

INSIGHTFUL GIST OF HEAVY METALS BIOREMEDIATION FOR ENVIRONMENTAL EQUITY

Abstract

The global accumulation of heavy metals in the environment represents a significant and widespread hazard. Exceeding established threshold levels of heavy metals such as mercury, lead, nickel, arsenic, chromium, zinc, cobalt, and aluminum has far-reaching consequences, deeply affecting soil quality, water purity, air quality, and the overall well-being of human populations. Heavy metal toxicants are at the peak of endangering nature. Toxic byproducts are released into the environment by both the Physical and chemical methods followed. Hence in recent times, research has been focused on the part using microbes for the remediation of heavy metal toxicants because of its eco-friendly approach. Toxic heavy metals and their by-products formed during the remediation process acts as energy reserve. This review hotspots about the potential use of various microbes for the bioremediation of heavy metal toxicants and also focuses on the resistance mechanism of microorganism that thrives in a toxic environment and its potential approaches as a key role for remediation techniques. This review review provides deep insight of knowledge into microbial remediation technology of hazardous heavy metal pollutants.

Keywords: Heavy metals, bioremediation, eco-friendly, toxicants, metalloids, carcinogens.

Authors

Preethi rathna R

Karpagam Academy of Higher Education
Coimbatore
Tamil Nadu, India.

Jayakala Devi R

Karpagam Academy of Higher Education
Coimbatore
Tamil Nadu, India.

Naveen Kishore S

Biosci Research Centre
Chennai
Tamil Nadu, India.

I. INTRODUCTION

Heavy metals, which are also broadly referred to as metals or metalloids, are characterized by their high density, typically exceeding 5 g/cm³, and atomic mass greater than 4000 kg m⁻³, surpassing that of water [1]. Among the notable heavy metals some are cadmium (Cd), cobalt (Co), arsenic (As), chromium (Cr), lead (Pb), nickel (Ni), vanadium (V), zinc (Zn), and mercury (Hg). This heavy metal toxic substance highly accumulates in soil, air, and water. These metals could be found in a variety of forms, including salt forms and insoluble compounds like carbonate, oxides, silicate, and sulfides [2]. However, heavy metals that are resilient in their ionic state—for example, Cd⁺², Hg⁺², Pb⁺², and As⁺³ exhibit the most toxic form as they combine with other toxicants and form a complex which degradation is quite complicated [3].

According to the research undergone by [4], The present global issue that cause more of the environmental issues is the heavy metal toxicants. Accumulations of heavy metals in soil, the marine ecosystem, and the atmosphere have caused disruptions. These disruptions have far-reaching effects, including the contamination of drinking water sources and the escalation of toxic metal pollutant levels in the food chain [5]. Metal pollution and emission from anthropogenic activities have scaled up with the tremendous growth of industrialization and urbanization. The environment and health impacts have been harmed by metal pollution brought on by mining, metal processing, and metal treatment, natural gas and oil, wastewater, road transport, and waste open dumping [6] (Figure 1). Lead, cadmium, mercury, and arsenic are regarded as the major toxic metals in the environment, as per the Environmental Protection Agency (EPA) [7]. Moreover, the United States Agency for Toxic Substances and Disease Registry (ATSDR) identifies over 20 heavy metals with significant toxicity, but it highlights four as particularly hazardous to human health: lead, arsenic, mercury, and cadmium. Among these, arsenic is the most frequent cause of acute heavy metal poisoning, followed by lead and cadmium. [8]. The heavy metal impact in the ecosystem is briefly described in (Table 1).

Microbial remediation is the optimal eco-friendly approach for restoring heavy metal contaminated sites into an invaluable form. Due to the often higher costs associated with traditional physical and chemical remediation methods, and as the concentration of heavy metal pollutants rises day by day, these methods can result in the generation of substantial amounts of toxic sludge. As a consequence, bioremediation is gradually being substituted by conventional techniques for reclaiming soils contaminated with heavy metals [9]. Heavy metals can be transformed from one oxidized form to another inorganic compound, but they cannot be broken down completely. Bioremediation is one of the most effective techniques for the cleanup of soil that is contaminated with toxic heavy metals based on their impact on the ecosystem and human well-being. To decontaminate or remove organic and inorganic xenobiotics from the environment, microorganisms play a prime role. This procedure restores and maintains the natural soil condition using a sustainable remediation technology. The emerging idea of environmentally friendly chemistry and engineering is cited among the new technologies or methodologies that include bioremediation. According to [10], microbial remediation is a technology that is rapidly developing and showing promising results. Interestingly, among the decontamination techniques, heavy metal microbial remediation has a wide-ranging progressive society. Concentrations of heavy metals can be tolerated by microorganisms, particularly soil microbes. However, certain microorganisms have specific metal requirements as micronutrients for their metabolic processes. For instance, while all

bacteria primarily use Fe³⁺, anaerobic bacteria rely on Fe²⁺. The primary mechanism governing the remediation of heavy metal ions by microorganisms is bioleaching. This process entails the mobilization of heavy metal ions from insoluble ores through dissolution or complexation, coupled with bio-oxidation.[12]. Biosorption also includes precipitation, chemical adsorption, ion exchange, surface precipitation, the formation of complexes with organic ligands, and redox reactions. Also bioaugmentation, and bioprecipitation mechanisms to resist the toxic heavy metals environment. The main aim of this review is to give the gist of the impacts of heavy metals which is remediated by microorganism through various bioremediation strategies. The benefits of employing microbial remediation for heavy metals are also emphasized.

