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Bacterial Extracellular Polymeric Substances Mediated Heavy Metals Removal of Soil

*MA Mubeenuddin, M Rajeshwar Malavath and M Shanti

Department of Soil Science and Agricultural Chemistry, College of Agriculture,
Rajendranagar, Professor Jayashankar Telangana Agricultural University,

Rajendranagar, Hyderabad, Telangana, India

*Corresponding Author's email: mubeenuddinma786@gmail.com

Heavy metals are defined as metals with an atomic number greater than 20 and a density exceeding 5 g/cm^3 . Based on their biological significance, they are classified as essential or non-essential. Essential trace elements, including Fe, Cu, Mn, Mo, Se and Zn, are required in small amounts for normal physiological and biochemical processes but become toxic at elevated concentrations. In contrast, non-essential heavy metals such as As, Cd, Pb, Hg, Pu, W and V have no known biological function and are highly toxic even at low exposure levels. Owing to their physico-chemical properties, particularly ionic charge, these metals can readily penetrate cells and tissues, posing severe risks to living organisms.

Bacterial Extracellular Polymeric Substances (EPS) are crucial for bioremediation, using a network of polysaccharides, proteins, and DNA to efficiently bind heavy metals in contaminated soil. EPS facilitates the removal of toxic metals like Cd, Pb, Hg and Cr through mechanisms such as biosorption, complexation, and precipitation, acting as a natural shield that immobilizes metals

Keywords: Heavy metals, Bacterial Extracellular Polymeric Substances, Biosorption complexation, and Precipitation

Introduction

Heavy metals are metals in the periodic table that have metallic properties, molecular weight is greater than 55 and an atomic number is more than 20. While this includes Mn and Fe heavy metals that are environmental concern are usually regarded as chromium (Cr), nickel (Ni), copper (Cu), zinc (Zn), arsenic (As), selenium (Se), strontium (Sr), molybdenum (Mo), technetium (Tc), cadmium (Cd), mercury (Hg) and lead (Pb). It should be apparent that these metals (Zn, Cu and Mo) are micronutrients absolutely required for plant growth. All cells need trace quantities of heavy metals. Heavy metals are often cofactors in enzyme-catalyzed reactions

Heavy metal pollution is a major global environmental and public health concern due to metal toxicity, environmental persistence and the ability to bioaccumulate and biomagnify within food chains (Ali *et al.*, 2019). Toxic heavy metals contaminate approximately 14–17% of global agricultural land, impairing soil microbial communities, reducing enzymatic activity and disrupting soil ecosystem functions (Dumitriu *et al.*, 2021). Crops grown on contaminated soils accumulate heavy metals, increasing the risk of food-chain transfer and associated health effects, including cancer, respiratory and digestive disorders.

Heavy metals are widely used in industries such as electronics, transportation and power generation and can be transported over long distances through atmospheric and aquatic pathways. According to the World Health Organization, over 10 million people worldwide are affected by heavy metal-contaminated drinking water, with arsenic exposure alone impacting nearly 140 million individuals. The United Nations Environment Programme

reports that approximately 70% of industrial wastewater containing heavy metals is discharged untreated in developing countries, significantly exacerbating environmental and health risks (UNEP, 2020). Conventional remediation methods are often costly and environmentally unsustainable, driving increasing interest in eco-friendly alternatives such as microbial bioremediation for effective heavy metal removal.

Extracellular polymeric substances (EPS) play a very important role in biosorption of heavy metals, and are produced by most bacteria. EPS comprise a mixture of polysaccharides, mucopolysaccharides and proteins, which depends on the strain and its culture conditions. They contain ionizable functional groups such as carboxyl, phosphoric, amine, and hydroxyl groups, which enable EPS to sequester heavy metals. Ion exchange, complexation with functional groups of negatively charged, adsorption and precipitation are the mechanisms involved in metal biosorption onto EPS (Wang *et al.*, 2023)



Permissible Limits of Heavy Metals Content in Water, Soil and Plant

The accumulation of heavy metals in agricultural soil and plants from different sources of soil contaminates, it may deteriorate the soil quality and that can increase risks to human health. The maximum allowable limits of heavy metals in irrigation water, soil and plants have been established by standard regulatory bodies such as World Health Organization (WHO), Food and Agricultural Organization (FAO) and Ewers standard Guidelines in Europe as shown in table 1

Heavy metals	Maximum permissible level in irrigation water(μgml^{-1})	Maximum permissible level in soils (μgg^{-1})	Maximum permissible level in plants (μgg^{-1})
Cadmium (Cd)	0.01	3	0.10
Lead (Pb)	0.065	100	0.30
Nickel (Ni)	1.40	50	67
Chromium (Cr)	0.55	100	-
Zinc (Zn)	0.20	300	100
Copper (Cu)	0.017	100	73
Iron (Fe)	0.50	50000	425
Manganese (Mn)	0.20	2000	500
Cobalt (Co)	0.05	50	50
Arsenic (As)	0.10	20	-

