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CADMIUM BIOREMEDIATION: A REVIEW

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ABSTRACT: The Increase in industrialization has raised the levels of heavy metal pollution in the country. Heavy metals are entering through the water supply, which is ultimately reaching into streams, lakes, rivers, and groundwater. So, ultimately, these heavy metals are causing environmental pollution, which is becoming a threat to humans as well as animals as the heavy metal pollution is linked with various life-threatening diseases as discussed below. It is very difficult to remove these potent and hazardous metals from the environment as they cannot be degraded chemically and biologically. Bioremediation offers an economical approach to remove these contaminants. Bioremediation uses both live microorganisms as well as plants (Phytoremediation) for the removal of heavy metals. Among toxic heavy metals, cadmium is one such metal which is harmful to flora and fauna. The study shows the present concrete scenario as well as the previous findings in this field of remediation of potent heavy metals, especially cadmium, which is very hazardous.

INTRODUCTION: Heavy metals are naturally present on earth: Lenntech, 2004 defined heavy metals as an element with high density. Mercury, cadmium, arsenic, chromium, and lead, etc. are few examples of heavy metals which are extremely toxic. Some metals are required for the metabolism of humans like copper *etc.* but at higher concentrations, they act as a toxin. Heavy metals are highly toxic because they start accumulating, and their breakdown is either slow, or they do not degenerate, resulting in many health problems. Increase in industrialization has raised the levels of heavy metal pollution in the country.

Heavy metals are entering into water supplies through waste material, which is releasing them into streams, lakes, and rivers. So ultimately these heavy metals are causing environmental pollution. It is very difficult to remove these potent and hazardous metals from the environment as they cannot be degraded chemically and biologically. The main metals which are hazardous are cadmium, lead, mercury, Zinc, Chromium, and copper. These heavy metals are extremely toxic and can mainly contaminate soil water *etc.*^{1, 2} The aquatic biota can be badly affected by the presence of heavy metals³. Nowadays their presence is also visible in the food chain, which is very harmful for mankind.

There are many industries that are releasing heavy metals in their effluent. Among all these potent metals the cadmium is one such non-essential heavy metal having high toxicity at even low dosage to all organisms.

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Cadmium can cause reproductive system failure, brain damage, kidney damage to human beings⁴, and also it is related to cancer⁵. Renal damage was also reported due to cadmium toxicity⁶. Cadmium can enter into aquatic life through sewage sludge. Discharges which contain cadmium should strictly control as they are highly toxic and tends to accumulate in tissues of organisms⁷. There is a lesser number of reports showing bioremediation of cadmium by microorganisms. Environmental pollution due to cadmium particulate from industries is increasing day by day. It is entering the food chain and ultimately reaching the environment. The purpose of the study is to completely remove the cadmium by using natural process, *i.e.* with the help of naturally occurring micro-organisms having potential to remove it completely from the environment.

Cadmium and its Utility: Cadmium is a silver-white colored metal and has a solubility in acids. Friedrich Strohmeyer, a German chemist, discovered cadmium in 1817. Cadmium has one interesting feature in it that it is noncorrosive, and that's why it is used in deposition on other metals as a protective layer. Cadmium belongs to Transition metal family of the periodic table with atomic number 48 and atomic mass 112.4g mol^{-1} . Its melting point is $321\text{ }^{\circ}\text{C}$, and the boiling point is $767\text{ }^{\circ}\text{C}$. By nature, Cadmium has high corrosive resistivity that's why it is used for plating other metals in electronics and aerospace industries⁸.

A very large amount of cadmium is released into the environment⁹. Cadmium is released into rivers through weathering of rocks, forest fires, *etc.* and also through human activities like electroplating, manufacturing, *etc.* Cadmium can cause many health problems like damage to the nervous system, immune system, psychological problems as well as reproductive failure and even infertility. So, it is essential to remove it to avoid human health problems and environmental deterioration. A most economical and eco-friendly way to treat this pollution is bioremediation.

Cadmium Toxicity:

Health Effects: The International Agency for Research on Cancer (IARC) has classified cadmium and cadmium compounds as carcinogenic to humans (IARC 1993 and 2009). According to

IPCS 1992 and WHO 2010 in humans, exposed to cadmium mainly occurs by consuming contaminated food and through inhalation by workers in the metal industries. Cadmium can easily bind to respiratory enzymes¹⁰ and can cause cancer¹¹.