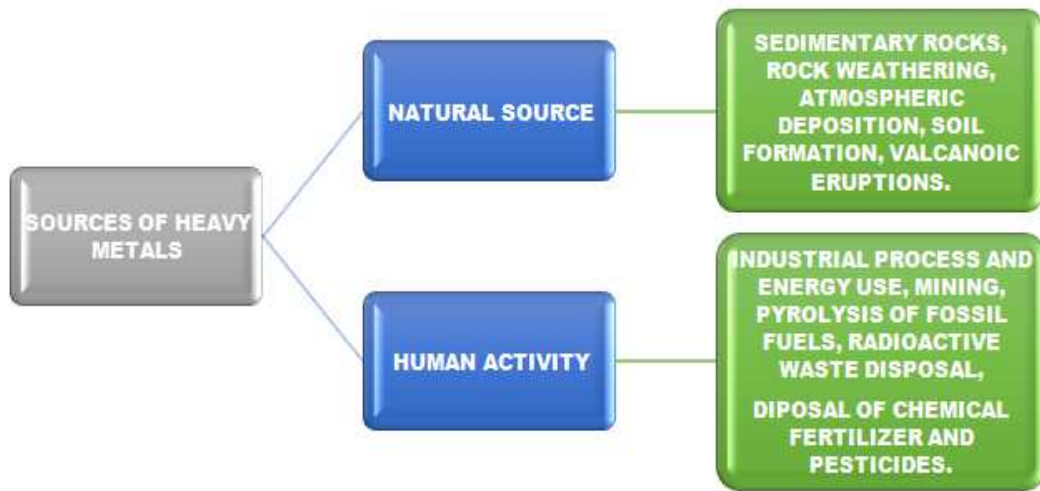


Figure 1: various modes of heavy metal contamination [13].

Table 1: Impact of Toxic Heavy Metals in the Environment.

Toxic Heavy Metals	Sources[14]	Impact In Soil	Impact In Water[15]	Impact In Human[16]
Arsenic	Industrial processing dust and manufacturing Automobile parts	Peroxidation of lipid and increase in Reactive Oxygen Species [17].	Cell toxicity for fishes.	DNA damage, epigenetic alterations and Neurotoxicity in humans.
Aluminium	Pesticides, insulation wires, and automobile parts	Peroxidation of lipid and increase in Reactive Oxygen Species [17].	Fish malformation.	-
Cadmium	Natural gas byproducts and chemical pigments from paints.	Reduction in biomass, stem conductivity and length of root, Inhibition of seed germination [18].	High blood pressure and cardiac illness in fish.	Fanconi-like syndrome, Neurotoxicity, phosphaturia, glucosuria, and aminoaciduria

Copper	Electroplating industries	Deformation in root and shoot of plants.[18].	Neurotoxicity for marine living beings.	Wilson's disease
Chromium	Leather, tanning, electroplating, chrome plating, textile industries.	Decrease in production of chlorophyll[19]	Bronchial, lymphocytosis, eosinophilia, anemia and renal injury.	Allergicdermatitis, lymphocyte proliferation is stopped.
Iron	Metal and engine parts	Above the limit, it inhibits the plant enzyme activity and impacts the soil pH [20].	Reduction in fertility and embryonic development in fish	Negative influence the neurodevelopment when exceeded its limit.
Zinc	Paints, dyes, ointments, and preservatives	Interveinal chlorosis and alternation in enzymatic activity [20].	Death, hypoxia	zinc-induced neurotoxicity
Lead	Mobile batteries, gasoline, pesticides.	DNA damage, reduction of chlorophyll and protein content causes foliage [20].	Fish malformation	Damages the DNA mechanism, structure of chromosomes and tumor-regulating genes.
Nickel	Metal refining, phosphate fertilizers, paints, batteries processing unit.	Decrease in production of chlorophyll [19]	Reduction in fertility and embryonic development in fish	nickel-induced hepatic dysfunction

II. MICROBIAL BIOREMEDIATION OF HEAVY METALS

Microbial bioremediation stands out as a crucial, appealing, cost-efficient, and eco-friendly approach. This method harnesses naturally occurring microorganisms already present in contaminated areas to effectively remove heavy metals. The use of microorganisms such as algae, bacteria, and fungi for detoxifying heavy metals in polluted sites has emerged as a promising solution. Through continuous exposure to pollutants, these microorganisms develop tolerance and demonstrate remarkable abilities to convert pollutants into sources of energy and raw materials. They can even genetically adapt to degrade contaminants, making them ideal candidates for an economical and environmentally friendly biological process [21]. Persistent exposure to metals enables microbes to acclimatize and build resistance against these metals. Hence, it becomes imperative to comprehend the dynamics of

interactions between microbes and metals. These interactions can be categorized into various types. (Fig:2).

