Pak. J. Bot., 43(4): 1909-1914, 2011.

TOXICITY AND TOLERANCE IN *SAMANEA SAMAN* (JACQ.) MERR. TO SOME METALS (Pb, Cd, Cu AND Zn)

M. KABIR, M. ZAFAR IQBAL AND M. SHAFIQ

Department of Botany,
University of Karachi, Karachi, 75270, Pakistan

Abstract

In this study toxicity and tolerance of *Samanea saman* (Jacq.) Merr. was investigated for Pb, Cd, Cu and Zn under lab conditions. Germination rate of *S. saman* showed that increased concentration of different metals from 25 to 100 ppm, significantly ($p<0.05$) reduced germination which was more prominent for Pb treatments. Seedling growth variables i.e. root and shoot length, seedling size, root/shoot ratio, seedling fresh and dry weights also declined significantly ($p<0.05$). Seedlings growth of *S. saman* gradually reduced with increasing in concentrations of metals especially Pb and Cd compared to Cu and Zn. The inhibitory effects of metals had the following order Pb>Cd>Cu>Zn of sequence at different concentrations. Tolerance indices determined for different metal illustrated that increasing concentrations of metals reduced the tolerance of *S. saman* but this reduction was more prominent for Pb and Cd as compared to Cu and Zn treatments.

Introduction

Increasing level of heavy metals in the environment of Karachi city is mainly due to solid refuse and domestic fuel burning. Transportation and industrial activities are the additional source of metals pollution in the environment. The indiscriminate discharge of pollutants in air, water and soil affects the growth performance of flora of the region.

Many studies have been conducted to identify plant species capable of accumulating undesirable toxic compounds such as heavy metals (Peralta-Videa *et al*., 2004). Reeves & Baker, (2000) compiled an exhaustive list of plant species that hyperaccumulated Cd, Cr, Ni, Pb, Se and Zn.

High levels of heavy metals were investigated in soil samples from various polluted areas of Karachi city (Khalid *et al*., 1996). Iqbal *et al*., (1998) carried out a survey of vegetation and trace metals (Cu, Zn and Pb) in soils along the super highway near Karachi city. Toxic metal ions enter cells by means of the same uptake processes that move essential micronutrient metal ions (Patra, *et al*., 2004). Plant and soils near the roadside had a higher concentration of Pb than at a distance of 4-6 meters. A good correlation existed between traffic volumes; total and extractable soil Pb and Pb content of roots and shoots of grass (*Cynodon dactylon)* in roadside of Delhi (Dutta & Mookerjee, 1981). Lead concentrations up to several thousands parts per million in street dirt and soils are frequently found in urban areas and near some industries (Barltrop *et al*., 1974). The major sources of lead available to plants has been the soil usually derived from weathered bedrock, parents material from lead mine, smelting operations, use of lead arsenate, use of tetraethyl and tetra methyl lead as antiknocks additive to petrol (Foy *et al*., 1978). Roadside trees in the city are under pressure and lost due to vehicular-traffic infrastructure and other community needs such as industrial products (Jim, 1998). Deposition of lead on the vegetation growing along the roads not only affected growth and germination but also caused a significant reduction in seed and fruit production of plants (Nasralla & Ali 1985; Ahmad *et al*., 2009). Foliar application of lead nitrate solution resulted in a reduction in various indices and yield parameters of wheat (Rashid & Mukherji 1993). Lead induces many biochemical and structural changes in biological systems (Minaii *et al*., 2008). Seedlings of tomato plants grown in pots, treated with lead nitrate solution at 500 and 1000 ppm showed pronounced effect in the root system as compared to leaves and stem (Jaffer *et al*., 1999).

Cadmium is strongly phytotoxic causing growth inhibition and even plant death, alterations in activated oxygen metabolism and cell disturbances (Sandalio *et al*., 2001). Plants grown under high levels of Cu normally showed reduced biomass and chlorotic symptoms (Yruela, 2005). Excess of Cu affects the aerial part as well as root growth, inhibiting cellular elongation due to the increase in plasmalemma permeability and cell wall lignification (Arduini *et al*., 1995). Inhibition of root growth is recognized as one of the most conspicuous symptoms of Cu toxicity (Kukkola *et al*., 2000), in which lateral development and elongation are more sensitive than root initiation (Kahle, 1993; Woolhouse, 1983). They reported that the toxicity and tolerance in plants is a response to heavy metals including cobalt and zinc. Higher concentrations of zinc reduced the germination and growth of *Pennisetum americanum* and *Parkinsonea aculeata*. High concentrations of Zn have been reported in some plant species growing along the Super Highway near Karachi (Iqbal & Shafiq, 1999).