Effects of heavy metals on soil microbial activity

Some heavy metals in the soil also have an effect on the growth of soil microbes (Buekers, 2007). Almost all soil biochemical reactions have the participation of soil microbes, which plays an important role in maintaining soil quality and plays an important role in the formation of soil organic matter and its decomposition of harmful substances, biochemical

cycles and the formation of soil structure. Heavy soil contaminated soils have a negative effect on soil microbial properties, such as the underlying soil respiration rate and enzyme activity that depends on soil pH, organic matter and other chemical properties. Studies have shown that, in most cases, low concentrations of heavy metal contaminated soil are conducive to the release of CO₂, high concentrations of heavy metal pollution conditions, significant inhibition of soil respiration, severe heavy metal pollution can inhibit soil microbial activity, seriously threatening the soil ecosystem function.

Effects of heavy metals on soil enzyme activities

As the concentration of heavy metals increases, the activity of most enzymes is significantly reduced and the decrease in their activity may be caused directly by the interaction between the enzyme and the heavy metals, which is not associated with a reduction in microbes (Shrivastava *et al.*, 2015). Heavy metals have a significant effect on soil enzyme activity. On the one hand, heavy metals have a direct effect on soil enzyme activity, so that the spatial structure of the active groups of the enzyme is destroyed. On the other hand, the growth and reproduction of microorganisms are inhibited, thus reducing the synthesis and metabolism of the microbial enzyme. There is a very close relationship between soil enzymes and soil microbes, and some microorganisms and enzymes secreted by microorganisms participate in the circulation of soil ecosystems and energy together.

Effects of heavy metals on soil microbial community composition

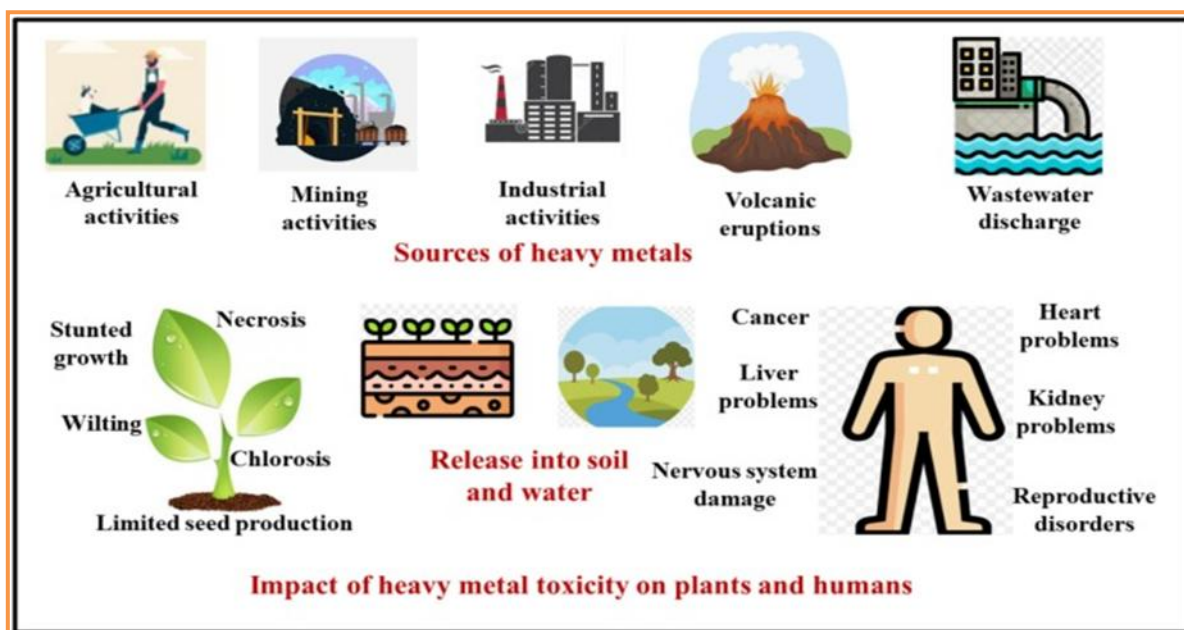
After the heavy metal into the soil, the primary impact is the amount of soil bacteria, fungi, actinomycetes and other microbial population. Heavy metals effect the microbial quality and quantity in the soil and at the same time, soil microbes on the heavy metal compounds with the reaction to the decomposition and the conversion. For example, Hg in the environment, there are a variety of valence (elemental mercury, inorganic Hg²⁺, organic mercury compounds), organic synthesis of mercury in addition to artificial synthesis, some bacteria also have the ability to synthesize organic mercury. Heavy metal contamination can produce different microbial community patterns. Even if many of the chemical and biological properties of the soil have changed greatly, there are many original microorganisms in the soil that are present in the microbial community .Long-term heavy metal contaminated soil will choose those who can specifically adapt to polluted soil microbial population. The higher the content of organic carbon in severely polluted soils, the lower the efficiency of microbial populations in organic mineralization. This can be a simple indication of the impact of heavy metal pollution on soil microbial communities (Kozdroj and Jdvan, 2001).