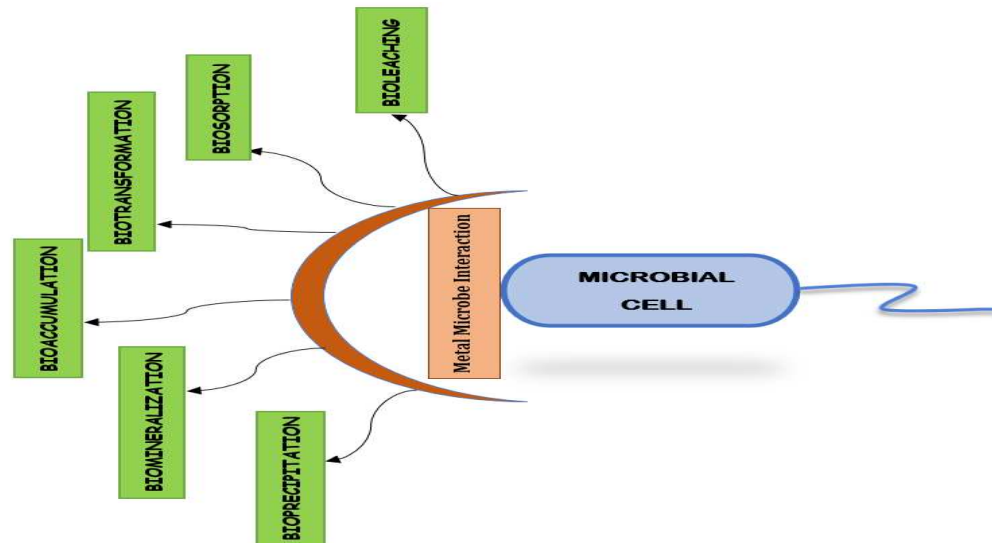


Figure 2: Illustration of diverse bacterial interactions with heavy metals contaminated soils [22].

III. MECHANISM OF HEAVY METALS DEGRADATION BY MICROORGANISM

Microbes have the ability to transform chemicals into vital sources of energy and raw materials to support their own growth and metabolism, creating a biological process that is inexpensive and ecologically sustainable. Heavy metals are now considered a significant environmental issue because of their extensive industrial use. Industrial activities and the burning of fuels are the primary contributors to the accumulation of toxic heavy metals in the food chain. This accumulation presents a significant threat to both the environment and human health. Heavy metals such as mercury, silver, lead, cadmium, and arsenic exert toxic effects on living cells. According to [73], in a polluted environment, nutrients such as nitrogen, phosphate, sulfur, iron, and potassium can serve as stimulants and essential supports for robust microbial growth and cellular metabolism. In their DNA, many different kinds of bacteria have genes that make them vulnerable to various cations and oxyanions of heavy metals. In order to deal with the uptake array of diverse heavy metal ions, bacteria go through an array of diverse mechanisms. Numerous mechanisms are involved in the interaction between microorganisms and heavy metals, including biosorption, entrapment, efflux, reduction, precipitation, and complexation [23].

IV. BIOSORPTION

Heavy metal ions exhibit non-specific binding to the polysaccharides and proteins found on the cell surface. Both living cells and deceased microbial biomass offer binding sites that can be utilized by heavy metals, even in highly dilute solutions [24].

Various microalgal strains, including *Spirulina platensis*, *Chlorella vulgaris*, *Oscillatoria* sp., and *Sargassum* sp., have been extensively studied for their interactions with heavy metals [25]. Research conducted by [26] indicates that algae can attract both positively

and negatively charged species of various heavy metal ions due to the presence of compounds like deprotonated sulfate, laminaran, and monomeric alcohols. Furthermore, [27] has reported the effectiveness of a lead-resistant bacterium, *Staphylococcus hominis* strain AMB-2, in biosorbing heavy metals such as lead and cadmium. Additionally, coral-associated solubilizing bacteria, specifically *Cronobacter muytjensii* KSCAS2, have demonstrated the ability to biosorb multiple heavy metals [28]. Biofilms produced by microorganisms can serve as effective adsorbents for heavy metals. According to a study conducted by [29], biofilms formed by *Staphylococcus aureus* have the capability to bio-precipitate U(IV). Moreover, the introduction of acid phosphatase further contributes to the remediation of U(IV). It's important to distinguish between absorption and adsorption. While absorption involves the penetration of a substance throughout the entire volume of another material, adsorption takes place only at the surface of the material. Many living organisms have been identified as potential biosorbents, including bacteria such as *Magnetospirillum gryphiswaldense* and *Bacillus subtilis*, fungi like *Rhizopus arrhizus*, yeast-like *Saccharomyces cerevisiae*, and algae such as marine microalgae and *Chaetomorpha* [30]. Several bacterial species, including *Bacillus* sp., *Pseudomonas* sp., and *Escherichia* sp., are adept at absorbing substances owing to their small size and adaptability to diverse environmental conditions [31]. In the biosorption process, the initial step involves a metal ion coming into contact with a microbial cell membrane. The metal ions attach to functional groups (such as amine, carboxyl, hydroxyl, phosphate, sulfate, and amine) located on the cell wall [32]. As indicated by [31], the process of heavy metal uptake involves the binding of metal ions to reactive groups present on the bacterial cell wall before these metal ions are incorporated into the cell. In fungal cell walls, it's noted that approximately 90% of the composition consists of polysaccharides. Various functional groups play a role in metal binding, including carboxyl, phosphate, uranic acids, proteins, nitrogen-containing ligands, and chitin or chitosan.

