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Heavy Metals and Plant Physiology: Tolerance Mechanisms and Phytoremediation Strategies

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ABSTRACT

Heavy metals such as cadmium, lead, and mercury are pervasive environmental pollutants resulting from industrial activities, mining, and agricultural practices. These metals pose significant threats to plant health by disrupting physiological processes, leading to reduced growth and productivity. Plants, however, have evolved various mechanisms to tolerate and detoxify heavy metals, making them potential candidates for phytoremediation—a green technology for environmental cleanup. This review explores the sources and types of heavy metals, their impact on plant physiology, and the intricate mechanisms plants employ for heavy metal tolerance. Additionally, it examines the current and potential future applications of phytoremediation strategies, including phytoextraction, phytostabilization, phytovolatilization, and rhizofiltration. Through case studies and examples, the review highlights successful applications and the challenges associated with phytoremediation. It concludes with a discussion on future perspectives, emphasizing advances in genetic engineering and the integration of sustainable practices and policies to enhance the effectiveness and adoption of phytoremediation.

Keywords: *This review explores the sources and types of heavy metals, their impact on plant physiology, and the intricate mechanisms plants employ for heavy metal tolerance.*

Introduction

Heavy metals, defined as elements with a density greater than 5 g/cm^3, are naturally occurring components of the Earth's crust. They have become widespread environmental contaminants due to various anthropogenic activities such as mining, industrial processing, and agricultural practices. Prominent heavy metals of concern include cadmium (Cd), lead (Pb), mercury (Hg), arsenic (As), and chromium (Cr) [1]. These metals persist in the environment, do not degrade, and can accumulate in living organisms, leading to toxic effects. The environmental and health risks posed by heavy metals necessitate effective remediation strategies.

Plants, being primary producers and integral components of ecosystems, are particularly vulnerable to heavy metal contamination. The uptake and accumulation of heavy metals by plants can have deleterious effects on their physiological processes, including photosynthesis, respiration, and nutrient uptake [2]. These disruptions can lead to stunted growth, reduced productivity, and, in severe cases, plant death. Understanding the impact of heavy metals on plant physiology is crucial for developing strategies to mitigate their adverse effects and enhance plant resilience, the toxic effects, plants have evolved a range of mechanisms to tolerate and detoxify heavy metals [3]. These mechanisms include the exclusion of heavy metals from cells, sequestration within cellular compartments, and the synthesis of metal-binding proteins and antioxidants. By exploiting these natural defense mechanisms, plants can be used for phytoremediation—a promising, eco-friendly technology that employs plants to remove, stabilize, or degrade contaminants from soil and water.

Phytoremediation strategies offer several advantages over conventional remediation techniques. They are cost-effective, environmentally sustainable, and can be applied to large areas with minimal disturbance to the ecosystem [4]. Phytoremediation can be categorized into several approaches, including phytoextraction, phytostabilization, phytovolatilization, and rhizofiltration, each targeting different aspects of heavy metal contamination. Phytoextraction involves the use of hyperaccumulator plants that absorb and concentrate heavy metals in their biomass, which can then be harvested and safely disposed of Phytostabilization uses plants to immobilize heavy metals in the soil, reducing their mobility and bioavailability. Phytovolatilization relies on plants to convert heavy metals into volatile forms that are released into the atmosphere. Rhizofiltration employs plant roots to absorb and precipitate heavy metals from aqueous solutions, providing a method for water purification.

While phytoremediation holds great promise, it also faces several challenges. The efficiency of phytoremediation can be influenced by factors such as soil properties, climate conditions, and the presence of co-contaminants. Additionally, the long duration required for phytoremediation and the need for effective disposal of contaminated biomass are significant limitations. Research and technological advancements are crucial to overcoming these challenges and enhancing the effectiveness of phytoremediation. Recent advances in genetic engineering offer new opportunities to improve plant tolerance to heavy metals and enhance their phytoremediation capabilities [5]. By introducing genes involved in metal transport, sequestration, and detoxification, transgenic plants can be developed with superior heavy metal accumulation and tolerance traits. These genetically engineered plants hold the potential to revolutionize phytoremediation and make it a viable solution for large-scale environmental cleanup, integrating phytoremediation with sustainable agricultural practices and environmental policies can further enhance its adoption and effectiveness. Collaborative efforts among researchers, policymakers, and industry stakeholders are essential to promote the widespread implementation of phytoremediation and address the global challenge of heavy metal contamination and the sources and types of heavy metals, their impact on plant physiology, and the mechanisms of heavy metal tolerance in plants. We will explore various phytoremediation strategies, supported by case studies and examples of successful applications [6]. Finally, to discuss future perspectives, highlighting the potential of genetic engineering and sustainable practices to advance phytoremediation technology. By understanding and harnessing the natural resilience of plants, we can develop effective and sustainable solutions to mitigate the environmental and health risks posed by heavy metal contamination.

