



HAL
open science

Effect of Metal Toxicity on Plant Growth and Metabolism: I. Zinc

Gyana Rout, Premananda Das

► **To cite this version:**

Gyana Rout, Premananda Das. Effect of Metal Toxicity on Plant Growth and Metabolism: I. Zinc. *Agronomie*, 2003, 23 (1), pp.3-11. 10.1051/agro:2002073 . hal-00885964

HAL Id: hal-00885964

<https://hal.science/hal-00885964v1>

Submitted on 11 May 2020

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

Effect of metal toxicity on plant growth and metabolism: I. Zinc

Gyana Ranjan ROUT*, Premananda DAS

Plant Biotechnology Division, Regional Plant Resource Centre, Nayapalli, Bhubaneswar – 751 015, India

(Received 2 March 2001; revised 21 December 2001; accepted 14 May 2002)

Abstract – Zinc toxicity and problems with regard to tolerance and ecological significance are briefly discussed. Differential tolerance of plant genotypes exposed to zinc toxicity is a more promising approach to enrich our understanding of zinc tolerance in plants. Knowledge concerning the physiology and biochemistry with regard to phytotoxicity, uptake and transport of zinc and tolerance and its characterization are also discussed. The cytotoxic effects of zinc on plants are elucidated. The major change was seen in the nucleus of the root tip cells due to zinc toxicity. The chromatin material was highly condensed and some of the cortical cells showed disruption and dilation of nuclear membrane in presence of 7.5 mM zinc. The cytoplasm became structureless, disintegration of cell organelles and the development of vacuoles were also observed. The number of nucleoli also increased in response to zinc resulting in the synthesis of new protein involved in heavy metal tolerance. This review may help in interdisciplinary studies to assess the ecological significance of metal stress.

phytotoxicity / tolerance / transport / uptake / accumulation / zinc

Résumé – Effet de la toxicité des métaux sur la croissance et le métabolisme des plantes : I. Zinc. La toxicité du zinc et les problèmes de tolérance ou de conséquence écologique liés sont brièvement discutés. L'approche en terme de tolérance différentielle des génotypes de plantes exposées à la toxicité du zinc est prometteuse pour l'enrichissement de notre compréhension de la tolérance des plantes au zinc. Les connaissances de la physiologie et la biochimie face à la phytotoxicité, à l'absorption et au transport du zinc, ainsi que la tolérance et sa caractérisation sont aussi discutées dans ce papier. Les effets cytotoxiques du zinc sur les plantes sont maintenant élucidés. La modification majeure concerne le noyau des cellules de l'extrémité des racines. La chromatine est fortement condensée et certaines des cellules corticales montrent la rupture et la dilatation de leur membrane nucléaire en présence de 7.5 mM de zinc. De plus, le cytoplasme perd sa structure, la désintégration d'organites et le développement de vacuoles sont aussi observés. Enfin, le nombre de nucléoles augmente en réponse au zinc. Ils résultent de la synthèse d'une nouvelle protéine impliquée dans la tolérance aux métaux lourds. Cette synthèse bibliographique pourra aider les études interdisciplinaires à évaluer les conséquences écologiques des stress dus aux métaux.

phytotoxicité / tolérance / transport / absorption / accumulation / zinc

1. INTRODUCTION

The occurrence of heavy metals in soils may be beneficial or toxic to the environment. Excess of metals may produce some common effects of individual metals on different plants (i.e. both macro- and microflora). The biota may require some of these elements in trace quantities but at higher concentrations there may be toxicity problems. Metal toxicity in plants have been reported by various authors [15, 18, 19, 41, 46]. Generally, zinc is an essential element which belongs to Group-II of the periodic table. It acts as a plant nutrient [101, 128] but at higher concentrations, it is toxic. Since it is

assimilated early by plants, it can be highly phytotoxic. Growth inhibition is a general phenomenon associated with zinc toxicity [28]. Zinc is also a constituent of metalloenzyme or a cofactor for several enzymes such as anhydrases, dehydrogenases, oxidases and peroxidases [55] and plays an important role in regulating the nitrogen metabolism, cell multiplication, photosynthesis and auxin synthesis in plants [101]. It also plays an important role in the synthesis of nucleic acid and proteins and helps in the utilization of phosphorous and nitrogen during seed formation. Hyper-accumulation of zinc has been observed in many plant species [9, 121], accumulations of 1% of the dry weight being

Communicated by Gérard Guyot (Avignon, France)

* Correspondence and reprints
grrout@hotmail.com

common among plants growing on zinc minewastes [90]. The toxic effects of zinc on plants have been reported by many researchers [11, 12, 31, 110, 127, 129, 130]. Genotoxicity of zinc in micro- and macroflora were also elucidated by various researchers [82, 108]. In addition, the toxic effects of the metals on photosynthesis in plants were elaborated [27, 44, 106, 118–120]. Recent progress in the study of toxic metals and their interactions with essential elements has greatly increased our understanding of the mechanism of toxicity at the biochemical level [1]. In this communication, the salient features of zinc toxicity, the effects on plant metabolism and its interaction with other essential elements are discussed; their possible implications in the plant ecosystem are highlighted.

2. ZINC TOXICITY

Zinc toxicity depends on pH, which controls the concentration of zinc in solution. High concentrations of zinc can cause toxicity in plants [33]. The general symptoms are stunting of shoot, curling and rolling of young leaves, death of leaf tips and chlorosis. Ye et al. [135] reported that the seedlings of *Typha latifolia* were chlorotic in the presence of ~ 80 μM zinc. Foliar chlorosis appeared on *Thlaspi ochroleucum* seedlings treated with 1 mM Zn after 4 days of culture as reported by Shen et al. [99].

2.1. Effect on germination

Zinc is an essential nutrient for plant growth, although elevated concentrations resulted in growth inhibition and toxicity symptoms. It does not affect seed germination but helps in plumule and radicle development. Baker [7] reported that the seeds of *Silene maritima* were germinated better and rapidly on calcium nitrate solutions containing different concentrations of zinc.

2.2. Effect on root

Zinc though an essential element for plant growth, showed toxicity symptoms at higher concentrations inhibiting root growth [7, 16]. Zinc toxicity was marked in root system particularly in root blunt, thickening and caused restraint on both cell division and cell elongation [6, 123]. Doyar and Van Hai Tang [36] reported that the nitrogen and phosphorous increased with the increase in zinc content in the roots. The concentration of zinc in the roots decreased with plant age as reported by Pearson and Rengel [86]. Sresty and Madhava Rao [104] based on transmission electron microscopy concluded that radicle elongation was more adversely affected than the plumule extension. The root elongation of *Cajanus cajan* cv. ICPL 87 was completely inhibited after 24 h treatment with 7.5 mM zinc. They also reported that root cortical cells were extensively damaged and major changes took place in the nucleus of the root tip cells treated with 7.5 mM zinc.

2.3. Effect on reproductive growth

Metal sensitive plants have great difficulties in reaching the reproductive phase when exposed to metals. The seeds of most plant species growing on heavy metal enriched soils have very low metal concentrations [38, 62]. Zinc-resistant genotypes of *Silene vulgaris* were stimulated by increased levels of zinc, not only in vegetative growth but also in seed production [40]. Surpassing a critical zinc concentration resulted in reduced growth and the no flower production.