Polysaccharides like alginic acid, chitin, xylan, and mannan contain functional groups (sulfate, hydroxyl, phosphate, imidazole, amino, and amine) known to serve as metal binding sites [31]. Two proposed mechanisms governing metal binding are ionic charge and covalent bonding [33]. Fungal cells' capacity to absorb substances can be altered through physical or chemical treatments, such as autoclaving, heating, or the use of dimethyl sulfoxide, laundry detergent, orthophosphoric acid, formaldehyde, glutaraldehyde, or NaOH. Macrofungi, naturally occurring in various environments such as forests, polluted soils, and water bodies, are capable of absorbing heavy metals. This process involves the desorption of metals from the biosorbent [34]. Microbes employ an import-storage system and remain metabolically active during the bioaccumulation of toxins. In this system, heavy metal ions are transported across the lipid bilayer of the cell membrane and into intracellular spaces or the cytoplasm with the assistance of transporter proteins. This process is referred to as active uptake or bioaccumulation. The bioaccumulation of heavy metals in the bacterial membrane is mediated by factors such as the extracellular environment, signal transduction, carrier-mediated transport, complex penetrability, and lipid penetrability [35].

V. BIOAUGMENTATION

Bioaugmentation is a technique employed for the remediation of heavy metal contamination in polluted sites, utilizing specific microorganisms or microbial populations to aid in the removal of these heavy metals. It is designed to enrich the microbial population and to make it more effective in downsizing the level of heavy metal contamination [36] (figure 3). In genetically modified *E. coli*, the expression of the Cd(II) adsorption protein EC20 on

the cell surface, along with the overall regulator gene *irrE* in the cytoplasm, has been utilized to enhance Cd(II) adsorption under high-salinity conditions. The research found that the maximum adsorption capacity was 30.79 mg/g at a Cd(II) concentration of 30.84 mg/L and with 2% NaCl in *E. coli* (EC20/*irrE*). According to the study conducted by [37], the removal percentages for both COD (Chemical Oxygen Demand) and Cd(II) reached notably high levels. Specifically, the removal percentages were 96.17% for COD and 97.60% for Cd(II), which are significantly higher when compared to the control group, which achieved removal percentages of approximately 79.15% for COD and 92.09% for Cd(II). Studies undergone by [38] state that Consortia of the three evaluated fungal species, *Geotrichum candidum*, *Aspergillus transmontanensis*, and *Cladosporium cladosporioides* influences in biomineralization of heavy metals contaminated soils. Maximum bio-removal capacities of the studied fungal species after 90 days were 72% for Cu, 99.8% for Co, 60.6% for Fe, 82.2% for Mn, and 100% for both Pb and Zn. According to [39], research has proved that the soil supplemented with fungal consortia, (*Perenniporia subtrophora*, *Daldinia starbaeckii*, *Padina conrescens*, *Fusarium equiseti*, *Polyporales sp.*, *Aspergillus niger*, *Purpureocillium lilacinus*, *Aspergillus fumigatus*) had an elevated metal removal capacity over the control which was resulted in mineralization of Mn (67%), Ni (67%), and Zn (66%) which are all the most removed metals.

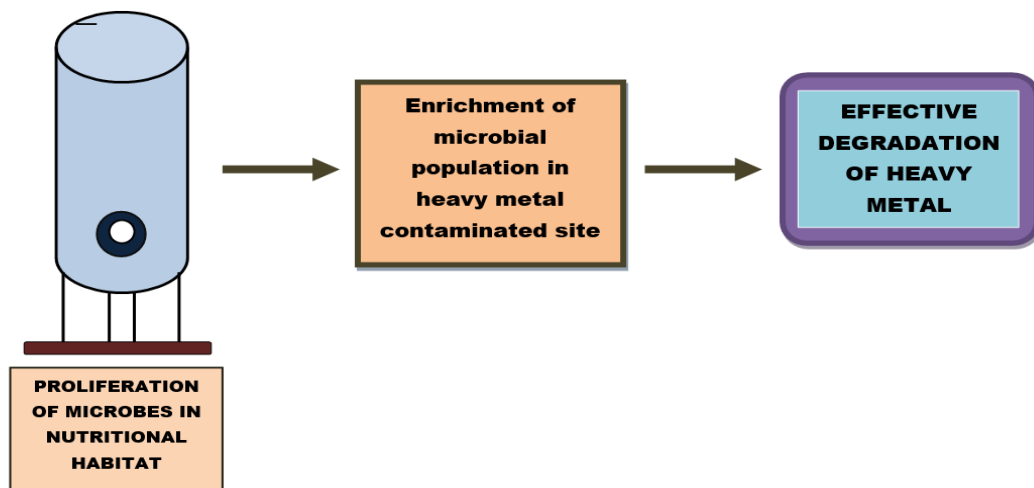


Figure 3: Illustration of process carried out during bioaugmentation.

