may contain heavy metals due to mineralization, forest fires, volcanic eruptions, or processes that create soil (Tchounwou, *et al*.,2012). Examples: arsenic, copper, lead, etc.

Anthropogenic sources: Air pollution, river sediments, human activities, mining, smelting, atmospheric deposition, improper disposal of industrial solid and liquid waste, metal piping, traffic, combustion byproducts from coal-burning power stations, pesticide and fertilizer use, and petrochemical plants are the main causes of heavy metal contamination. Other sources include foundries, smelters, oil refineries, petrochemical plants, chemical industry, and pesticide production (Rajkumar *et al.*,2023).

Atomic absorption spectroscopy (AAS), inductively coupled plasma (ICP)-mass spectrometry, X-ray fluorescence, and other spectroscopic methods are available for use in heavy metal analysis (Jin et al.,2020). Due to its ability to provide exact quantitative determination, AAS stands far above the other methods described. Applied analytical chemistry (AAS) is a method for determining the concentration of trace elements in soil or other sample materials (Briffa *et al.,* 2020).

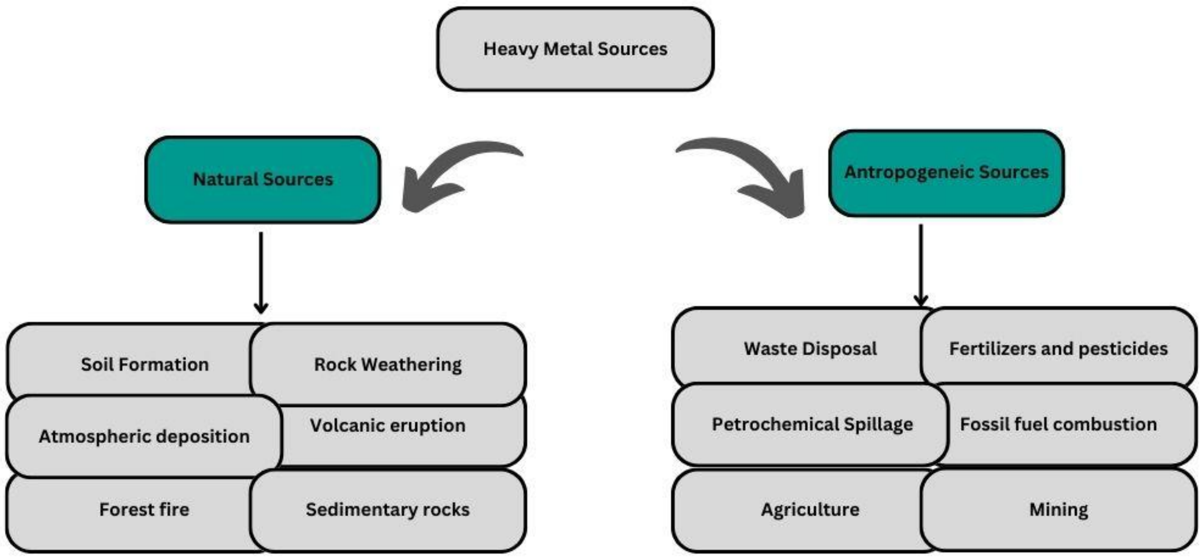

Figure 1: Various sources of heavy metals. Heavy metals may be found in natural or man-made environments; either way, they are hazardous to living things.

1.2. TOXICITY BY HEAVY METAL IN CONTAMINATED SOILS

Trace amounts of some heavy metals (e.g., cobalt, copper, iron, manganese, molybdenum, vanadium, silver, and zinc) are necessary for human and plant health and appropriate functions (Calabrese & Agathokleous, 2021). Nevertheless, even a slight increase in their threshold limit may lead to toxicity and noncommunicable illnesses in all living beings (Tchounwou et al.,2012). See **Table 1**. Some nonessential heavy metals (e.g., Cd, As, Pb, and Hg) are classified as strong carcinogens according to studies conducted by the International Agency for Research on Cancers (IARC) and the United States Environmental Protection Agency (USEPA) (Rajkumar et al. 2023).

Heavy metals have physicochemical characteristics comparable to those of physiologically active metals; they may wreak havoc in biological systems by interfering with enzyme activities, disrupting the function of subcellular structures, and triggering free radical processes (Ballatori, 2002). For example, metal ions can attach easily to functional groups such as -SH, -OH, and $-NH_2$ in the cytoplasm of cells. This leads to conformational changes in proteins, which in turn reduces their biological activity and ultimately causes cell death (Balali-Mood, *et al*., 2021; Ballatori, 2002).

Heavy metals have varying degrees of harmful effects on plants, depending on their concentration. When plants are exposed to high concentrations of heavy metals, their metabolism is disrupted, which may lead to physiological changes and even death (Moustakas, 2023). It has been noted that agricultural plants may experience chlorosis, low biomass accumulation, photosynthesis, growth inhibition, nutrient absorption, and ultimately plant mortality as a result of exposure to both necessary and nonessential metals.(Moustakas, 2023) Metal poisoning has a detrimental impact on plant development because it reduces water absorption and translocation, increases oxidative damage, and decreases nutrient uptake and translocation(Moustakas, 2023). Metallic elements such as lead and aluminum may hinder the formation of adenosine triphosphate (ATP), increase the generation of reactive oxygen species (ROS), and damage DNA (Tang, et al., 2023). Additionally, transcriptome sequencing revealed that these metals can considerably slow root elongation, plant development, germination, and chlorophyll synthesis (Moustakas, 2023). The World Health Organization reports that the maximum concentrations of cadmium, nickel, copper, zinc, lead, and chromium in clean soil are 0.8, 35, 36, 50, 85, and 100 mg/kg, respectively (Wieczorek, et al., 2023). Growing maize and soybeans on soils polluted with copper and lead resulted in fewer photosynthetic pigments, poorer biomass, and poor stomatal conductance (Ghuge, *et al*., 2023, Wang, *et al*., 2020; Wieczorek *et al*.,2023).

Table 1: Table indicating the effects of heavy metals on humans. Heavy metals, if consumed above the permissible limit, can cause severe damage to humans.

