REDUCTION OF GERMINATION AND SEEDLING GROWTH OF *Leucaena leucocephala* CAUSED BY LEAD AND CADMIUM INDIVIDUALLY AND IN COMBINATIONS

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Abstract

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Seed germination, root, shoot, seedling length and seedling dry weights of *Leucaena leucocephala* were reduced in most of the treatments of lead and cadmium individually and in combination. The intensity of effects of lead and cadmium on the growth of *Leucaena leucocephala* was different at different levels and combination of treatment. In root length, the highest reduction was found at 300 ppm of lead, while in shoot and seedling length, the highest reduction was recorded at 300 ppm Pb + 300 ppm. Cd 100 ppm of lead treatment also showed reduction in seedling dry weight more than any of the other treatments. The investigation showed that 300 ppm Cd in combination with 300 ppm Pb showed comparatively adverse effects on seedling growth of *Leucaena leucocephala*. At the same combination of lead and cadmium, the lowest tolerance was observed in *Leucaena leucocephala*.

Key words: cadmium, city, germination, growth, lead, plant and toxicity

Introduction

Pollution of the environment is mainly caused by the activities of human. In general term, pollution causes degradation or damage to natural environment, which affect vegetation, animals, crops, soil, water, properties etc. Pollution can alter the environment by changing the growth rate of species, interferes with food chain and produced recognizable toxic effects. The human civilization achieved a lot from science, technology and agriculture but due to continued economic growth, population increase, urbanization and technical development man has also introduced a significant amount of pollutants in the environment.

The concentrations of heavy metals in soil increases with the excessive use of fertilizers, pesticides and careless handling of sewage wastes. The waste water from industries posses different heavy metals which pose hazards to plants. A number of heavy metals e.g. lead, cadmium, copper, zinc, nickel etc. have been found to accumulate in toxic concentrations in soil, especially in the urban areas (Ademorti, 1986; Ho, Tal, 1988; Yousafzai, 1991; Ali et al., 1992; Davis et al., 1992; Ara et al., 1996; Khalid et al., 1996). The excessive uptake of these elements from soil reduced the yield of plants because of reduction in the metabolic and nutritional processes (Burzynski, Mereck, 1990; Hailing et al., 1991; Chugh et al., 1992). The studies are available that, absorption of heavy metals inhibit the activities of several enzymes (Burzynski, Mereck, 1990; Hailing et al., 1991), seed germination and seedling growth (Singh, Srivastava, 1991; Iqbal, Siddiqui, 1992), anatomical changes (Rashid, Mukherjee, 1990) and decrease in nitrate reductase activities (Chugh et al., 1992).

With the development of leaded gasoline in 20th century and tremendous increase in the number of motor vehicles, the atmosphere has become an increasing significant source of lead in plants growing in the industrial regions and near motorways (Ara et al., 1996; Khalid et al., 1996). Tetra-ethyle lead which is an anti knocking additive is added in the gasoline which escapes from auto exhaust and goes into soil (Yousafzai, 1991) and, thereby contaminating plants (Ara et al., 1996; Khalid et al., 1996).

Karachi is the main industrial center and the only port city of Pakistan; consequently it requires large fleet of vehicles for transportation, that is why Karachi is faced with a serious exhaust emission hazards from vehicles and industries. The smoke and unburned fuel discharged from faulty automobiles have polluted the air (Yousafzai, 1991) to such an extent that the deposited particulate matters have changed the color of the tree leaves from green to gray and contaminated the plants with lead and other metals (Khalid et al., 1996; Ara et al., 1996).

Lead and cadmium are non-essential toxic elements. The harmful effects of Pb and Cd have been demonstrated by many workers (Javed, Sahar, 1987; Burzynski, Merek, 1990; Breckle, Kahile, 1992; Chugh et al., 1992; Lerda, 1992). Presence of single metal in the environment is rarely happen. More than one metals are mostly present in the environment and their effects on plants growth may be synergistic or antagonistic. In the recent years there have been a number of reports on synergistic effects of Pb and Cd in the inhibition of plants growth (Carlson, Bazzaz, 1977). A synergistic toxic effect of Pb plus Cd combination was observed by Wong and Chuae (1985) on Brassica chinensis. Breckle, Kahile (1992) identified the toxicity of Pb + Cd on roots of *Fagus sylvatica.*

The objective of this study was to determine the individual and combined effects of Pb and Cd on *Leucaena leucocephala,* an important roadside plants.