2.4. Effect on plant physiology and morphology

The physiology of metal toxicity in plants was mainly concerned with metal movement from soil to root and metal absorption and translocation. Plant availability of a metal in the soil depends on soil adsorption strength as well as plant effectors such as root exudates for metal chelation or reduction. Lindsay [64] and others developed models for the chemical activity of metal ions in soil. The chemical activity was usually dependent on soil pH, CO_2 concentration and redox potential of the soil [57, 58, 64, 65]. The concept of metal equilibrium in soil, and of metal movement from soil to root were well documented [41]. Studies on the physiological and biochemical metal toxicity were almost unknown. The activity of a metal in the cytoplasm was affected by chelating carboxylic and aminoacids and the pH. Zinc inhibited Fe translocation in some cases [3] and the young chlorotic leaves showing Zn^{2+} toxicity generally contained more than 100 ppm Fe; Fe deficiency generally occurred at concentrations lower than 40 ppm Fe in the young leaves. The metal toxicity resulted with the increased metal supplied to the root which affect the disintegration of cell organelles, disruption of membranes and condensation of chromatin material and increase in number of nucleoli were major events during zinc toxicity [104]. Ernst [40] elaborated details on mechanism of heavy metal toxicity in plants at the cellular and organism level. He also reported that the plasma membrane was the cell compartment which regulated metal entry into the cell, in addition its proteins, especially the SH groups might be affected in their activity causing damage to membrane stability due to zinc toxicity. As soon as heavy metals passed through the plasma membrane, they could immediately interact with all metabolic processes in the cytosol. Godbold and Huttermann [48] reported that increasing zinc levels in culture solution decreased the shoot to root ratios and translocation of Zn, Fe, Mg, K, P and Ca and caused accumulation of these nutrients in the root. Pearson and Rengel [86] indicated that higher concentration of zinc affected the leaf and the root morphology. They suggested that the zinc supply from the roots into the leaves of different ages might be determined by the relative transpiration rate of the leaves. Further, Malea et al. [72] tested the effect of zinc on leaf cell mortality of *Halophila stipulacea*. The mortality of the cells increased along with the increase of zinc concentrations (10^{-7} to 10^{-4} M) and the increase of incubation time (2nd–12th days). Necrosis was evident in the epidermal cells at all concentrations on the 12th day of culture. Necrosis was also detectable in the mesophyll cells at the highest concentration (10^{-4} M) after the 8th day. Zinc toxicity was primarily associated with alteration of root physiology, thereby inhibiting the root elongation [134].

3. DIFFERENTIAL ZINC TOLERANCE IN PLANTS

Differential tolerance in vitro and in vivo

Zinc toxicity and differential zinc tolerance in various plant groups were reported [3, 18, 37, 88, 109, 130]. Earley [37] reported that different soybean cultivars responded differentially to toxic Zn in sand culture. White [131] studied the differential varietal tolerance in soybean and found that they were associated with differences in susceptibility to zinc-induced Fe deficiency, Zn uptake and translocation, and susceptibility to toxicity unrelated to Fe deficiency. Gregory and Bradshaw [49] developed Zn-tolerant efficiency in bentgrasses. Further, Ernst [39] and Mathys [75] accepted the compartmentalization hypothesis and suggested that malate might chelate zinc in the vacuoles of tolerant plants [76]. The fact that high malate, Zn-tolerant ecotypes were not tolerant to Cu and was difficult to understand since malate would be expected to bind Cu much more strongly than it does in Zn. Mathys [77] and Ernst [39] conducted detailed studies on metal tolerant enzymes in tolerant and non-tolerant ecotypes. However, in in vitro studies, the activities of several enzymes remained at normal levels in tolerant ecotypes as metal supply increased, but activities fell markedly in nontolerant ecotypes. They also confirmed that tolerant plants were able to maintain the normal metabolic processes when subjected to metal stress which severely interfered with the metabolism of nontolerant plants. Hertstein and Jager [54] studied the tolerances of different populations of three grass species (*Agrostis tenuis*, *Festuca rubra* and *Anthoxanthum odoratum*) to zinc and other metals. They observed that all populations originating from metal contaminated habitats possessed multiple-metal-tolerance. Genecological and physiological aspects of tolerance were studied extensively with regard to zinc and other metals tolerance [5, 134]. Baker [7] selected zinc tolerant *Silene maritima* populations in water-culture experiments. Subsequently, Amado-Filho et al. [4] screened six seaweed species for zinc tolerance for a period of 21 days using rapid hydroponic methods. They reported that all species died at 76 μM of zinc, two species (*Ulva lactuca* and *Enteromorpha flexuosa*) died at 152 μM and one, *Hypnea musciformis* died at 0.152×10^{-2} mM. Ye et al. [135] tested four populations of *Typha latifolia* collected from metal-contaminated and uncontaminated sites grown in nutrient solution containing 1.0–5.0 $\mu\text{g/ml}$ Zn under controlled environmental conditions. They also reported that seedlings from metal contaminated populations showed three times more tolerance to zinc as compared to the uncontaminated population. Shen et al. [99] compared the growth of hyperaccumulator species *Thlaspi caerulescens* and non-hyperaccumulator species *Thlaspi ochroleucum* in nutrient culture experiment with zinc. They reported that *Thlaspi caerulescens* was able to tolerate 0.5 mM Zn in solution without growth reduction, and up to 1 mM Zn without showing visible toxic symptoms but with a 25% decrease in dry matter yield. Sresty and Madhava Rao [104] and Madhava Rao and Sresty [71] tested two genotypes of pigeonpea (*Cajanus cajan*) in nutrient solution containing zinc (2.5, 5.0 and 7.5 mM) to assess Zn tolerance in term of root and shoot

tolerance index, metal uptake and dry mass population. Rout et al. [96] selected Zn tolerant callus lines of *Brassica* species using in vitro techniques. They assessed various growth parameters such as fresh and dry weight of callus, growth tolerance index and metal uptake to determine the tolerant line. The tolerant calli had the maximum peroxidase and catalase activity and higher rate of metal uptake as compared to non-tolerant calli. Subsequently, Samantaray et al. [97] developed zinc tolerant calli and regenerated of plantlets from tolerant calli of *Setaria italica* using in vitro technique. The tolerant plants showed luxuriant growth in zinc rich minespoil. Rengel [93] observed that the ecotypes of *Holcus lanatus* tolerant to zinc toxicity also tolerated Zn-deficiency better than the Zn-sensitive ecotypes because of their greater ability to taking up zinc from Zn-deficient soil.

4. EFFECT OF ZINC ON NUCLEAR ACTIVITY

Growth of plants was mainly dependent on cell division. Fujii [43] found that zinc played an important role in mitosis. Gebhart [45] observed higher frequency of chromosome aberrations in presence of heavy metals. Heavy metals, which constituted one of the important groups of environmental pollutants, were mostly genotoxic [98]. Further study indicated that the heavy metals like nickel, cadmium and zinc had genotoxic activity through oxidative pathways involving free radicals [80, 85]. Sresty and Madhava Rao [104] observed a major change in the nucleus of the root tip cells in response to zinc. The chromatin material was highly condensed and some of the cortical cells showed disruption and dilation of nuclear membrane in presence of 7.5 mM zinc. The cytoplasm became structureless; disintegration of cell organelles and the development of vacuoles were also observed. They also noted that the number of nucleoli increased in response to zinc resulting in the synthesis of new proteins involved in heavy metal tolerance. Further, the information of SH-rich phytochelatins appears to play a role in heavy metal detoxification in different organisms [50, 92, 95, 111]. Ernst [40] compared the nuclear activity and cell division in Zn-sensitive and Zn-resistant ecotypes of the perennial grass *Festuca rubra* in presence of different level of zinc. He observed that the nuclear volume decreased by 30% and doubled the length of the cell cycle when Zn-sensitive ecotypes were exposed to 3 μM Zn for 4 days [89]; the nuclear volume increased by 50% in Zn-resistant ecotypes exposed to same Zn level.