Heavy metals	**Possible disorders in human due to excessive heavy metals**	**Permissible Level (mg/L)**	**Reference**
Cadmium (Cd)	Cardiovascular, osteoporosis, bronchitis, Kidney damage, lung cancer, and destruction of testicular tissue.	0.06	Gunnar, et al , 2017
Chromium (Cr)	Genomic instability, Damage to the nervous system, carcinogenicity, respiration problems, rapid hair loss, skin irritation	0.05	Marina Tumolo, et al 2020
Copper (Cu)	Failure of the brain and kidney, intestinal and stomach irritation, anemia, liver toxicity	0.1	Alicia A,et al 2020
Lead (Pb)	Mental retardation in children, chronic damage to the nervous system, kidneys, and liver	0.1	Rajat et al,2022
Mercury (Hg)	Tremor, nephritic syndrome, hypersensitivity	0.01	Zhushan Fu et al 2020
Nickel (Ni)	Dermatitis, nasopharyngeal tumors, reduced sperm count, lung fibrosis, and lung and nasal cancer	0.2	Giuseppe Genchi et al 2020
Zinc (Zn)	Depression, lethargy, and damage to the nervous system	0.80	Ahmed et al 2021

1.3. REMEDIATION OF SOILS CONTAMINATED WITH HEAVY METALS

1.3.1. Conventional approaches for heavy metal removal

Heavy metals have been removed from polluted environments using a variety of clean-up procedures, including chemical, physical, and biological approaches (Arteaga, et al., 2022; Gu et al.,2022). Some examples of more conventional methods include chemical extraction, electrolysis, ion exchange, leaching, hydrolysis, polymer microencapsulation, and, of course, archaic methods of excavation and landfilling (Gu et al., 2022; Kumar et al., 2023). Because of their toxicity and mutagen properties, they appear to pose significant risks to human and environmental health (Kumar et al.,2023). Vapor extraction, stabilization, solidification, verification, and membrane technology are some of the ways in which heavy metal ions may be removed from contaminated regions (Kumar *et al.*,2023). However, there are drawbacks because most of these techniques are very expensive and require constant monitoring, which can be dangerous (Ma *et al.*,2023).

1.3.2. Bioremediation

The use of plants for bioremediation is known as phytoremediation (Gupta et al., 2013; Heredia *et al., 2022)* or microorganism-based remediation (Atuchin *et al., 2023;* Hlihor *et al., 2017)*.

The process of microorganism remediation involves cleaning polluted areas by harnessing the metabolic power of microbes (Atuchin *et al., 2023;* Y. Wang *et al.,* 2021). Even in the most hostile environments, microorganisms can thrive (**Fig. 2**). Their ability to transform virtually all forms of organic material makes them attractive organisms for bioremediation (Wang *et al.,* 2022). Microorganisms introduced to the soil must compete for resources with native species; they must also avoid natural hazards such as microbial toxins and predation (Pande *et al*., 2022).

There are two main approaches to microbial bioremediation: in situ and *ex situ* (Akhtar & Mannan, 2020; Atuchin et al., 2023). When soil is excavated and placed in a lined treatment area above ground, a biological process known as *ex situ* bioremediation (ESB) occurs. This approach involves aeration and processing to improve the ability of the native microbial population to degrade organic pollutants by increasing the breakdown of organic components after processing. One method of removing pollutants from underground water, often from groundwater, is known as in situ bioremediation (ISB). Environmental restoration of polluted soil and water habitats is best accomplished via in situ bioremediation techniques rather than *ex situ* techniques, the effectiveness of which is largely dependent on microbial metabolism.

Through a variety of physicochemical and biological processes, microbes impact the environmental destiny of hazardous metals by altering the transitions between soluble and insoluble phases (Atuchin et al., 2023; Pande et al., 2022; Wang et al., 2022). Due to their intricate structure, microbes can take up metal ions in a variety of ways (Atuchin et al., .2023). Depending on the metabolic activity of the cell, metal transport across the cell membrane causes intracellular buildup. The buildup of harmful metals is often associated with a sufficient defense system for microbes (Fig. 2). The absorption of metals occurs when the molecular structure of the metal binds to the surface of microbes in a process called biosorption, which is not reliant on metabolism. (Sarker *et al.,* 2023).

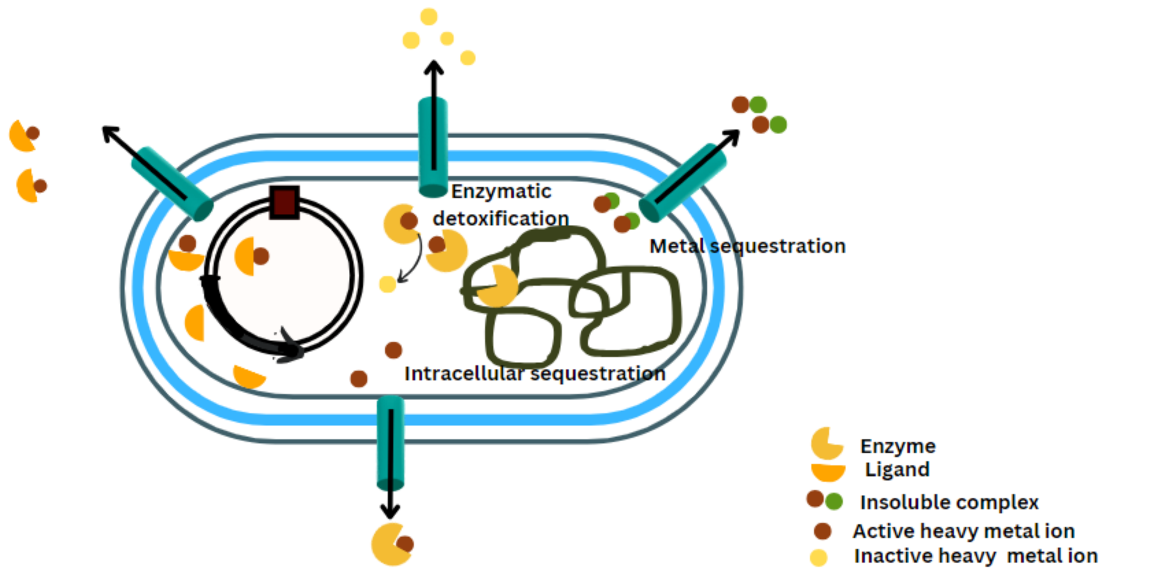

Figure 2: Microbial mechanisms to detoxify heavy metals. Enzyme detoxification, metal sequestration, and intracellular sequestration are three of the ways in which bacteria might lessen the harmful effects of heavy metals in soil.

1.4. CURRENT BIOREMEDIATION APPROACHES

Technological progress has also given people more freedom to destroy the environment and deplete natural resources (Ghuge et al., 2023). Bioremediation is the latest and most revolutionary method for environmental decontamination that utilizes biological systems; it is the best option for reducing pollution (Sarker *et al.,*2023). Even though this cutting-edge technology draws from a variety of fields, microbiology remains its backbone. Some examples of this technique include phytoremediation, bioaccumulation, biosorption, bioaugmentation, and biostimulation, all of which involve promoting the growth of native, viable microbial populations (plants) (Sarker *et al.,*2023; Wrobel *et al.,* 2023).