VI. BIOLEACHING

In mining and biohydrometallurgy, vital metals are been retrieved from low-grade ore with the help of different microbes. This method is termed bioleaching (or biomining). By releasing organic acids, microbial species diffuse carbonaceous material on metal. Organic acids interact with metals through three primary mechanisms: acidolysis, complexolysis, bioaccumulation, and chelate formation; these processes are seen in *Bacillus* sp., *Penicillium* sp., *Aspergillus* sp., and some species of actinobacteria. The environmental mechanisms of numerous microorganisms are influenced by various biotic and abiotic stress conditions. They have the power to impose cellular reactions that can block particular metabolic pathways, designed to protect microorganisms from internal or external stress conditions [40].

Depending on the target metal, cyanogenic bacteria like *Chromobacterium violaceum* or chemolithoautotroph bacteria are the quite often used species of bacteria for the bioleaching of PCBs (common e-waste) [41]. Citric acid served as a chelating agent in [42] proposed hybrid bioleaching and hydrometallurgy approach, which augmented base metal recovery from e-waste. *Acidithiobacillus ferroxidans* produce exopolymeric substances more efficaciously when using citric acid, and jarosite formation is reduced. In their study of monazite dissolution at pH 1.8 - 4.0 using HCl and different organic acids, [43] the study revealed a notable correlation between the extraction of rare earth elements and pH, indicating that proton-promoted dissolution plays a significant role. In the context of heterotrophic organisms, the provision of an organic carbon source significantly contributes to the expenses associated with bioleaching rare earth elements. Economic analyses conducted on the bioleaching of rare earth elements using *Gluconobacter oxydans* demonstrated that both the cost of the medium component and the overall process cost were primarily attributed to the carbon source, specifically glucose. [44].

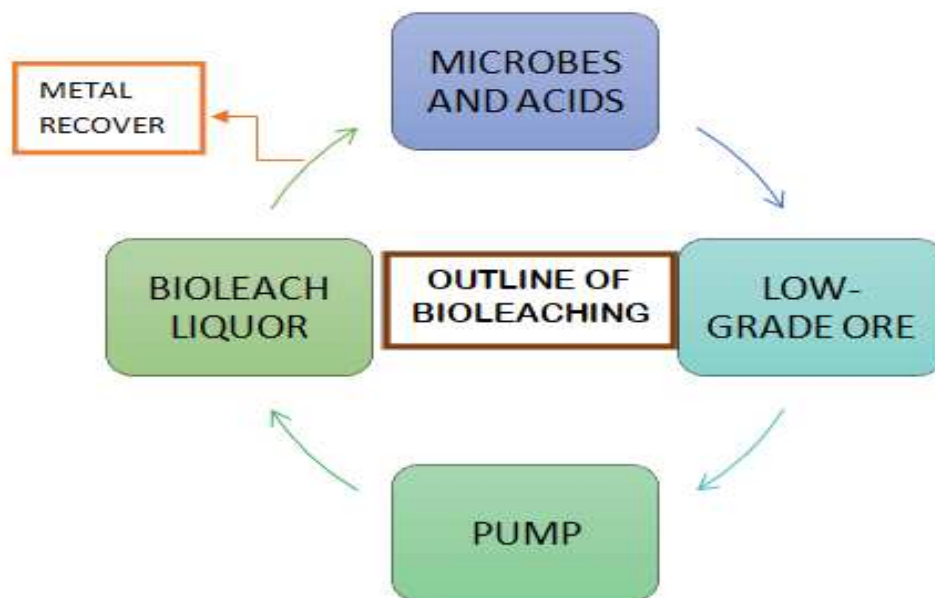


Figure 4: Outline of bioleaching process (modified from Itam *et al.*, 2019)

Many different types of microbes can perform bioleaching, but acidophiles stand out among them. Acidophiles are chemolithotrophs that thrive in acidic environments, especially pH range of 2.0 and below, and oxidize Fe (II) to Fe (III) and also reduce sulfur to sulfuric acid. Sulfuric acid plays a crucial role in the extraction process by generating ferric ions and protons. These components are effective in dissolving metal sulfides and iron oxide found in ore deposits, thus separating metals in the solid phase from those that are more water-soluble. This separation process enhances the efficiency of metal extraction. Heavy metals can also be extracted and recovered using bioleaching, which it employs microorganisms as reduction agents [47]. According to reports in the most recent literature by [48], bioleaching microorganisms produce oxidative compounds and sulfuric acids that help hydrolyze heavy metals from contaminated soils. For bioleaching, organic acids, siderophores, heterotrophic microbes that generate biosurfactants, and bacteria that disintegrate Fe/Mn have all been identified. In order to oxidize, and intricate toxic metals in soil, the bioleaching process primarily relies on metabolic activity and the production of primary products.

