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Heavy metal stress in plants: a review

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Abstract

Heavy metals, such as cadmium, copper, lead, chromium and mercury are major environmental pollutants, particularly in areas with high anthropogenic pressure. Heavy metal accumulation in soils is of great concern in agricultural production due to the adverse effects on food safety and marketability, crop growth due to phytotoxicity, and environmental health of soil organisms. The influence of plants and their metabolic activities affects the geological and biological redistribution of heavy metals through pollution of the air, water and soil. A common consequence of heavy metal toxicity is the excessive accumulation of reactive oxygen species (ROS) and methylglyoxal (MG), both of which can cause peroxidation of lipids, oxidation of protein, inactivation of enzymes, DNA damage and/or interact with other vital constituents of plant cells. This review focuses on effect of heavy metals on plant growth, yield and their mode of toxic effects in plants.

INTRODUCTION

Naturally plants are exposed with many adverse environmental conditions like biotic and abiotic stress. Despite all others stresses heavy metal stress is one of great importance which has a notable adverse effects on crop productivity and growth. Heavy metal stress triggers different responses in plants, ranging from biochemical responses to crop yield. The term "heavy metals" refers to any metallic element that has a relatively high density and is toxic or poisonous even at low concentration (Lenntech Water Treatment and Air Purification, 2004). "Heavy metals" in a general collective term, applies to the group of metals and metalloids with atomic density greater than 4 g/cm^3, or 5 times or more, greater than water (Hawkes, 1997). However, chemical properties of the heavy metals are the most influencing factors compared to their density.

Heavy metals include lead (Pb), cadmium (Cd), nickel (Ni), cobalt (Co), iron (Fe), zinc (Zn), chromium (Cr), iron (Fe), arsenic (As), silver (Ag) and the platinum group elements. Heavy metals are largely found in dispersed form in rock formations. Industrialization and urbanization have increased the anthropogenic contribution of heavy metals in biosphere. Heavy metals have largest availability in soil and aquatic ecosystems and to a relatively smaller proportion in atmosphere as particulate or vapors. Heavy metal toxicity in plants varies with plant species, specific metal, concentration, chemical form and soil composition and pH, as many heavy metals are considered to be essential for plant growth. Some of these heavy metals like Cu and Zn either serve as cofactor and activators of enzyme reactions e.g., in forming enzymes/substrate metal complex (Mildvan, 1970) or exert a catalytic property such as prosthetic group in metalloproteins. These essential trace metal nutrients take part in redox reactions, electron transfer and structural functions in nucleic acid metabolism. Some of the heavy metal such as Cd, Hg and As are strongly poisonous to metal-sensitive enzymes, resulting in growth inhibition and death of organisms.

An alternative classification of metals based on their coordination chemistry, categorizes heavy metals as class B metals that come under non-essential trace elements, which are highly toxic elements such as Hg, Ag, Pb, Ni (Nieboer and Richardson, 1980). Some of these heavy metals are bioaccumulative, and they neither break down in the environment nor easily metabolized. Such metals accumulate in ecological food chain through uptake at primary producer level and then through consumption at consumer levels. Plants are stationary, and roots of a plant are the primary contact site for heavy metal ions. In aquatic systems, whole plant body is exposed to these ions. Heavy metals are also absorbed directly to the leaves due to particles deposited on the foliar surfaces.

Source of contamination

There are different sources of heavy metals in the environment such as: natural, agricultural, industrial, domestic effluent, atmospheric sources and other sources. Activities such as mining and smelting operations and agriculture have contaminated extensive areas of world such as Japan, Indonesia and China mostly by heavy metals such as Cd, Cu and Zn (Herawati et al., 2000).

Natural sources of heavy metals

Heavy metals originate within the Earth’s crust; hence their natural occurrence in soil is simply a product of weathering process. The composition and concentration of heavy metals depend on the rock type and environmental conditions, activating the weathering process. The geologic plant materials generally have high concentrations of Cr, Mn, Co, Ni, Cu, Zn, Cd, Sn, Hg and Pb. However, class-wise the heavy metal concentrations vary with in the rocks. Soil formation takes place mostly from sedimentary rock, but is only a small source of heavy metals, since it is not generally or easily weathered. However, many igneous rocks such as olivine, augite and hornblende contribute considerable amounts of Mn, Co, Ni, Cu and Zn to the soils. Within the class of sedimentary rocks, shale has highest concentrations of Cr, Mn, Co, Ni, Cu, Zn, Cd, Sn, Hg and Pb followed by limestone and sand stone. Volcanoes have been reported to emit high levels of Al, Zn, Mn, Pb, Ni, Cu and Hg along with toxic and harmful gases (Seaward and Richardson, 1990). Wind dust, which arises from desert region such as Sahara, has high levels of Fe and lesser amounts of Mn, Zn, Cr, Ni and Pb (Ross, 1994). Marine aerosols and forest fires also exert a major influence in the transport of some heavy metals in many environments. While the long range transport of dusts, particularly from the Sahara, has received considerable recent attention. Volatile heavy metals such as Hg and Se are part of carbonaceous matter produced during the fire. Natural vegetation emits heavy metals into the soil and atmosphere through leaching from leaves and stems, decomposition and volatilization. Many heavy metals have been detected in inland coastal areas due to sea sprays and aerosols produced in oceanic activities.