1.5. Biostimulation

Biostimulation is an environmental modification that promotes the growth of existing microorganisms with bioremediation capabilities (Nivetha *et al.,* 2023; Zhang *et al.,* 2022). Phosphorus, nitrogen, oxygen, and carbon are some of the electron acceptors and limiting nutrients that may help accomplish this. The main benefit of biostimulation is that it allows natural microorganisms to bioremediate. These microbes are already present in the subsurface and are adapted to the subterranean environment (Nivetha *et al., 2023)*. The geology of the subsurface determines the magnitude of the issue of efficiently delivering additives to subsurface microorganisms so that they may readily use them (Narayanan, *et al*., 2023).

1.6. Bioaugmentation

Bioaugmentation offers the possibility of inoculating inhabited areas with bacteria that possess the necessary enzymatic capabilities, thereby increasing their biodegradative capacities (Tyagi, et al., 2011). However, since considerable amounts of bacteria have been introduced into polluted locations, their influence on ecology has yet to be determined. The impact of germs on people and the environment must be defined (Azubuike, et al., 2016). Incorporating a preadapted pure bacterial strain or introducing genes relevant to biodegradation into native microorganisms via conjugation are the two main techniques for implementing bioaugmentation (Pande et al., 2022). A prior understanding of the microbial communities present at the target location is needed to determine the optimal approach for choosing competent microorganisms (Lin *et al*., 2022).

1.7. Bioaccumulation

Bioaccumulation is the gradual buildup of contaminants, such as heavy metals, in the biological tissues of aquatic organisms from their entry points into the food chain, which includes water, food, and suspended sediment particles (Nawab et al., 2015). Accumulation occurs in living organisms when metals are absorbed and retained at a rate greater than their metabolism or excretion. Protecting humans and other species against metal exposure may be greatly improved by gaining a better understanding of this process and its dynamics (Glavac *et al*., 2017).

1.8. Biosorption

For some types of biomass to passively deposit and bind contaminants onto their cellular structure, a natural physiochemical mechanism called biosorption is at work (Edulamud *et al*., 2023; Wang *et al., 2020)* (**Fig. 3**). Organic and inorganic substances, as well as soluble and insoluble substances, may be completely eliminated using this procedure (Dhanwal *et al., 2018;* Edulamudi *et al., 2023)*. The biosorption process exhibits two phases (Dhanwal *et al.,* 2018). Adsorption occurs in two distinct phases: solid and liquid (**Fig. 3**). The other contains the dissolved species that need to be adsorbed. The solid phase is generally biomass or biological material, and the liquid phase generally contains water as a solvent. The biosorption process is often quicker than the bioaccumulation process (Dhanwal *et al., 2018)*.

To recover cadmium, zinc, copper, and lead from water-based solutions, researchers have examined the adsorption ability of six distinct types of algae, including red, brown, and green varieties (Murphy, et al., 2008; Shahi et al., 2022). The brown algae had the lowest concentrations of metals in the fluid, whereas Fucus spiralis produced the best results*(Murphy et al., 2008)*.

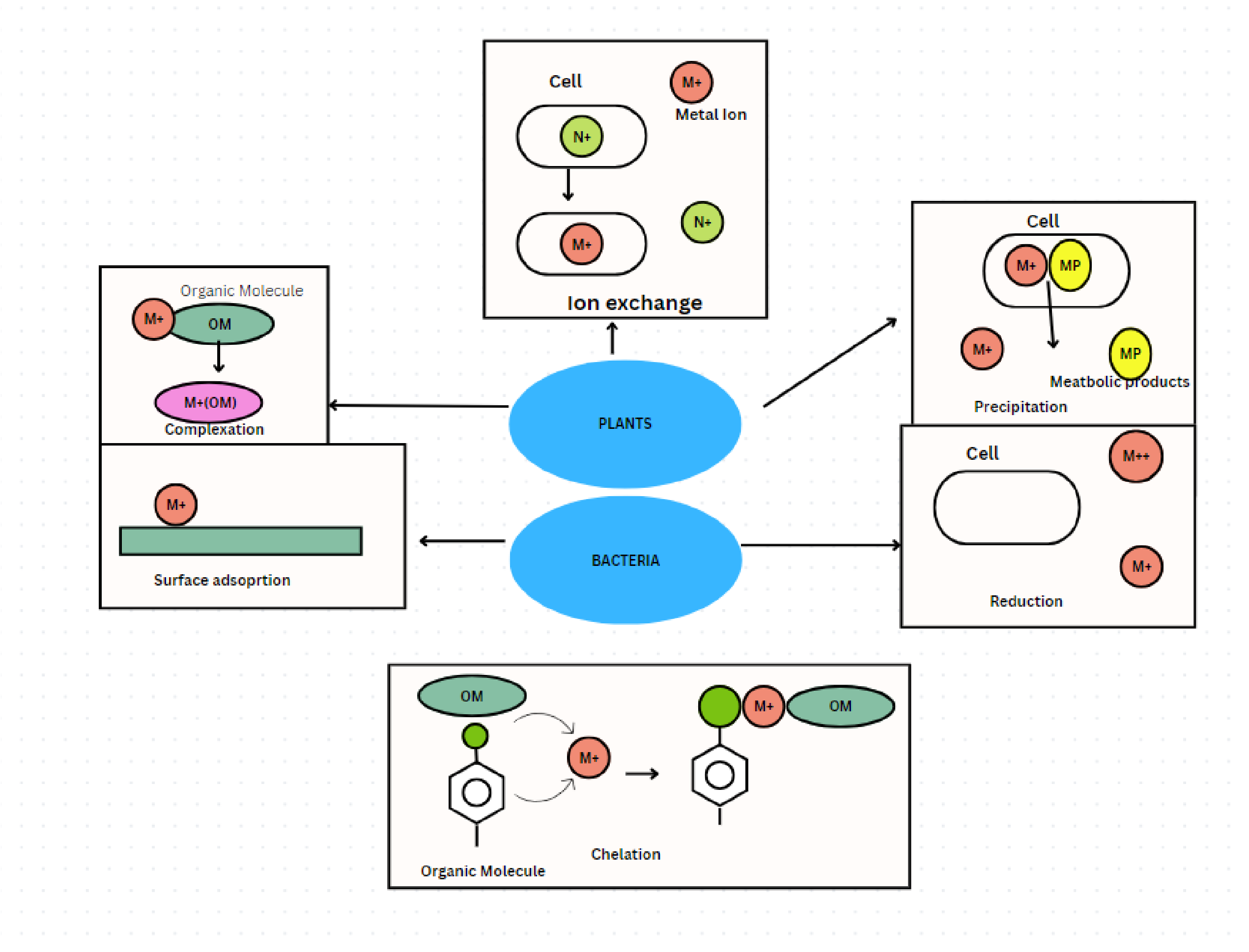

Figure 3: Mechanism for heavy metal biosorption. Biosorbent organisms (plants and bacteria) interact with heavy metals via different mechanisms, including complexation, ion exchange, surface absorption, chelation, reduction, and/or precipitation. OM, organic molecule, metal ion (M+), metabolic products (MP).