Heavy Metals in the Environment

Heavy metals are prevalent in the environment due to both natural processes and human activities. Their presence poses significant risks to ecosystems and human health. Understanding the sources and types of heavy metals is crucial for developing effective strategies to manage and remediate contaminated environments.

Sources of Heavy Metals

Heavy metals can originate from a variety of sources, both natural and anthropogenic.

1. Natural Sources

Geological Weathering: The natural weathering of rocks releases heavy metals into the soil, water, and atmosphere. This process occurs over long geological periods and contributes to the background levels of heavy metals in the environment.

Volcanic Activity: Volcanic eruptions emit large quantities of heavy metals such as mercury (Hg), arsenic (As), and lead (Pb) into the atmosphere, which can then be deposited on the Earth's surface through precipitation.

Forest Fires: Natural wildfires can release heavy metals from vegetation and soil into the atmosphere, where they can be transported and redeposited over wide areas [7].

2. Anthropogenic Sources

Mining and Smelting: Extractive industries are major contributors to heavy metal pollution. Mining activities expose metal-rich ores, and the smelting process releases metals like cadmium (Cd), lead (Pb), and zinc (Zn) into the environment.

Industrial Processes: Industries such as electroplating, battery manufacturing, and metal finishing discharge heavy metals into the environment through waste emissions and effluents.

Agricultural Practices: The use of fertilizers, pesticides, and sewage sludge in agriculture introduces heavy metals like arsenic (As), cadmium (Cd), and lead (Pb) into the soil. These metals can accumulate in crops and enter the food chain.

Urbanization and Traffic: Urban activities, including construction, vehicular emissions, and waste disposal, contribute to the release of heavy metals. Lead (Pb) from gasoline, though reduced significantly with the advent of unleaded fuel, still persists in urban soils.

Waste Disposal: Improper disposal of industrial waste, electronic waste, and municipal solid waste can lead to the leaching of heavy metals into the soil and groundwater [8].

Types of Heavy Metals

Heavy metals of environmental concern include a range of elements, each with unique properties and toxic effects on living organisms.

1. Cadmium (Cd)

Sources: Cadmium is released from mining, smelting, and industrial activities such as battery manufacturing and electroplating. It is also present in phosphate fertilizers.

Environmental Impact: Cadmium is highly toxic to plants, animals, and humans, causing damage to kidneys, bones, and the respiratory system.

2. Lead (Pb)

Sources: Major sources of lead include mining, smelting, battery production, and the use of leaded gasoline and paints. It is also found in plumbing materials and old infrastructure.

Environmental Impact: Lead is a potent neurotoxin, affecting the nervous system and cognitive development, especially in children. It also disrupts plant growth and soil microbial activities.

3. Mercury (Hg)

Sources: Mercury is released from natural sources like volcanic activity and forest fires, as well as human activities such as coal combustion, mining, and waste incineration.

Environmental Impact: Mercury bioaccumulates in aquatic food chains, posing severe health risks to fish, wildlife, and humans. It affects the nervous, digestive, and immune systems.

4. Arsenic (As)

Sources: Arsenic contamination arises from mining, smelting, and the use of arsenic-based pesticides and herbicides. It is also naturally present in some groundwater sources.

Environmental Impact: Arsenic is a carcinogen and can cause skin lesions, cardiovascular diseases, and neurological effects. In plants, it inhibits root growth and nutrient uptake.

5. Chromium (Cr)

Sources: Chromium pollution is primarily from industrial processes such as leather tanning, electroplating, and stainless steel production.