5. EFFECT OF ZINC ON METABOLISM

In general, a number of plant species were resistant to certain amounts of metals. This was probably achieved through trapping of these metals with in the metal-binding proteins. Zinc was known to be a constituent of many enzymes which stimulated various metabolic activities such as nucleic acid metabolism, protein synthesis, photosynthesis, respiration and carbohydrate metabolism [102]. Zinc was easily assimilated by plants, and could also be strongly phytotoxic; growth inhibition was a general phenomenon to

zinc toxicity [28]. Many authors observed inhibition of photosynthesis by heavy metal ions but the mechanism of action was not known in details [106, 112]. Van Assche [116] reported that the high concentration of zinc inhibited metabolic activity. In some cases, the concentration ranged from 0.4–1.0 mM inhibited photosystem [10, 53, 112, 118]. Lorimer [67] and Lorimer and Mizioroko [68] reported that bivalent cations (Zn^{2+}) played a major role in the activation of the key enzyme of the Calvin cycle and in the equilibrium between CO_2 and O_2 binding by protein and inhibition of photosynthesis reactions localized in the thylakoid membranes of chloroplasts [117]. Interference of zinc in photochemical reactions of chloroplast was intensively studied [118]. Garty et al. [44] studied the effects of low pH and Zn on chlorophyll degradation in lichen (*Ramalina duriaei* Bagl.). They found that Zn decreased photosynthesis in lichens having chlorophycean photobionts under pH 2.0. Verkleij and Schat [121] and Meharg [79] reported that the plasma membrane was the compartment of the cell which might at least partially regulate the entry of a heavy metal ion into the cell. This direct exposure might have consequences for the adaptation and selection of plant species and genotypes. Marschner [73] indicated that cation and anion uptake by the cell could be actively regulated by electrogenic proton pumps ($H^+ - ATPase$), transmembrane redox pumps (NAD (P) oxidase), and ion channels [74]. Davis et al. [32] indicated that the plants exposed to higher concentrations of zinc disturbed the mitochondrial structure and reduces the energy.

6. ZINC UPTAKE AND TRANSPORT

Zinc an essential element for the normal growth and metabolism of plants played very important role in enzyme activation and was also involved in the biosynthesis of some enzymes and growth hormones [34, 83]. The transport and distribution of zinc and other metals in plants were reported by various researchers [66, 73, 122]. The accumulation of zinc in the roots in the tolerance races were reported [87, 114, 115]. Baker [7] reported that the populations of *Silene maritima* accumulated zinc to a high degree in the roots relative to the shoots. Turner and Marshall [114, 115] detected a correlation between the uptake of ^{65}Zn by cell wall and mitochondrial fractions from the roots of a range of *Agrostis tenuis* populations and the indices of zinc tolerance. Wahbeh [122] reported that the distribution and abundance of zinc in various vegetative tissues of the sea grasses *Halophila ovalis*, *Halophila stipulacea* and *Halodule uninervis*. He also indicated that both the root and the leaf absorbed metals, and that translocation was low [69]. Availability of heavy metals in soil and their uptake by plants not only was dependant on the total metal content in the soil but also upon other factors i.e. soil organic matter and cation exchange capacity [14, 70, 81, 107]. Hinesly et al. [56] found that the uptake of heavy metals (Zn and Cd) by *Zea mays* generally decreased as the soil pH increased. They also assumed that the increased solubility of Zn in the alkaline pH range was due to the dispersion of organic matter containing

complexed zinc form. Adriano [2] demonstrated that the distribution of metals in the fractions depended on the soil properties, and, for most mineral soils, the largest amounts of zinc were present in the residual fraction. Whitehead [133] found that the accumulations of zinc (Zn^{+2}) varied in different plant species. The ratio between the accumulation of root and the shoot was always greater than 1.0 in ryegrass and clover. McKenna et al. [78] found a complex interaction between Zn^{+2} and Cd^{+2} on the accumulation in the root and leaf of lettuce and spinach; the interactions were dependent on the relative concentration of those metals in the growth medium. Zinc accumulation was higher in the younger than the older leaves. Shetty et al. [100] reported that the application of mycorrhizal fungi helped in higher accumulation of zinc in the roots than in the shoots. The influence of arbuscular mycorrhizae (AM) on plant growth and zinc uptake by *Lygeum spartum* and *Anthyllis cytisoides* was studied in soils with different levels of heavy metals [35]. Rout et al. [96] and Samantaray et al. [97] conducted in vitro studies on zinc accumulation in tolerant and non-tolerant calli of *Setaria italica* and *Echinochloa colona* respectively and observed that the uptake and accumulation was more in tolerant calli than the non-tolerant ones. Bert et al. [13] investigated zinc tolerance in five populations of *Arabidopsis halleri* raised from seed collected from contaminated and *Arabidopsis thaliana* from non-contaminated sites. They observed that the populations of *Arabidopsis thaliana* from non-contaminated area accumulated zinc in shoots and roots more quickly than the populations from the contaminated sites. Frey et al. [42] measured zinc concentration in shoot which was higher and reached a maximum value of $83 \text{ mM} \cdot \text{kg}^{-1}$ dry mass whereas total concentration of zinc in roots were lower up to $13 \text{ mM} \cdot \text{kg}^{-1}$. They observed that the distribution of zinc in *Thlaspi caerulescens*; Zn got mainly accumulated in the vacuoles of epidermal leaf cells and was totally absent from the vacuoles of the cells from the stomatal complex, thereby protecting the guard and subsidiary cells from high zinc concentrations. They concluded that zinc also got accumulated in high concentrations in both the cell walls of epidermal cells and in the mesophyll cells, indicating that apoplastic compartmentation was another important mechanism involved in zinc tolerance in the leaves of *T. caerulescens*. Chardonnens et al. [23] demonstrated that the tonoplast vesicles derived from Zn-tolerant ecotype of *Silene vulgaris* accumulated more Zn than the Zn-sensitive ecotype. They also characterized the tonoplast-transport system that caused this difference in the uptake and demonstrated its genetic correlation. The most prominent differences being its insensitivity to protonophores and ortho-vanadate and its stimulation by Mg-CTP. They concluded that in both Zn-tolerant and Zn-sensitive ecotypes, Zn was actively transported across the tonoplast (temperature coefficient > 1.6), most likely as a free ion, since citrate did not accumulate in vesicles. Lasat et al. [61] reported the molecular physiology and zinc transport in the Zn-hyperaccumulator *Thlaspi caerulescens* and a non-accumulator. They reported that Zn transport was stimulated at a number of sites in *T. caerulescens* contributing to the hyperaccumulation trait. The transport processes that were stimulated included Zn influx into both root and leaf cells, and Zn loading into the xylem. They also hypothesized that the stimulation of Zn influx was 4 to 5 fold into the root because

of an increased abundance of Zn transporters in *Thlaspi caerulescens* root cells. Zinc was sequestered in the vacuoles of non accumulator *T. arvense* root cells which retarded Zn translocation to the shoot. Subsequently, they characterized the Zn transport genes in *T. caerulescens* and reported that ZNT1 (Zn transporter) gene was highly expressed in roots and shoots of *T. caerulescens*. But in *T. arvense*, ZNT1 was expressed at far lower levels and this expression was stimulated by imposition of Zn deficiency. Further, Whiting et al. [132] indicated the positive responses to Zn and Cd by roots of the Zn and Cd hyperaccumulator *Thlaspi caerulescens*. Hacısalihoglu et al. [51] reported the high and low-affinity zinc transport systems and their possible role in zinc efficiency in wheat (*Triticum aestivum*). The low-affinity system showed apparent Km values similar to those previously reported for wheat (2–5 mM). High-affinity Zn transport system with apparent Km values were found in the range of 0.6 to 2 nM. Because it functioned in the low range of the available Zn level found in most soils, this novel high affinity uptake system was likely to be the predominant Zn²⁺ uptake system. Zn²⁺ uptake was similar for cv. Dagdas and cv. BDME-10 over both the high- and low-affinity Zn²⁺ activity ranges.