Adaptation is a change in the structure or functioning of an organism that makes it better appropriate to environmental stresses (Jules & Shaw, 1994). Lead (Pb) and copper (Cu) mines produce tons of tailings (wastes) containing excess of elements that are toxic for living organisms, even at low concentrations. However, certain plants as bent grass (*Agrotis tenuis*), that grows in mine wastes, evolved tolerance to heavy metals in 400 years of mining (Jules & Shaw, 1994). Plants have both constitutive (present in most phenotypes) and adaptive (present only in tolerant types) mechanisms for coping with elevated metal concentrations (Mehrag, 1994). Metal tolerance is an evolutionary phenomenon that can be demonstrated by comparing the growth of mine plants with non-mine plants in non-contaminated soil (Haque *et al*., 2009).

Samanea saman (Jacq.) Merr. belongs to family Leguminosae and sub-family Mimosoideae, normally known as rain tree commonly having 25 m height with rough, fissured bark, bipinnately compound, alternate leaves and commonly planted for shade and wood. In Fiji, it is found from near sea level to an elevation of 700 m, cultivated and sometimes abundantly naturalized along roadsides, on river banks and in forests (Smith, 1985).

*E-mail: kabir_botany82@yahoo.com

The purpose of this study was to examine the effect of heavy metals on seed germination, seedling growth and seedling dry weight under normal and stress conditions in *S. saman*.

Materials and Methods

Healthy seeds of *Samanea saman* were collected randomly from Karachi University Campus. The top ends of seeds were slightly cut with a clean scissor to remove any possible seed coat dormancy. Then seeds were surface sterilized with 30% dilute solution of sodium-hypo chlorite to prevent any fungal contamination. Petri dishes and filter papers were also sterilized in an autoclave to reduce the chances of fungal growth. Ten seeds were placed on filter paper (Whatman No. 42) in petri dishes (90 mm diameter). Metal treatments were prepared using lead nitrate, cadmium nitrate, copper sulfate and zinc nitrate having 25, 50, 75 and 100 ppm concentrations.

At the start of the experiment, 3 ml of distilled water and 3 ml of each metal solution of lead nitrate, cadmium nitrate, copper sulfate and zinc nitrate were added to each set of respective treatment. Later on every alternate day fresh treatment of 2 ml solution was given to each set of petri dishes while, distilled water was applied to control treatment before removing the old solution in order to avoid turgidity of seed. The experiment was completely randomized based on 3-replicates. Petri dishes were kept at room temperature (20±2°C) with 250 lux light intensity and the experiment lasted for 12 days. Seed germination, root, shoot, seedling lengths, seedling fresh weight were recorded and dry biomass was determined by placing the seedling in an oven at 80°C for 24 hours. Seedling fresh and dry biomass was measured by electrical balance. Seedling vigor index (S.V.I) was determined as per the formula given by Bewly & Black (1982). Tolerance indices (T.I.) were determined as mentioned by Iqbal & Rahmati (1992). The seed germination and seedling growth were statistically analyzed at $p<0.05$ level of significance on personal computers SPSS version 13.

Results

Metals treatments reduced seed germination and seedling growth of *S. saman* at different concentrations. At 100 ppm of lead, cadmium, copper and zinc treatments, the rate of germination percentage was 53 %, 56%, 60% and 66%. Seedling size (root + shoot length), seedling fresh and dry weight showed reduction when treated with increased concentration of lead from 25 to 100 ppm (Table 1 & Fig. 1). The seed germination of *S. saman* was significantly ($p<0.05$) reduced at 25 ppm lead treatment while, reduction was more prominent at 100 ppm of lead treatment. Similar reductions were also noted for root dry weight and seedling dry weight of *S. saman*. Seedling length and dry weight at 100 ppm lead treatment was 5.76 cm and 262 mg, respectively.