1.9. Bioprecipitation

The bioprecipitation process decreases the absorption and toxicity of metals by converting their concentrations into insoluble complexes (Azubuike et al., 2016). Contaminants such as lead, cadmium, chromium, and iron are precipitated by microorganisms via catalyzed oxidative and reductive reactions (Wang et al., 2021; Zeng et al., 2023). Some microbes have been shown to release phosphates and accelerate the precipitation of metal phosphates, whereas other bacteria may generate alkanes and precipitate hydroxides or carbonates (Zhang *et al*., 2021).

2. PHYTOREMEDIATION

When plants and bacteria are present in polluted environments, this process is called phytoremediation (Bhadrecha et al., 2023; Placido & Lee, 2022; Yan et al.,2020). This method takes advantage of the ways in which plants and microorganisms in their rhizosphere absorb and store contaminants, both organic and inorganic (Bhadrecha et al., 2023). See **Table 2**. One effective method for removing contaminants caused by both organic and inorganic substances is phytoremediation (Ghuge *et al.,* 2023; Pande *et al.,* 2022). To eliminate heavy metals, photorremedial procedures might involve a variety of approaches, each tailored to the specific nature of the pollutant: eradicating all traces of heavy metals, reducing their concentration, or using a hybrid model (Gupta *et al., 2013;* Tripathi *et al.,* 2022).

Aromatic plants such as vetiver, palmarosa, citronella, geranium mint, tulsi, and *Cymbopogon winterianus* are attractive options for phytoremediation since they are both environmentally friendly and efficient (Gupta *et al.,* 2013; Wei, et al., 2018). The high value and minimal resource requirements of certain stress-tolerant and perennial aromatic grasses make this approach a safe, economical and environmentally friendly method for soil bioremediation (Boros-Lajszner, *et al.,* 2021; Calabrese & Agathokleous, 2021). Plants rely on endophytic fungi for survival and adaptive processes. There are a variety of endophytic fungi that may enhance heavy metal stress tolerance, including dark septate endophytic fungi, arbuscular mycorrhizal fungi, and endophytic fungi that promote plant development (Ahammed et al., 2023). Given that beneficial microbial symbionts may confer tolerance to exogenous heavy metal stressors in plants, they may find use in phytoremediation approaches for soils polluted with these pollutants (Ma, et al., 2019).

The combination of endophytic fungi and phytoremediation might enhance plant adaptability to heavy metal stress and ecological restoration efficiency.

Plants have developed several internal and exterior regulatory systems in response to heavy metal stress; however, these processes differ between heavy metals and plant species (Yan et al., 2020). *Brassica juncea*, a hyperaccumulator that can take in a large amount of lead from its roots to its shoots, is a suitable candidate for use in internal regulatory systems (Gao et al., 2023; Q. Wang et al., 2019). Heavy metals are less hazardous to organs and have less transmembrane transport in a few plants because of a process that mixes them with organic acids, polysaccharides, and proteins (Wang *et al.,* 2019).

Table 2: Lists of plant species used for phytoremediation: A variety of plants may be used depending on a multitude of criteria, including the kind and concentration of the contaminant.

Heavy Metal	Plants species	Reference
Cd	*Ricinus communis*	Hanzhi et al., 2016
Cd, Pb,Zn	*Zea mays*	R.A. Wuana et al., 2010
Hg	*Populus deltoides*	Xuan Zhang et al., 2022
Se	*Brassica juncea, Astragalus bisulcatus*	Francesca Dalla et al., 2023
Zn	*Populus canescens*	Wen Guang et al., 2014
Cd, Cu, Ni, Pb	*Jatropha*	Olamilekan et al., 2019
Cd, Cu, Pb, Zn	*Salix species*	El-Sayed et al., 2019
As	*Pteris vittate*	Huili Yan et al., 2019
Ni	*Alyssum bertolonii*	Sharareh Dehghani et al ., 2021

Phytoremediation has several advantages, such as environmental friendliness, low maintenance, ease of maintenance, and cost effectiveness (Yan et al., 2020). However, there are also limitations to phytoremediation. The cleanup-mediating plants must be physically present at the site of the contamination and possess the ability to degrade it. Therefore, weather, soil characteristics, and toxin levels are the three main factors that determine growth (Boros-Lajszner *et al.,*2021). One of the other drawbacks concerns the length and depth of the roots because the plants must be able to reach contaminants (**Fig. 4**). Enhancing the efficiency of this process requires a deeper understanding of key factors, including the rhizosphere, chelation, volatilization, and pollutant availability (Bai, *et al*., 2023; Moustakas, 2023).

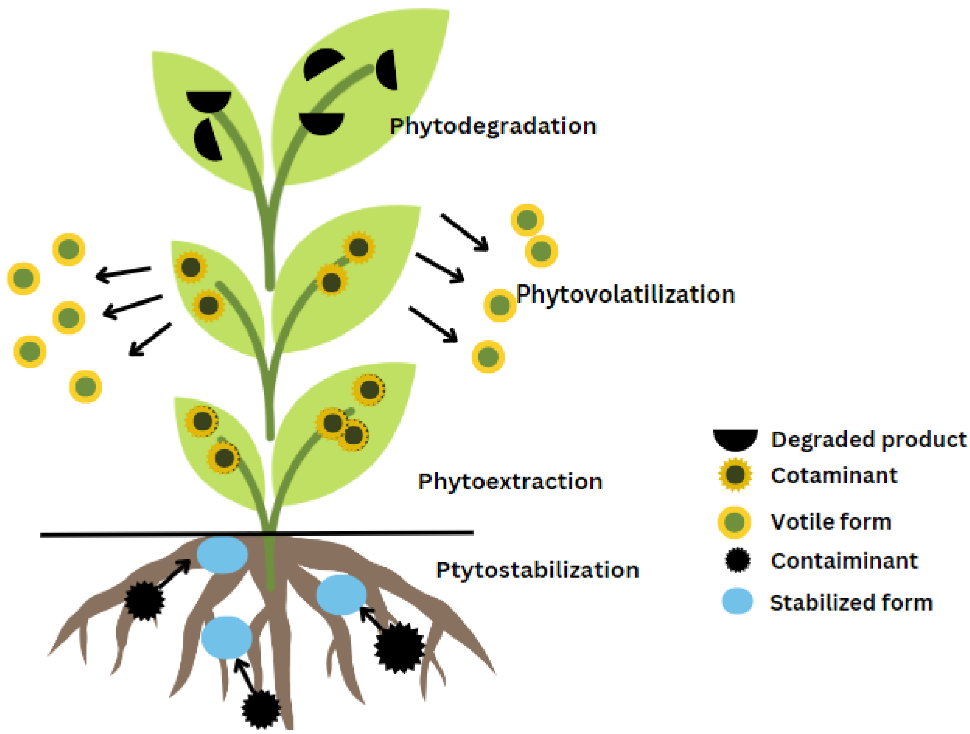

Figure 4: Mechanism of phytoremediation. Phytoremediation can be achieved by phytodegradation (to uptake organic contaminants), phytovolatilization (to transform contaminants into volatile compounds), phytoextraction (to extract contaminants from the soil and concentrate them in plant tissue), or phytostabilization (to decrease the availability of pollutants in the soil).