Mechanisms involved in Zn tolerance

Mechanism of metal tolerance by plants has been studied by various authors [8, 30, 41]. At first, exclusion of metals from roots seemed a likely mechanism because root cell walls could bind metals; the extent of binding by the cell wall was related to the degree of tolerance to a specific metal [113–115]. Jones et al. [59] found that Zn complex was formed in roots of Zn-tolerant bentgrass after digesting the roots with cellulase. Although binding of metals to root cell walls might contribute to metal tolerance, it was not adequate enough to enable the plants to prevent metal transport to their leaves [124]. Woolhouse [134] felt that there existed different specific mechanisms of tolerance within each of the populations which were related to the contamination of their original habitat. The mechanism of tolerance depended possibly on three major factors i.e. an exclusion of heavy metals from uptake, i.e. an ‘avoidance’ [63] or translocation to the shoots [7], a protection of sensitive structures in the cytoplasm either by immobilization of metals in the cell wall [114, 115] and metal complexation with soluble compounds, e.g. organic acids and aminoacids [54] or by binding to specific proteins [91]. Cumming and Taylor [29] described the signal transduction and acclimation mechanisms on metal tolerance in plants. They also indicated that the mechanism of metal tolerance were due to exclusion and external metal detoxification mechanism, internal metal detoxification mechanisms, and multiple mechanisms and co-tolerances. The production of metal-chelating ligands high in thiol groups might also render co-tolerance to Zn²⁺. Further, Neumann et al. [84] found by electron microscopy that most of the heavy metals were tightly bound to the cell wall in tolerant plant populations as compared to nontolerant ones. Frey et al. [42] elucidated tolerance mechanism at the cellular and subcellular level for the detoxification of the accumulated zinc within the leaf of hyperaccumulator *Thlaspi caerulescens*. They also observed that there

was apoplastic compartmentation in the leaves of *T. caerulescens*. Hall [52] reported the range of potential cellular mechanisms that may be involved in the detoxification of heavy metals and thus tolerance to metal stress. These includes the binding to cell wall and extracellular exudates, reduced uptake or efflux pumping of metals at the plasma membrane, chelation of metals in the cytosol by peptides such as phytochelatins, repair of stress-damaged proteins and the compartmentation of metals in the vacuole by tonoplast located transporters.

7. PHYTOTOXICITY

To evaluate meaningful physiological and biochemical effects of toxicity, one must know the metals which are phytotoxic in nature and interactions with other metals [30]. Before starting a phytotoxicity experiment one should be fully aware of the movement of the metal including its absorption and translocation in the plant system. Availability of metal in the soil depends on soil adsorption strength as well as plant effectors such as root exudates for metal chelation or reduction. Metal phytotoxicity can result only if metals can move from the soil to root systems [41]. Phytotoxicity levels of zinc in different crop plants were reported by many workers [21, 105]. The most significant phytotoxicity symptoms were stunting of growth, chlorosis and reduction in biomass yield. The phytotoxicity caused by a wide variety of metals has been well documented; however, models designed to quantify the relationship between exposure to metal ions and progressive yield losses are lacking [110].

7.1. Phytotoxicity and its interaction with other nutrients

Ambler et al. [3] indicated that zinc induced inhibition of Fe translocation from root to tops which causes chlorosis in plants. Zinc causes phytotoxicity like chlorosis at low pH level as reported by Chaney et al. [22]. Chavan and Banerjee [25] reported that Zn toxicity appear to be due to Fe deficiency. Cayton et al. [20] reported that the absorption and translocation of plant nutrients like Fe, Mg, K, P and Ca depended on Zn concentration in soil. They reported that zinc was an antagonist to Cu at the primary absorption site, in contrast with its action on Fe, P, Mg, K and Ca. Zinc interfered at the loading site of the roots and decreased the rate of translocation or absorption of essential nutrients to plants or caused mineral imbalances [17, 24, 25, 125, 126]. White et al. [131] observed that increased levels of Zn in soil greatly increased translocation of Mn to tops which indicate the appearance of chlorosis. They hypothesized that the Zn and Mn interfere with Fe utilization in the leaves for chlorophyll synthesis. Accumulation of zinc in the roots or shoots was generally accompanied by accumulation of calcium [7]. He also compared the interaction of zinc and calcium with regard to uptake by zinc-tolerant and non-tolerant population of *Silene maritima* by conducting solution culture experiment. The result indicated that the total zinc uptake was not affected by calcium level in the tolerant population but was decreased significantly in the non-tolerant population. The stimulation

by calcium of zinc uptake in the tolerant plants may reflect an involvement of calcium in the zinc tolerance mechanism sited in the roots. Davies and Parker [31] reported that zinc toxicity were highly correlated with Ca:Zn ratio and reduced stem biomass. Shetty et al. [100] pointed out that growth inhibition was due to interference of zinc with phosphorous uptake by plants. They also reported that the application of vesicular-arbuscular mycorrhizae (VAM) fungi at zinc contaminated sites increased plant biomass even at elevated levels of zinc in the soil. Synergism was observed between boron and zinc when both were in excess together as excess accelerated the effect of high zinc by lowering the biomass, economic yield and carbonic anhydrase activity in mustard [103]. Gianquinto et al. [47] concluded that the Zn concentration in leaf of *Phaseolus vulgaris* cv. Borlotto nano was reduced by the addition of phosphorous to plants grown at low Zn supply. Further, Rengel and Romheld [94] reported that the zinc deficiency depressed the rate of Fe transport to shoots at early stages of *Triticum aestivum*. Kaya and Higgs [60] tested the effectiveness of phosphorus and iron supplemented in nutrient solution on growth of tomato plant at high zinc (77 μM) concentration. They also reported that the application of supplementary P and Fe in the nutrient solution resulted in increase of both dry weight and chlorophyll concentration in *Lycopersicon esculentum* cvs. Blizzard, Liberto, Calypso and also decreased zinc concentration in the leaves and roots of plants grown at high Zn treatment.

7.2. Phytotoxicity and its interaction with other heavy metals

Zinc and cadmium have many physical and chemical similarities as they both belong to Group II of the Periodic Table. They are usually found together in the ores and compete with each other for various ligands. Thus interaction between Zn and Cd in the biological system is likely to be similar. The fact that cadmium is a toxic heavy metal and zinc is an essential element which makes this association interesting as it raises the possibility that the toxic effects of cadmium may be preventable or treatable by zinc [26]. Hinsely et al. [56] suggested that both cadmium and zinc uptake by plants were dependent on the pH of the growing media. The basic mechanisms of Cd-Zn in the tissues induced the synthesis of different types of metallothionein, binding characteristics of metallothionein, alteration in absorption and tissue distribution of one metal by another, and competition at the level of zinc containing metallozymes known to be involved in the interactions. McKenna et al. [78] reported the interactions between zinc and cadmium and the concentration and tissue distribution in spinach and lettuce. They observed that the cadmium concentration in young leaves of lettuce and spinach decreased exponentially in the solution containing Zn at low (0.0316 μM) but not at high (0.316 μM) concentration of Cd. The Zn:Cd concentration ratios in young leaves of lettuce and spinach grown at 0.316 μM Cd was greater as the concentration of Zn increased. Cayton et al. [20] indicated that Zn enhanced translocation of Mn and Cu but decreased Cu absorption by the root.