Cadmium, copper and zinc treatments with different concentrations also reduced seedling growth parameters. Significant differences in seed germination and seedling growth were noted for Cd treatment at 25 ppm in *S. saman* as compared to control (Table 2). Cd concentrations at 50 ppm markedly decreased shoot growth, root/shoot ratio and seedling growth as compared to control. Cd treatment at 75 ppm found responsible for significantly ($p<0.05$) reduction in roots growth. It was noted that with an application of Cd at 100 ppm, 45 % reduction in seedling fresh and dry weights was observed as compared to control.

Toxicity of copper treatment for *S. saman* is summarized in Table 3. In control, root length was recorded as 5.46 cm which was significantly ($p<0.05$) reduced to 3.40 and 2.60 cm at 75 and 100 ppm Cu treatments, respectively. Similarly, shoot length, seedling size, root/shoot ratio, seedling fresh and dry weights also declined when concentration of Cu was increased from 25 to 100 ppm.

Significant ($p<0.05$) reduction in seed germination and seedling growth of *S. saman* were noted with 25 ppm Zn as compared to control. An increase in zinc concentration up to 50 ppm markedly decreased shoot growth (Table 4). Further increase in Zn treatment up to 75 ppm found responsible for reduction in roots growth (4.18 cm). Results illustrated that application of Zn at 100 ppm high percentage of reduction (58 %) in seedling dry weight was observed as compared to others metal treatment with same concentration.

Seedling vigor index (SVI) is the potential of seed germination and seedling size against the toxicity and tolerance of metals. Results indicated that *S. saman* showed reduction in SVI with increasing concentration of different metal treatments but this reduction was more prominent for Pb and Cd as compared to Cu and Zn. Increased concentrations of Pb and Cd from 25 to 100 ppm caused high reduction in SVI of *S. saman*. Seedling of *S. saman* showed low percentage of tolerance to Pb, Cd, Cu and Zn treatment as compared to control. The order of decrease in tolerance indices was recorded as Pb>Cd>Cu>Zn (Fig. 2).

Discussion

Excess level of heavy metals in the environment can produce toxic effects to plants. Lead, cadmium, copper and zinc toxicity has become important due to their constant increase in the environment. Present study revealed that metal treatments produced toxic effects on seed germination and seedling growth of *S. saman*.

The plant species under stress conditions are most likely to be adversely affected by metals. Metal sensitivity and toxicity are influenced by the concentration range of the toxicant, the length of the exposure period, and the life-stage or biological process (Ernst & Nelissen, 2000). It is illustrated from the results that *S. saman* showed higher sensitivity to Pb toxicity as compared to other metals treatment (Tables 1-4). This sensitivity might be due to low tolerance to Pb and more accumulation of the substrate. Our study is also supported by the work of some others workers as increasing concentrations have deleterious affects on plant growth. Farooqi *et al*., (2009) reported that lead treatments at 10, 30, 50, 70 and 90 μmol^{-L} concentrations produced significant ($p<0.05$) effects on seed germination and seedling length of *Albizia lebbeck* while lead treatment at 50 μmol^{-L} significantly affected root growth and seedling dry biomass as compared to control.

Table 1. Effects of lead on different growth variables and Seedling Vigor Index (SVI) of *Samanea saman*.

Treatment lead (Pb) ppm	Germination (%)	Root length (cm)	Shoot length (cm)	Seedling size (cm)	Root/shoot ratio	Seedling fresh weight (mg)	Seedling dry weight (mg)	SVI
00	83.33 ± 3.33a	5.46 ± 0.12a	7.56 ± 0.48a	11.70 ± 0.50a	0.72 ± 0.03a	697 ± 97.75a	461 ± 38.55a	974
25	70.00 ± 5.77ab	3.53 ± 0.46b	5.80 ± 0.26b	9.33 ± 0.33b	0.61 ± 0.09ab	493 ± 16.07b	372 ± 5.04ab	653
50	66.66 ± 6.66bc	2.73 ± 0.34bc	5.73 ± 0.52b	8.46 ± 0.68bc	0.48 ± 0.06bc	464 ± 8.09b	345 ± 6.17b	563
75	63.33 ± 3.33bc	2.23 ± 0.09cd	5.23 ± 0.20bc	7.46 ± 0.24cd	0.43 ± 0.02c	408 ± 97.14b	299 ± 63.26b	472
100	53.33 ± 3.33c	1.70 ± 0.26d	4.06 ± 0.55c	5.76 ± 0.82d	0.41 ± 0.03c	360 ± 19.34b	262 ± 7.05b	307

Table 2. Effects of cadmium on different growth variables and Seedling Vigor Index (SVI) of *Samanea saman*.

