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Lead (Pb) Toxicity in Crop Plants: Mechanisms of Damage, Tolerance, and Remediation Strategies

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Abstract: Lead (Pb) is a non-essential heavy metal that is highly toxic and is continually being added to agricultural soils from mining, smelting, industrial emissions, sewage sludge, phosphate fertilizers, paint residues and vehicle-related deposition. In crop plants, Pb entry occurs mainly through roots and it is tightly retained in root tissues but a small fraction can be translocated to shoots and edible organs. Pb disrupts seed germination, root architecture, water relations, mineral nutrition, photosynthesis, carbohydrate metabolism and reproductive development. At cellular level, Pb induces oxidative stress via overproduction of reactive oxygen species (ROS), disrupts membranes, proteins, nucleic acids and chloroplasts and disturbs essential metal homeostasis and enzyme function. Plants react to Pb stress by exclusion in the rhizosphere, binding to cell walls, efflux at the plasma membrane, chelation with organic acids and thiol compounds, detoxification mediated by phytochelatins and glutathione, sequestration in vacuoles and activation of enzymatic and non-enzymatic antioxidants. Remediation needs source control and risk-based soil management. In situ immobilization using lime, phosphate, biochar, compost, iron oxides, and other amendments can reduce Pb bioavailability and crop uptake. Phytoextraction, phytostabilization, microbe-assisted remediation, and chelate-assisted approaches provide biological options with a range of trade-offs. Pb has low mobility and limited phytoextractability. The most reliable approach to protect crop productivity and human health is through integrated strategies such as contamination assessment, amendment-based stabilization, tolerant crop selection, food-safety monitoring, and, when necessary, excavation or capping.

Keywords: Lead, Heavy-Metal Stress, Oxidative Stress, Phytoremediation, Biochar, Soil Amendments, Food Safety.

Review Paper

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1. INTRODUCTION

Lead is one of the most ubiquitous and persistent inorganic pollutants in agricultural environments. Unlike essential micronutrients such as Zn, Cu, Fe, Mn and Ni, Pb has no beneficial role in plant metabolism and even low internal concentrations of Pb can inhibit growth and development (Kabata-Pendias, 2011; Nagajyoti *et al.,* 2010). Peri-urban and industrial farming systems, old mining districts, roadside fields and areas receiving untreated waste water or industrial effluents are of particular concern for Pb contamination. Pb is persistent in soil due to its stable complex formation with organic matter, clay minerals, iron and manganese oxides and phosphate compounds, leading to long residence times and continued exposure risks (Alloway, 2013; Shahid *et al*., 2012).

Crop plants represent an important pathway for the transfer of soil Pb into the human food chain. The majority of Pb taken up by plants is stored in the roots, but depending on soil chemistry, plant species, cultivar and management practices, Pb can be translocated to leaves, grains, tubers and other edible plant tissues (Islam *et al.,* 2007; Pourrut *et al*., 2011). Humans are exposed by consuming contaminated vegetables and grains, direct ingestion of soil, and inhalation of dust and children are particularly vulnerable to neurodevelopmental effects (Nriagu, 1996).

The degree of Pb toxicity to crops is influenced by total and bioavailable Pb in the soil, pH, organic matter, cation exchange capacity, phosphorus status, redox status, microbial activity and presence of competing ions such as Ca, Zn, Fe and phosphate (Shahid *et al*., 2012; Kabata-Pendias, 2011). Pb toxicity is also greatly dependent on time and concentration: short-term exposure can reduce germination and root elongation, whereas long-term exposure may inhibit biomass, photosynthesis, nutrient uptake and yield (Kopittke *et al*., 2007; Sharma and Dubey, 2005).

This review covers (i) major sources and behavior of Pb in agricultural soils; (ii) Pb uptake, transport and compartmentalization in crop plants; (iii) physiological, biochemical and molecular mechanisms of Pb toxicity; (iv) plant tolerance and detoxification mechanisms; and (v) physical, chemical and biological remediation strategies. The goal is to create an integrated management strategy for Pb-contaminated croplands, ensuring both crop productivity and food safety.