Agricultural sources of heavy metals

The inorganic and organic fertilizers (Fertilizer is a substance added to soil to improve plants growth and yield.) are the most important sources of heavy metals to agricultural soil include liming, sewage sludge, irrigation waters and pesticides, sources of heavy metals in the agricultural soils. Others, particularly fungicides, inorganic fertilizers and phosphate fertilizers have variable levels of Cd, Cr, Ni, Pb and Zn depending on their sources. Cadmium is of particular concern in plants since it accumulates in leaves at very high levels, which may be consumed by animals or human being. Cadmium enrichment also occurs due to the application of sewage sludge, manure and limes (Yanqun et al., 2005). Although the levels of heavy metals in agricultural soil are very small, but repeated use of phosphate fertilizer and the long persistence, time for metals, there may be dangerously high accumulation of some metals.

Animal manure enriches the soil by the addition of Mn, Zn, Cu and Co and sewage sludge by Zn, Cr, Pb, Ni, Cd and Cu (Verkleji, 1993). The increase in heavy metal contamination of agricultural soil depends on the rate of application of the contributors with its elemental concentration and soil characteristics to which it is applied. Heavy metal accumulation in soil is also due to application of soil amendments such as compost refusing and nitrate fertilizers (Ross, 1994). Liming increases the heavy metal levels in the soil more than the nitrate fertilizers and compost refuse. Sewage sludge is one of the most important sources of heavy metal contamination to the soil (Ross, 1994). Several heavy metal based pesticides are used to control the diseases of grain and fruit crops and vegetables and are sources of heavy metal pollution to the soil (Ross, 1994). The orchards where these compounds have been used frequently resulted into contamination of orchard soil with high levels of heavy metals such as Cu, As, Pb, Zn, Fe, Mn and Hg (Ross, 1994). Pesticides such as lead arsenate were used in Canadian orchards for more than six decades and were found to be enriched with Pb, As and Zn having greater consequences for food contamination. Continued irrigation of agricultural soil can lead to accumulation of heavy metals such as Pb and Cd (Ross, 1994).

Industrial sources of heavy metals

Mining operation emits different heavy metals depending on the type of mining. For example, coalmines are sources of As, Cd, Fe, etc., which enrich the soil around the coalfield directly or indirectly. The utilization of Hg in gold mining and the mobilization of significantly high amounts of Hg from old mines have become a significant source of this pollutant to the environment (Lacerda, 1997). High temperature processing of metals such as smelting and castings emit metals in particulate and vapor forms. Vapor form of heavy metals such as As, Cd, Cu, Pb, Sn and Zn combine with water in the atmosphere to form aerosols. These may be either dispersed by wind (dry deposition) or precipitated in rainfall (wet deposition) causing contamination of soil or water bodies. Contamination of soil and water bodies can also occur through runoff from erosion of mine wastes, dusts produced during the transport of crude ores, corrosion of metals and leaching of heavy metals to soil and ground water. Soil contamination of heavy metals occurs due to different types of processing in refineries. Energy-supplying power stations such as coal burning power plants, petroleum combustion, nuclear power stations and high tension lines contribute many heavy metals such as Se, B, Cd, Cu, Zn, Cs and Ni to the environment (Verkleji, 1993). Other industrial sources include processing of plastics, textiles, microelectronics, wood preservation and paper processing. Contamination of plants growing beneath the power line with high concentration of Cu is reported to be toxic to the grazing animals (Kraal and Ernst, 1976).

Domestic effluents

These waste waters probably constitute the largest single source of elevated metal values in rivers and lakes. Domestic effluents may consists of (1) untreated or solely mechanically treated waste waters (2) substances which have passed through the filters of biological treatment plants (3) waste substances passed over sewage outfalls and discharged to receiving water bodies often end up into the sea from coastal residential areas. The use of detergents creates a possible pollution hazard, since common house hold detergent products can affect the water quality. Angino et al. (1970) found that most enzyme detergents contained trace amounts of the elements Fe, Mn, Cr, Co, Zn, Sr and B.