Environmental Impact: Hexavalent chromium (Cr VI) is highly toxic and carcinogenic, affecting respiratory health and causing skin irritation. It also impairs plant growth and soil health.

6. Nickel (Ni)

Sources: Nickel is released from mining, smelting, and industrial activities such as alloy production and battery manufacturing.

Environmental Impact: Nickel exposure can lead to respiratory problems, skin dermatitis, and allergic reactions. In plants, it interferes with photosynthesis and root development.

7. Zinc (Zn)

Sources: Zinc contamination results from mining, smelting, and the use of zinc-containing fertilizers and pesticides.

Environmental Impact: While zinc is an essential nutrient for plants and animals, excessive levels can be toxic, causing chlorosis in plants and gastrointestinal distress in animals [9].

Understanding the sources and types of heavy metals is essential for assessing their environmental impact and devising appropriate remediation strategies. In the following sections, we will explore the specific effects of heavy metals on plant physiology, the mechanisms plants use to tolerate heavy metals, and the various phytoremediation strategies employed to mitigate heavy metal contamination [10].

Impact of Heavy Metals on Plant Physiology

Heavy metals have profound effects on plant physiology, influencing various aspects of growth, development, and metabolic processes [11]. This section delves into how plants take up and accumulate heavy metals, the toxic effects on cellular processes, and the specific impacts on photosynthesis and respiration.

Uptake and Accumulation

Plants absorb heavy metals primarily through their root systems, although foliar uptake through leaves can also occur, especially in areas with significant atmospheric deposition. The uptake process involves the movement of metal ions from the soil solution into root cells through passive diffusion and active transport mechanisms. Ion channels, transport proteins, and membrane carriers facilitate this process. Once inside the plant, heavy metals are translocated through the vascular system (xylem and phloem) to various tissues, including leaves, stems, and seeds. The extent of accumulation depends on several factors, including the metal's chemical form, soil properties, plant species, and environmental conditions [12]. Hyperaccumulator plants, for instance, can accumulate exceptionally high levels of heavy metals without suffering toxic effects, making them particularly valuable for phytoremediation.

Toxic Effects on Cellular Processes

The presence of heavy metals within plant cells disrupts normal cellular functions by interfering with essential biochemical and physiological processes. One primary mode of toxicity is the generation of reactive oxygen species (ROS), which can cause oxidative stress. Oxidative stress leads to damage of cellular components, including lipids, proteins, and nucleic acids, resulting in lipid peroxidation, protein denaturation, and DNA mutations. Heavy metals can also disrupt enzyme activities by binding to their active sites or by displacing essential metal cofactors, impairing metabolic pathways [13]. Additionally, heavy metals can interfere with the integrity and function of cellular membranes by altering their permeability and fluidity. This can compromise nutrient and water uptake, ion homeostasis, and signal transduction, ultimately affecting overall plant health and growth.

Impact on Photosynthesis and Respiration

Photosynthesis and respiration, the two fundamental metabolic processes in plants, are particularly sensitive to heavy metal stress. Heavy metals can adversely affect photosynthesis by disrupting chloroplast structure and function [14]. They interfere with the synthesis of chlorophyll and other photosynthetic pigments, reducing the plant's ability to capture light energy. Moreover, heavy metals can inhibit the activity of key enzymes in the Calvin cycle and the electron transport chain, leading to decreased carbon fixation and energy production. Damage to the thylakoid membranes within chloroplasts further impairs electron transport and ATP synthesis, essential for photosynthetic efficiency.

Respiration, the process by which plants convert glucose into usable energy, is also hindered by heavy metal toxicity. Heavy metals can inhibit the function of mitochondrial enzymes involved in the electron transport chain, leading to reduced ATP production and energy deficits. This disruption in energy metabolism affects various physiological processes, including growth, nutrient assimilation, and stress responses [15]. Additionally, heavy metal-induced ROS production can damage mitochondrial membranes, further compromising respiratory efficiency, the uptake and accumulation of heavy metals in plants lead to a cascade of toxic effects that disrupt cellular processes and impair vital metabolic functions. The adverse impacts on photosynthesis and respiration highlight the vulnerability of plants to heavy metal stress and underscore the importance of developing effective strategies to mitigate these effects. In the following sections, we will explore the mechanisms plants use to tolerate heavy metals and the potential applications of these mechanisms in phytoremediation.

