Effects of lead on seedling growth of *Thespesia populnea* L.

M. Kabir, M.Z. Iqbal, M. Shafiq, Z.R. Farooqi

Department of Botany, University of Karachi, Karachi, Pakistan

ABSTRACT

The effects of lead on root, shoot and seedling length, leaf area, number of leaves, plant circumference, seedling dry weight, root/shoot and leaf area ratios of *Thespesia populnea* L. were determined in greenhouse under natural environmental conditions with and without phytotoxic metal ions at 5, 10, 15, 20, and 25 µmol/l. Lead treatments have a strong influence on the growth and development of *T. populnea* by reducing significantly ($P < 0.05$) all the above parameters. Lead treatment at 5–25 µmol/l produced significant ($P < 0.05$) effects on seedling and root length, plant circumference and seedling dry weight of *T. populnea,* while lead treatment at 10–25 µmol/l produced significant ($P < 0.05$) effects on shoot length, number of leaves and leaf area as compared to control. Tolerance in *T. populnea* seedling at 25 µmol/l of lead treatment was lowest as compared to all other treatments.

Keywords: lead; lead pollution; *Thespesia populnea*; toxicity

Heavy metals play an important but dual role in plant metabolism. On one hand, some of them are essential micronutrients acting, for example, as co-factors of key metabolic enzymes. On the other hand, when exceeding their critical concentrations, the same metals become the most toxic pollutants in the soil (Stobrawa and Lorenc-Plucińska 2007). Pollution by toxic elements is a worldwide problem. Toxic pollutants are discharged in the air by man made activities (Nriagu and Pacyna 1988). Mining operations, metal smelting, electroplating, gas exhaust, energy and fuel production power lines, intense agriculture are some of the numerous human activities that contain quantities of toxic metals (Kumar et al. 1995). Once released into the environment they are not broken-down into harmless components. The main sources of heavy metals in plants are their growth media, nutrients, agro inputs and soil. Other sources may include pesticides and fertilizers (Seenivasan et al. 2008). The hazard and fast industrial growth is causing an enormous environmental pollution affecting plants. Vegetation differs from area to area depending on climatic factors and soil characteristics. Polluted soil can alter plant growth and quality, and the effects are often destructive (Sagar et al. 1982).

In industrial city like Karachi, vehicular emission is also a major source of atmospheric pollution (Qadir and Iqbal 1991). Dense traffic releases detrimental exhaust gases and toxic pollutants like unburnt and partially burnt hydrocarbons and other elements that are contained in petrol polluting the city environment (Iqbal et al. 2001). These air pollutants may interfere with the biological processes relating to general metabolism, photosynthetic activities, mitochondrial respiration and stomatal clogging of plants (Miles et al. 1972, Ahmed and Qadir 1975, Abdullah and Iqbal 1991).

Among the heavy metals Pb is a highly toxic pollutant. It is generally added to the environment through automobile exhaust (Lagerwerff and Specht 1970), fertilizer impurities (William and David 1973) and through industrial effluents (Campbell 1976). Lead is the heaviest of the non-radioactive metals that also naturally occur in substantial quantities in the earth's surface. It is present in all soils, rivers, lakes and sea water. It is also a component of dust, rubber, paints, metal products (steel and brass) and lead batteries. Plants absorb lead through soil, water and air. Besides the uptake from the soil and water, Pb may enter the plant surface through aerial parts including leaf surface. Deposition of lead on the vegetation growing along the roads not only effects growth and germination but also causes a significant reduction in seed and fruit production of plants

(Nasralla and Ali 1985). Foliar application of lead nitrate solution resulted in a reduction in various indices and yield parameters of wheat (Rashid and Mukherji 1993). Lead is a toxic environmental contaminant that induces many biochemical and structural changes in biological systems (Minaii et al. 2008).

Thespesia populnea is a member of family Malvaceae with basically tropical and subtropical distribution. It is used as a shade tree and as a wind break in order to control the soil erosion.

In view of the destructive role of heavy metals derived from the vehicular emission and metal processing industries, an investigation was carried out to determine the toxic effects of lead on the seedling growth of *T. populnea*.