Treatment lead (Pb) ppm	Germination (%)	Root length (cm)	Shoot length (cm)	Seedling size (cm)	Root/shoot ratio	Seedling fresh weight (mg)	Seedling dry weight (mg)	SVI
00	83.33 ± 3.33a	5.46 ± 0.12a	7.56 ± 0.48a	11.70 ± 0.50a	0.72 ± 0.03a	697 ± 97.75a	461 ± 38.55a	974
25	73.33 ± 3.33ab	4.06 ± 0.32b	6.78 ± 0.08b	10.84 ± 0.29b	0.58 ± 0.06b	723 ± 15.93b	526 ± 4.91aa	795
50	70.00 ± 5.77b	3.33 ± 0.23c	6.43 ± 0.18b	9.76 ± 0.41b	0.52 ± 0.04b	657 ± 12.89c	458 ± 10.83b	683
75	66.66 ± 3.33bc	2.33 ± 0.09d	5.21 ± 0.15c	7.54 ± 0.23c	0.47 ± 0.04b	563 ± 8.98d	417 ± 8.18b	503
100	56.66 ± 3.33c	2.30 ± 0.06d	5.16 ± 0.14c	7.46 ± 0.18c	0.44 ± 0.01b	484 ± 14.81e	339 ± 34.02c	423

Fig. 1. Effects of lead, Cadmium, Copper and Zinc on seed germination and seedling growth of *Samanea saman* at different concentrations in lab conditions.

Table 3. Effects of copper on different growth variables and Seedling Vigor Index (SVI) of *Samanea saman*.

Treatment Copper (Cu) ppm	Germination (%)	Root length (cm)	Shoot length (cm)	Seedling size (cm)	Root/shoot ratio	Seedling fresh weight (mg)	Seedling dry weight (mg)	SVI
00	83.33 ± 3.33a	5.46 ± 0.12a	7.56 ± 0.48a	11.70 ± 0.50a	0.72 ± 0.03a	697 ± 97.75a	461 ± 38.55a	974
25	76.66 ± 3.33a	4.70 ± 0.06b	6.87 ± 0.06b	11.57 ± 0.10b	0.68 ± 0.05a	729 ± 15.09b	531 ± 4.70a	886
50	73.33 ± 3.33ab	3.87 ± 0.03c	6.71 ± 0.16b	10.58 ± 0.15c	0.57 ± 0.02b	662 ± 12.41c	463 ± 10.81b	775
75	70.00 ± 5.77ab	3.40 ± 0.06d	6.41 ± 0.11b	9.81 ± 0.15d	0.53 ± 0.05b	569 ± 9.56d	421 ± 9.06b	686
100	60.00 ± 5.77b	2.60 ± 0.05e	5.17 ± 0.14c	7.77 ± 0.13e	0.50 ± 0.21b	494 ± 14.57e	350 ± 33.22c	466

Table 4. Effects of zinc on different growth variables and Seedling Vigor Index (SVI) of *Samanea saman*.

Treatment Zinc (Zn) ppm	Germination (%)	Root length (cm)	Shoot length (cm)	Seedling size (cm)	Root/shoot ratio	Seedling fresh weight (mg)	Seedling dry weight (mg)	SVI
00	83.33 ± 3.33a	5.46 ± 0.12a	7.56 ± 0.48a	11.70 ± 0.50a	0.72 ± 0.03a	697 ± 97.75a	461 ± 38.55a	974
25	81.23 ± 3.33ab	4.74 ± 0.57b	6.91 ± 0.05b	11.65 ± 0.09b	0.68 ± 0.003b	724 ± 13.53b	527 ± 5.57a	970
50	80.00 ± 5.77ab	4.33 ± 0.05c	6.60 ± 0.09c	11.00 ± 0.11c	0.65 ± 0.003c	659 ± 12.85c	458 ± 10.26b	880
75	73.33 ± 3.33bc	4.18 ± 0.03c	6.42 ± 0.13c	10.60 ± 0.15d	0.65 ± 0.01c	566 ± 8.56d	417 ± 8.65b	777
100	66.66 ± 3.33c	3.42 ± 0.06d	5.37 ± 0.11d	8.79 ± 0.17e	0.63 ± 0.003c	491 ± 14.25e	345 ± 34.00c	585