2. Sources and Soil Behavior of Lead

2.1 Sources of Anthropogenic and Geologic Origin

Pb enters agricultural systems from natural and anthropogenic sources. Geogenic Pb originates from weathering of Pb-bearing minerals but anthropogenic inputs dominate in most of the contaminated farmlands (Alloway, 2013; Nriagu, 1996). Major sources are mining and ore-processing operations, smelting, battery production, metal recycling, coal combustion, past use of leaded gasoline, lead-based paints, irrigation with contaminated wastewater, sewage sludge, some phosphate fertilizers and atmospheric deposition from industrial emissions and traffic (Alloway, 2013; Kabata-Pendias, 2011). Lead contamination in mining landscapes is frequently highly heterogeneous and severe, with food crops taking up Pb and posing long-term exposure risks to local communities (Nriagu, 1996; Islam *et al*., 2007).

2.2 Pb Fractions and Bioavailability

Total soil Pb concentration is not a good predictor for plant uptake. Pb is found in operationally defined fractions: water-soluble and exchangeable Pb, carbonate-bound Pb, Fe–Mn oxide-bound Pb, organic-matter-bound Pb and residual Pb in mineral lattices (Shahid *et al*., 2012; Kabata-Pendias, 2011). The exchangeable and loosely adsorbed fractions are generally the most bioavailable to plants and soil organisms, while residual Pb is relatively inert.Pb solubility is mainly governed by soil pH. Acidic soils increase Pb mobility and availability to plants, while liming increases pH, enhances adsorption and precipitation, and generally decreases Pb uptake (Kabata-Pendias, 2011; Shahid *et al*., 2012). Phosphate amendments result in the precipitation of insoluble lead phosphates such as pyromorphite-like minerals, thus reducing bioavailable Pb (McGrath and Zhao, 2003). Organic matter can immobilize Pb by complexation or, in the presence of a high abundance of decomposing organic acids, can temporarily increase mobility (Shahid *et al*., 2015). Pb is also adsorbed by oxides of iron and manganese, clay minerals and biochar, thereby lowering its concentration in solution (Alloway, 2013). Pb is tightly held by soil constituents and its vertical mobility is generally limited. However, erosion, dust and surface runoff and transport of contaminated particles can redistribute Pb across landscapes and into the water bodies (Alloway, 2013; Shahid *et al*., 2012).

3. Lead Uptake, Transport, and Distribution in Crop Plants

3.1 Root Uptake Pathways

Pb is mainly taken up by the roots via apoplastic and symplastic pathways. In the apoplastic pathway Pb travels through cell walls and intercellular spaces until it reaches the endodermis, where the Casparian strip blocks further passive movement. In the symplastic route, Pb crosses plasma membranes and enters the cytoplasm, often via transport systems or channels that are normally involved in the uptake of essential cations (Pourrut *et al*., 2011; Seregin and Ivanov, 2001). Mechanisms proposed for Pb uptake into root cells include nonselective cation channels, Ca^{2+} channels and other cation-permeable pathways, aquaporin-mediated pathways for Pb complexes, endocytosis of Pb-bound molecules and carrier-mediated transport involving proteins that normally transport divalent cations (Clemens, 2001; Pourrut *et al*., 2011). Pb uptake depends on rhizosphere pH, root exudates, microbial activity, Ca^{2+} concentration, phosphate availability and the speciation of Pb in the soil solution. Pb can be chelated by root exudates such as citrate, malate, oxalate and amino acids, affecting its availability. Depending on the ligand and concentration, this can lead to a reduction in free Pb 2+ or an increase in soluble Pb complexes that can be taken up (Shahid *et al*., 2015; Pourrut *et al*., 2011).

3.2 Radial Transport and Xylem Loading

Pb has to cross the endodermis and reach the stele to be loaded into the xylem after entering the root cortex. The Casparian strip and the root apoplast are important barriers. Thus, Pb accumulation is usually greatest in the root epidermis, cortex and cell walls, with concentrations much lower in the stems, leaves and seeds (Seregin and Kozhevnikova, 2008; Jiang *et al.,* 2000). Transpiration-driven transport may occur, but Pb xylem loading is limited. Pb may be complexed with organic acids, amino acids, glutathione or phytochelatins in the xylem (Clemens, 2001; Hall, 2002). Phloem-mediated redistribution is usually limited, and this is why fruits and seeds generally contain less Pb than leaves or roots, though this depends on the species and the contamination scenario (Pourrut *et al.,* 2011).

3.3 Compartmentation and Tissue Distribution

Plants reduce Pb toxicity by limiting its mobility and by sequestering it in less sensitive compartments. The root cell walls, plasma membrane and apoplast, vacuoles and trichomes or epidermal tissues are the major sites of detoxification (Clemens, 2001; Hall, 2002). The low translocation factor of most crop plants is desirable for food safety, but it is a problem for phytoextraction, because successful extraction requires not only uptake, but also significant translocation of Pb to harvestable aboveground biomass (McGrath and Zhao, 2003; Lasat, 2002).