8. CONCLUSION

This review mostly concerns with the role of zinc toxicity in micro- and macroflora. Zinc acts as a plant nutrient [99, 126]; but at higher concentrations it is toxic. Zinc toxicity in plants is clearly visible with the inhibition of growth and decrease in biomass production; severe toxicity can also be fatal. Zn-toxicity might be the result of complex interactions of the major toxic ions i.e. Cd^{+2} , Cu^{+2} , Pb^{+2} with Ca, Mg, Fe and P and other environmental factors. Zinc toxicity was almost certainly involved with metabolism through competition for uptake, inactivation of enzymes, displacement of essential elements from functional sites. Generally, Zn toxicity caused chlorosis and inhibited Fe translocation in some cases [3]. The physiology and biochemistry of zinc toxicity have been less studied in intact plants.

REFERENCES

- [1] Abdulla M., Nair B.M., Chandra R.K., Health effects and interactions of essential and toxic elements, *Nutr. Res.* 1 (1985) 1–751.
- [2] Adriano D.C., Trace elements in the terrestrial environment, Springer Verlag, New York, 1970.
- [3] Ambler J.E., Brown J.C., Gauch H.G., Effect of zinc on translocation of iron in soybean plants, *Plant Physiol.* 46 (1970) 320–323.
- [4] Amado-Filho G.M., Karez C.S., Andrade L.R., Yoneshigue-Valentin Y., Pfeiffer W.C., Effects on growth and accumulation of zinc in six seaweed species, *Ecotoxicol., Environ. Safety* 37 (1997) 223–228.
- [5] Antonovics J., Bradshaw A.D., Turner R.G., Heavy metal tolerance in plants, *Adv. Ecol. Res.* 7 (1971) 1–85.
- [6] Barcelo J., Poschenrieder Ch., Plant water relations as affected by heavy metal stress: a review, *J. Plant Nutr.* 13 (1990) 1–37.
- [7] Baker A.J.M., Ecophysiological aspects of zinc tolerance in *Silene maritima*, *New Phytol.* 80 (1978) 635–642.
- [8] Baker A.J.M., Metal tolerance, *New Phytol.* 106 (1987) 93–111.
- [9] Baker A.J.M., Walker P.L., Ecophysiology of metal uptake by tolerant plants, in: Shaw A.J. (Ed.), Heavy metal tolerance in plants: Evolutionary aspects, CRC Press, Boca Raton, Fla, 1990, pp. 155–172.
- [10] Baker N.R., Fernyhough P., Meek I.F., Light dependent inhibition of photosynthetic electron transport by zinc, *Physiol. Plant.* 56 (1982) 217–222.
- [11] Beckett P.H.T., Davis R.D., Upper critical levels of toxic elements in plants, *New Phytol.* 79 (1977) 95–106.
- [12] Beckett P.H.T., Davis, R.D., The additivity of the toxic effects of Cu, Ni and zinc in young barley, *New Phytol.* 81 (1978) 155–173.
- [13] Bert V., MacNair M.R., DeLaguerie P., Saumitoulaprade., Petit D., Zinc tolerance and accumulation in metallicolous and nonmetallicolous populations of *Arabidopsis halleri* (Brassicaceae), *New Phytol.* 146 (2000) 225–233.
- [14] Bjerre G.K., Schierup H.H., Uptake of six heavy metals by Oat as influenced by soil type and additions of cadmium, lead, zinc and copper, *Plant and Soil* 88 (1985) 57–69.
- [15] Bollard E.G., Butler, G.W., Mineral nutrition of plants, *Annu. Rev. Plant Physiol.* 17 (1966) 77–112.
- [16] Bradshaw A.D., McNeilly T., Evolution and Pollution, Edward Arnold, London (1981).
- [17] Brown J.C., Effect of Zn stress on factors affecting Fe uptake in navy beans, *J. Plant Nutr.* 1 (1979) 171–183.
- [18] Brown J.C., Jones W.E., Heavy metal toxicity in plants. I. A crisis in embryo, *Commun. Soil Sci. Plant Anal.* 6 (1975) 421–438.