Materials and methods

Healthy seeds of *Leucaena leucocephala* (L.) were collected from the University campus. The seeds were air tightly stored at room temperature before treatments with metal solution. The seeds were rubbed with sandpaper for softening the seed coat and were soaked in 0.2% solution of mercuric chloride for one minute to avoid any

Table 1. Effect of different concentrations of Pb and Cd on germination and seedling growth of *Leucaena leucocephala*

Treatments [ppm]	Germination [%]	Root length [cm]	Shoot length [cm]	Seedling length [cm]	Dry weight [gm]	Root /Shoot ratio
Control	100±0.00	4.52±0.30 a	6.30±0.68 a	10.38±1.17 a	0.23±0.02 a	0.16±0.02 a
100 Pb	83.34±9.14	3.64±0.46 b	3.64±0.39 b	7.28±0.81 b	0.17±0.02 b	0.97±0.11 b
100 Cd	93.34±10.00	1.61±0.17 c	4.70±0.50 c	6.31±0.68 c	0.21±0.02 c	0.35±0.04 c
300 Pb	93.34±10.94	1.03±0.11 d	3.61±0.39 b	4.64±0.49 e	0.18±0.02 b	0.28±0.03 b
300Cd	93.34±10.04	1.27±0.14 c	4.33±0.47 c	5.60±0.61 d	0.22±0.02 c	0.56±0.06 d
100 Pb+100 Cd	96.67±10.69	1.95±0.21 d	5.11±0.55 d	7.09±0.77 b	0.22±0.02 c	0.38±0.04 c
300 Pb+300 Cd	100.00±0.00	1.23±0.13	3.28±0.35	4.52±0.48 e	0.22±0.02 c	0.38±0.04 c

Statistical significance determined by analysis of variance, Number followed by the same letters in the same column are not significantly different ($p < 0.05$) according to Student Newman Keuls Multiple Range Test. ± standard error.

type of fungal contamination during seed germination. The seeds were washed with distilled water and were placed in 90 mm diameter glass petridishes on the filter paper. Twenty seeds were taken in each petridish for each treatment and were replicated three times. Metal treatments of Pb and Cd were prepared with lead nitrate and cadmium nitrate and were applied at 100 and 300 ppm of Pb and Cd along with 100 ppm Pb +100 ppm Cd and 300 ppm Pb + 300 ppm Cd, respectively. 2ml of respective treatment was added to each petridish and at every alternate day, the old solution was replaced with a new solution. All the petridishes were kept at room temperature (25 ± 2°C) with six hourly light period provided by three mercury tubes (120 W). After ten days, percentage germination was recorded. Maximum root, shoot and seedling length were obtained and the data was subjected to analysis of variance (ANOVA) and Student Multiple Range Test (SMRT).

Results

Seed germination of *Leucaena leucocephala* was reduced in most of the treatment of Pb and Cd but the difference was not significant (Table 1) On the other hand, root, shoot and seedling length and dry weight were significantly ($p < 0.05$) reduced in most of the individual and combined treatments of Pb and Cd. In root length although 100 ppm Cd, 300 ppm Cd, 100 ppm Pb+100 ppm Cd and 300 ppm Pb + 300 ppm Cd showed reduction in root length as compared to control, but the highest reduction

Table 2. Decreased percentage in seed germination and seedling growth of *Leucaena leucocephala* at different concentrations of Pb and Cd

Treatments (ppm)+B89	Germination [%]	Root length [cm]	Shoot length [cm]	Seedling length [cm]	Dry weight [%]
100 Pb	16.66	19.46	42.22	32.77	35.29
100 Cd	6.66	64.38	25.39	41.73	8.69
300 Pb	6.66	77.21	42.69	57.15	21.73
300Cd	6.66	71.90	31.26	48.29	4.34
100 Pb+100 Cd	3.36	56.85	18.88	34.53	4.34
300 Pb+300 Cd	0.00	72.78	47.93	58.26	4.34

was observed at 300 ppm of Pb. In shoot and seedling length, the highest reduction was recorded in 300 ppm Pb +300 ppm Cd. In dry weight, 100 ppm Pb showed the highest reduction as compared to all other treatments. The differences in root/shoot ratio are related to root and shoot length of *L. leucocephala* at various treatments.