3. NANOTECHNOLOGY IN BIOREMEDIATION

Progress in nanobiotechnology has provided opportunities for tackling heavy metal pollutants by nanomaterials that are produced by eco-friendly methods (Rather *et al.,*2023). See **Fig. 5**.

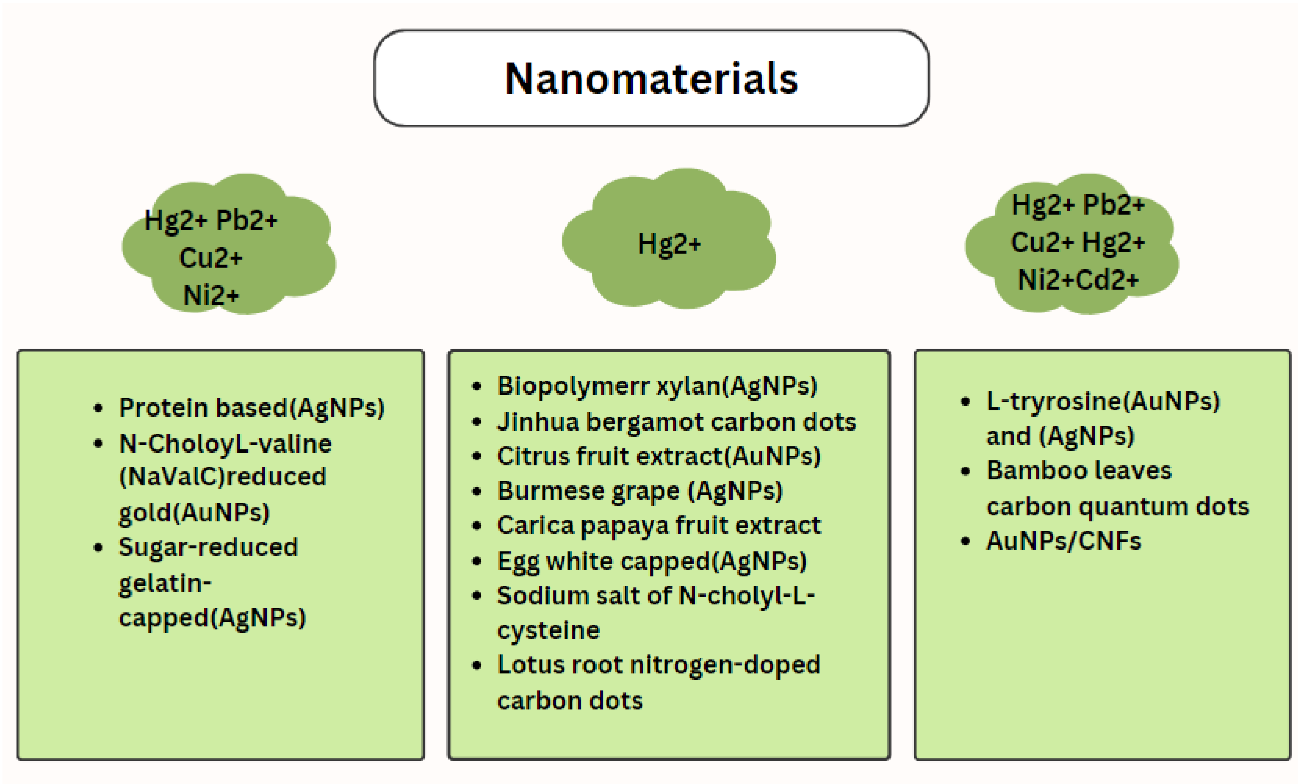

Figure 5: Nanomaterials used for the removal and detection of heavy metals in soils. Heavy metals may be identified and removed from polluted soils using nanomaterials, biopolymers, and fruit extracts. AgNPs, silver nanoparticles, gold nanoparticles (AuNPs), carbon nanofibers (CNFs)

4. STATE-OF-THE-ART BIOREMEDIATION APPROACHES

A staggering array of chemical compounds that have contributed to the modernization of our lives were created in the 20th century because of explosive research in the chemical industry (Hou *et al.,*2023). Globally, environmental quality has been declining because of the surge in the industrial-scale manufacturing of several chemical substances. (Esteves-Aguilar *et al.,*2023). However, compounds such as heavy metals, pesticides, and toxic gases, which have chemical structures different from those of natural organic compounds and cause toxicity, are resistant to biodegradation and biomagnification (Ghuge *et al., 2023)*.

Mycoremediation, cyanoremediation, genoremediation, hyperaccumulation, dendroremediation, rhizoremediation, biomineralization, and phytostabilization are among the more recent biotechnology methods used for bioremediation (Azubuike et al.,2016). The collaboration, integration, and synthesis of such biotechnological developments are needed to restore the ecosystem.

4.1. Biomineralization

Biomineralization refers to a process by which living organisms internally or externally form inorganic minerals (Hoffmann, *et al*., 2021; Maqsood *et al.,* 2023; Xing *et al.,* 2022). The role of fungi in this process stems from the ability of their hyphae to traverse diverse substrates and their organic matter degradation through either saprotrophy or parasitic and pathogenic interactions (Gadd, 2021). Typically, the biodegradation of contaminants involves the collaboration of multiple microorganisms (Gajewska *et al*., 2022). After 48 hours of incubation, heavy metal removal rates ranging from 88% to 95% were achieved by introducing soil bacteria to a heavy metal solution that included urea. The organisms UR47, UR31, and *Terrabacter tumescens* were shown to have the greatest removal rates for Cu, Pb, Co, Zn, Ni, and Cd, respectively (Zhao *et al.,* 2019).