- [19] Brown J.C., Ambler J.E., Chaney R.L., Foy C.D., Differential responses of plant genotypes to micronutrients, in: Mortvedt J.J., Giordano P.M., Lindsay W.L. (Eds.), *Micronutrients in Agriculture*, Madison, Wisconsin, Soil Sci. Soc. Am., 1972, pp. 389–418.
- [20] Cayton M.T.C., Reyes E.D., Neue H.U., Effect of zinc fertilization on the mineral nutrition of rice differing in tolerance to zinc deficiency, *Plant and Soil* 87 (1985) 319–327.
- [21] Chardonens A.N., Koevoets P.L.M., van Zanten A., Schat H., Verkleij J.A.C., Properties of enhanced tonoplast zinc transport in naturally selected zinc-tolerant *Silene vulgaris*, *Plant Physiol.* 120 (1999) 779–785.
- [22] Chavan A.S., Banerjee N.K., Fe-Zn interaction in a black loamy soil as studied on rice crop, *Ind. Soc. Soil Sci.* 28 (1980) 203–205.
- [23] Chaney P.L., White M.C., Simon P.W., Plant uptake of heavy metals from sewage sludge applied to land, in: *Proc. Natl. Conf. Munic Sludge Manage.*, pp. 167–178 (1975), Rockville, USA.
- [24] Chaney R.L., Metals in plants absorption mechanisms, accumulation, and tolerance, *Proc. Symp. Metals Biosphere*, Univ. of Guelph, Ontario, 1975, pp. 79–99.
- [25] Chaudhry F.M., Alam S.M., Rashid A., Latif A., Mechanism of differential susceptibility of two rice varieties to Zn deficiency, *Plant and Soil* 46 (1977) 637–642.
- [26] Chowdhury B.A., Chandra R.K., Biological and Health implications of toxic heavy metal and essential trace element interactions, *Progr. Food Nutr. Sci.* 11 (1987) 55–113.
- [27] Clijsters H., VanAssche F., Inhibition of photosynthesis by heavy metals, *Photosynth. Res.* 7 (1985) 31–40.
- [28] Collins J.C., Zinc, in: Lepp N.W. (Ed.), *The effect of heavy metal pollution on plants*, Vol. 1, Applied Science Publishers, 1981, pp. 145–170.
- [29] Cumming J.R., Taylor G.J., Mechanism of metal tolerance in plants: Adaptation for Exclusion of Metal ions from the cytoplasm, in: Alscher R.G., Cumming J.R. (Eds.), *Stress Responses in Plants: Adaptation and Acclimation*. Wiley-Liss, pp. 329–356 (1990), New York.
- [30] Cunnigham J.D., Keenay D.R., Ryan J.A., Phytotoxicity and uptake of metals added to soils as inorganic salts or in sewage sludge, *J. Environ. Qual.* 4 (1975) 460–462.
- [31] Davis J.G., Parker M.B., Zinc toxicity symptom development and partitioning of biomass and zinc in peanut plants, *J. Plant Nutr.* 16 (1993) 2353–2369.
- [32] Davis K.L., Davies M.S., Francis D., The effects of zinc on cell viability and on mitochondrial structure in contrasting cultivars of *Festuca rubra* L. – A rapid test for zinc tolerance, *Environ. Pollut.* 88 (1995) 109–113.
- [33] Daviscarter J.G., Shuman L.M., Influence of texture and pH of baolinitic soils on zinc fractions and zinc uptake by peanuts, *Soil Sci.* 155 (1993) 376–384.
- [34] Devlin R.M., *Plant Physiology*, Reinhold, New York, 1967, pp. 564–580.
- [35] Diaz G., Azcon-Aquilar C., Honrubia M., Influence of arbuscular mycorrhizae on heavy metal (Zn and Pb) uptake and growth of *Lygeum spartum* and *Anthyllis cytisoides*, *Plant and Soil* 180 (1996) 241–249.
- [36] Doyar M.A., Van Hai Tang, Effect of P, N and HCO₃ levels in the nutrient solution on rate of zinc absorption by rice roots and zinc content in plants, *Z. Pflanzenphysiol.* 98 (1980) 203–212.
- [37] Earley E.M., Minor element studies with soybeans. I. Varietal reactions to concentrations of zinc in excess of the nutritional requirement, *J. Am. Soc. Agron.* 35 (1943) 1012–1023.
- [38] Ernst W.H.O., *Schwermetallvegetation der Erde*, Stuttgart, G. Fischer Verlag (1974).
- [39] Ernst W.H.O., Physiology of heavy metal resistance in plants, *Proc. Int. Conf. Heavy Metals in Environ.* 94 (1977) 121–136.
- [40] Ernst W.H.O., Effects of heavy metals in plants at the cellular and organismic level, in: Schuurmann G., Markert B. (Eds.), *Bioaccumulation and Biological effects of chemicals*, John Wiley and Sons Inc. and Spektrum Akademischer Verlags, 1998, pp. 587–620.
- [41] Foy C.D., Chaney R.L., White M.C., The physiology of metal toxicity in plants, *Annu. Rev. Plant Physiol.* 29 (1978) 511–566.
- [42] Frey B., Keller C., Zierold K., Schulin R., Distribution of Zn in functionally different leaf epidermal cells of the hyperaccumulator *Thlaspi caerulescens*, *Plant Cell Environ.* 23 (2000) 675–687.
- [43] Fujii T., Presence of zinc in nucleoli and its possible role in mitosis, *Nature* 174 (1954) 1108–1109.
- [44] Garty J., Karary Y., Harel J., Effect of low pH, heavy metals and anions on chlorophyll degradation in the lichen *Ramalina duriaei* (De Not) Bagl, *Environ. Exp. Bot.* 32 (1992) 229–241.
- [45] Gebhart E., Chromosome damage in individuals exposed to heavy metals, *Toxicol. Environ. Chem.* 8 (1984) 253–266.
- [46] Gerloff G.C., Comparative mineral Nutrition of plants, *Annu. Rev. Plant Physiol.* 14 (1963) 107–124.
- [47] Gianquinto G., AbuRayyan A., Ditola L., Piccotino D., Pezzarossa B., Interaction effects of phosphorous and zinc on photosynthesis, growth and yield of dwarf bean grown in two environments, *Plant and Soil* 220 (2000) 219–228.
- [48] Godbold D.L., Huttermann A., Effect of zinc, cadmium and mercury on root elongation of *Picea abies* (Karst.) seedlings, and the significance of these metals to forest die-back, *Environ. Pollut. (Series A)* 38 (1985) 375–381.
- [49] Gregory R.P.G., Bradshaw A.D., Heavy metal tolerance in populations of *Agrostis tenuis* Sibth. and other grasses, *New Phytol.* 64 (1965) 131–143.
- [50] Grill E., Winnacker E.L., Zenk M.H., Phytochelatins, a class of heavy metal binding peptides from plants, are functionally analogous to metallothioneins, *Proc. Natl. Acad. Sci., USA* 84 (1987) 439–443.
- [51] Hacısalihoglu G., Hart J.J., Kochian L.V., High- and low-affinity zinc transport systems and their possible role in zinc efficiency in bread wheat, *Plant Physiol.* 125 (2001) 456–463.
- [52] Hall J.L., Cellular mechanisms for heavy metal detoxification and tolerance, *J. Exp. Bot.* 53 (2002) 1–11.
- [53] Hampp R., Beulron K., Ziegler H., Effects of zinc and cadmium on photosynthetic CO₂-fixation and Hill activity of isolated spinach chloroplasts, *Z. Pflanzenphysiol.* 73 (1976) 336–344.
- [54] Hertstein U., Jager H.J., Tolerances of different populations of three grass species to cadmium and other metals, *Environ. Exp. Bot.* 26 (1986) 309–319.
- [55] Hewitt E.J., in: *Metals and micronutrients: uptake and utilization by plants*, in: Robb D.A., Pierpoint W.S. (Eds.), Academic Press, London, 1983, pp. 277–300.
- [56] Hinesly T.D., Redborg K.E., Pietz R.I., Ziegler E.L., Cadmium and zinc uptake by Corn (*Zea mays* L.) with repeated applications of sewage sludge, *J. Agric. Food Chem.* 32 (1984) 155–163.
- [57] Hodgson J.F., Chemistry of the micronutrients elements in soil, *Adv. Agron.* 15 (1963) 119–159.
- [58] Hodgson J.F., Contribution of metal-organic complexing agents to the transport of metals to roots, *Soil Sci. Soc. Am. Proc.* 33 (1969) 68–75.
- [59] Jones R.G.W., Sutcliffe M., Marshall C., Physiological and biochemical basis for heavy metal tolerance in clones of *Agrostis tenuis*, in: Samish R.M. (Ed.), *Recent Advances in Plant Nutrition*, Vol. 2, New York: Gordon & Breach, 1971, pp. 275–281.
- [60] Kaya C., Higgs, D., Growth enhancement by supplementary phosphorous and iron in tomato cultivars grown hydroponically at high zinc, *J. Plant Nutr.* 24 (2001) 1861–1870.
- [61] Lasat M.M., Pence N.S., Garvin D.F., Ebbs S.D., Kochian L.V., Molecular physiology of zinc transport in the Zn hyperaccumulator *Thlaspi caerulescens*, *J. Exp. Bot.* 51 (2000) 71–79.
- [62] Lepp N.W., Dickison, N.M., Fungicide-derived copper on plantation crops, in: Ross S.M. (Ed.), *Toxic metals in soil-plant systems*, Wiley Chichester, L. K., 1994, pp. 367–393.
- [63] Levitt J., Response of plants to environmental stress, Academic Press, New York (1980).