- 1. Autotrophic Bioleaching:** As reported by [49] that sulfuroxidizing bacteria are *Acidithiobacillus thiooxidans*, *Acidithiobacillus caldus*, *Sulfobacillus benefaciens* and iron-oxidizing bacteria are *Acidithiobacillus ferrooxidans*, *Leptospirillum ferrooxidans*. The bacteria most thoroughly investigated for autotrophic bioleaching belong to the genus *Acidithiobacillus*. Because of their remarkable resistance to the heavy metals and their need for small nutrients for metal mobilization, they are predominantly used in bioleaching [50]. By oxidizing sulfur and iron, these bacteria cause metal sulfides to disintegrate, which significantly reduces pH and promotes the solubilization of other metal compounds. The oxidized ferrous ions, thiosulfate, and elemental sulfur do provide the energy for *Acidithiobacillusferrooxidans*, resulting in the production of ferric ions and sulfuric acids for the biosorption of metals. Since it produces inorganic acids (sulfuric acids), *Acidithiobacillus thiooxidans*, quickly oxidizes elemental sulfur. As a consequence, Fe^{2+} and sulfur reducing species form in the solution and act as substrates for protozoans. Fe^{2+} and sulfur species solutions are converted to Fe^{3+} and sulfuric acid, respectively, by planktonic cells. In addition, Fe ion serves as an electron carrier, preventing Fe ion from being oxidized by direct contact. The indirect mechanism takes place when microbes are not in direct contact with mineral surfaces. Bacteria's sole purpose is to ramp up the reoxidation of Fe^{2+} [51]. Microorganisms can significantly raise leaching efficiency in two aspects: one is by adhering to the soil medium's surface, lowering mass-transfer restrictions by producing bio lixivants (such as chelators and organic acids) directly on the surfaces of the metals; and the other by an accumulation of heavy metals, complexation by released chemicals or adsorption leading to changes in the equilibriumgrowth of the strain. However, the experimental evidence in contact leaching supports how the microorganisms displace metal sulfidesis still underwhelming. In contrast, optimization of the microbial leaching agent of metal sulfides by indirect bioleaching studied separately. It's important to note that bacteria through contact and non-contact mechanisms oxidize to sulfur compounds from Fe (III) ion, by production of the oxidizing agent, created by the mineral dissolution [44].
- 2. Heterotrophic Bioleaching:** In the indirect process of heterotrophic bioleaching of toxic metals in polluted sites are solubilized by microorganisms with production of certain biosurfactants, organic acids and other metabolites [52]. Oxalic, isocitric, gluconic, acetic, lactic, succinic, malonic, pyruvic, and formic acids are a few examples of organic acids secreted during the metabolic process of degradation of heavy metals [53]. To eliminate metals from complex substrates by electron transfer and maintain the lower pH required for effective bioleaching, these organic acids produced by heterotrophic microorganisms are essential for the solubilization of metal ions [54]. Because of their strong affinity for Fe(III) and Mn(II), siderophores with carbonyl structures are crucial for transferring iron in media with low levels of feasible Fe(III). Using organic acids, they can chelate these metals as well. These metabolites not only dissolve metal ions from minerals but also synthesize chelates and soluble metal complexes that do the same for soil [55]. The capacity to excrete significant amounts of organic acids like citric acid, lactic acid, gluconic acid, oxalic acid, and siderophore. The heterotrophic fungal species *Penicillium simplicissimum* and *Aspergillus niger* are the most frequently used for bioleaching [56]. The stability of the compound and the complexity of the organic acid both affect the rate of leaching. This suggests that the difficulty of the ligand's retention or adsorption by the soil, as well as its strength, both significantly increase the leaching efficiency [57]. Different types of heterotrophic microorganisms that play a role in complexolysis include

Bacillus megaterium, *Pseudomonas aeruginosa*, *Chromobacterium violaceum*, and *Pseudomonas fluorescens*.

VII. BIOTRANSFORMATION

According to [58], biotransformation is the process of structurally altering a chemical compound to produce a more polar molecular. In other words, toxic metals and organic compounds are changed by this interaction of metal and microorganisms into a form that is relatively less dangerous. The microorganisms adapt to environmental changes as a result of developing this mechanism. The production of new carbon structure, isomerization, the addition of new functional groups, reduction, oxidation, hydrolysis, condensation, methylation, and demethylation are all reaction happens as a resultant in microbial transformations. By reduction, oxidation, methylation, demethylation, and complexation, microorganisms can interact with heavy metals and affect how they are biotransformed [59]. The physical and chemical characteristics of metal ions and its geochemical conditions of polluted areas determine how microbial transformation affects accumulation of heavy metal and transformation [60]. The majority of mineral components contain high concentrations of non-biodegradable, soluble metals. In these circumstances, subsequent transformation of insoluble metal species can be controlled by microbial mobilization. When it comes to soluble metal, microbial transformation is crucial in altering or modifying the reduction/oxidation states of heavy metals ions in the sediments, as well as their solubility, mobility, bioavailability, and toxic nature. A number of biological mechanisms, including the direct reduction of Cr(VI) to Cr(III) and the indirect reduction of Cr(VI) to Cr(III) by biologically produced Fe(II), have been linked to the bioreduction of Cr(VI) [61].

The solubilization of the metal ions occurs as a result of a series of redox reactions that occur when intracellular heavy metals in many microbes bind to the metallothionein complex [62]. A protein with a high cysteine is primarily found in higher microbes, cyanobacteria, and microorganisms which have a strong affinity for metals like cadmium, zinc, copper, and mercury. Additionally, the *Pseudomonas* sp., has metals reducing genes that are conserved for the production of metallothionein, which helps these organisms survive in toxic environments [63]. Cellular superoxide and hydroxyl radicals are directly impacted by the metallothionein complexes' redox potentials.

VIII. BIOACCUMULATION

The influx and accumulation of metals within bacterial membranes is known as bioaccumulation. Pathways are α -helical proteins that allow heavy metals to passively diffuse across the membrane along a gradient of concentration. A wide variety of bacteria, including *Corynebacterium* sp., *E. coli*, *Serratia* sp., *Streptomyces coelicolor*, and *Pseudomonas* sp., have been found to transport these channel proteins, which are members of the Major Intrinsic Proteins Super Family [64].