Plants act as a carrier of various cadmium salts (Especially those having great solubility in water) into the food chain¹². Plants which are growing in polluted soil can easily take up heavy metals that may then enter the food chain¹³. In selected vegetables, Cd concentration in soils and food crops was above permissible limits¹⁴. So from their report, it can be concluded that consuming such vegetables may lead to cadmium toxicity in humans as well as animals. People who are living near the industrial area are more prone to toxicity, which can severely damage their lungs. People who like fishes such as shellfish in their diet regularly are at more risk of cadmium exposure (ATSDR 2008) **Table 1**.

TABLE 1: SHOWING HEALTH EFFECTS OF CADMIUM

S. no.	Problems	References
1	Bone fracture	15
2	Reproductive failure and possibly even infertility	16
3	Kidney Damage	17
4	Shortness of breath, lung edema	18
5	Cancer development	19
6	Psychological disorders	20
7	Hypertension, renal damage, testicular atrophy	6

Environmental Effects: cadmium exposure to the environment is a threat now a day's. This exposure is mainly from the mining industries **Table 2**.

TABLE 2: SHOWING EFFECTS OF CADMIUM ON GROWTH OF PLANTS

S. no.	Problems	References
1	Effects tissues of aerial parts of plants	21
3	Inhibits cell division in leaf	22
4	Causes abnormal mitosis division in plant	23

Bioremediation: Bioremediation is an act of removing pollutants from a contaminated site with the help of microorganisms as well as plants. Using microorganisms for the treatment of polluted soils is a more eco-friendly and economical approach in the field of bioremediation.

According to the EPA, bioremediation is a “technique which uses naturally occurring microorganisms to break down hazardous substances into less toxic or non-toxic substances.” There are two techniques of bioremediation *in situ* and *ex-situ* Fig. 1.

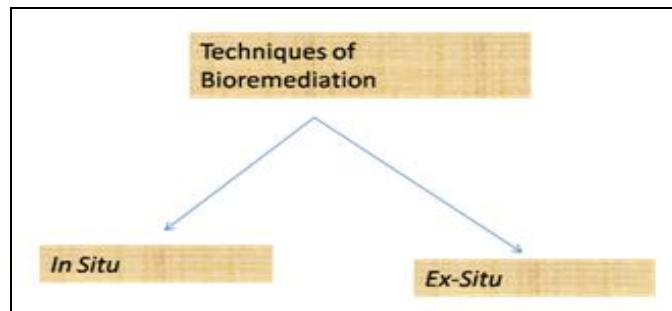


FIG. 1: TECHNIQUES OF BIOREMEDIATION

In-situ Bioremediation: Involves the onsite treatment of the contaminated effluent. We need not remove soil or water samples from the site. The pollution by heavy metal on the site can be directly removed. In this type of remediation naturally occurring microorganisms are stimulated to show their activity against the heavy metals^{24, 25}. Since it uses naturally occurring microorganisms so it is not cheaper as well as harmless nor it produces harmful byproducts in the environment. It mainly involves the onsite treatment of polluted substances. These techniques are very cheap than *ex-situ* bioremediation techniques, as no disturbance to its structure. Dyes, heavy metals, chlorinated solvents can be treated with the help of this technique^{26, 27}. The suitable conditions required for successful *in-situ* bioremediation are moisture content, pH, temperature, nutrient availability, the status of electron acceptor, *etc.*²⁸ The advantages of *in-situ* bioremediation involves minimal exposure of public and site personnel, minimal site disruption, low cost, simultaneous treatment of contaminated water in soil Fig. 2.

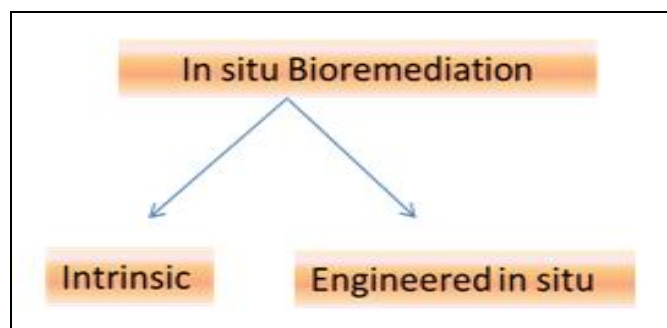


FIG. 2: TYPES OF *IN-SITU* BIOREMEDIATION

Intrinsic bioremediation is simply enhancing microflora on the site of remediation where pollution of heavy metals occurred.

Engineered *in-situ* is introduced the systems have to be incorporated to enhance the microflora on the site.