Atmospheric sources

Natural and man-made processes have been shown to result in metal containing airborne particulates. Depending on prevailing climatic conditions, these particulates may become wind-blown over great distances; nonetheless, they are subjected to the fate that they are ultimately returned to the lithosphere as precipitations by rain or snowfall. Additional sources of atmospheric metal enrichment, such as the high temperature anthropogenic sources, are of special importance on a global scale. Geothermal sources, such as volcanic eruptions, have caused significant atmospheric pollution (Eshleman et al., 1971).

Other sources

Other sources of heavy metals include refuse incineration, landfills and transportation (automobiles, diesel-powered vehicles and aircraft). Two main anthropogenic sources that contaminate the soil are fly ash produced due to coal burning and the corrosion of commercial waste products, which add Cr, Cu, Pb and galvanized metals (primarily Zn) into the environment. Coal burning adds heavy metals such as Cd, Hg, Mn, Ni, Al, Fe and Ti into the soils (Verkleji, 1993). Oil burning contributes V, Fe, Pb and Ni to the environment. Metal emission during the transportation of vehicles includes Ni and Zn from tires, Al from catalyst, Cd and Cu primarily from diesel engines and Ni and Zn from aerosol emissions. Lubricants, which are antiwear protectants for vehicles, emit Cd, Cr, Hg, Ni, Pb and Zn, particularly incase of inefficient engines. The burning of leaded gasoline has been an important source of Pb in the environment. Incinerations of municipal wastes generate significant concentrations of Zn, Pb, Al, Sn, Fe and Cu.

Effects of heavy metals on plants

The exposure of plants to toxic levels of heavy metals triggers a wide range of physiological and metabolic alterations (Dubey, 2011; Villiers et al., 2011). However, as different heavy metals have different sites of action within the plant, the overall visual toxic response differs between heavy metals. The most widespread visual evidence of heavy metal toxicity is a reduction in plant growth (Sharma and Dubey, 2007) including leaf chlorosis, necrosis, turgor loss, a decrease in the rate of seed germination, and a crippled photosynthetic apparatus, often correlated with progressing senescence processes or with plant death (Dalcarso et al., 2010; Carrier et al., 2003). All these effects are related to ultrastructural, biochemical, and molecular changes in plant tissues and cells brought about by the presence of heavy metals (Gamalero et al., 2009). Contamination of agricultural soil by heavy metals has become a critical environmental concern due to their potential adverse ecological effects. Such toxic elements are considered as soil pollutants due to their widespread occurrence and their acute and chronic toxic effect on plants grown of such soils.

Zinc effects on plants

Zinc (Zn) is an essential micronutrient that affects several metabolic processes of plants (Rout and Dass, 2003) and has a long biological half-life. The phytotoxicity of Zn and Cd is indicated by decrease in growth and development, metabolism and an induction of oxidative damage in various plant species such as Phaseolus vulgaris (Cakmak and Marshner, 1993), Brassica juncea (Prasad and Hagmeyer, 1999) and Tobacco (Tkalec et al., 2014). Cd and Zn have reported to cause alternation in catalytic efficiency of enzymes in Phaseolus vulgaris

(Van Assche et al., 1988; Somasekharaiah et al., 1992) and pea plants (Romero-Puertas et al., 2004). Concentrations of Zn found in contaminated soils frequently exceed to those required as nutrients and may cause phytotoxicity. Zn concentrations in the range of 150–300 mg/kg have been measured in polluted soils (Devries et al., 2002; Warne et al. 2008). High levels of Zn in soil inhibit many plant metabolic functions, result in retarded growth and cause senescence. Zinc toxicity in plants limited the growth of both root and shoot (Malik et al., 2011). Zinc toxicity also causes chlorosis in the younger leaves, which can extend to older leaves after prolonged exposure to high soil Zn levels (Ebbs and Kochian, 1997). The chlorosis may arise partly from an induced iron (Fe) deficiency as hydrated Zn^{2+} and Fe^{2+} ions have similar radii (Marschner, 1986). Excess Zn can also give rise to manganese (Mn) and copper (Cu) deficiencies in plant shoots. Such deficiencies have been ascribed to a hindered transfer of these micronutrients from root to shoot. This hindrance is based on the fact that the Fe and Mn concentrations in plants grown in Zn-rich media are greater in the root than in the shoot (Ebbs and Kochian, 1997). Another typical effect of Zn toxicity is the appearance of a purplish-red color in leaves, which is ascribed to phosphorus (P) deficiency (Lee et al., 1996).