- [64] Lindsay W.L., Inorganic phase equilibria of micronutrients in soils, in: Mortvedt J.J., Giordano P.M., Lindsay W.L. (Eds.), *Micronutrients in Agriculture*, Madison, Wisconsin, Soil. Sci. Soc. Am., 1972, pp. 389–418.
- [65] Lindsay W.L., Role of chelatin in micronutrient availability, in: Carson E.W. (Ed.), *The plant Root and its environment*. Charlottesville, University Press Virginia, 1974, pp. 508–524.
- [66] Longnecker N.E., Robson A.D., Distribution and transport of zinc in plants, in: Robson A.D. (Ed.), *Zinc in soils and plants*, Kluwer Academic Publ., Dordrecht, The Netherlands, 1993, pp. 79–91.
- [67] Lorimer G.H., The carboxylation and oxygenation of ribulose – 1,5-bisphosphate: the primary events in photosynthesis and photorespiration, *Annu. Rev. Plant Physiol.* 32 (1981) 349–383.
- [68] Lorimer G.H., Mizioro H.M., RuBP carboxylase: the mechanism of activation and its relation to catalysis, in: Akoyunoglou G. (Ed.), *Regulation of carbon metabolism. Photosynthesis. IV*. Balaban Int. Sci. Serv., Philadelphia, 1981, pp. 3–16.
- [69] Lyngby J.E., Brix H., Schierup H.H., Absorption and translocation of Zn in algrass (*Zostera marina* L.), *J. Exp. Mar. Biol. Ecol.* 58 (1982) 259–270.
- [70] MacLean A.J., Cadmium in different plant species and its availability in soils as influenced by organic matter and additions of lime, P, Cd and Zn, *Can. J. Soil Sci.* 56 (1976) 129–138.
- [71] Madhava Rao K.V., Sresty T.V.S., Antioxidative parameters in the seedlings of pigeonpea (*Cajanus cajan* (L.) Millspaugh) in response to Zn and Ni stresses, *Plant Sci.* 157 (2000) 113–128.
- [72] Malea P., Kevrekidis T., Haritonidis S., The short-term uptake of zinc and cell mortality of the sea grass *Halophila stipuleacea* (Forsk.) Aschers, *Israel J. Plant Sci.* 43 (1995) 21–30.
- [73] Marschner H., *Mineral Nutrition of Higher Plants*, Academic Press, London, 1986, pp. 300–312.
- [74] Marschner H., *Mineral Nutrition of Higher Plants*, 2nd ed., Academic Press, London, 1995.
- [75] Mathys W., Comparative investigations of the uptake of zinc by resistant and sensitive populations of *Agrostis tenuis* Sibth, *Flora* 162 (1973) 492–499.
- [76] Mathys W., The role of malate, oxalate, and mustard oil glucosides in the evolution of zinc resistance in herbage plants, *Physiol. Plant.* 40 (1977) 130–136.
- [77] Mathys W., The role of malate in zinc tolerance, *Proc. Int. Conf. Heavy Metals in Environ.* 2 (1977) 97–120.
- [78] McKenna J.M., Chaney R.L., Williams F.M., The effects of cadmium and zinc interactions on the accumulation and tissue distribution of zinc and cadmium in lettuce and Spinach, *Environ. Pollut.* 79 (1993) 113–120.
- [79] Meharg A.A., The role of plasmalemma in metal tolerance in angiosperms, *Physiol. Plant.* 88 (1993) 191–198.
- [80] Michaelis A., Takehisa R.S., Rieger R., Aurich, O., Ammonium chloride and zinc sulfate pretreatments reduce the yield of chromatid aberrations induced by TEM and maleic hydrazide in *Vicia faba*, *Mutat. Res.* 173 (1986) 187–191.
- [81] Miner G.S., Gutierrez R., King L.D., Soil factors affecting plant concentrations of cadmium, copper and zinc on sludge-amended soils, *J. Environ. Qual.* 26 (1997) 989–994.
- [82] Mukherjee A., Sharma A., Effect of pretreatment on metal cytotoxicity in plants, *Indian Bot. Repr.* 4 (1985) 76–78.
- [83] Nanson A., McElroy W.D., Modes of action of the essential mineral elements, *Plant Physiology: A Treatise*, in: Steward F.C. (Ed.), Vol. 3, Academic Press, New York, 1963, pp. 451–521.
- [84] Neumann D., Zur Nieden U., Schwieger W., Leopold I., Lichtenberger O., Heavy metal tolerance of *Minuartia verna*, *J. Plant Physiol.* 151 (1997) 101–108.
- [85] Ochi T., Ishigura T., Osakawa M., Participation of active oxygen species in the induction of DNA single-strand scissions by cadmium chloride in cultured chinese hamster cells, *Mutat. Res.* 122 (1983) 169–175.
- [86] Pearson J.N., Rengel Z., Uptake and distribution of ^{65}Zn and ^{54}Mn in wheat grown at sufficient and deficient levels of Zn and Mn. I. During vegetative growth, *J. Exp. Bot.* 46 (1995) 833–839.
- [87] Peterson P.J., The distribution of zinc in *Agrostis tenuis* Sibth and *A.stolonifera* L. tissues, *J. Exp. Bot.* 20 (1969) 863–865.
- [88] Polson D.E., Adams M.W., Differential response of navy beans to zinc. I. Differential growth and elemental composition at excessive zinc levels, *Agron. J.* 62 (1970) 557–560.
- [89] Powell M.J., Davies M.S., Francis D., Effects of zinc on cell, nuclear and nuclear size and on RNA and protein content in the root meristem of a zinc-tolerant and a non-tolerant cultivars of *Festuca rubra* L., *New Phytol.* 104 (1986) 671–679.
- [90] Rascio N., Metal accumulation by some plants growing on zinc mine deposits, *Oikos* 37 (1977) 250–255.
- [91] Rauser W.E., Estimating metallothionein in small root samples of *Agrostis gigantea* and *Zea mays* exposed to cadmium, *J. Plant Physiol.* 116 (1984) 253–260.
- [92] Rauser W.E., Phytochelatin and related peptides, *Plant Physiol.* 109 (1995) 1141–1149.
- [93] Rengel Z., Ecotypes of *Holcus lanatus* tolerant to zinc toxicity also tolerate zinc deficiency, *Ann. Bot.* 86 (2000) 1119–1126.
- [94] Rengel Z., Romheld V., Root exudation and Fe uptake and transport in wheat genotypes differing in tolerance to Zn deficiency, *Plant and Soil* 222 (2000) 25–34.
- [95] Robinson N.J., Ratcliff R.L., Anderson P.J., Delhaize E., Berger J.M., Jackson P.J., Biosynthesis of poly (Y-glutamyl-cysteinyl) glycines in cadmium resistant *Datura innoxia* cells, *Plant Sci.* 56 (1988) 197–204.
- [96] Rout G.R., Samantaray S., Das P., In vitro selection and biochemical characterization of zinc and manganese adapted callus lines in *Brassica* spp., *Plant Sci.* 137 (1999) 89–100.
- [97] Samantaray S., Rout G.R., Das P., (1999) In vitro selection and regeneration of zinc tolerant calli from *Setaria italica* L., *Plant Sci.* 143 (2000) 201–209.
- [98] Sharma A., Talukdar G., Effects of metals on chromosomes of higher organisms, *Environ. Mutagen.* 9 (1987) 191–226.
- [99] Shen Z.G., Zhao F.J., McGrath S.P., Uptake and transport of zinc in the hyperaccumulator *Thlaspi caerulescens* and the non-hyperaccumulator *Thlaspi ochroleucum*, *Plant Cell Environ.* 20 (1997) 898–906.
- [100] Shetty K.G., Hetrick B.A.D., Figge D.A.H., Schwab A.P., Effects of mycorrhizae and other soil microbes on revegetation of heavy metal contaminated mine spoil, *Environ. Pollut.* 86 (1994) 181–188.
- [101] Shier W.T., Metals as toxins in plants, *J. Toxicol.-Toxin Rev.* 13 (1994) 205–216.
- [102] Shkolnik M.Y., *Trace elements in plants*, Elsevier Science Publishers, New York, 1984, pp. 140–171.
- [103] Sinha P., Jain R., Chatterjee C., Interactive effect of boron and zinc on growth and metabolism of mustard, *Comm. Soil Sci. Plant Anal.* 31 (2000) 41–49.
- [104] Sresty T.V.S., Madhava Rao K.V., Ultrastructural alterations in response to zinc and nickel stress in the root cells of pigeonpea, *Environ. Exp. Bot.* 41 (1999) 3–13.
- [105] Staker E.V., Cummings R.W., The influence of zinc on the productivity of certain New York peat soils, *Soil Sci. Soc. Am. Proc.* 6 (1941) 207–214.
- [106] Stiborova M., Hromadkova R., Leblova S., Effect of ions of heavy metals on the photosynthesis characteristics of maize (*Zea mays* L.), *Biologia* 41 (1986) 1221–1228.
- [107] Strickland R.C., Chaney W.R., Lamoreaux R.J., Organic matter influences phytotoxicity of cadmium to soybeans, *Plant and Soil* 52 (1979) 393–402.
- [108] Subhadra A.V., Panda B.B., Metal induced genotoxic adaptation in barley (*Hordeum vulgare* L.) to maleic hydrazide and methyl mercuric chloride, *Mutat. Res.* 321 (1994) 93–102.