Ex-situ Bioremediation: As the name suggests, *ex-situ* means outside, so it involves the removal of the contaminated effluent to be treated elsewhere. The technique of land farming was demonstrated by the US Army Corps of Engineers, which showed the enhanced remediation of petroleum contaminated soils. Microorganisms which are used to perform the function of bioremediation are well known as bioremediation.

Types:

Phytoremediation: It is an emerging technique that mainly depends on the use of physical, chemical, and biochemical interactions of plants in contaminated sites for the treatment/mineralization of pollutants. There are several mechanisms which are involved in phytoremediation. These mechanisms mainly depend upon the type of pollutant as a different process is needed for different contaminants. The pollutants like toxic heavy metals are removed by extraction, while pollutants like hydrocarbons and chlorinated compounds are removed by degradation, rhizoremediation, and volatilization^{29, 30}. There are many important factors that should be kept in mind while choosing a plant as a phytoremediator like root system – which may be fibrous or tap depending on the toxicity of pollutant, to which extent the pollutant is toxic to plant, what will be plant growth rate in the presence of pollutant, and how much time is required to achieve the desired level of remediation. The use of Plant Growth Promoting Rhizobacteria was reported, which might play an important role in phytoremediation. Phytoremediation of cadmium was done with the help of three indigenous plants *Eleusine indica*, L. (KB-1), *Ageratum conyzoides*, L. (KB-2), and *Euphorbia hirta* (KB-3) and subsequently, 58.8, 52.2, 51.8% of reduction of cadmium concentration was observed.

Phytoextraction: Phytoextraction involves a different a mechanism of plants which remove

toxic elements, *i.e.* heavy metals from soil or water, metals whose density is very high and may be toxic to organisms when present in low concentrations. The mechanism mainly depends upon the ability of plants to translocate the pollutants from roots to aboveground shoots or leaves. Natural ability and efficiency of a plant is the basis of Phytoextraction^{31, 32}. Phytoextraction of cadmium was performed with the help of plant *Brassica juncea*, and 60% reduction was observed in Cd concentration³³.

Microorganisms for Bioremediation: Besides plants, the minute species which cannot be seen by the naked eye (visible in microscopy) can be used as bioremediation to remove toxic pollutants from the environment. The naturally present microorganisms which are indigenous or extraneous, are used as agents for bioremediation. The nature of micro mainly relies on the type of polluting agent. Proper selection criteria of organisms should be selected as different species show different survival rate in different concentrations of pollutants. Bioremediation can be achieved by mainly two processes, *i.e.* either naturally or through intervention process, as reported by³⁴.

Classification of Microbes- Microbes involved in bioremediation are –

(1) Aerobic: Aerobic bacteria are those who need oxygen for their growth and survival. The aerobic bacteria which are involved in bioremediation are *Mycobacterium*, *Pseudomonas putida*, *P. diminuts*, *Sphingomonas*, *Rhodococcus*, *etc.* These microbes decontaminate the sites by using the pollutants like heavy metals, pesticides, and hydrocarbon compounds as their carbon and energy sources.

(2) Anaerobic: Anaerobic bacteria are those who cannot grow in the presence of oxygen and also the use of these bacteria is not as frequent as aerobic ones for bioremediation purpose. The main approach, which is used for anaerobic bioremediation is *in-situ*³⁵.

(3) Methylophils: These bacteria come under the class of Aerobic bacteria that grow utilize methane as a source of energy and carbon. It has a wide range of substrate and reported against a huge range of pollutants. In Methylophils the enzyme methane mono-oxygenase was reported for its

activity and stability against a wide range of pollutants

(4) Ligninolytic Fungi: The fungi also can degrade a wide range of toxic pollutants, for example, White rot fungus such as *Phanaerochaete chrysosporium*. White rot fungi degrade organic pollutants in soil and effluents. *Candida* degrades formaldehyde. Common substrates which are used are straw, corn cobs, or sawdust.

Mycoremediation (Using Fungi): Myco-remediation is bioremediation in which fungi species are used to decontaminate the pollutant or to target a specific pollutant. Scientists experimented in 1999; a plot of diesel oil contaminated soil was inoculated with mycelia and succeeded in its decontamination. Fungi like white rot fungus can degrade toxic pollutants³⁶. Fungi of the genera *Penicillium*, *Aspergillus*, and *Rhizopus* have been studied as an efficient bioremediator for the removal of heavy metals and. Sometimes microfiltration term is used in this process in the mycelium of fungi is used to filtrate the toxic compounds from water in the soil^{37, 38}. In **Table 3**, few of the microorganism has been shown through which bioremediation has been achieved against heavy metal³⁶. Ligninolytic Fungi such as *Phaneraero chaetechrysosprrium* can degrade a wide range of pollutants³⁹ **Table 3**.