Decreased percentage in seed germination of *L. leucocephala* showed that 100 ppm Pb had the highest reduction (16.67%) followed by other treatments (Table 2). In root length, the highest reduction (77.21%) was observed in 300 ppm Pb followed by 300 ppm Pb + 300 ppm Cd (72.78%), 300 ppm Cd (71.9%), 100 ppm Cd (64.38%), 100 ppm Pb + 100 ppm Cd (56.85%) and 100 ppm Pb (19.49%), respectively. Shoot and seedling length showed almost similar pattern of reduction. The highest percentage reduction in shoot and seedling length was recorded in 300 ppm Pb + 300 ppm Cd treatment. The highest percentage reduction in dry weight (35.29%) of *L. leucocephala* was found in 100 ppm Pb followed by 300 ppm Pb (21.73%) and 100 ppm Cd (8.69%), respectively. 300 ppm Cd, 100 ppm Pb + 100 ppm Cd and 300 ppm Pb + 300 ppm Cd showed similar percentage of reduction in dry weight (4.34%).

The percentage tolerace (Table 3) of *L. leucocephala* showed that the highest tolerance (80.53%) was recorded at 100 ppm Pb treatments whereas, the lowest tolerance (7.21%) was obtained at 300 ppm Pb + 300 ppm Cd tretment. At other levels of treatments, the percentage tolerance was in between 100 ppm Pb and 300 ppm Pb + 300 ppm Cd.

Table 3. Percentage tolerance of *Leucaena leucocephala* to different concentrations of Pb and Cd

Treatment [ppm]	Percentage tolerance [%]
100 Pb	80.53
100 Cd	35.61
300 Pb	22.78
300 Cd	28.09
100 Pb +100 Cd	43.14
300 Pb + 300 Cd	7.21

Discussion

Lead and cadmium in different combinations reduced the seed germination, root, shoot and seedling length. The results showed that both metals, Pb and Cd in different combinations reduced the seed germination, root, shoot and seedling length. Similarly, Dalal, Bairgi

(1985) have found reduction in seed germination, root, shoot and seedling length of jute varieties, *Corchorus olitorius* cv JRO 524 and *C. capsular* JRC 321 at different levels of Pb, particularly at 20 mg/l. Javed, Sahar (1987) have also found reduction in seed germination and seedling growth in maize at 5 to 100 mM lead nitrate treatment. Iqbal et al. (1991) have studied the influence of Cd toxicity on germination and growth on some common trees. Iqbal, Mahmood (1989) reported that seeds of *Albizia lebbeck* and *Dalbergia sissoo* from polluted areas of Karachi showed significant reduction in seed germination due to presence of Pb in the vehicular exhaust. The reduction in root length in *L. leucocephala* was greater in different concentrations of Pb and Cd treatments as compared to shoot and seedling length. This was also confirmed by Stefan et al. (1991) in some others species. The reduction in root length may be due to accumulation of metals within the root which reduced the rate of mitosis in meristematic zones of roots specially blocking the metaphase in meristematic cells, therefore, root showed reduction in length as demonstrated by Goldbold, Kettner (1991) and Sharifah, Hishashi (1992). Lerda (1992) observed that 50, 100 and 200 ppm Pb stopped the growing processes in plants after 24 hours. These findings confirm that Pb causes inhibition in root growth.

Cadmium has been found to inhibit the seed germination, root, shoot and seedling length greatly in *L. leucocephala* in combination with lead. It was found that Cd was comparatively more toxic to seed germination and seedling growth than Pb. Breckle, Kahile (1992) have also studied the single and combined effects of Pb and Cd on growth of *Fagus sylvatica* and found that the root elongation was sensitive parameter; 20.0 ppm Pb + 1.0 Cd greatly reduced the root growth. Carlson, Bazzaz (1977) found synergistic growth reduction in seedling of American sycamore in an agricultural soil treated with various levels of Pb and Cd chlorides. Root, shoot and seedling length treated with 300 ppm Pb + 300 ppm Cd showed highest reduction than any other combinations. This decrease in growth of plants might be due to reduction in enzymatic activities (Burzynski, Mereck, 1990) or reduction in mitotic cell division in the meritematic zones of roots (Breckle, Kahile, 1992).