4. Mechanisms of Lead Toxicity in Crop Plants

4.1 Inhibition of Germination and Seedling Growth

Pb exposure often reduces seed germination, radicle elongation, shoot growth and seedling establishment. The early root system is especially sensitive because roots are the first organ to be in direct contact with soil Pb inhibits cell division and elongation in root tips, damages root cap cells, reduces mitotic activity and changes root hair formation (Kopittke *et al.,* 2007; Liu *et al.,* 2003). The reduced root growth has cascading effects: plants explore a smaller volume of soil, absorb less water and nutrients, and become more susceptible to drought and other stresses (Sharma and Dubey, 2005).

4.2 Disruption of Mineral Nutrition and Water Relations

Pb competes with essential cations for transport sites and binding domains, thereby disrupting the uptake and distribution of Ca, Mg, Zn, Fe, Mn, Cu and K. Pb is also capable of inhibiting root plasma-membrane H+-ATPase activity and thus decreasing the proton gradients required for nutrient uptake (Sharma and Dubey, 2005; Hall, 2002). Pb stress often causes reduction in leaf water potential, relative water content and stomatal conductance. Stomatal closure is a protective measure to decrease transpiration and metal uptake, but also restricts CO2 uptake and photosynthesis (Kopittke *et al.,* 2008; Ashraf and Harris, 2013).

4.3 Photosynthesis and CHLOROPLAST DAMAGE

Pb affects many processes, of which sunlight is one of the most sensitive. Pb reduces the photosynthetic performance by decrease in chlorophyll synthesis, distortion of chloroplasts and thylakoid membranes, inhibition of photosystem II activity and electron transport, reduction in carotenoid and plastoquinone levels, suppression of Calvin cycle enzymes, and reduction in stomatal conductance (Sharma and Dubey, 2005; Ashraf and Harris, 2013). These effects lead to a lower net photosynthetic rate, change in chlorophyll fluorescence parameters, lower assimilate supply and lower biomass accumulation (Kaur *et al.,* 2013; Mittler, 2002).

4.4 Oxidative Stress and ROS Production

Lead (Pb) phytotoxicity is mediated mainly by oxidative stress. Pb is not a redox-active metal like Fe or Cu, but it indirectly promotes the accumulation of reactive oxygen species including superoxide radical, hydrogen peroxide, hydroxyl radical and singlet oxygen (Gill and Tuteja, 2010; Apel and Hirt, 2004). Too much ROS causes oxidation of lipid, protein, pigment and nucleic acid. Lipid peroxidation, frequently assessed by measuring the increase in malondialdehyde content, causes membrane damage and increased electrolyte leakage (Mittler, 2002; Kaur et al., 2013).

4.5 Enzyme Inhibition and Metabolic Disruption

Pb has a high affinity for sulfhydryl groups in proteins, and can displace essential metal cofactors from enzymes. Pb accordingly inhibits enzymes of photosynthesis, respiration, nitrogen metabolism, antioxidant defense and cell wall synthesis (Sharma and Dubey, 2005; Hall, 2002). Especially important is the inhibition of antioxidant enzymes which reduces the plant's ability to detoxify ROS, although, at moderate Pb exposure, some antioxidant enzymes increase at the first instance as an adaptive response (Gill and Tuteja, 2010; Ahmad *et al.,* 2010).

4.6 Effects on Reproduction, Yield, and Grain Quality

Pb toxicity causes reduced crop yield due to reduced biomass, reduced tillers or branches, reduced flowering, poor pollen and ovule viability, reduced fruit set and smaller grains or tubers (Kopittke *et al*., 2007; Islam *et al*., 2007). Pb may also affect the grain composition including starch, protein, mineral and antioxidant profiles. However, Pb accumulation in edible tissues may make produce unsafe or unfit for market even when visible injury is modest (Islam *et al*., 2007; Nagajyoti *et al*., 2010).

5. Plant Tolerance and Detoxification Mechanisms

5.1 Avoidance and Exclusion

Avoidance is the first line of defense: plants can restrict Pb entry to roots or translocation to sensitive tissues. Such mechanisms comprise immobilization of Pb in the rhizosphere by root exudates, binding of Pb to root cell walls and mucilage, reduced permeability of the root apoplast, development of endodermal barriers, efflux of Pb from root cells, and restricted xylem loading and limited translocation to shoots (Pourrut et al., 2011; Seregin and Kozhevnikova, 2008).