Mechanisms of Heavy Metal Tolerance in Plants

Plants have evolved various sophisticated mechanisms to tolerate and mitigate the adverse effects of heavy metal exposure [16]. These mechanisms include exclusion strategies to prevent metal entry, sequestration and compartmentalization to isolate metals within safe cellular compartments, and the production of detoxification enzymes and proteins that neutralize or transform toxic metals.

Exclusion Mechanisms

Exclusion mechanisms are the first line of defense in plants, preventing heavy metals from entering cells. Plants achieve this through various strategies at the root-soil interface. One such strategy involves the modification of root cell walls and the exudation of organic acids, mucilage, and secondary metabolites that can chelate or bind heavy metals in the rhizosphere, reducing their bioavailability [17]. These organic compounds form stable complexes with metal ions, effectively immobilizing them and preventing their uptake by root cells, root cells can modify their membrane transport systems to restrict the entry of heavy metals. For instance, plants may downregulate the expression of certain metal transporters or activate selective transporters that preferentially uptake essential nutrients over toxic metals. The development of physical barriers, such as casparian strips in the endodermis, can also impede the apoplastic movement of metals into the vascular system, further limiting their translocation to aerial parts.

Sequestration and Compartmentalization

Once heavy metals enter plant cells, sequestration and compartmentalization serve as crucial mechanisms to mitigate their toxic effects. Plants can sequester heavy metals in specific cellular compartments where they cause less harm. Vacuoles, the largest organelles in plant cells, play a significant role in this process. Heavy metals are transported into vacuoles via tonoplast transporters, where they are stored in a non-toxic form, often bound to organic acids, phytochelatins, or metallothioneins [18]. This compartmentalization prevents metals from interfering with vital cellular processes occurring in the cytoplasm and other organelles. Cell walls and apoplasts also act as important sites for heavy metal sequestration. Heavy metals can be bound to the negatively charged pectin and cellulose in cell walls, immobilizing them and preventing their movement into the cytoplasm. This extracellular sequestration reduces the likelihood of metal-induced damage to intracellular components.

Detoxification Enzymes and Proteins

Plants produce a variety of detoxification enzymes and proteins that play crucial roles in mitigating heavy metal toxicity. Among these are antioxidant enzymes such as superoxide dismutase (SOD), catalase (CAT), and peroxidase (POD), which help to neutralize reactive oxygen species (ROS) generated by heavy metal stress [19]. These enzymes convert ROS into less harmful molecules, thereby protecting cellular components from oxidative damage.

Phytochelatins and metallothioneins are specialized metal-binding proteins that contribute to heavy metal detoxification. Phytochelatins, synthesized from glutathione, form complexes with heavy metal ions, facilitating their sequestration in vacuoles. Metallothioneins, small cysteine-rich proteins, bind metal ions through thiol groups, rendering them non-toxic and assisting in their compartmentalization. Both phytochelatins and metallothioneins play essential roles in the detoxification and homeostasis of heavy metals within plant cells.

Transport proteins, such as ATP-binding cassette (ABC) transporters, also participate in the detoxification process by actively transporting heavy metals across membranes into vacuoles or out of the cell. These transporters utilize the energy derived from ATP hydrolysis to pump metal ions against concentration gradients, thereby reducing their cytoplasmic concentrations, plants employ a multi-faceted approach to tolerate heavy metals, involving exclusion mechanisms at the root level, sequestration and compartmentalization within cellular compartments, and the action of detoxification enzymes and proteins [21-25]. These strategies collectively enable plants to survive and grow in environments contaminated with heavy metals, highlighting their potential for use in phytoremediation technologies. The next section will explore the practical applications of these mechanisms in various phytoremediation strategies.