In this investigation, different treatments of Pb and Cd individually and in combination had little effects on seed germination of *Leucaena leucocephala* but the effects were significant on root, shoot and seedling length particularly at 300 ppm Pb + 300 ppm Cd.

Translated by the authors

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References

Ademorti, C.M.A., 1986: Levels of heavy metals in bark and fruit of trees in Bennin City, Nigeria. Environmental pollution (Ser. Biochem. Physiol.), *11*, p. 241-254.

Ali, E.A., Ibrahim, H.Y., Nasralla, M.M., 1992: Contamination of agricultural land due to industrial activities in South of greater Cario. Environmental Science and Engineering, *27*, p. 1293-1304.

Ara, F., Iqbal, M.Z., Qureshi, M.S., 1996: Determination of heavy metals contamination of trees and soils due to vehicular emission in Karachi city. Karachi University Journal of Science, *24*, p. 80-84.

Breckle, S.W., Kahile, H., 1992: Effects of toxic heavy metals Cd, Pb on growth and mineral nutrition of beech Vegetatio., *101*, p. 43-53.

Burzynski, S., Merek, K., 1990: Effect of Pb and Cd on enzymes of nitrate assimilation in cucumber seedling. Acta Physiology Plantarum, *12*, p. 105-110.

Carlson, R.W., Bazzaz, F.A., 1977: Growth reduction in American sycamore (*Plantanus occidentalis* L.) caused by Pb-Cd interaction. Environmental pollution, *12*, p. 243-253.

Chugh, L.K., Virinder, K.G., Surinder, K.S., 1992: Effects of cadmium on enzymes of nitrogen metabolism in pea seedlings. Phytochemistry, *31*, p. 395-400.

Dalal, T., Bairgi, P., 1985: Effect of Hg, As and Pb on germination and seedling growth of two jute varieties. Environmental Ecology, *3*, p. 403-407.

Davis, B.E., 1992: Inter-relationships between soil particles and the uptake of cadmium, copper, lead and zinc from contaminated soil by radish. Water, Air and soil pollution, *63*, p. 331-342.

Goldbold, D.L., Kettner, C., 1991: Lead influences root growth and mineral nutrition of *Picea abies* seedlings. Plant physiology, *139*, p. 95-99.

Hailing, L.I.U., Qing, L.I., Peng, Y., 1991: Effect of cadmium on seed germination, seedling growth and oxidase enzyme in crops. Chinese Journal of Environmental Science, *12*, p. 29-31.

Ho, Y.B., Tal, M.K., 1988: Elevated levels of lead and other metals in roadside and grass. Environmental pollution, *49*, p. 37-52.

Iqbal, M.Z., Siddiqui, D.A., 1992: Effect of lead toxicity on seed germination and seedling growth of some tree species. Pakistan Journal of Scientific and Industrial Research, *35*, p. 139-41.

Iqbal, M.Z., Mahmood, T., 1989: Impact of vehicular emission on seed geremination of some roadside trees. Pakistan Journal of Scientific and Industrial Research, *32*, p. 752-756.

Iqbal, M.Z., Mahmood, T., Firdous, A., 1991: Influence of cadmium toxicity on germination and growth of some common trees. Pakistan Journal of Scientific and Industrial Research, *34*, p. 140-142.

Javed, I., Sahar, M., 1987: Effect of lead on germination, early seedling growth, soluble protein and acid phosphatase content in *Zea mays* L. Pakistan Journal of Scientific and Industrial Research, *30*, p. 853-856.

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

Lerda, D., 1992: The effect of lead on *Allium cepa*. Mutation Research, *231*, p. 80-92.

Rashid, P., Mukherjee, A., 1990: Effect of lead nitrate on some anatomical features of mung bean. Bangladesh Journal of Botany, *19*, p. 149-154.

Sharifah, B.A., Hishashi, O., 1992: Effects of lead, cadmium and zinc on the cell elongation of *Impatiens balsamina*. Environ. Experi. Bot., *32*, p. 439-448.