- [109] Symeonidis L., McNeilly T., Bradshaw A.D., Differential tolerance of three cultivars of *Agrostis capillaris* L. to Cd, Cu, Pb, Ni and Zinc, *New Phytol.* 101 (1985) 309–315.
- [110] Taylor G.J., Stadt K.J., Dale M.R.T., Modelling the phytotoxicity of aluminium, cadmium, copper, manganese, nickel and zinc using the Weibull frequency distribution, *Can J. Bot.* 69 (1991) 359–267.
- [111] Tomsett A.M., Thurman D.A., Molecular biology of metal tolerance of plants, *Plant Cell Environ.* 11 (1998) 383–394.
- [112] Tripathy B.C., Mohanty P., Zinc inhibited electron transport of photosynthesis isolated barley chloroplasts, *Plant Physiol.* 66 (1980) 1174–1178.
- [113] Turner R.G., The subcellular distribution of zinc and copper within the roots of metal-tolerant clones of *Agrostis tenuis* Sibth, *New Phytol.* 69 (1970) 725–731.
- [114] Turner R.G., Marshall C., The accumulation of ⁶⁵Zn by root homogenates of zinc tolerant and non-tolerant clones of *Agrostis tenuis* Sibth, *New Phytol.* 70 (1972a) 539–545.
- [115] Turner R.G., Marshall C., The accumulation of zinc by subcellular fractions of roots of *Agrostis tenuis* Sibth. In relation to zinc tolerance, *New Phytol.* 71 (1972b) 671–676.
- [116] Van Assche F., Physiological study of zinc toxicity on photosynthesis, Ph.D. thesis, Univ. Instelling Antwerpen, Germany, 1973.
- [117] Van Assche F., Clijsters H., Interaction of zinc at the protein level with photosynthetic light and dark reactions in *Phaseolus vulgaris*, in: Sybesma C. (Ed.), *Advances in Photosynthesis Research. Part-IV*, Nijhoff/Junk Publisher, The Hague, 1984, pp. 431–434.
- [118] Van Assche F., Clijsters H., Inhibition of photosynthesis in *Phaseolus vulgaris* by treatment with toxic concentration of zinc: effect on ribulose – 1,5-bisphosphate carboxylase/oxygenase, *J. Plant Physiol.* 125 (1986) 355–360.
- [119] Van Assche F., Clijsters H., Marcelle R., Photosynthesis in *Phaseolus vulgaris* L., as influenced by Supra-Optimal zinc nutrition, in: Marcelle R. (Ed.), *Photosynthesis and Plant Development*, W. Junk Publishers, The Hague, 1979, pp. 175–184.
- [120] Van Assche F., Ceulemans R., Clijsters H., Zinc mediated effects on leaf CO₂ diffusion conductances and net photosynthesis in *Phaseolus vulgaris* L., *Photosynth. Res.* 1 (1980) 171–180.
- [121] Verkleij J.A.C., Schat H., Mechanisms of Metal Tolerance in Plants, in: Shaw A.J. (Ed.), *Heavy Metal Tolerance in Plants: Evolutionary Aspects*, CRC Press, Boca Raton, Fla, 1990, pp. 179–193.
- [122] Wahbeh M.I., Levels of zinc, manganese, magnesium, iron and cadmium in three species of seagrass from Aqaba (Jordan), *Aquat. Bot.* 20 (1984) 179–183.
- [123] Wainwright S.J., Woolhouse H.W., Physiological mechanisms of Heavy metal tolerance, in: Chadwicks M.J., Goodman G.T. (Eds.), *The Ecology of Resource Degradation and Renewal*, Br. Ecol. Soc. Symp., Blackwell Publishers, Oxford, 15, 1976, pp. 231–257.
- [124] Wainwright S.J., Woolhouse H.W., Some physiological aspects of copper and zinc tolerance in *Agrostis tenuis* Sibth: cell elongation and membrane damage, *J. Exp. Bot.* 28 (1977) 1029–1036.
- [125] Warnock R.E., Micronutrient uptake and mobility within the corn plants (*Zea mays* L.) in relation to P-induced Zn deficiency, *Soil Sci. Soc. Am. Proc.* 34 (1970) 765–769.
- [126] Watanabe F.S., Lindsay W.L., Olsen S.K., Nutrient balance involving P, Fe and Zn, *Soil Sci. Soc. Am. Proc.* 29 (1965) 562–565.
- [127] Webber J., Trace elements in Agriculture, in: Lepp N.W. (Ed.), *Effect of Heavy Metal Pollution on Plants*, Vol. 2, Applied Sci. Publishers Ltd., London, 1981, pp. 159–184.
- [128] Welch R.M., Micronutrient nutrition of plants, *Crit. Rev. Plant Sci.* 14 (1995) 49–82.
- [129] Wheeler D.M., Power I.L., Comparison of plant uptake and plant toxicity of various ions in wheat, *Plant and Soil* 172 (1995) 167–173.
- [130] White M.C., Chaney R.L., Decker A.M., Differential varietal tolerance in soybean to toxic levels of zinc in sassafras sandy loam, *Agron. Abstr.* (1974) 144–145.
- [131] White R.E., Studies on the mineral ion absorption by plants. III. The interaction of aluminium phosphate and pH on the growth of *Medicago sativa*, *Plant and Soil* 46 (1976) 195–208.
- [132] Whiting S.N., Leake J.R., McGrath S.P., Baker A.J.M., Positive responses to Zn and Cd by roots of the Zn and Cd hyperaccumulator *Thlaspi caerulescens*, *New Phytol.* 145 (2000) 199–210.
- [133] Whitehead D.C., Some soil-plant and root-shoot relationships of copper, zinc and manganese in white clover and perennial ryegrass, *Plant and Soil* 97 (1987) 47–56.
- [134] Woolhouse H.W., Toxicity and tolerance in the responses of plants to metals, in: Lange O.L. (Ed.), *Encyclopedia of Plant Physiology*, New Series, Vol. 12, Part-C, *Physiological Plant Ecology – III*, Springer-Verlag, Berlin, 1983, pp. 245–300.
- [135] Ye Z.H., Baker A.J.M., Wong M.H., Willis A.J., Zinc, lead and cadmium tolerance, uptake and accumulation by *Typha latifolia*, *New Phytol.* 136 (1997) 469–480.