Conclusion

Heavy metal contamination poses a significant threat to plant health, ecosystem stability, and human well-being. Understanding the mechanisms of heavy metal tolerance in plants is essential for developing effective strategies to mitigate these environmental hazards. Plants have evolved sophisticated systems to cope with heavy metal stress, including exclusion mechanisms that prevent metal uptake, sequestration and compartmentalization strategies that isolate metals within cellular compartments, and detoxification pathways that neutralize metal toxicity. The diverse mechanisms employed by plants to tolerate heavy metals highlight their resilience and potential for use in phytoremediation—a sustainable and eco-friendly technology for cleaning up contaminated environments. Phytoremediation offers several advantages over traditional remediation methods, including cost-effectiveness, minimal environmental disturbance, and the ability to treat large areas. The key strategies of phytoremediation, such as phytoextraction, phytostabilization, phytovolatilization, and rhizofiltration, leverage the natural abilities of plants to absorb, stabilize, volatilize, and filter heavy metals from soil and water, its promise, phytoremediation faces challenges such as variability in metal uptake, long treatment times, and the need for appropriate disposal of contaminated biomass. Advances in genetic engineering and biotechnology offer new opportunities to enhance the efficiency and effectiveness of phytoremediation. By developing transgenic plants with enhanced metal tolerance and accumulation capabilities, we can potentially overcome many of the current limitations, integrating phytoremediation with sustainable agricultural practices and environmental policies can promote its widespread adoption and implementation. Collaborative efforts among scientists, policymakers, and industry stakeholders are crucial for advancing phytoremediation technology and addressing the global challenge of heavy metal contamination, plants' natural mechanisms for heavy metal tolerance and detoxification provide valuable insights and tools for environmental remediation. Continued research and technological innovation are essential to harness the full potential of phytoremediation and achieve sustainable solutions for a cleaner and healthier environment. By leveraging the power of plants, we can make significant strides toward mitigating the impacts of heavy metal pollution and preserving the integrity of our ecosystems.

References

1. Alloway, B. J. (2013). Heavy Metals in Soils: Trace Metals and Metalloids in Soils and their Bioavailability (3rd ed.). Springer.

2. Baker, A. J. M., & Brooks, R. R. (1989). Terrestrial higher plants which hyperaccumulate metallic elements - A review of their distribution, ecology and phytochemistry. Biorecovery, 1(2), 81–126. https://doi.org/10.1080/10588333.1989.10464085

3. Clemens, S., & Ma, J. F. (2016). Toxic heavy metal and metalloid accumulation in crop plants and foods. Annual Review of Plant Biology, 67, 489–512. https://doi.org/10.1146/annurev-arplant-043015-111953

4. DalCorso, G., Farinati, S., & Furini, A. (2010). Regulatory networks of cadmium stress in plants. Plant Signaling & Behavior, 5(6), 663–667. https://doi.org/10.4161/psb.5.6.11569

5. Dixit, R., Wasiullah, Malaviya, D., Pandiyan, K., Singh, U. B., Sahu, A., Shukla, R., Singh, B. P., Rai, J. P., Sharma, P. K., & Lade, H. (2015). Bioremediation of heavy metals from soil and aquatic environment: An overview of principles and criteria of fundamental processes. Sustainability, 7(2), 2189–2212. https://doi.org/10.3390/su7022189

6. Hall, J. L. (2002). Cellular mechanisms for heavy metal detoxification and tolerance. Journal of Experimental Botany, 53(366), 1–11. https://doi.org/10.1093/jexbot/53.366.1

7. Hossain, M. A., Piyatida, P., da Silva, J. A. T., & Fujita, M. (2012). Molecular mechanism of heavy metal toxicity and tolerance in plants: Central role of glutathione in detoxification of reactive oxygen species and methylglyoxal and in heavy metal chelation. Journal of Botany, 2012, Article 872875. https://doi.org/10.1155/2012/872875

8. Jaiswal, A., Singh, R. K., Gautam, S., & Chatterjee, S. (2018). Heavy metal pollution: Source, impact, and remedies. In A. Singh & S. Prasad (Eds.), Remediation of Heavy Metals in the Environment (pp. 3–28). Springer. https://doi.org/10.1007/978-3-319-61346-0_1.