5.2 Cell-Wall Binding and Apoplastic Immobilization

In cell walls, negatively charged groups such as carboxyl groups in pectins bind divalent cations such as Pb 2+. Pb can also induce callose deposition, lignification and changes in cell-wall composition,

thereby reinforcing the barrier function of the apoplast (Hall, 2002; Seregin and Ivanov, 2001). The immobilization in cell walls restricts the access of Pb to the protoplast and is considered a major tolerance mechanism (Clemens, 2001).

5.3 Chelation, Glutathione, and Phytochelatins

Upon entering the cytosol, Pb is detoxified by plants via complexation with ligands. Phytochelatins are synthesized from glutathione which binds Pb. Phytochelatin synthase polymerizes glutathione to phytochelatins that bind Pb and other soft metals in stable complexes that are transported to vacuoles, where they are stabilized and sequestered away from metabolism (Grill *et al.,* 1989; Cobbett and Goldsbrough, 2002). Organic acids, amino acids and phenolic compounds are also involved in Pb binding (Rauser, 1995; Shahid *et al.,* 2015).

5.4 Antioxidant Defense System

Plants respond to Pb-induced ROS through a coordinated antioxidant network consisting of enzymatic antioxidants such as SOD, CAT, APX, GPX, GR and GST and non-enzymatic antioxidants including ascorbate, glutathione, tocopherols, carotenoids, phenolics and flavonoids (Gill and Tuteja, 2010; Ahmad et al., 2010). Superoxide dismutase converts the superoxide into hydrogen peroxide, which is then decomposed by catalase, ascorbate peroxidase and guaiacol peroxidase (Apel and Hirt, 2004). Proline, soluble sugars and phenolic compounds can also stabilize membranes and proteins under Pb stress (Ashraf and Harris, 2013).

5.5 Molecular Regulation and Signaling

Stress caused by Pb triggers signaling cascades involving calcium, mitogen-activated protein kinases, phytohormones and transcription factors. Abscisic acid is generally increased under Pb stress and is also involved in the stomatal closure and gene expression in response to stress. Jasmonic acid, salicylic acid, ethylene and nitric oxide can also modulate antioxidant defense, metal binding and programd cell death pathways (Gill and Tuteja, 2010; Clemens, 2001). Gene families encoding metal transporters, such as the ABC, HMA, NRAMP, YSL, and CDF/MTP families, may be involved in metal homeostasis and compartmentation (Clemens, 2001; Hall, 2002).

6. Remediation of Lead-Contaminated Croplands

6.1 Principles of Remediation

Effective Pb management should be the starting point for site characterization. Total Pb, bioavailable Pb, soil pH, organic matter, texture, phosphorus status, land use, crop type, and exposure pathways (Alloway, 2013; Kabata-Pendias, 2011). The goals of remediation vary by land use. For soils used for food production, the priority is to reduce Pb transfer to edible tissues. For heavily contaminated industrial or mining sites, the priority may be containment, stabilization or removal (McGrath and Zhao, 2003; Lasat, 2002).

6.2 Excavation, Soil Replacement, and Containment

For small, heavily contaminated areas, such as former battery sites, smelter "hot spots" or residential gardens, the most reliable, but also most expensive, option is to dig up and dispose of the contaminated soil and then replace it with clean soil. Capping with clean soil, geotextile barriers, mulch or pavement can reduce direct contact and dust exposure (Alloway, 2013; McGrath and Zhao, 2003).

6.3 Chemical Immobilization and Soil Amendments

In situ immobilization does not remove the Pb, but reduces its bioavailable fraction. Common amendments include lime, phosphate materials, biochar, compost and manure, iron oxides, clay minerals and biosolids (Kabata-Pendias, 2011; Shahid *et al.,* 2012). Biochar and other organic amendments are particularly attractive as they can both immobilize Pb as well as improve soil fertility, enhance water retention and support microbial activity (Rizwan *et al.,* 2017; Rizwan *et al.,* 2016). The phosphate-based stabilization is effective because of high insolubility of Pb phosphate minerals. However, over application of phosphorus can increase the runoff and contribute to water-quality problems (McGrath and Zhao, 2003).