TABLE 3: MICROORGANISMS HAVING the ABILITY TO DEGRADE HEAVY METALS (SOURCE: VIDALI 2001)

S. no.	Elements	Microorganisms
1	Zn, Cu, Cd	<i>Bacillus sp. Pseudomonas aeruginosa</i> ⁴⁰
2	CO, Ni, Cd	<i>Citrobacter sp.</i> ⁴⁰
3	Cd, Pb	<i>Phormidium valderium</i> ^{41, 42}
4	Pb, Au, Zn, Cd	<i>Chlorella vulgaris</i> ⁴⁰
5	Cd, Zn, Ag	<i>Aspergillus niger</i> ⁴⁰
6	Zn, Cu, Cd	<i>Pleurotus ostreatu sp.</i> ⁴⁰
7	Hg, Ag, Cd	<i>Rhizopus arrhizus</i> ⁴³

The specific features of microorganisms like survival and growth at extreme environmental conditions like temperature, heat, and desert conditions make them suitable for the removal of toxic pollutants from the environment. Many fungi and bacteria are known to be used for bioremediation of Industrial effluent. Different species of *Pseudomonas*, *Bacillus*, *Aspergillus*, *Sporophyticus* have been reported as efficient

chromium and nickel reducers. The first patent for the strain *Pseudomonas putida* for bioremediation was filed in 1974 which was able to degrade petroleum³⁴. Some microorganisms have resistance against the heavy metals and can grow in the heavy metal-rich environment. A metal tolerant bacterium that has the capability of removing Copper, Chromium, and arsenic from treated wood was isolated⁴⁴. These microorganisms have different mechanisms from each other to tolerate the uptake of heavy metals. Studies were conducted on biosorption of cadmium by *Pseudomonas* and concluded that this species could tolerate up to 50 mg/L of cadmium⁴⁵.

Bioremediation in the rice plant by metal resistance *Ochrobactrum* sp. and *Bacillus* sp was done recently⁴⁶. *Ralstonia eutropha* has been modified genetically to decrease the toxic effect of the Cd (II) in the contaminated sites³⁴. For the efficient elimination of Zn(II) and Cd (II), *Saccharomyces cerevisiae* acts as a bio sorbent^{47, 48}. *Cunninghamella elegans* was reported as promising organism against heavy metals released by textile wastewater⁴⁹. Recently cadmium and chromium metals are checked for their bioremediation with the help of Earthworm *Eisenia fetida* who showed the tolerance up to 0.06 and 0.14 mg/l respectively⁵⁰. Among the wide range of microorganisms used for bioremediation, there are many fungal strains that are used for the purpose. *A. niger* is used for the bioremediation of chromium from the tannery industry⁵¹. *Aspergillus fumigates* has excellent efficiency in removing heavy metals⁵².

Different groups all over the world are involved in search of microbial strains that can be helpful for bioremediation. In Brazil, China and other countries where Industrialization is increasing with the higher pace the hazardous contaminants are also increasing at a similar pace. In addition to the development of various biosensors, the work has also been started where the organism's resistant to specific metal is checked to show resistant against other heavy metals also. Cadmium resistant *Stenotrophomonas* sp. showed resistant to Selenium and Copper⁵³.

Recently Heavy metal resistant bacterial strain from Industrial and Agricultural areas in Mauritius was isolated, which are helpful in bioremediation

⁵⁴. A wide range of accumulation of cadmium was regulated by *Escherichia coli*⁵⁵. *Pseudomonas* strain was modified with the pMR68 plasmid with novel genes (mer) made that strain resistant to mercury⁵⁶. *Klebsiella pneumonia* M426 was modified to precipitate the mercury⁵⁷.

Many laboratories throughout the nation are involved in isolation and identification of heavy metal resistant bacteria. Physiochemical methods were reported to be used as an approach to reduce the heavy metals from contaminated site⁵⁸. The strain has successfully reduced Cr and Ni. Bacterial strains of *Pseudomonasputida*, *Pseudomonas aeruginosa*, and *Bacillus* sp. were found to reduce Chromium (VI) content of the textile Industry effluent⁵⁹.

In addition to bacterial strains, many fungal strains have also been reported showing heavy metal absorption efficiency. Fungal strains from Sugar mill of Cheyyiar Cooperative Sugar Mill, Thiruvannamalai district were isolated and utilized them for the bioremediation of sugar mill effluent⁶⁰.