Singh, D.N., Srivastava, S., 1991: Effect of cadmium on seed germination and seedling growth of *Zea mays*. Biol. Sci., *61*, p. 245-247.

Stefan, A., Arundin, I., Onnes, A., 1991: Germination and initial growth in presence of heavy metals. Ann. Bot. Fenn., *28*, p. 37-43.

Wong, M.K., Chuae, K.P., 1985: Interactive effects of lead, cadmium and copper combinations in the uptake of metals and growth of *Brasica chinensis*. Eniviron. Experi. Bot., *26*, p. 331-339.

Yousafzai, A.H.K., 1991: Lead and heavy metals in the street dust of Metropolitan City of Karachi. Pakistan Journal of Scientific and Industrial Research, *34*, p. 157-172.

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Iqbal M.Z., Shazia Y.: **Redukcia klíčenia a rastu semenáčika *Leucaena leucocephala* zapríčinenej olovom a kadmiom a ich kombináciami.**

Klíčenie semien, dĺžka koreňa, výhonu, semenáčika a sušina semenáčika u *Leucaena leucocephala* sa najviac redukovala pri aplikácii olova a kadmia jednotlivo, ako aj pri ich kombinácii. Intenzita vplyvu olova a kadmia na rast *Leuceana leucocephala* bola rozdielna pri rôznych úrovniach a kombináciách aplikácie. V dĺžke koreňa najvyššia redukcia bola pri 30 ppm olova. Kým v dĺžke výhonku a semenáčika sa najvyššia redukcia dosiahla pri 300 ppm Pb + 30 ppm. Cd 100 ppm aplikácie olova preukázalo aj redukciu v sušine semenáčika viacej ako pri iných aplikáciách. Z výskumu vidieť, že 300 ppm Cd v kombinácii s 300 ppm Pb preukázalo pomerne škodlivý účinok na rast semenáčika *Leucaena leucocephala.* Najnižšiu toleranciu sme zistili u *Leucaena leucocephala* pri rovnakej kombinácii olova a kadmia.

BIOMONITORING OF POLLUTION LOAD LOWERING WITH CHANGE OF THE ALUMINIUM PRODUCTION TECHNOLOGY

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Abstract

Maňkovská B.: Biomonitoring of pollution load lowering with change of the aluminium production technology. Ekológia (Bratislava), Vol. 23, No. 2, 169-183, 2004.

Anthropogenic loading of all tree species in Žiarska valley, expressed by means of the coefficient of loading by air pollutants K_z has decreasing trend. Total loading of all tree species by air pollutants in Žiarska valley (pollution zones A, B, C) expressed by means of K_z coefficient represents in 2001 only 5.9 times higher compared with 7.9 higher loading compared with 2000; 19.4 times higher loading compared with in 1997; 22.9 times higher loading compared with 1996 and 29.5 times higher loading compared with 1995. Total loading by air pollutants for all tree species (pollution zones A, B, C, D) within 50 km from SLOVALCO, a.s. given by K_z represents 3.7 times higher loading. We found statistically significant difference for K_z in coniferous tree species (1 and 2 years old) and broadleaved tree species in A zone in comparison with the zone B, C and D. We also found statistically insignificant difference in coniferous tree species (1 and 2 y) and in broadleaved tree species in the zones B, C and D except for broadleaved tree species, for which we found statistically significant increase between the zone B and D.

Key words: biomonitoring, air pollution, mapping of F, S, N

Introduction

Problems of the environment pollution in Žiarska basin are connected with the operation of Aluminium plant from 1953. Abolishment of a municipality Horné Opatovce, which was situated near to the plant, proves of an injurious effect of this emission. Aluminium plant is situated only 2 km from town Žiar nad Hronom. A new chapter in the history started on 29 February 1996. The technology of production was supplied by Norwegian firm Hydro Aluminium. Emission of dust Al_2O_3 dropped from 766 to 243 $t.year^{-1}$, emission of fluorides dropped from 847 to 65.9 $t.year^{-1}$ and emission of SO_2 have increased from 716 to 974 $t.year^{-1}$. On that day, the last electrolyser from the old series based on Söderberg technology of aluminium production was