MATERIALS AND METHODS

The experiment was conducted in a greenhouse under the uniform natural environmental conditions at the Department of Botany, University of Karachi during March and April, 2007. Healthy and uniform size seeds of *Thespesia populnea* were collected from the University Campus. The top ends of seeds were slightly cut with the clean scissor to break dormancy and were sown in garden soil at 1 cm depth in large pots and watered regularly. After two weeks of their germination, uniform sized seedlings were transplanted in pots of 20 cm diameter and 9.8 cm in depth in garden soil. A metal treatment of Pb was applied as lead nitrate at 0, 5, 10, 15, 20 and 25 µmol/l, respectively. There were five replicates for each treatment and experiment was completely randomized. Only one seedling was grown in each pot and the plants were regularly treated with 3 ml of their respective treatment. Every week reshuffling of pots was also carried to avoid light/shade or any other green house effects. Daily climatic data, as average atmospheric temperature, atmosphere relative humidity, sun shine and weather outlook were recorded (Table 1). Seedling height, number of leaves, leaf area and plant circumference were noted after every week. After eight weeks of growth, seedlings were taken out from pots, their roots were washed with water and root length, shoot length, seedlings length (which included length of root and shoot), plant circumference, number of leaves and leaf area were measured. Seedlings were dried in an oven at 80°C for 24 h and oven-dry weights of seedlings were determined. Tolerance indices (T.I.) were determined as mentioned by Iqbal and Rahmati (1992).

T.I. = (Mean root length in metal solution/Mean root length in distilled water) × 100

RESULTS

Seedling growth of *Thespesia populnea* decreased with phytotoxic metal ions treatments (Pb) at 5, 10, 15, 20 and 25 µmol/l as compared to control. Data records on seedling growth of *T. populnea* exposed to different concentrations of Pb are presented in Table 2. The results clearly indicate that all the growth parameters were suppressed gradually with the increase in concentrations of lead. A marked inhibition in seedling length (17.32 cm) became evident at 25 µmol/l concentration of Pb.

Table 1. Weekly climatic data of Karachi during the Growth of *Thespenia populnea* (1.3.2007–30.4.2007)

Dates		Maximum temp. (°C)	Minimum temp. (°C)	Average temp. (°C)	Humidity (%)	Sun shine (h)	Weather (out look)
01	7.3.07	30	19	25	38	11:45	fair/hazy morning
08	14.3.07	29	18	24	42	11:54	fair/hazy morning
15	21.3.07	26	16	21	32	12:04	fair/partly cloudy
22	28.3.07	28	18	23	28	12:16	fair/partly cloudy
29	4.4.07	34	21	28	38	12:25	hot and dry
5	11.4.07	38	24	31	39	12:36	hot and dry
12	18.4.07	35	25	30	39	12:45	warm and humid
19	25.4.07	30	24	30	53	12:55	warm and humid
26	30.4.07	34	26	30	53	13:04	warm and humid

Source: Daily Dawn news paper 2007

Table 2. Effect of different concentrations of lead on seedling growth of *Thespesia populnea*

Pb treatment (µmol/l)	Shoot length (cm)	Root length (cm)	Seedling length (cm)	Numbers of leaves	Leaf area (cm^2)	Circumference (cm)	Seedling dry weight (g)	Root/shoot ratio	Leaf area ratio (cm^2/g)
0	7.30 ± 0.28^{b}	12.62 ± 0.47^{c}	19.92 ± 0.13^{d}	8.80 ± 1.30^{a}	15.06 ± 2.07^{c}	13.80 ± 0.37^{c}	1.75 ± 0.03^{e}	1.83 ± 0.08^{b}	8.68 ± 1.23^{a}
5	6.82 ± 0.21^{ab}	12.46 ± 0.32^{c}	19.48 ± 0.27^{d}	8.60 ± 1.81^{a}	11.29 ± 0.93^{b}	11.20 ± 0.20^{b}	1.56 ± 0.02^{de}	1.73 ± 0.26^{c}	7.25 ± 2.18^{c}
10	6.66 ± 0.29^{ab}	11.88 ± 0.48^{b}	18.54 ± 0.24^{c}	8.0 ± 1.73^{a}	7.91 ± 0.42^{a}	10.80 ± 0.48^{b}	1.45 ± 0.15^{cd}	1.78 ± 0.15^{a}	6.46 ± 2.41^{b}
15	6.68 ± 0.15^{ab}	11.66 ± 0.39^{b}	18.14 ± 0.31^{bc}	8.40 ± 1.67^{a}	7.88 ± 0.55^{a}	10.60 ± 0.24^{b}	1.27 ± 0.1^{c}	1.75 ± 0.12^{b}	6.70 ± 2.03^{c}
20	6.76 ± 0.27^{ab}	11.54 ± 0.51^{b}	17.68 ± 0.29^{ab}	7.40 ± 0.89^{a}	7.69 ± 0.27^{a}	10.40 ± 0.24^{b}	1.05 ± 0.3^{b}	1.71 ± 0.31^{c}	5.33 ± 2.24^{c}
25	6.52 ± 0.13^{a}	10.84 ± 0.32^{a}	17.32 ± 0.19^{a}	6.80 ± 0.83^{a}	7.12 ± 0.79^{a}	9.60 ± 0.24^{a}	0.77 ± 0.4^{a}	1.66 ± 0.40^{c}	5.72 ± 2.36^{b}