An appealing alternative to the physical and chemical technique used to treat heavy metal accumulation by use of the microorganisms. Most significant bacterial species used in bioaccumulation processes are *Bacillus* sp., *Staphylococcus* sp., *Corynebacterium* sp., *Enterobacter* sp., *Escherichia* sp., *Aeromonas* sp., *Pseudomonas* sp., *Klebsiella* sp., *Vibrio* sp., *Arthrobacter* sp., *Brevibacterium* sp., *Deinococcus* sp., *Erwinia* sp., *Micrococcus* sp., *Nocardia* sp., *Serratia* sp., and *Thiobacillus* sp., [13]. Through Bacterial cell wall detoxification

processes, it can continuously remove metals when exposed to it. The various defense mechanisms used by bacteria to overcome metal toxicity include methylation, sequestration by metal-organic complexation, biotransformation and reduction of metals using metal reducing enzymes, and production of metal chelators and metallothioneins [65].

According to the USEPA (2010), the microorganisms absorb pollutants by direct contact with polluted media or indirect ingestion of the pollutant. The rate of pollutant removal is outpaced by the amount of absorption and bioaccumulation. The pollutant consequently gets stuck inside the organism and builds up [66]. The chemical toxic metals or its substances bond inside of a microbial cell structure through a process called heavy metal bioaccumulation. The various exposure pathways (diet and solution) and geochemical effects on bioavailability affect the bioaccumulation of metal.

However, Microbial accumulation is a useful integrative exposure indicator of chemicals in contaminated sites [66]. Bioaccumulation depends on the bioactivity of the biomass. The metabolic processes in microbial cells are active during adsorption of heavy metal pollutants through the bioaccumulation technique [67]. The same pathways that allow nutrients to enter cells in living things also allow metals to do so. Metals and vital nutrients, such as calcium and magnesium, are absorbed by microbes.

Microbial bioaccumulation involves several stages. In the initial stage, metal ions are bonded to the microbial cell surface. This initial stage has no metabolic activity. The cell is then supplied with metal ions. Only when the cells are metabolically active can the second stage of this procedure be initiated. If the second stage in the ideal conditions for organism growth are sustained, biomass production rises. This makes it possible for larger concentrations of metal ions to bind [68] and solubilize the metal ions more effectively.

IX. BIOPRECIPITATION

Microbial bioprecipitation transform soluble metals and metalloids into insoluble precipitates. According to research by [69], *Desulfomicrobium norvegicum*'s biofilm can precipitate elemental selenium and sulfur. A review conducted by [70] thorough the mechanism of bacterial strains involved in the reduction of uranium. *Citrobacter* sp. cells were immobilized and precipitated the cadmium, copper, uranium and lead from supplemented glycerol-2-phosphate. When glycerol-2-phosphate was broken down in this instance by phosphatase, hydrogen phosphates were released. These precipitated metals are excreted out the cell in the form of insoluble metal phosphates. When zirconium was mineralized by *Citrobacter* sp., a mixture of $Zr(HPO_4)_2$ and hydrated zirconia (ZrO_2) was produced [71].

Numerous microbes are known to produce insoluble but non-crystalline metal species. The biological immobilization of heavy metals like zinc, calcium, selenium, and nickel, has also been studied. Bioprecipitation of toxic heavy metals is best understood for arsenic and iron. Utilizing microbes' capacity to precipitate extremely maneuverable, soluble metals allows for bioremediation of heavy metal accumulated sites. The *Desulfuromonas* sp., and *Geobacter* sp., are being used in experiments to use this capability, such as to precipitate uranium from contaminated aquifers [72]. Acetate act as an electron donor, these bacteria can convert soluble hexavalent to insoluble tetravalent uranium. To pinpoint the precise physiological conditions required for bacterial growth, more research is still needed before

these bacterial abilities can be used in situ. Another intriguing example for bioremediation studies is the microbial formation of insoluble arsenic species. According to recent studies, from the sediment of freshwater isolated the organism *Desulfotomaculum auripigmentum*, which can form arsenic trisulphide by reducing arsenate and sulphate to arsenite and sulphide then oxidizing lactate to carbon dioxide (Equation 1). An iron-sulfur cluster is thought to be the enzyme's active center based on how sensitively the reaction responds to inhibition of molybdate by microbial enzymes. Where abiotic formation is impossible, when minerals are produced by extra- and intracellular in microbial metabolism.