Extracellular polymeric substances

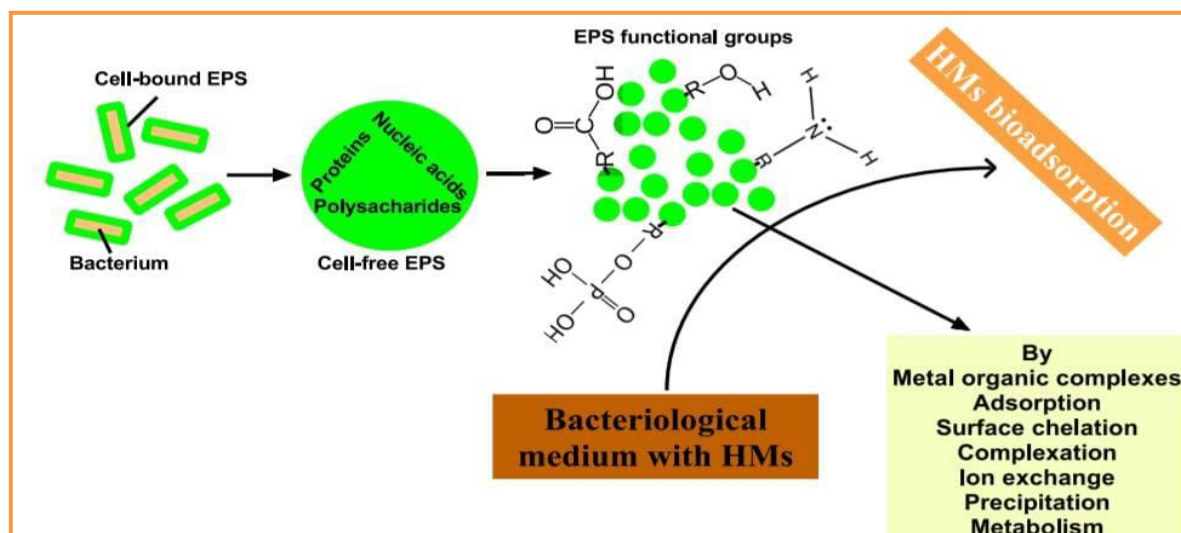
Extracellular polysaccharides, also known as EPS, are high molecular weight bacterial byproducts that have various application. Exopolysaccharides are various macromolecules found in microbes and are composed of various macromolecules. They have a crucial part in biofilm generation and the existence of microbes. To extract EPS safely and efficiently, various techniques have been developed. EPS is a lucrative and sustainable substitute for conventional methods for removing heavy metal impurities. EPS is acidic due to the presence of organic, and inorganic components, which make it capable of tolerating heavy metal stress. EPS has multiple functions such as promoting cell attachment, having a long-lasting biofilm structure, and water retention. Eradication of metal varies from capsular polysaccharides to mucoid polysaccharides released by bacteria have greater elimination (Wei *et al.*, 2017)

Mechanism of extracellular polymeric substance-based bioremediation

- ✓ **Adsorption:** Extracellular Polymeric Substances (EPS) adsorb heavy metals primarily through passive, non-metabolic processes involving electrostatic attraction, complexation, and ion exchange. Negatively charged functional groups—such as carboxylic (–COOH), phosphoric (–PO₄³⁻), hydroxyl (–OH) and amino (–NH₂) groups in EPS proteins and polysaccharides—bind positively charged metal cations like Pb²⁺, Cu²⁺, Zn²⁺, and Cd²⁺



- ✓ **Electrostatic Interaction:** Negatively charged functional groups on the EPS surface strongly attract cationic heavy metals.
- ✓ **Complexation:** Metal ions form complexes with active sites, primarily carboxyl and hydroxyl groups within the EPS structure.
- ✓ **Ion Exchange:** Metal ions replace lighter metal ions or protons previously associated with the acidic functional groups in the EPS.
- ✓ **Surface Precipitation:** The metal ions are trapped and accumulated within the matrix of the extracellular polymer EPS can create a microenvironment that promotes the precipitation of heavy metals, often mediated by sulfhydryl groups or as metal sulfides.



Conclusion

- Bacterial extracellular polymeric substances can effectively remove and immobilize heavy metals from contaminated soils.
- Mechanisms include complexation, ion exchange, precipitation and redox reactions. extracellular polymeric substances enhance microbial survival and improve soil microbial community structure.
- Microbial extracellular polymeric substances is an environmentally friendly, effective and cost-efficient approach for immobilizing and removing heavy metals from wastewater and soil.

- Integration with phytoremediation, nanotechnology, recombinant DNA and EPS modification can further increase heavy metal detoxification and scalability of remediation strategies.

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