Cadmium effects on plants

The regulatory limit of cadmium (Cd) in agricultural soil is 100 mg/kg soil (Salt et al., 1995). Plants grown in soil containing high levels of Cd show visible symptoms of injury reflected in terms of chlorosis, growth inhibition, browning of root tips and finally death (Wojcik and Tukiendorf, 2004; Mohanpuria et al., 2007; Guo et al., 2008). The inhibition of root Fe (III) reductase induced by Cd led to Fe (II) deficiency, and it seriously affected photosynthesis (Alcantara et al., 1994). In general, Cd has been shown to interfere with the uptake, transport and use of several elements (Ca, Mg, P and K) and water by plants (Das et al., 1997). Cd also reduced the absorption of nitrate and its transport from roots to shoots, by inhibiting the nitrate reductase activity in the shoots (Hernandez et al., 1996). Appreciable inhibition of the nitrate reductase activity was also found in plants of Silene cucubalus (Mathys, 1975). Nitrogen fixation and primary ammonia assimilation decreased in nodules of soybean plants during Cd treatments (Balestrasse et al., 2003). Metal toxicity can affect the plasma membrane permeability, causing a reduction in water content; in particular, Cd has been reported to interact with the water balance (Costa and Morel, 1994). Cadmium treatments have been shown to reduce ATPase activity of the plasma membrane fraction of wheat and sunflower roots (Fodor et al., 1995). Cadmium produces alterations in the functionality of membranes by inducing lipid peroxidation (Fodor et al., 1995) and disturbances in chloroplast metabolism by inhibiting chlorophyll biosynthesis and reducing the activity of enzymes involved in CO_2 fixation (Raziuddin et al., 2011).

Copper effects on plants

Copper (Cu) is considered as a micronutrient for plants (Gang et al., 2013) and plays important role in CO_2 assimilation and ATP synthesis. Cu is also an essential component of various proteins like plastocyanin of photosynthetic system and cytochrome oxidase of respiratory electron transport chain (Demirevska-kepova et al., 2004). But enhanced industrial and mining activities have contributed to the increasing occurrence of Cu in ecosystems. Cu is also added to soils from different human activities including mining and smelting of Cu-containing ores. Mining activities generate a large amount of waste rocks and tailings, which get deposited at the surface. Excess of Cu in soil plays a cytotoxic role, induces stress and causes injury to plants. This leads to plant growth retardation and leaf chlorosis (Lewis et al., 2001). Exposure of plants to excess Cu generates oxidative stress and ROS (Stadtman and Oliver, 1991). Oxidative stress causes disturbance of metabolic pathways and damage to macromolecules (He-gedus et al., 2001). Copper toxicity affected the growth of Alyssum montanum (Ouzounidou, 1994) and Cd of cucumber (Moreno-Caselles et al., 2000) and Brassica juncea (Singh and Tewari, 2003). Copper and Cd in combination have affected adversely the germination, seedling length and number of lateral roots in Solanum melongena (Neelima and Reddy, 2002).

Mercury effects on plants

Hg is a unique metal due to its existence in different forms e.g., HgS, Hg^{2+} , Hg and methyl-Hg. However, in agricultural soil, ionic form (Hg^{2+}) is predominant (Han et al., 2006). Hg released to the soil mainly remains in solid phase through adsorption onto sulfides, clay particles and organic matters. Increasing evidence has shown that Hg^{2+} can readily accumulate in higher and aquatic plants (Kamal et al., 2004; Wang and Greger, 2004; Israr et al., 2006). High level of Hg^{2+} is strongly phytotoxic to plant cells. Toxic level of Hg^{2+} can induce visible injuries and physiological disorders in plants (Zhou et al., 2007). For example, Hg^{2+}can bind to water channel proteins, thus inducing leaf stomata to close and physical obstruction of water flow in plants (Zhang and Tyerman, 1999). High level of Hg^{2+} interfere the mitochondrial activity and induces oxidative stress by triggering the generation of ROS. This leads to the disruption of biomembrane lipids and cellular metabolism in plants (Messer et al., 2005; Cargnelutti et al., 2006).