Numbers followed by the same column are not significantly different according to Duncan Multiple Range Test at $P < 0.05$ level (± standard error)

Application of Pb at 5, 10, 15 and 20 µmol/l suppressed the seedling length (19.48, 18.54, 18.14, 17.68 cm, respectively) as compared to control. The assessment of early growth in terms of root and shoot length followed the similar pattern as observed for seedling growth in various applied concentrations of Pb. Root length was consistently reduced with increasing concentrations of Pb at 20 µmol/l but high reduction was observed at 25 µmol/l as compared to control and other treatments. Similarly, a gradual decline in shoot length was recorded with enhanced Pb concentration. However, maximum and statistically significant ($P < 0.05$) depression in shoot length (6.52 cm) was evident at the highest concentration of Pb at 25 µmol/l. Number of leaves, leaf area and circumference of plant were also reduced in different concentrations of lead as compared to control.

The data recorded on seedling dry weight revealed that treatment of Pb at 5, 10, 15 and 20 µmol/l concentrations suppressed the seedling dry weight significantly to 1.56, 1.45, 1.27 and 1.05 g respectively as compared to control (1.75 g). Seedling dry weight (0.77 g) at 25 µmol/l of Pb was significantly decreased as compared to rest of all concentrations. Root/shoot ratio was lowered in treatment as compared to control and it decreased continuously with increase in concentration of Pb (Table 2). It was noted that inhibitory effect of Pb was greater at 25 µmol/l concentration of Pb than other treatments. Percentage decrease in different parameters of *T. populnea* was also calculated which increased along with increased concentrations of Pb (Figure 1).

The seedlings of *T. populnea* were tested for tolerance to lead toxicity (Figure 1). Increased

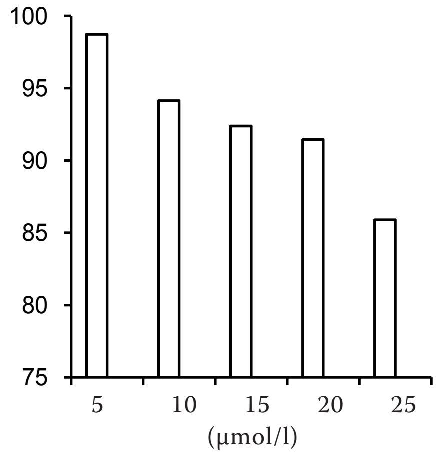

Figure 1. Indices of tolerance of *Thespesia populnea* L. in different concentrations of lead

concentration of lead (5–25 µmol/l) gradually decreased tolerance of *T. populnea*. Lead treatment at 5, 10, 15, 20 and 25 µmol/l produced 98.73, 94.14, 92.39, 91.44 and 85.90% tolerance in *T. populnea*, respectively.

DISCUSSION

Heavy metal toxicity in the environment is of great concern because of its effects on plants growth. The plants under stress conditions are most likely to be adversely affected by high concentration of heavy metals. Lead toxicity has become important because of its constant increase in the environment. High concentration of Pb may lead to the reduction in root hair development and stunted growth due to reduced photosynthetic rate in plants, which is the result of stomata closure by the deposition of Pb (Sarkar and Jana 1986). Lead exerts deleterious effects on morphology, growth and photosynthetic processes of plants and causes inhibition of enzyme activities, water imbalance, alterations in membrane permeability and disturbs mineral nutrition (Singh et al. 1997, Sharma and Dubey 2005, Israr and Sahi 2008). Seed germination inhibitions by heavy metals have been reported by some workers (Morzek and Funiceli 1982, Brown and Wilkins 1986). Seedling length of *Thespesia populnea* showed reduction with increased concentrations of lead. The reduction in plant height could be attributed to the adverse affect of metal treatments on the cell elongation and cell expansion. The effects of toxic substances on plants are dependent on the amount of toxic substance taken up from a given environment. The toxicity of some metals may be so high that plant growth is retarded before a large quantity of an element can be translocated (Haghiri 1973). The inhibition in seedling growth of *T. populnea* appeared to be due to toxicity of Pb solution. Lead produced toxic effects on germination, seedling growth and biomass of *Albizia lebbeck* L. Benth. at 10, 30, 50, 70 and 90 µmol/l concentrations (Farooqi et al. 2009). Exposure to lead (Pb) as well as other heavy metals in the environment is still a matter of public health concern (Rinderknecht et al. 2005).