The use of potent microorganisms that are resistant to activities of heavy metal is very important⁶¹. During past years strains have been isolated like *Staphylococcus aureus* especially for cadmium^{62, 63}. Cadmium resistant bacteria, i.e. *Pseudomonas aeruginosa* from waste processing plant sludge effluent was isolated and was subjected to induction of mutation by acridine orange and acriflavine and 94.7% of cadmium removal was achieved⁶⁴. The strains *E. coli* and *P. syringae* were studied for the removal of cadmium by respectively^{65, 66}.

Many studies have been carried out in the past for the isolation and identification of bacterial strains that are resistant to cadmium. *Pseudomonas putida*, *Pseudomonas aeruginosa*, *Pseudomonas fluorescens*, *E. coli*, *Comamonas testosteroni*, *Klebsiella planticola*, *Staphylococcus aureus*, *Bacillus subtilis*, *Lactobacillus plantarium*, *Staphylococcus lugdunensis*, *Ralstonia metallidurans*, *Alcaligenes eutrophus*, *Serratia liquefaciens*, and *Bacillus thuringiensis* were the common species of bacteria that have been studied most regarding their cadmium accumulation ability⁶⁷.

Pseudomonas putida degrades about more than 80% of cadmium in less than 5 min at pH from 5.0 - 7.5⁶⁸. *Alcaligenes eutrophus* CH₃₄ bacterial strain was grown on a pilot - plant. It degrades up to 90% of the cadmium⁶⁹. The bacterial strains were able to degrade cadmium, but their efficiency depends upon the induced proteins that are produced in the cells⁷⁰. *Streptomyces rimosus* has a cell wall which contains anionic groups such as phosphate amino, carboxyl, etc. whose ability to adsorb cadmium is very high⁷¹.

Microorganisms used biosorption method to remove the toxic metals which are present in water and streams. *Bacillus licheniformis* and *Bacillus laterosporus* were two bacterial strains that remove cadmium from water. The temperature of 60 °C did not affect the ability of these strains to degrade cadmium. The maximum capacities of non- living cells of these strains to remove cadmium were 142.7 and 159.5 mg/g respectively⁷². Bacterial strain *Rhodotorula* sp. Y11 was able to degrade cadmium. Its capacity was about 11.38 mg/g⁷³.

Recent studies revealed that *Staphylococcus xylosus* and *Pseudomonas* sp. were the bacterial strains whose ability to degrade cadmium was 278 and 250 mg/g respectively⁷⁴. A bacterial strain KUCd1 was isolated, which closely resembled with *Pseudomonas aeruginosa*. *Pseudomonas aeruginosa* strain KUCd1 showed its ability to remove about 75% and 89% of cadmium⁷⁵. *Spirogyra* and *Oscillatoria* sp. were studied for sequestration of cadmium from industrial wastewater⁷⁶.

Phytoremediation: Besides of microorganisms plants can also be used to clean the residues of toxic heavy metals⁷⁷ **Table 4.**

TABLE 4: LIST OF PLANTS USED FOR REMEDIATION OF HEAVY METALS

S. no.	Name of Plants	Heavy metals
1	<i>Brassica juncea</i>	Lead and cadmium ⁷⁸
2	<i>Claytonia perfoliata</i>	Cadmium ⁷⁹
3	<i>Cerastium arvense</i>	Cadmium ⁷⁹
4	<i>Stellaria calycantha</i>	Cadmium ⁷⁹
5	<i>Lupinus Albus</i>	Arsenic and Cadmium ⁸⁰
6	<i>Allium schoenoprasum</i>	Cadmium ⁸¹
7	<i>Digitalis purpurea</i>	Cadmium ⁸²
8	<i>Pteris vittata</i>	Arsenic and cadmium ⁸³
9	<i>Melilotus officinalis</i>	Hydrocarbons ⁸⁴
10	<i>Populus termula</i>	Lead and cadmium ⁸²

CONCLUSION: The presence of heavy metals in the food chain has become a threat to human and wildlife as it can cause many severe diseases as discussed above. The heavy metals as fumes are also degrading the environment. So to remove all these threats, there is a need for the implication of proper treatment processes which can minimize the health and environmental impacts of toxic heavy metals.

There are many other physical processes by which above said a goal could be achieved, but they are expensive and have low efficiency for the removal of compounds. The eco-friendly techniques such as bioremediation, phytoremediation, Phytoextraction, and Mycoremediation, etc. offer a great choice to eliminate these contaminants permanently from the environment and clean it.

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