4.2. Phytostabilization

Phytostabilization is a method that reduces the bioavailability of heavy metals by immobilizing them underground using metal-tolerant plant species. Reducing the possibility of contaminants entering the food chain is another benefit of this strategy. (Barba-Brioso, *et al*., 2023; Heredia *et al*., 2022). A variety of processes, including heavy metal precipitation or valence reduction in the rhizosphere, absorption and sequestration in root tissues, and adsorption onto root cell walls, contribute to phytostabilization (Salt et al., 1995). To ensure that this occurs, it is crucial to choose the right plant species. To achieve optimal phytostabilization, it is essential that the selected species exhibit resilience under heavy metal conditions (Rahman *et al., 2022)*. Since roots immobilize heavy metals and stabilize soil structure to avoid erosion, rooting systems are an essential feature of plants (Yan et al., 2020). For a plant cover to be established in a given area in a timely manner, it must be able to grow rapidly and generate large amounts of biomass, all while being simple to care for in the field.

4.3. Hyperaccumulation

Metals may be removed from soil and water sources by plants, which can hyperaccumulate metals (Wang et al., 2019). By creating phytochelatins in their roots, plants may store and transfer metal ions to their shoots. To clean soil and water, people often turn to plants, which may hyperaccumulate metals (Wang et al., 2019). Plants may store metal ions in phytochelatins they produce in their roots and then transfer them to their shoots (Maqsood *et al.,* 2023; Rahman *et al.,*2022). This could lead to the development of transgenic plants characterized by significant biomass, rapid growth, and critical qualities for effectively removing heavy metals. Research has been carried out to identify plants that may accumulate metals in the industrial states of Islamabad and Rawalpindi, which are in polluted regions (Wang et al., 2019; Yasir et al., 2021). Species may only be considered for remediation procedures if their bioconcentration factor, biological accumulation coefficient, and biological transfer coefficient are all greater than one. For the 23 different plant species that were tested, 43 samples were examined. It was determined that none of them were hyperaccumulators of metals. However, among the plants with BCFs, BACs, or BTCSs greater than 1, the most efficient metal-absorbing plants were *Brachiaria reptans, Cannabis sativa, Parthenium hysterophorus, and Polygonum barbatum* (Ahsan *et al., 2019)*.

4.4. Dendoremediation

According to González-Oreja et al., dendroremediation was a promising approach for metal phytoextraction in 2008, especially when rapidly growing tree species such as poplars (Populus sp. pi) and (Salix sp. pi) were used (Heredia *et al.*, 2022). Dendoremediation is a contemporary approach that utilizes trees to remove, sequester, or chemically breakdown contaminants from inorganic contaminated soils (Budzynska, *et al*., 2019). Dendroremediation has been demonstrated to be beneficial in soils contaminated with explosives, crude oil, metals, pesticides, solvents, and landfills. (Calabrese & Agathokleous, 2021). For fast-growing woody tree species with strong metal resistance potential, such as willows, oaks, birches, and poplars, metal phytoextraction (Populus sp.) is a promising approach (Budzynska, *et al*., 2019).

4.5. Rhizoremediation

The process of rhizoremediation involves breaking down organic pollutants in the soil area immediately adjacent to plant roots, known as the rhizosphere. Degradation often occurs because plant roots stimulate the catalytic activity of microorganisms (Chen *et al.,* 2023). It combines the two strategies of phytoremediation and bioaugmentation (Podar & Maathuis, 2022). Plant roots have the ability to distribute bacteria throughout the soil and penetrate solid soil layers. In regard to phytoremediation and bioaugmentation, the use of bacteria that breakdown pollutants in plant seeds could be a major breakthrough (Podar & Maathuis, 2022). One intriguing approach is rhizoremediation, which uses the remarkable capacity of root-associated microbes to biotransform harmful metals into less harmful compounds and breakdown organic contaminants (Podar & Maathuis, 2022; Wang et al., 2023). Plant-based in situ phytorestoration is a proven, efficient, and cost-effective method that is easy to use in the field.

Researchers found 11 bacterial strains that can withstand cadmium in mining waste, sewage sludge, and the root zones of Indian mustard plants grown on soils treated with cadmium in their study. Zn, Cu, Ni, and Co were also shown to have greater resistance to the bacteria. The isolated bacteria, which included *Flavobacterium* sp., *Rhodococcus* sp., and *Variovorax paradoxus*, may cause detrimental Cd concentrations or not in *Brassica juncea* seedlings by causing root elongation (Zhang et al., 2022). The detected bacteria could improve the development of *Brassica juncea*, a plant that accumulates metals, even in environments with toxic levels of Cd. They might also be used as plant-inoculant systems for the phytoremediation of contaminated soils (Niu *et al.,* 2023).

4.6. Mycoremediation

Mycoremediation can be performed in the presence of both filamentous fungus (molds) and macrofungi (mushrooms), both of which include enzymes for the breakdown of a wide range of contaminants. Mycoremediation involves the use of fungi in bioremediation to eliminate harmful substances (Akhtar & Mannan, 2020). Decomposers such as fungi are recognized as being able to significantly decrease and breakdown persistent and highly hazardous contaminants. The enhancement of mycoremediation can be achieved by introducing carbon sources into polluted areas and creating ideal conditions to accelerate the degradation process (Chaurasia *et al., 2023;* Gadd, 2021; Passarini *et al., 2022)*. According to recent research, *Aspergillus fumigatus* (M3Ai) had the highest bioleaching capacity (0.40 mg/g) for Cr in CYE media. The findings show that *A. fumigatus* M3Ai and *A. niger* M1DGR might be used to create novel approaches to treat soil polluted with heavy metals (Cd and Cr) by mycoremediation, either in situ or ex situ (Khan *et al., 2019)*.

4.7. Cyanoremediation

After thriving in challenging environments, Cyanobacteria can contribute to a cleaner environment. Their phototropic nature and ability to produce bioenergy make them potential candidates for a sustainable future. This approach to bioremediation using cyanobacteria is known as cyanoremediation (Ciani & Adessi, 2023; Sharma *et al., 2022)*. The capacities of cyanobacteria to remediate wastewater, accumulate excess food, and produce valuable biomass for multiple uses demonstrate their versatility. Furthermore, as photosynthetic autotrophs, they contribute to the improvement of water quality (Ciani & Adessi, 2023).

4.7.1. Genoremediation

Molecular genetic principles are used in phytoremediation to enhance metal tolerance. The transition of this technology from the laboratory to the field has accelerated through genetic engineering and the development of plants with inherited resistance to metals. The adoption of procedures for this process has already begun worldwide (Rai et al., 2020). It is possible to improve metal accumulation and tolerance by overexpressing naturally occurring or engineered genes that encode antioxidant enzymes (Pande et al., 2022; Podar & Maathuis, 2022). According to Ojuederie and Babalola (2017), transgenic *Brassica juncea* plants outperform their wild-type counterparts in terms of biomass and Se and Cd accumulation. These examples motivate researchers to conduct further studies on hybridization, selective breeding, and gene transfer (Chakdar et al., 2022; Gavrilescu, 2022; Rahman et al., 2022; Yadav et al., 2023).