Number followed by the same letters in the same column are not significantly different according to Duncan Multiple Range Test at $p<0.05$ level.
± Standard Error

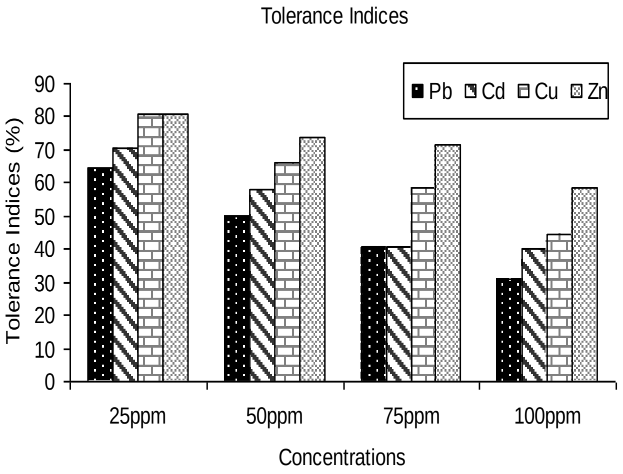

Fig. 2. Tolerance indices (%) in *Samanea saman* at different concentration of Pb, Cd, Cu and Zn.

Nriagu & Pacgana (1988) reported that about 220,00 tones of cadmium is globally discharged every year in to the soil. Cadmium is one of the most toxic elements and it affects plant growth adversely. Our results illustrated that Cd tretment adversely affected the germination and growth of *S. saman*. Cadmium is easily taken up by plants and translocated to different plant parts due to which it causes significant reduction in different growth parameters. In an investigation, Cd treatment at 10 μmol^{-L} concentration produced toxic effects on seed germination, seedling and root growth. Increase in cadmium concentration up to 50 μmol^{-L} produced a significant reduction in seedling dry weight of *Thespesia populnea* as compared to control (Kabir *et al*., 2008). Some other investigations were carried out by a number of workers for Cd uptake and toxicity which are in support of our findings. Patterson (1977) reported that plant species and even genotypes differ greatly in their ability to take up, transport and accumulate Cd within the plant. Muramoto *et al*., (1990) reported that root and shoot weights of rice were reduced 32% and 21% by 100 mg Cd kg^{-1}. Genotypical variations in tolerance to Cd toxicity are well documented in the literature (Grant *et al*., 1998; Ozturk *et al*., 2003). Results of similar nature were also found on *Brassica juncea*, genotypes (Varuna and DHR-9504) in relation to the effect of cadmium (Cd) on growth, yield and levels of cadmium (Cd) in different plant parts. Increased Cd supply in the form of $CdCl_2$ @ 0, 20, 40, 60, 80 and 100 mg Cd kg^{-1} soil showed Cd phytotoxicity by growth retardation of Varuna and DHR-9504. Varuna showed greater sensitivity to Cd toxicity than DHR-9504 (Chaturvedi, 2004). Marschner *et al.,* 1988 also demonstrated Cd toxicity by growth retardation and leaf chlorosis in Chinese cabbage. The reduction observed in dry matter yield by Cd application is in agreement with the findings of Lehoczky *et al*., (1996). Yield reductions in mustard plants have been attributed to the direct effect of higher Cd concentrations in plant tissue and not through an indirectly induced deficiency of other nutrients (Wilson, 1992). Reduction in shoot and root growth of *S. saman* might be due to low tolerance limit to cadmium stress. Marschner *et al*., (1988) and Ozturk *et al*. (2003) also found reduction in shoot and root dry matter to a similar extent due to Cd treatment.