Chromium effects on plants

Chromium (Cr) compounds are highly toxic to plants and are detrimental to their growth and development (Davies et al., 2002). Since seed germination is the first physiological process affected by Cr, the ability of a seed to germinate in a medium containing Cr would be indicative of its level of tolerance to this metal (Peralta et al., 2001). Seed germination of the weed Echinochloa colona was reduced to 25% with 20ppm Cr (Rout et al., 2000). High levels (500 ppm) of hexavalent Cr in soil reduced germination up to 48% in the bush bean Phaseolus vulgaris (Parr and Taylor, 1982). Peralta et al., (2001) found that 40 ppm of Cr(VI) reduced by 23% the ability of seeds of Lucerne (Medicago sativa cv. Malone) to germinate and grow in the contaminated medium. Reductions of 32–57% in sugarcane bud germination were observed with 20 and 80 ppm Cr, respectively (Jain et al., 2000). The reduced germination of seeds under Cr stress could be a depressive effect of Cr on the activity of amylases and on the subsequent transport of sugars to the embryo axes (Zeid, 2001). Protease activity, on the other hand, increases with the Cr treatment, which could also contribute to the reduction in germination of Cr-treated seeds (Zeid, 2001). Decrease in root growth is a well-documented effect due to heavy metals in trees and crops (Tang et al., 2001). Prasad et al. (2001) reported that the order of metal toxicity to new root primordia in Salix viminalis is Cd <Cr <Pb, whereas root length was more affected by Cr than by other heavy metals studied. Chromium stress is one of the important factors that affect photosynthesis in terms of CO_2 fixation, electron transport, photophosphorylation and enzyme activities (Clijsters and Van Assche, 1985). In higher plants and trees, the effect of Cr on photosynthesis is well documented (Van Assche and Clijsters, 1983). However, it is not well understood to what extent Cr-induced inhibition of photosynthesis is due to disorganization of chloroplast’s ultra structure (Vazques et al., 1987), inhibition of electron transport or the influence of Cr on the enzymes of the Calvin cycle. Chromate is used as a Hill reagent by isolated chloroplast (Desmet et al., 1975). The more pronounced effect of Cr (VI) on PS I than on PS II activity in isolated chloroplasts has been reported by Bishnoi et al., (1993 b) in peas. Nevertheless, in whole plants, both the photosystems were affected. Chromium stress can induce three possible types of metabolic modification in plants: (i) alteration in the production of pigments, which are involved in the life sustenance of plants (e.g., chlorophyll, anthocyanin) (Boonyapookana et al., 2002) (ii) increased production of metabolites (e.g., glutathione, ascorbic acid) as a direct response to Cr stress, which may cause damage to the plants (Shanker et al., 2003b) and (iii) alterations in the metabolic pool to channelise the production of new biochemically related metabolites, which may confer resistance or tolerance to Cr stress (e.g., phytochelatins, histidine) (Schmfger, 2001). Induction activation of superoxide dismutase (SOD) and of antioxidant catalase are some of the major metal detoxification mechanisms in plants (Shanker et al., 2003a). Gwozdz et al., (1997) found that at lower heavy metal concentrations, activity of antioxidant enzymes increased, whereas at higher concentrations, the SOD activity did not increase further and catalase activity decreased. Effect of Cr stress also reported in allium (Nematshahi et al., 2012).

Lead effects on plants

Lead (Pb) is one of the ubiquitously distributed most abundant toxic elements in the soil. It exerts adverse effect on morphology, growth and photosynthetic processes of plants. Lead is known to inhibit seed germination of Spartiana alterniflora (Morzck and Funicclli, 1982), Pinus helipensis (Nakos, 1979). Inhibition of germination may result from the interference of lead with important enzymes. Lead also inhibited root and stem elongation and leaf expansion in Allium species (Gruenhage and Jager, 1985) and barley (Juwarkar and Shende, 1986). The degree to which root elongation is inhibited depends upon the concentration of lead and ionic composition and pH of the medium (Goldbold and Hutterman, 1986). Concentration-dependent inhibition of root growth has been observed in *Sesamum indicum* (Kumar et al., 1992). A high lead level in soil induces abnormal morphology in many plant species. For example, lead causes irregular radial thickening in pea roots, cell walls of the endodermis and lignification of cortical parenchyma (Paivoke, 1983). Lead also induces proliferation effects on the repair process of vascular plants (Kaji et al., 1995). Lead administrated to potted sugar beet plants at rates of 100–200 ppm caused chlorosis and growth reduction (Hewilt, 1953). Low amounts of lead (0.005 ppm) caused significant reduction in growth of lettuce and carrot roots (Baker, 1972). Inhibitory effects of Pb^{2+} on growth and biomass production may possibly derive from effects on metabolic plant processes (Sharma and Dubey, 2005). The primary cause of cell growth inhibition arises from a lead-induced simulation of indole-3-acetic acid (IAA) oxidation. Lead is also known to affect photosynthesis by inhibiting activity of carboxylating enzymes (Stiborova et al., 1987). High level of Pb also causes inhibition of enzyme activities (Sinha et al., 1988a, b), water imbalance, alterations in membrane permeability and disturbs mineral nutrition (Sharma and Dubey, 2005). Pb inhibits the activity of enzymes at cellular level by reacting with their sulfhydril groups. High Pb concentration also induces oxidative stress by increasing the production of ROS in plants (Reddy et al., 2005).