Heavy metals also inhibit the seedling growth of *Phaseolus aureus* (Sharma 1982). The reason of reduced shoot and seedling length in metal treatments could be the reduction in meristematic cells present in this region and some enzyme contained in cotyledon and endosperm. Cells become active and begin to digest and store nutrients which are converted into the soluble form and transported to the radicle and plumule tips e.g. enzyme amylase converts starch into sugar and protease acts on protein. So, when the activities of different enzymes were affected, nutrients did not reach to the radicle and plumule and in this way shoot and seedling length were affected. Pb treatment had toxic effects on the root growth for seed collected from the polluted and clean areas. The reason for reduced root length in metal treatments could be due to reduction in mitotic cells in meristematic zone of root as suggested by Lerda (1992) on *Allium cepa*. He observed that 50, 100 and 200 ppm of Pb stopped the growing processes in plants after 24 h. These findings confirm that Pb reduced the frequency of mitotic cell in meristematic zone and Pb causes inhibition of root growth. Cho and Wang (1990) showed that percentage length of infected roots and dry biomass of maize plant were reduced by the addition of heavy metals. Dalal and Bairgi (1985) found reduction in seed germination, root, shoot and seedling length of jute verities, *Corchorus olitorius* cv. JRO 524 and C*apsular corchorus* JRC 321 at different levels of Pb, particularly at 20 mg/l. The reduction in root length in *T. populnea* was more prominent in different concentrations of Pb treatments as compared to shoot and seedling length. The influence of increasing concentrations of lead on 14 day-old seedlings of *Triticum aestivum* cv. Vergina was observed (Karataglis et al. 2008). They found that plants grown in 1/10 strength Rorison's nutrient solution plus increasing concentrations of lead depressed shoot growth and the most evident symptoms were found on roots. These effects demonstrate the diverse modes of metal action resulting in different degrees of toxicity.

Mineral nutrients are important for the normal growth of plants. Presence of unbalance nutrients in soil can cause disturbance in the uptake of certain elements, which are necessary for plant growth (Iqbal and Shafiq 2003). Some elements, such as Cu, Fe, Mn etc., are essential mineral nutrients. Others, such as Cd and Pb, however, have no known physiological activity (Lasat 2002). Plants differ in the level of tolerance to different elements for growth but excessive amount can lead to toxicity. These differences may be attributed to variable ion translocation to the aerial parts of the plants. Dry weight is an important indicator of adaptations mechanism. The seedling of *T. populnea* showed a gradual decrease in dry weight with increasing concentrations of lead. The lead treatments showed more effects on seedling dry weight which is evident from the poor growth of root and aerial parts. Root/shoot and leaf area ratios indicate that increased concentrations of Pb are more toxic, reduce root/shoot ratio and other parameters of growth.

The percentage decrease in seedling length, root length, shoot length, number of leaves, leaf area, plant circumference and seedling dry weight was calculated. These findings confirmed that increased concentrations of Pb showed higher percentage reductions indicating its toxicity. Tolerance to Pb in *T. populnea* was relatively low. The reason for reduced tolerance to Pb may be changes in the physiological association of the tolerance mechanism and seedling growth of plant. Results of the present finding appear to be a useful indicator establishing the toxic nature and tolerance indices for Pb in *T. populnea*. Plants differ in the level of tolerance to metal stresses. This information can be considered a contributing step in exploring and finding the tolerance limit of *T. populnea* at different concentrations of lead treatments.

REFERENCES

Abdullah C.M., Iqbal M.Z. (1991): Response of automobiles, stone and cement particulate matters on stomatal clogging of plants. Geobios, *18*: 196–201.

Ahmed Z., Qadir S.A. (1975): The effects of air pollution on stomata clogging, carbohydrates and chlorophyll contents in certain roadside plants. Pakistan Journal of Botany, *7*: 81–84.

Brown M.T., Wilkins D.A. (1986): The effect of zinc on germination, survival and growth of *Betula*. Environmental Pollution – (Series A): Ecological and Biological, *41*: 53–61.

Campbell J.A. (1976): A mathematical treatment of variation of heavy metal content of lichen with distance from a source. Lichnologist, *8*: 83–86.