S. saman also showed reduction in seed germination and seedling growth due to toxicity of copper treatments. Phototoxic responses to Cu have been observed for many plant species (Woolhouse & Walker 1981) and the toxic symptoms and yield reductions observed are thought to be a consequence of tissue damage, alteration of membrane permeability, peroxidation of chloroplast membrane lipids and inhibition of photosynthetic electron transport, and disruptions to carbohydrate metabolism and protein synthesis (Droppa & Horvath, 1990; Woolhouse, 1983). Zinc is an important element for plant growth. Excess Zn reaching the shoot may also be excreted via the salt glands on the leaves (MacFarlane & Burchett, 1999). Seed germination and seedling growth responses of *S. saman* with excess Zn were also observed. High tolerance to zinc treatment in seedling of *S. saman* indicated that it has better absorbing ability than lead, cadmium and copper.

Results indicated that overall, more reduction was observed in the seed germination and seedling growth of *S. saman* when treated with Pb and Cd as compared to Cu and Zn. Root length was found more susceptible to all the metal treatments as compared to shoot. Results illustrated that tolerance mechanism of *S. saman* to different metals decreased as Zn>Cu>Cd>Pb. Reduced seedling vigor index was possessed by *S. saman* with increasing concentration of different metals from 25 to 100 ppm probably due to less tolerance.

Conclusion

Heavy metal stress is one of the major problems affecting productivity of plants. Natural flora show relative differences in their heavy metal tolerance capacity. Some plants grow well in a soil enriched with toxic levels of heavy metals while others could not grow

(Yadev, 2010). Plants have proven particularly affective for both bioremediation of metal contaminated sites and for studying the uptake, transport and toxicity of metals in plants (Tug *et al.*, 2010; Hogan, 2011). It is concluded that Pb, Cd, Cu and Zn treatment was strongly phytotoxic to seed germination and seedling growth of *S. saman*. High concentrations of metal treatment found responsible for decreasing the percentage of tolerance indices in seedling of *S. saman.* The level of plant tolerance to heavy metals is related to the balance between the rate at which metal ions are taken up and the efficiency with which they are detoxified within the plant (Antosiewicz, 2005; Azmat *et al.*, 2009; Malik *et al.*, 2010). Metal uptake and translocation to shoots might be the cause of reduction in seedling growth and biomass production.

Acknowledgements

We are highly grateful to Higher Education Commission (HEC) of Pakistan for funding this study which is a part of project No. 20-1073/07. The assistance of Mr. Zia-ur-Rehman Farooqi for collection and analysis of data is also acknowledged.

References

Ahmad, M.S.A., M., Hussain, M., Ashraf, R. Ahmad, and M.Y. Ashraf. 2009. Effect of nickel on seed germinability of some elite sunflower (*Helianthus annuus* L.) cultivars. *Pak. J. Bot.*, 41(4): 1871-1882.

Antosiewicz, D.M. 2005. Study of calcium-dependent lead-tolerance on plants differing in their level of Ca-deficiency tolerance. *Environmental Pollution,* 134 (1): 23-34.

Arduini, I., D.L. Godbold and A. Onnis. 1995. Influence of copper on root growth and morphology of *Pinus pinea* L. and *Pinus pinaster* Ait. seedlings. *Tree Physiology*, 15: 411-415.

Azmat, R., S. Haider, H. Nasreen, F. Aziz and M. Riaz. 2009. A viable alternative mechanism in adapting the plants to heavy metal environment. *Pak. J. Bot.,* 41(6): 2729-2738.

Barltrop, D., D.D. Strehlow, I. Thornton and J.S. Webb. 1974. Significance of high soil lead concentrations for childhood lead burdens. *Environment Health,* 7: 75-82.

Bewly, J.D. and B.M. Black. 1982. Germination of seeds. In: A.A. Khan. (Ed.), Physiology and biochemistry of seed germination. Springer Verlag, New York, pp. 40-80.

Chaturvedi, I. 2004. Phytotoxicity of cadmium and its effect on two genotypes of *Brassica juncea* L. *American Journal of Agricultural Science*, 16(2): 01-08.

Droppa, M. and G. Horvath. 1990. The role of Cu in photosynthesis. *Critical reviews in Plant Sciences*, 9(2): 111-124.

Duncan, D.B. 1955. Multiple ranges and multiple F-tests. *Biometrics,* 11: 1-42.

Dutta, I. and A. Mookerjee. 1981. Lead in the soil and grass along roadsides of Delhi, India. *Proceeding of Indian National Science Academy* (*Biological Science),* 47: 58-64.