Arsenic effects on plants

Arsenate (As) is an analog of phosphate (P) and competes forth same uptake carriers in the root plasmalemma of plants (Meharg and Macnair, 1992). The As tolerance has been identified in a number of plant species (Meharg, 1994; Sharples et al., 2000). The As tolerance in grasses results from suppression of a high-affinity P/As uptake system (Meharg and Macnair, 1992). This suppression reduces As influx to a level at which plant can easily detoxify it, presumably by constitutive mechanisms (Meharg, 1994). The As tolerance is achieved by a single gene encoding for the suppressed P/As transport (Meharg and Macnair, 1992). The As also undergoes transformation within plant cells to other less phytotoxic As species (Meharg, 1994). In phytoplankton and macro algae, As is converted to arsenite, dimethylarsinic acid (DMA) and mono methyl arsenicacid (MMA). Such methylated forms of As are then metabolized to organo phospholipids and arsenosugars (Phillips, 1990).

Cobalt effects on plants

Cobalt (Co) naturally occurs in the earth's crust as cobaltite [CoAsS], erythrite [$Co_3(AsO_4)_2$] and smaltite [$CoAs_2$]. Plants can accumulate small amount of Co from the soil. The uptake and distribution of Coin plants is species- dependent and controlled by different mechanisms (Kukier et al., 2004; Li et al., 2004; Bakkaus et al., 2005). Very little information is available regarding the phytotoxic effect of excess Co. Phytotoxicity study of Coin barley (Hordeum vulgare L.), oilseed rape (Brassica napus L.) and tomato (Lycopersicon esculentum L.) has recently shown the adverse effect on shoot growth and biomass (Li et al., 2009). In addition to biomass, excess of Co restricted the concentration of Fe, chlorophyll, protein and catalase activity in leaves of cauliflower. Further, high level of Co also affected the translocation of P, S, Mn, Zn and Cu from roots to tops in cauliflower. In contrast to excess Cu or Cr, Co significantly decreased water potential and transpiration rate. While diffusive resistance and relative water content increased in leaves of cauliflower upon exposure to excess Co (Chatterjee and Chatterjee, 2000).

Nickel effects on plants

Nickel (Ni) is a transition metal and found in natural soils at trace concentrations except in ultramafic or serpentinic soils. However, Ni^{2+}concentration is increasing in certain areas by human activities such as mining works, emission of smelters, burning of coal and oil, sewage, phosphate fertilizers and pesticides (Gimeno-Garcia et al., 1996). Ni^{2+} concentration in polluted soil may range from 20 to 30- fold (200–26,000 mg/kg) higher than the overall range (10–1,000 mg/kg) found in natural soil (Izosimova, 2005). Excess of Ni^{2+} in soil causes various physiological alterations and diverse toxicity symptoms such as chlorosis and necrosis in different plant species (Pandey and Sharma, 2002; Rahman et al. 2005), including rice (Das et al., 1997). Plants grown in high Ni^{2+}containing soil showed impairment of nutrient balance and resulted in disorder of cell membrane functions. Thus, Ni^{2+} affected the lipid composition and H-ATPase activity of the plasma membrane as reported in Oryza sativa shoots (Ros et al. 1992). Gonnelli et al., (2001) reported an increase in MDA concentration of Ni^{2+} sensitive plants compared to a Ni^{2+} -tolerant saline. Such changes might disturb membrane functionality and ion balance in the cytoplasm, particularly of K^{2+}, the most mobile ion across plant cell membrane. High uptake of Ni^{2+} induced a decline in water content of dicot and monocot plant species. The decrease in water uptake is used as an indicator of the progression of Ni^{2+} toxicity in plants (Pandey and Sharma, 2002; Gajewska et al., 2006).