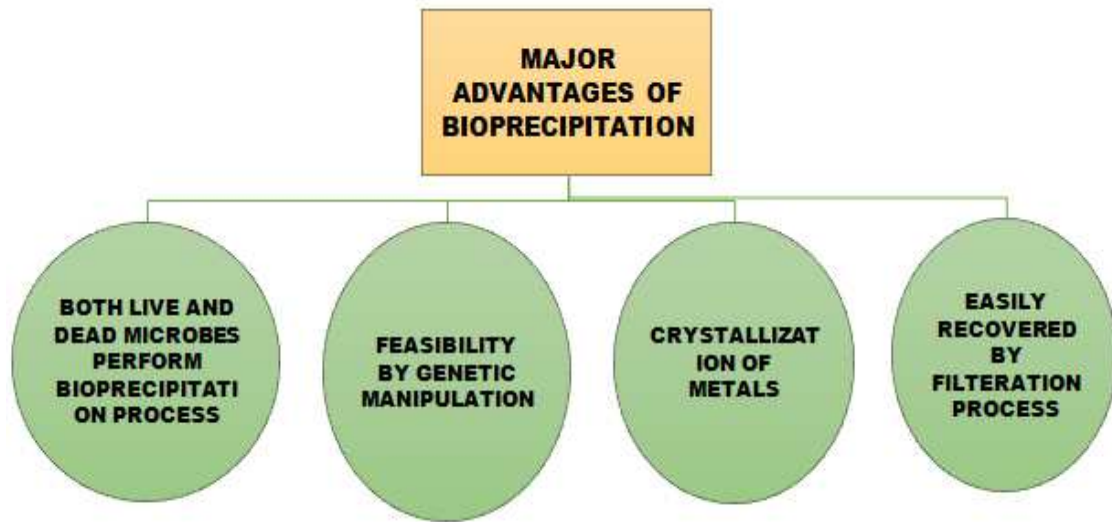
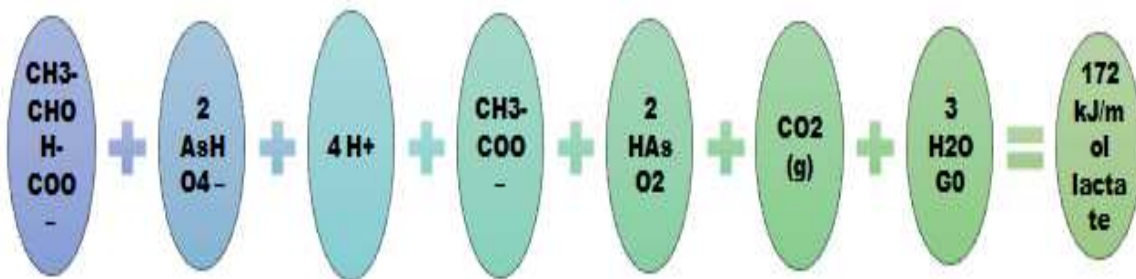


Figure 5: Advantages of bioprecipitation for heavy metals remediation



Equation 1 bioprecipitation of heavy metals. Although the exact mechanism by which orpiment is transported from within the cell to the outside is unknown, experimental data points to the possibility that orpiment adhere to the membrane and passes into the cell wall. Arsenate is primarily reduced in the presence of sulphate and arsenate because this interaction provides the microbe with much more energy. To maintain a low concentration outside the cell membrane of arsenite and a high energy yield during the reduction of arsenate, sulphate must be reduced first, followed by the precipitation of orpiment. If *Desulfotomaculum auripigmentum* has an arsenite utilization pathway like other bacteria does not yet appear to be known. On the other hand *Agrobacterium albertimagni*, appear to be unable to bio-precipitate arsenate by the microbes.

X. ADVANTAGES OF DEGRADATION OF HEAVY METALS BY MICROORGANISM:

Bioremediation requires very little work, requires less labor, is affordable, environmentally friendly, sustainable, and generally simple to implement. The number of microbes that break down the contaminant rises, and they produce harmless byproducts. Typically, the products left behind after treatment are nontoxic substances like carbon dioxide, water, and cell biomass. Compared with other chemical and physical methods for removal of toxic heavy metals, microbial bioremediation requires a lot less work, requires less labor, and is less expensive. The prior removal of toxic heavy metals by microbes will help to hamper the ecological and financial factors. The best microbial processes should be chosen for the bioremediation of toxic heavy metals for not to harm the environment while the remediation processes. Sometime the site of contamination can be cleaned up a particular period without large transport or labor cost.

XI. CONCLUSION

Heavy metals originating from Natural and artificial sources become the major pollutant in the environmental ecosystem. As they take long time for degradation and negative impacts to the environment. Industrial wastewater that has been accumulated in the environment like rivers and soil—needs immediate government involvement, regular monitoring and technological rehabilitation. Approaches by traditional treatment have their limitations and ought to be replaced with more effective, economical, and eco-friendly options by microbial remediation. To obtain the benefits of these organisms' ability to minimize to heavy-metal pollution, it is necessary to thoroughly examine their potential and carry out in-depth research. While using microbes for bioremediation, various abiotic factors like pH, temperature, concentration, and biomass contact time must be given consideration. Determining the outcome of pollutants accumulated microbial cell is crucial, as well as making sure that microbial biomass doesn't synthesize toxic substances to the food chain to animal and human health. This entails determining the best organisms and environmental factors for the heavy metals bioremediation as well as creating ecofriendly and sustainable processes for handling and getting rid of the toxic pollutants. This article lists various forms of bioremediation and discusses their benefits as well as their applicability to various sectors. The review pinpoints about the microbial mechanisms of action of various bioremediation techniques, as well as the microbes that are significant and the potential influences on the bioremediation of heavy metals, as well as how recent developed technologies can make microbial bioremediation more effective. This study will help the researchers to fill the research gap in the heavy metal biodegradation.

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