6.4 Phytoremediation Approaches

Phytoremediation is the use of plants and associated microbes to remove, immobilize or reduce the hazard of contaminants. The main strategies are phytoextraction, phytostabilization, rhizofiltration, phytoexclusion and assisted phytoremediation (Salt *et al.,* 1998; Raskin *et al.,* 1997). Phytoremediation of Pb with high-biomass plants such as some grasses, hemp, flax, mustard, sunflower and trees has been explored (Zaier *et al.,* 2010; Kumar *et al.,* 1995). Fiber crops such as hemp and flax are of interest because biomass can be used for non-food purposes and therefore reduce the risks to the food chain (Kumar *et al.,* 2012).

6.5 Microbe-Assisted Remediation

Plant growth-promoting rhizobacteria, arbuscular mycorrhizal fungi and other beneficial microbes can influence Pb remediation through Pb immobilization, mobilization by organic acids and siderophores, improved plant growth, enhanced antioxidant activity and alteration of rhizosphere pH and redox conditions (Gisbert *et al.,* 2003; Rizwan *et al.,* 2016). Microbial inoculants are promising since they can increase plant biomass without necessarily increasing Pb translocation to the edible tissues (Rizwan *et al.,* 2016).

6.6 Chelate-Assisted Phytoextraction

Synthetic chelators like EDTA can increase the solubility of Pb and enhance its uptake by certain plants. The chelate-assisted phytoextraction, however, has

important limitations. The chelators may reduce the plant biomass and increase the phytotoxicity and mobilize Pb downwards through the soil profile, raising the leaching and groundwater risks (Ehsan *et al*., 2014; McGrath and Zhao, 2003). Biodegradable chelators, low molecular weight organic acids and appropriate timing of applications can minimize some of these risks, but they still need site-specific consideration (Zaier *et al*., 2010).

6.7 Agronomic Management for Safer Crop Production

In cases of incomplete immediate remediation, agronomic practices can reduce transfer to the food chain: liming to raise soil pH, adequate phosphorus nutrition, increasing soil organic matter, selection of low-Pb-accumulating cultivars, avoidance of leafy vegetables and root crops in highly contaminated soils, use of raised beds with clean imported soil, washing of produce, prevention of dust and erosion, and monitoring of Pb in soil and edible produce (Islam *et al*., 2007; Kabata-Pendias, 2011). These measures do not remove Pb from soil but may substantially decrease the exposure while longer term remediation is applied (Shahid *et al.*, 2015).

7. Research Gaps and Future Directions

Several gaps exist that prevent translation of Pb-toxicity research to field practice. Further studies should link soil Pb fractions and speciation with actual Pb accumulation in specific crops under realistic field conditions (Shahid *et al.,* 2012). Breeding programs should focus on identification of crop genotypes maintaining yield with minimum Pb in edible organs (Kumar *et al.,* 2012). Research should link Pb transporters, cell-wall chemistry, thiol metabolism, hormone signaling, and antioxidant responses along the soil–root–shoot axis (Clemens, 2001; Hall, 2002). The durability of biochar, phosphate, lime and organic amendments needs to be evaluated over multiple cropping seasons (Rizwan *et al.,* 2017). Microbiome-based solutions and synthetic microbial communities may improve stabilization and crop tolerance, but field-scale reproducibility is uncertain (Gisbert *et al*., 2003). Finally, remediation studies should measure Pb in edible tissues and assess dietary exposure, not just total soil Pb or shoot concentration (Islam *et al.,* 2007).

8. CONCLUSION

Lead toxicity to crop plants is a cascade of effects starting from root uptake and ending with disrupted nutrition, water relations, photosynthesis, metabolism, growth and yield (Sharma and Dubey, 2005; Kopittke *et al*., 2007). Although Pb is not readily translocated in most crops, its accumulation in roots and in some cases edible tissues, is a serious concern for food safety (Pourrut *et al.,* 2011; Islam *et al*., 2007). Plants cope with Pb by exclusion, binding to cell wall, chelation, sequestration into vacuole and antioxidant defense but these mechanisms are not sufficient in case of high or prolonged exposure (Clemens, 2001; Gill and Tuteja, 2010). Because Pb is highly retained in soils and poorly taken up by most plants, remediation should be aimed at reducing bioavailability and exposure. The most feasible route for contaminated croplands is integrated management, i.e., source control, pH and nutrient management, clean organic amendments and biochar, phosphate-based stabilization where environmentally appropriate, tolerant crop selection, and microbial assistance (McGrath and Zhao, 2003; Rizwan *et al.,* 2017). Severe hotspots still require engineering controls or soil replacement (Alloway, 2013).

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