Ernst, W.H.O., J.A.C. Verkleij and H. Schat. 1992. Metal tolerance in plants. *Acta Botanica Neerlandica*, 41: 229-248.

Farooqi, Z.R., M. Z. Iqbal, M. Kabir and M. Shafiq. 2009. Toxic effects of lead and cadmium on germination and seedling growth of *Albizia lebbeck* (L.) Benth. *Pakistan Journal of Botany,* 41(1): 27-33.

Foy, C.D., R.L. Chaney and M.C. White. 1978. The physiology of metal toxicity in plants. *Annual Review of Plant Physiology,* 29: 511-566.

Grant C.A., W.T. Buckley, L.D. Bailey and F. Selles. 1998. Cadmium accumulation in crops. *Canadian Journal of Plant Science,* 78: 1-17.

Haque, N., J.R. Peralta-Videa, M. Duarte-Gardea and J.L. Gardea-Torresdey. 2009. Differential effect of metals/metalloids on the growth and element uptake of mesquite plants obtained from plants grown at a copper mine tailing and commercial seeds. *Bio-resource Technology,* 100 (24): 6177-6182.

Hogan, C.M. 2011. Heavy metals in plants. In "The Encyclopedia of Earth". http://www. eoearth.org/ article/Heavy_metal?topic=49498. Retrieved on 07-03-2011.

Iqbal, M.Z. and K. Rahmati. 1992. Tolerance of *Albizia lebbeck* to Cu and Fe application. *Ekologia*, 11: 427-430.

Iqbal, M.Z. and M. Shafiq. 1999. Impact of vehicular emission on germination and growth of Neem (*Azadirachta indica*) tree. *Hamdard Medicus* 42(4): 65-69.

Iqbal, M.Z., A.K. Sherwani and M. Shafiq. 1998. Vegetation characteristics and trace metals (Cu, Zn and Pb) in soils along the super highways near Karachi, Pakistan. *Studia Botanica Hungarica,* 29: 79-86.

Jaffer, T.M.R., E.A. Eltayeb, S.A. Farooq and S.A. Albahry. 1999. Lead pollution levels in Sultanate of Oman and its effect on plant growth and development. *Pakistan Journal of Biological Sciences*, 2: 25-30.

Jim, C.Y. 1998. Roadside trees in urban Hong Kong: Part II species composition. *Arboricultural Journal,* 20: 279-298.

Jules, E.S. and A.J. Shaw. 1994. Adaptation to metal contaminated soils in populations of the moss, *Ceratodon purpureus*: vegetative growth and reproductive expression, *American Journal of Botany,* 81: 791-797.

Kabir, M., M. Z. Iqbal, M. Shafiq and Z.R. Farooqi. 2008. Reduction in germination and seedling growth of *Thespesia populnea* L., caused by lead and cadmium treatments. *Pak. J. Bot*., 40(6): 2419-2426.

Kahle, H. 1993. Response of roots of trees to heavy metals. *Environmental Experimental Botany*, 33: 99-119.

Khalid, F., M.Z. Iqbal and M.S. Qureshi. 1996. Concentration of heavy metals determined in leaves and soil from various areas of Karachi city. *Environmental Science,* 4: 213-219.

Kukkola, E., P. Rauti and S. Huttunen. 2000. Stress indicators in copper- and nickel-exposed Scots pine seedlings. *Environmental Experimental Botany*, 43: 197-210.

Lehoczky, E., I. Szabados, and P. Martha. 1996. Cadmium content of plants as affected by soil cadmium concentration. *Soil Science and Plant Analysis*, 27: 1765-77.

MacFarlane, G. R. and M. D. Burchett. 1999. Zn distribution and excretion in the leaves of the Grey Mangrove *Avicennia marina* (Forsk.) Veirh. *Environmental and Experimental Botany*, 41: 167-175.

Malik, R.N., S.Z. Husain and I. Nazir. 2010. Heavy metal contamination and accumulation in soil and wild plant species from industrial area of Islamabad, Pakistan. *Pak. J. Bot.*, 42(1): 291-301.