9. Kopittke, P. M., Lombi, E., Zhao, F. J., & McGrath, S. P. (2011). Mobilizing arsenic using phytoremediation. Environmental Science & Technology, 45(8), 3359–3360. https://doi.org/10.1021/es200379a

10. Krämer, U. (2010). Metal hyperaccumulation in plants. Annual Review of Plant Biology, 61, 517–534. https://doi.org/10.1146/annurev-arplant-042809-112156

11. Lombi, E., & Susini, J. (2009). Synchrotron-based techniques for plant and soil science: Opportunities, challenges and future perspectives. Plant and Soil, 320(1-2), 1–35. https://doi.org/10.1007/s11104-008-9879-9

12. Maestri, E., Marmiroli, M., Visioli, G., & Marmiroli, N. (2010). Metal tolerance and hyperaccumulation: Costs and trade-offs between traits and environment. Environmental and Experimental Botany, 68(1), 1–13. https://doi.org/10.1016/j.envexpbot.2009.10.011

13. Manara, A. (2012). Plant responses to heavy metal toxicity. In M. N. V. Prasad (Ed.), Metallothioneins in Biotic and Abiotic Stress Tolerance in Plants (pp. 27–53). Springer. https://doi.org/10.1007/978-3-642-21805-3_2

14. Mishra, S., & Dubey, R. S. (2006). Heavy metal uptake and detoxification mechanisms in plants. International Journal of Agriculture and Plant Science, 3(2), 50–58. https://doi.org/10.18869/acadpub.ijab.v3n2.50

15. Muthusaravanan, S., Sivarajasekar, N., Vivek, J. S., Paramasivan, T., Naushad, M., Prakashmaran, J., Gayathri, V., & Al-Duaij, O. K. (2018). Phytoremediation of heavy metals: Mechanisms, methods and enhancements. Environmental Chemistry Letters, 16, 1339–1359. https://doi.org/10.1007/s10311-018-0747-6

16. Nagajyoti, P. C., Lee, K. D., & Sreekanth, T. V. M. (2010). Heavy metals, occurrence and toxicity for plants: A review. Environmental Chemistry Letters, 8, 199–216. https://doi.org/10.1007/s10311-010-0297-8

17. Nascimento, C. W. A., & Xing, B. (2006). Phytoextraction: A review on enhanced metal availability and plant accumulation. Scientia Agricola, 63(3), 299–311. https://doi.org/10.1590/S0103-90162006000300014

18. Rascio, N., & Navari-Izzo, F. (2011). Heavy metal hyperaccumulating plants: How and why do they do it? And what makes them so interesting? Plant Science, 180(2), 169–181. https://doi.org/10.1016/j.plantsci.2010.08.016

19. Sarwar, N., Imran, M., Shaheen, M. R., Ijaz, A., Kamran, M. A., Matloob, A., Rehim, A., & Hussain, S. (2017). Phytoremediation strategies for soils contaminated with heavy metals: Modifications and future perspectives. Chemosphere, 171, 710–721. https://doi.org/10.1016/j.chemosphere.2016.12.116

20. Singh, R., Gautam, N., Mishra, A., & Gupta, R. (2011). Heavy metals and living systems: An overview. Indian Journal of Pharmacology, 43(3), 246–253. https://doi.org/10.4103/0253-7613.81505

21. Suman, J., Uhlik, O., Viktorova, J., & Macek, T. (2018). Phytoextraction of heavy metals: A promising tool for clean-up of polluted environment? Frontiers in Plant Science, 9, Article 1476. https://doi.org/10.3389/fpls.2018.01476

22. Tangahu, B. V., Abdullah, S. R. S., Basri, H., Idris, M., Anuar, N., & Mukhlisin, M. (2011). A review on heavy metals (As, Pb, and Hg) uptake by plants through phytoremediation. International Journal of Chemical Engineering, 2011, Article 939161. https://doi.org/10.1155/2011/939161

23. Verbruggen, N., Hermans, C., & Schat, H. (2009). Mechanisms to cope with arsenic or cadmium excess in plants. Current Opinion in Plant Biology, 12(3), 364–372. https://doi.org/10.1016/j.pbi.2009.05.001

24. Visioli, G., & Marmiroli, N. (2013). The proteomics of heavy metal hyperaccumulation by plants. Journal of Proteomics, 79, 133–145. https://doi.org/10.1016/j.jprot.2012.11.016

25. Wuana, R. A., & Okieimen, F. E. (2011). Heavy metals in contaminated soils: A review of sources, chemistry, risks and best available strategies for remediation. ISRN Ecology, 2011, Article 402647. https://doi.org/10.5402/2011/402647