4.8. SYSTEM BIOLOGY AND BIOINFORMATICS IN ADVANCED BIOREMEDIATION TECHNIQUES

To understand how an organism degrades a given contaminant, bioinformatics technologies are crucial. These tools extract information from a wide variety of biological databases, including those that store chemical formulas, microbial degradation pathways, organic compounds, catalytic enzymes, structural and compositional databases, expression databases for RNA and proteins, and comparative genomics (Shi et al., 2023; Yadav et al., 2023) (**Fig. 6**). Few bioremediation applications have occurred because researchers do not yet know enough about the factors that regulate the development and metabolism of bacteria that may be useful in these contexts (Chakdar et al., 2022; Rahman et al., 2022).

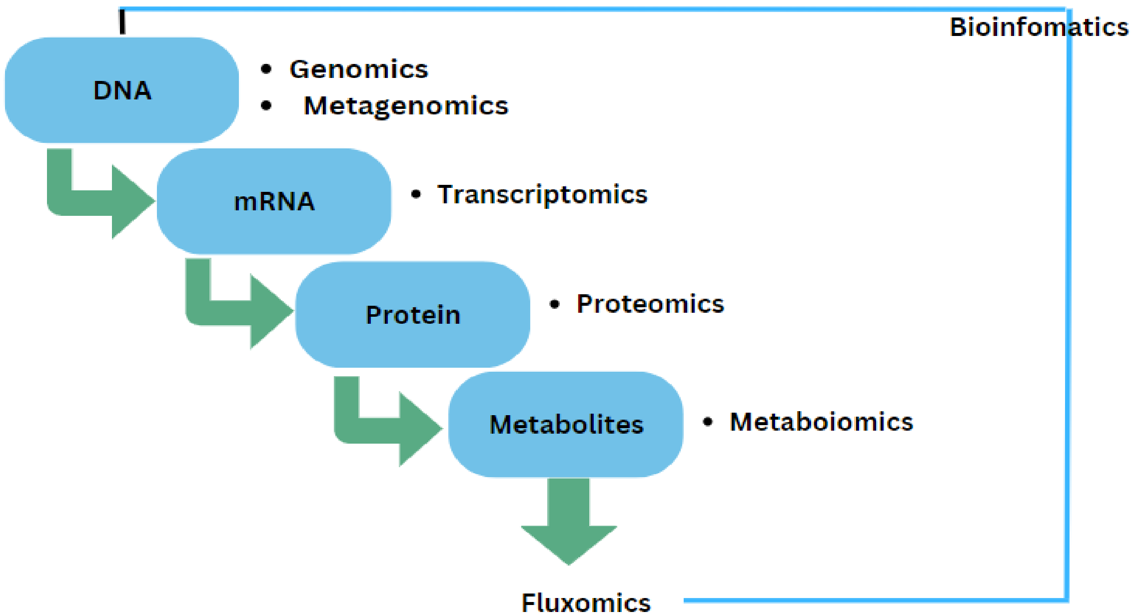

Figure 6: System biology approaches for bioremediation. The fields of genomics, metagenomics, transcriptomics, proteomics, metabolomics, and fluxomics are used in these methods. Plant–microbe interactions during heavy metal bioremediation in polluted soils may be better understood via the use of these omics methods in conjunction with interactomics.

The development of molecular methods for environmental management, including transcriptomics, metagenomics, proteomics, and associated omics approaches, has opened new possibilities (Rahman et al.,2022; Sharma et al.,2022; Yadav et al., 2023). While these methods eliminate the need for culture-based technologies, they have sped up the investigation of microbiome organization (Pande *et al.,*2022; Passarini *et al.,*2022; Shi *et al.,* 2023; Wang *et al.,* 2022; Zhu *et al.,* 2022).

4.8.1. Genomics-based bioremediation tools

The evaluation of pure cultures, which are crucial models for important bioremediation processes, has undergone an initial revolutionary change due to the use of genomes in bioremediation (Chakdar et al., 2022; Mathur et al., 2023). Mathur et al. (2023) and Pande et al. (2022) reported that bacteria with potential applications in bioremediation and whose physiology has not been well investigated before may greatly benefit from whole-genome sequencing (**Fig. 7**). To better understand biodegradation, scientists may employ genomic technologies, including QPCR, isotope distribution analysis, DNA hybridization, molecular connection, interactomics, metabolic footprinting, and metabolic engineering (Atuchin *et al.,* 2023; Gavrilescu, 2022; Rahman *et al.,* 2022; Sharma, et*al*., 2022).