Manganese effects on plants

Accumulation of excessive manganese (Mn) in leaves causes a reduction of photosynthetic rate (Kitao et al., 1997 b). Mn is readily transported from root to shoot through the transpiration stream, but not readily remobilized through phloem to other organs after reaching the leaves (Loneragan, 1988). Necrotic brown spotting on leaves, petioles and stems is a common symptom of Mn toxicity (Wu, 1994). This spotting starts on the lower leaves and progresses with time toward the upper leaves (Horiguchi, 1988). With time, the speckles can increase in both number and size resulting in necrotic lesions, leaf browning and death (Elamin and Wilcox, 1986a). General leaf bronzing and shortening of internodes has been documented in Cucumis sativus (Crawford et al,. 1989). Another common symptom is known as "crinkle- leaf", and it occurs in the youngest leaf, stem and petiole tissue. It is also associated with chlorosis and browning of these tissues (Wu, 1994; Bachman and Miller, 1995). Roots exhibiting Mn toxicity are commonly brown in color (Le Bot et al., 1990; Foy et al., 1995) and sometimes crack (Foy et al., 1995). Chlorosis in younger leaves by Mn toxicity is thought to be caused through Mn-induced Fe deficiency (Horst, 1988). Excess Mn is reported to inhibit synthesis of chlorophyll by blocking a Fe-concerning process (Clarimont et al., 1986). Manganese toxicity in some species starts with chlorosis of older leaves moving toward the younger leaves with time (Bachman and Miller, 1995). This symptom starts at the leaf margins progressing to the interveinal areas and if the toxicity is acute, the symptom progresses to marginal and interveinal necrosis of leaves (Bachman and Miller, 1995).

Iron effects on plants

Iron as an essential element for all plants has many important biological roles in the processes as diverse as photosynthesis, chloroplast development and chlorophyll biosynthesis. Iron is a major constituent of the cell redox systems such as heme proteins including cytochromes, catalase, peroxidase and leghemoglobin and iron sulfur proteins including ferredoxin, acontiase and superoxide disumutase (SOD) (Marschner, 1995).

Although most mineral soils are rich in iron, the expression of iron toxicity symptoms in leaf tissues occurs only under flooded conditions, which involves the microbial reduction of insoluble Fe^{3+} to insoluble Fe^{2+} (Becker and Asch, 2005). The appearance of iron toxicity in plants is related to high Fe^{2+} uptake by roots and its transportation to leaves and via transpiration stream. The Fe^{2+} excess causes free radical production that impairs cellular structure irreversibly and damages membranes, DNA and proteins (Arora et al., 2002; de Dorlodot et al., 2005). Iron toxicity in tobacco, canola, soybean and Hydrilla verticillata are accompanied with reduction of plant photosynthesis and yield and the increase in oxidative stress and ascorbate peroxidise activity (Sinha et al., 1997).

Mode of Action of Toxic heavy metals in Plant Cells

The toxicity of heavy metals is manifested in many ways when plant cells accumulate them at high levels. Heavy metals can be divided into two groups: redox active (Fe, Cu, Cr, Co) and redox inactive (Cd, Zn, Ni, Al, etc.). The redox active heavy metals are directly involved in the redox reaction in cells and result in the formation of $O^{2\bullet-}$ and subsequently in H_2O_2 and $^{\bullet}OH$ production via the Haber-Weiss and Fenton reactions (Schutzendubel and Polle, 2002). Exposure of plants to redox inactive heavy metals also results in oxidative stress through indirect mechanisms such as interaction with the antioxidant defense system, disruption of the electron transport chain, or induction of lipid peroxidation. The latter can be due to an heavy metal-induced increase in lipoxygenase (LOX) activity.