Cho C., Wang Y. (1990): Effects of heavy metals on the infection of vesicular-arbuscular mycorrhizae and the growth of maize. New Series, *152*: 34–45.

Dalal T., Bairgi P. (1985): Effect of Hg, As and Pb on germination and seedling growth of two Jute varieties. Environment and Ecology, *3*: 403–407.

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

Haghiri F. (1973): Cadmium uptake by plants. Journal of Environmental Quality, *2*: 93–96.

Iqbal M.Z., Shafiq M., Kausar A.S. (2001): Toxic effects of lead and cadmium individually and in combination on germination and growth of *Leucaena leucocephala*. Pakistan Journal of Botany, *33*: 551–557.

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

Iqbal M.Z., Shafiq M. (2003): Effects of aluminium on germination and growth of two different wheat cultivars. Acta Botanica Hungarica, *45*: 315–321.

Israr M., Sahi S.V. (2008): Promising role of plant hormones in translocation of lead in *Sesbania drummondii* shoots. Environmental Pollution, *153*: 29–36.

Karataglis S., Symeonidis L., Moustakas M. (2008): Effect of Toxic Metals on the Multiple Forms of Esterases of *Triticum aestivum* cv. Vergina. Journal of Agronomy and Crop Science, *160*: 106–112.

Kumar P.B.A.N., Dushenkov V., Motto H., Raskin I. (1995): Phytoextraction: the use of plants to remediation of soils. Environmental Science and Technology, *29*: 1232–1238.

Lasat M.M. (2002): Phytoextraction of toxic metals. A review of biological mechanism. Journal of Environmental Quality, *31*: 109–120.

Lagerwerff J.W., Specht A.W. (1970): Contamination of road side vegetation with cadmium, nickel, lead and zinc. Environmental Science and Technology, *4*: 583–586.

Lerda D. (1992): The effect of lead on *Allium cepa*. Mutation Research, *231*: 80–92.

Miles C.D., Brandle J.S., Danial D.J., Chur-Der O., Schnare P.D., Uhlik D.J. (1972): Inhibition of photosystem II in isolated chloroplast by lead. Plant Physiology Lancaster, *49*: 820–825.

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

Morzeck J.R.E., Funicelli N.A. (1982): Effect of zinc and lead on germination of S*partina alterniflora*, Loisel. seeds at various salinities. Environmental and Experimental Botany, *22*: 23–32.

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

Nriagu J.O., Pacyna J.M. (1988): Quantitative assessment of worldwide contamination of air, water and soil by trace metals. Nature, *333*: 134–139.

Qadir N., Iqbal M.Z. (1991): Growth of some plants raised from polluted and unpolluted seeds. International Journal of Environmental Studies, *39*: 95–99.

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

Rinderknecht A.L., Kleinman M.T., Ericson J.E. (2005): Pb enamel biomarker: deposition of pre- and postnatal Pb isotope injection in reconstructed time points along rat enamel transect. Environmental Research, *99*: 169–176.

Sagar V.K., Gregory C.P., Paul S.J. (1982): Air pollution: an important issue in plant health. Plant Disease, *66*: 429–434.

Sarkar A., Jana S. (1986): Heavy metal pollution tolerance. Water, Air and Soil Pollution, *27*: 15–18.

Seenivasan S., Manikandan N., Muraleedharan N.N., Selvasundaram R. (2008): Heavy metal content of black teas from south India. Food Control, *19*: 746–749.

Sharma S.S. (1982): Effects of mercury on germination and seedling growth of different varieties of *Phaseolus aureus*. Indian Journal of Ecology, *9*: 79–81.

Sharma P., Dubey R.S. (2005): Lead toxicity in plants. Brazil Journal of Plant Physiology, *17*: 35–52.

Singh R.P., Tripathi R.D., Sinha S.K., Maheshwari R., Srivastava H.S. (1997): Response of higher plants to lead contaminated environment. Chemosphere, *34*: 2467–2493.

Stobrawa K., Lorenc-Plucińska G. (2007): Changes in carbohydrate metabolism in fine roots of the native European black poplar (*Populus nigra* L.) in a heavy-metal-polluted environment. Science of the Total Environment, *373*: 157–165.

William C.H., David D.J. (1973): The effects of super phosphate on the cadmium content of soil and plants. Australian Journal of Soil Research, *11*: 43–46.

Received on July 10, 2009

Corresponding author:

Dr. Muhammad Kabir, Department of Botany, University of Karachi, Karachi, Pakistan
e-mail: kabir_botany82@yahoo.com