Marschner, A., G.R. Andersen and J.K. Mason. 1988. Yield and uptake of cadmium and zinc by vegetables grown in soil polluted with heavy metals. *Swedish Journal of Agricultural Research,* 8: 74-79.

Mehrag, A.A. 1994. Integrated tolerance mechanisms: constitutive and adaptive plant responses to elevated metal concentrations in the environment, *Plant Cell Environment,* 17: 989-993.

Minaii B., M. Abdollahi and Z. Towfighi. 2008. Toxicity of lead acetate on rabbit arteries: A histological evaluation. *Toxicology,* 180: 53.

Muramoto, S., H. Nishizaki and I. Aoyama. 1990. The critical levels and the maximum metal uptake for wheat and rice plants when applying metal oxides to soil. *Journal of Environmental Science and Health, Part B.,* 25(2): 273-280.

Nasralla, M.M. and E.A. Ali. 1985. Lead accumulation in edible proteins of crops grown near Egyptian traffic roads. *Agriculture, Ecosystems and Environment*, 13: 73-82.

Nriagu, J.O. and J.M. Pacgana. 1988. Quantitative assessment of worldwide contamination of air, water and soils of trace metals. *Nature,* 333: 134-139.

Ozturk, L., S. Eker and F. Ozkutlu. 2003. Effect of cadmium on growth and concentrations of cadmium, ascorbic acid and sulphydryl groups in durum wheat cultivars. *Turkish Journal of Agriculture,* 27: 161- 168.

Patra, M., N. Bhowmik, B. Bandopadhyay and A. Sharma. 2004. Comparison of mercury, lead and arsenic with respect to genotoxic effects on plant systems and the development of genetic tolerance. *Environmental and Experimental Botany,* 52(3): 199-223.

Patterson, O. 1977. Differences in cadmium uptake between plant species and cultivars. *Swedish Journal of Agricultural Research,* 7: 21-24.

Peralta-Videa, J.R., G. D. Rosa, J. H. Gonzalez and J. L. Gardea-Torresdey. 2004. Effects of the growth stage on the heavy metal tolerance of alfalfa plants. *Advances in Environmental Research,* 8(3-4): 679-685.

Prasad, M.N.V. 1995. Cadmium toxicity and tolerance in vascular plants. *Environmental and Experimental Botany*, 35(4): 525-545.

Rashid P. and S. Mukherji. 1993. Effect of foliar application of lead on the growth and yield parameters of wheat. *Pakistan Journal of Scientific and Industrial Research*, 36: 473-475.

Reeves, R.D. and A.J.M. Baker. 2000. Metal-accumulating plants. In: *Phytoremediation Toxic Metals: Using Plants Clean Up Environment*, (Eds.): I. Raskin and B.D. Ensley. John Wiley and Sons, New York pp. 193-229.

Sandalio, L.M., H.C. Dalurzo, M. Gómez, M.C. Romero-Puertas and L.A. del Río. 2001. Cadmium-induced changes in the growth and oxidative metabolism of pea plants, *Journal of Experimental Botany*, 52: 2115-2126.

Smith, C. A. 1985. Flora Vitiensis nova: a new flora of Fiji. *National Tropical Botanical Garden, Lawai, Kauai, Hawaii*, 3-758 pp.

Tug G.N. and F. Duman. 2010. Heavy metal accumulation in soils around a salt lake in Turkey. *Pak. J. Bot.*, 42(4): 2327-2333.

Wilson, N.J. 1992. Accumulation of cadmium in crop plants and its consequences to human health. *Agronomy,* 51: 173-212.

Woolhouse, H. M. and S. Walker. 1981. The physiological basis of copper toxicity and copper tolerance in higher plants. In: *Copper in soils and plants*. (Eds.): J.F. Loneragan, A.D. Robson, & R.D. Graham. Pp. 235. New York: Academic Press.

Woolhouse, H.W. 1983. Toxicity and tolerance of plants to heavy metals. *Encyclopedia of Plant Physiology*, 12: 246-300.

Yadav, S.K. 2010. Heavy metals toxicity in plants: An overview on the role of glutathione and phytochelatins in heavy metal stress tolerance of plants. *South African Journal of Botany,* 76(2): 167-179.

Yruela, I. 2005. Copper in plants. *Brazilian Journal of Plant Physiology*, 17: 145-156.

(Received for publication 16 June 2010)