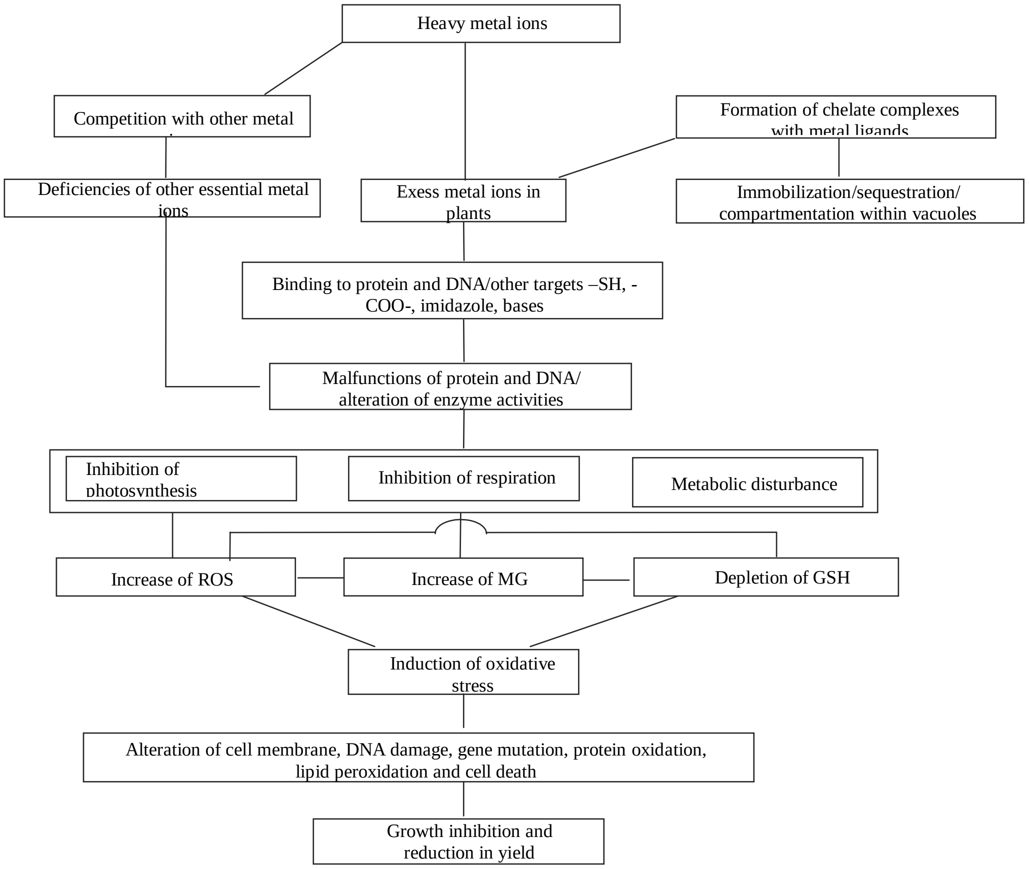

Figure 1: Possible biochemical and molecular mechanisms of heavy metal-mediated ROS induction and damage to the development of higher plants.

Another important mechanism of heavy metal toxicity is the ability of heavy metals to bind strongly to oxygen, nitrogen, and sulphur atoms. This binding affinity is related to free enthalpy of the formation of the product of the Heavy metal and ligand with low solubility of these products. Because of these features, heavy metals can inactivate enzymes by binding to cysteine residues. For example, Cd binding to sulfhydryl groups of structural proteins and enzymes leads to misfolding and inhibition of activity and/or interference with redox-enzymatic regulation (Dalcorso et al., 2008; Hall, 2002). Many enzymes need cofactors to work properly for both heavy metal ions (such as Fe^{2+}, Mg^{2+}, Cu^{2+}, Ca^{2+}) and organic molecules (such as haem, biotin, FAD, NAD, or coenzyme A). The displacement of one heavy metal ion by another leads to the inhibition or loss of enzyme activities. Divalent cations such as Co^{2+}, Ni^{2+}, and Zn^{2+} displace Mg^{2+} in ribulose-1,5- bisphosphate-carboxylase/oxygenase (RuBisCO) and result in a loss of activity . Displacement of Ca^{2+} by Cd^{2+} in calmodulin, an important protein in cellular signaling, led to the inhibition of calmodulin-dependent phosphodiesterase activity in radish (Rivetta et al., 1997).

Additionally, heavy metals cause membrane damage through various mechanisms, including the oxidation of and cross-linking with protein thiols, inhibition of key membrane protein such as H^+-ATPase, or causing changes in the composition and fluidity of membrane lipids (Merag, 1993). Accumulation of methyl glyoxal, a cytotoxic compound, was found to increase in response to heavy metal stress in plants due to impairment of the glyoxalase system that finally elicits oxidative stress by reducing the GSH content (Hossain et al., 2009, Hossain et al., 2010 and Singla et al., 2006). Based on the aforementioned, it can be concluded that heavy metal toxicity is attributed to three mean reasons: (a) stimulation of ROS and MG production by auto-oxidation and the Fenton reaction or by modification of the antioxidant defense system and the glyoxalase system, (b) direct interaction with proteins due to their affinities for thioyl-, histidyl-, and carboxyl-groups, causing the heavy metals to target structural, catalytic, and transport sites of the cell, and (c) displacement of essential metal ions from specific binding sites, causing function to collapse (Sharma and Dietz, 2009). The possible sequential events of ROS-induced damage development in sensitive plants in response to HM stress are summarized in Figure 1.

Thus, it is evident from the several research reports that the presence of heavy metals have many toxic effects on plants. Therefore, it is well needed to intensify the research programmes for better understanding of heavy metal toxicity on plants and allied areas to maintain the ecological harmony of the globe.

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