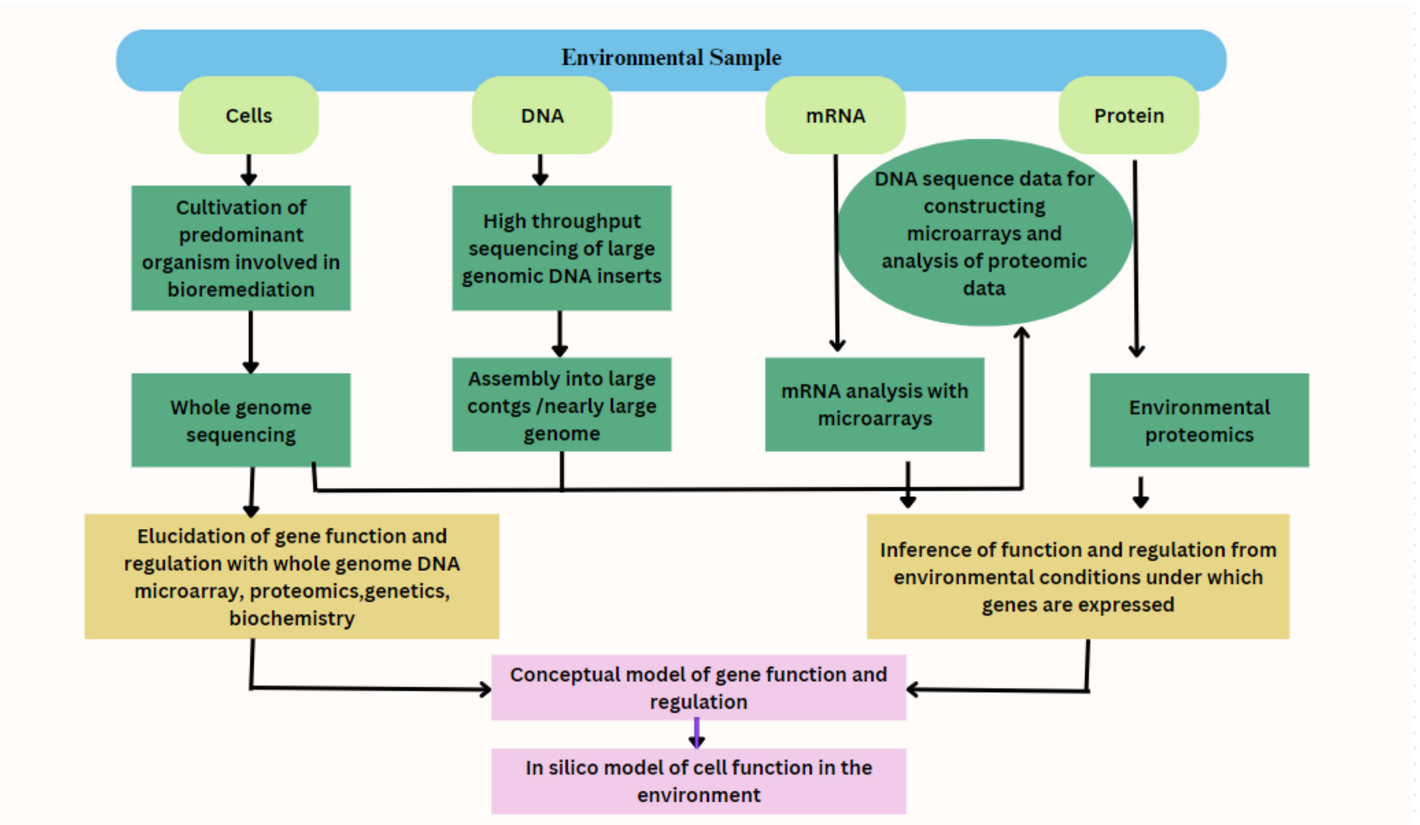

Figure 7: Contribution of OMICs and bioinformatics to the understanding of the bioremediation mechanism. To develop new methods for the environmentally safe bioremediation of polluted soils, scientists are using genomic, metagenomic, epigenomic,

transcriptomic, proteomic, metabolomic, and interactive methods to identify genes, microbes, and plant-microbial interactions.

4.8.2. *Transcriptomics*

More "multiomics" technologies have been used than any other, except transcriptomics. Bioremediation has also been extensively studied (Chen et al., 2023; Ghuge et al., 2023; Huang et al., 2022; Singh et al., 2022). The transcriptome has found widespread application in investigating bacterial involvement in the bioremediation process. By capturing the set of genes actively transcribed under specific conditions, the transcriptome functions as a crucial bridge connecting the cellular phenotype, interactome, genome, and proteome. This compressive approach enhances our understanding of bacterial activities during bioremediation, offering valuable insights into the intricate relationships among gene expression, cellular function and environmental response (Huang et al., 2022). Controlling gene expression is essential for adjusting to environmental changes and guaranteeing survival (Chen *et al.,* 2023; Sarker *et al., 2023;* Singh *et al., 2022;*Tiwari & Lata, 2018). Transcriptome analysis was performed using microarray and sequencing methods. Gene expression is evaluated with the use of microarrays, and the quantity of RNAs in a sample is determined with RNA sequencing using next-generation sequencing (Huang et al., 2022). Predesigned probes are used in microarray technology; this method is more effective, less expensive, and more suitable for analyzing protein expression (Huang et al., 2022). RNA sequencing is expansive in scope, as it enables extensive discovery research and offers significantly enhanced coverage of diverse RNA types (Huang et al., 2022).

4.9. *Proteomics and metabolomics*

The field of proteomics is concerned with studying the whole repertoire of proteins that are expressed by a cell at a given time and place. In contrast, metabolomics looks at an organism's total metabolite output across a certain time or place (Phurailatpam, et al., 2021; Zhang et al., 2023). Protein abundance and compositional variations may be studied using proteomics, which has also helped uncover key proteins linked to microorganisms. Metabolite profiling, foot printing, and target analysis are just a few of the many tools available in the microbial metabolomics toolbox that may be used to detect and measure the myriad of biological byproducts found in living organisms (Rahman et al., 2022). For cell-free bioremediation, information from both the proteome and metabolome will be helpful (Raklami et al., 2022). To investigate and comprehend the stressors caused by substrates, such as intermediates, hazardous dead-end products, and other environmental factors, metabolite analysis is crucial (Raklami et al., 2022). Proteomic analysis aids in the decoding of internal cellular molecular mechanisms, metabolic pathways, posttranslational changes, etc. In microbes, it has made it possible to follow and analyze the ubiquitous spatial expression of proteins (Hlihor et al., 2017; Phurailatpam et al., 2021; Raklami et al., 2022).

5. LIMITATIONS OF BIOREMEDIATION

To be able to bioremediate, the chemicals must be biodegradable. Complete and rapid breakdown is not possible for all chemicals.

Biodegradation raises additional concerns about the potential for byproducts to be much more dangerous or long-lasting than the initial substance. It is not easy to move from small-scale studies on benches and pilots to large-scale operations.

Research is essential for advancing bioremediation technologies tailored to environments characterized by intricate combinations of toxins that are unevenly distributed across the ecosystem. The challenges may include contaminants present in various forms, such as solids, liquids, and gases.

The applications of recombinant DNA technologies in the environment are presently limited.

When phytoremediation is used, nonedible plant products are produced.

6. CONCLUSION

Bioremediation has been suggested as the most environmentally benign method for removing heavy metals from soils. It stands out as a desirable, cost-effective alternative that is also simple to adopt and manage, may be conducted on-site or off-site, and minimizes the quantity of waste that is contributing to the landfill. This study delves into the fundamental principles of bioremediation and explores recent scientific breakthroughs enabling the effective application of biotechnological tools for environmental management. The focus is on mitigating global soil contamination from heavy metals. To address the significant environmental and societal challenges arising from such detrimental incidents, it becomes imperative to extend our exploration beyond traditional physical and chemical technologies. The most modern bioremediation methods, including heavy metal detoxification, root–microbe interactions, genomic and proteomic treatments for pollutants, and nanoparticles, have all shown promise for cleaning contaminated soils. New organomineral materials and promising genetically enhanced organisms (microbial strains and plants) that can more effectively absorb heavy metals have been created because of ongoing advancements in biotechnology, genetic engineering, and materials science. These developments have furthered the bioremediation process.

The rapid adoption and application of recent biotechnology are imperative for advances in soil bioremediation. With an emphasis on the affordability, suitability, and sustainability of the techniques to mitigate the effects of environmental change, contamination of food products and biological systems, the impact of anthropogenic activities on the environment, and exploration of the aforementioned opportunities along with new initiatives for the restoration of the environment, there is currently a wide range of scientific innovations.

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Conflict of interest declaration

The authors have no conflicts of interest to declare.

Author´s contribution

HKC, SS, and YEN prepared the draft of the manuscript. HKC, RDC, GMSE, HB, RBS, SS, and MYK edited and discussed the information included in this manuscript. All the authors agreed with the contents and final version of this document.

Data availability statement

Not applicable

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