



## Effects of lead on growth and mineral nutrition of *Amaranthus gangeticus* L. and *Amaranthus oleracea* L.

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### Abstract

Pot experiments were conducted to study the effects of lead (Pb) on growth and nutrient uptake of *Amaranthus gangeticus* L. and *Amaranthus oleracea* L. The levels of Pb used in the experiments were 0, 20, 40, 60, 80, 100 mg kg<sup>-1</sup>. Shoot and root weight of *A. gangeticus* declined by 28 and 53% and *A. oleracea* by 46 and 37%, respectively over control at the highest rate of Pb application. Lead application in soil significantly decreased N and P concentration in shoots as well as Ca, Zn and Mn in both shoots and roots of *A. gangeticus*. Phosphorus, K and Fe in roots of *A. gangeticus* increased with increasing rates of Pb. The contents of P, Fe and Mn in shoots and Ca, Zn and Mn in roots of *A. oleracea* decreased with increased rates of Pb application. On the other hand, an increase of N, K and Zn concentration in shoots and K and Fe concentration in roots of *A. oleracea* were observed. Lead application in soil significantly increased Mg concentration in both shoots and roots of *A. gangeticus* and *A. oleracea*.

**Keywords:** Lead, *amaranthus gangeticus*, *amaranthus oleracea*, concentration, shoot, root

### Introduction

A wide variety of contaminants enter into our environment due to extensive industrial production, energy and fuel production and intensive agriculture. Heavy metals are one of the most dangerous of these contaminants. Among heavy metals, lead is an element that is easily accumulated in soil and sediments. The level of Pb in the environment is currently of great concern. Although lead is not an essential element for plants, it is absorbed and accumulated (Kabata Pendias and Pendias, 1999; Kibria *et al.*, 2006, 2007). The level of Pb found in plants is often correlated with the level present in the environment (Vesk and Allaway, 1997). For example, Salim *et al.* (1993) showed that concentration of Pb increased in radish plants when treated with an increasing concentration of the metal. The absorption of metals from the soil by plants is influenced by a variety of factors, including pH, temperature, soil ions, cation exchange capacity of soil, organic matter content of the soil, the type and concentration of metal and the species of plant (Antosiewicz, 1992; Salim *et al.*, 1993).

Lead is one of the most widely distributed heavy metals and is very toxic to plants (Kosobrukhov *et al.*, 2004). In the whole plant, Pb can affect photosynthesis at the stomata level, mesophyll cells, pigment content and light and dark reactions. It interferes with nutritional elements of seedlings and plants, thus causing deficiencies or adverse ion distribution within the plant (Trivedi and Erdei, 1992) as well as growth inhibition (Woźny and Jerezyńska, 1991; Malkowski *et al.*, 2002). Soil contaminated with heavy metals often reduces and

sometimes disables the production of quality food products and animal feeds (Kabata Pendias, 2001). Crops harvested from heavy metal polluted areas are usually tested for heavy metal concentration while the concentrations of essential macro and micro elements are often neglected. Experiments on the effects of lead on contents of macro and micro elements are scarce. The present study is an attempt to determine the effects of soil contamination with Pb on the contents of N, P, K, Ca, Mg, Zn Fe and Mn in *A. gangeticus* and *A. oleracea*.

### Materials and Methods

#### Pot experiments

Two separate pot experiments with *Amaranthus gangeticus* L. and *Amaranthus Oleracea* L. were carried out in net house of the Department of Soil Science, University of Chittagong in order to study the effects of Pb on nutrient uptake by these plants.

Soils were collected from agricultural field near Shahid Minar of Chittagong University from a depth of 0-15 cm. Dry roots, grasses and other particulate materials were discarded from the air dry soils and processed for pot experiment. A portion of the soils passed through 2 mm sieve was retained for laboratory analyses. The soil contained 57% sand, 22% silt, 23% clay; pH, 5.5; organic carbon, 0.83%; cation exchange capacity, 6.93 cmol kg<sup>-1</sup> soil; total nitrogen, 0.086%; total phosphorus, 0.029%; total potassium, 0.24%, and aqua regia digestible Pb 7.8 mg kg<sup>-1</sup>.

Eight kg soil was taken in each earthen pot of 30 cm diameter and 28 cm height. Nitrogen, phosphorus and potassium were applied in both the experiments at the rate of 90, 25 and 30 kg ha<sup>-1</sup> from Urea, Triple Super Phosphate

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and Muriate of Potash, respectively. All phosphorus and potassium and one-half nitrogen were applied and mixed with soil during preparation of the pot according to BARC (1997) recommendations. Remaining N was applied in two equal installments at 10 and 25 days after sowing (DAS). Lead at the rate of 0, 20, 40, 60, 80 and 100 mg kg<sup>-1</sup> soil were added in pots in solution form as lead nitrate [Pb(NO<sub>3</sub>)<sub>2</sub>]. Each treatment was replicated three times. Soils mixed with Pb were allowed to equilibrate for 15 days and then seeds of *A. gangeticus* and *A. oleracea* were sown in the pots. At 15 days after sowing, plants were thinned to keep 5 plants in each pot. The pots were arranged according to a randomized complete block design. Plants were harvested 45 days after sowing (DAS). During harvesting, the heights of plants were measured. The shoots and roots were collected separately. The roots were collected carefully and washed thoroughly to remove adhering soil particles.

### Soil analysis

The particle size distribution was determined by hydrometer method of Day (1965). Soil pH was measured in a 1:2.5 soil/water suspension with glass electrode pH meter. The potassium dichromate wet-oxidation method of Jackson (1973) was used for the determination of organic carbon followed by multiplying the values with 1.724 to calculate the organic matter contents. The cation exchange capacity was determined by saturation with 1N ammonium acetate at pH 7.0 (Jackson, 1973). The soil samples were digested with aqua regia (Jackson, 1973) on a sand bath for the determination of total Cd, Pb, Zn, Fe, Mn, P and K. Total nitrogen was determined by micro - Kjeldahl method as described by Jackson (1973). Phosphorus was determined by vanadomolybdo phosphoric yellow color method in nitric acid system according to Jackson (1973). Potassium was measured by flame photometer and Fe, Mn, Zn, Cd and Pb were determined by atomic absorption spectrophotometer (Varian spectra AA-220).

### Plant analysis

Oven dried (65<sup>o</sup> C to constant weight) and ground plant samples were digested with a mixture of H<sub>2</sub>SO<sub>4</sub>, H<sub>2</sub>O<sub>2</sub> and lithium sulfate for the determination of N, P, K, Ca, Mg, Zn, Fe and Mn in the plant tissues (Allen *et al.*, 1986). The concentrations of Ca, Mg, Zn, Fe and Mn in the digest were measured by atomic absorption spectrophotometer (Varian Spectra AA 220). Micro-Kjeldahl method as described by Jackson (1973) was used for the determination of nitrogen. Phosphorus was determined by vanadomolybdo phosphoric yellow color method in nitric acid system according to Jackson (1973). Potassium was measured by flame photometer (Helios γ).

### Data analysis

The significance of differences between the means of the treatments was evaluated by one way analysis of variance followed by Duncan's Multiple Range Test at the significance level of 5%. The statistical software Excel (Excel Inc., 2003) and SPSS version 12 (SPSS Inc., 2003) were used for the analysis.

## Results and Discussion

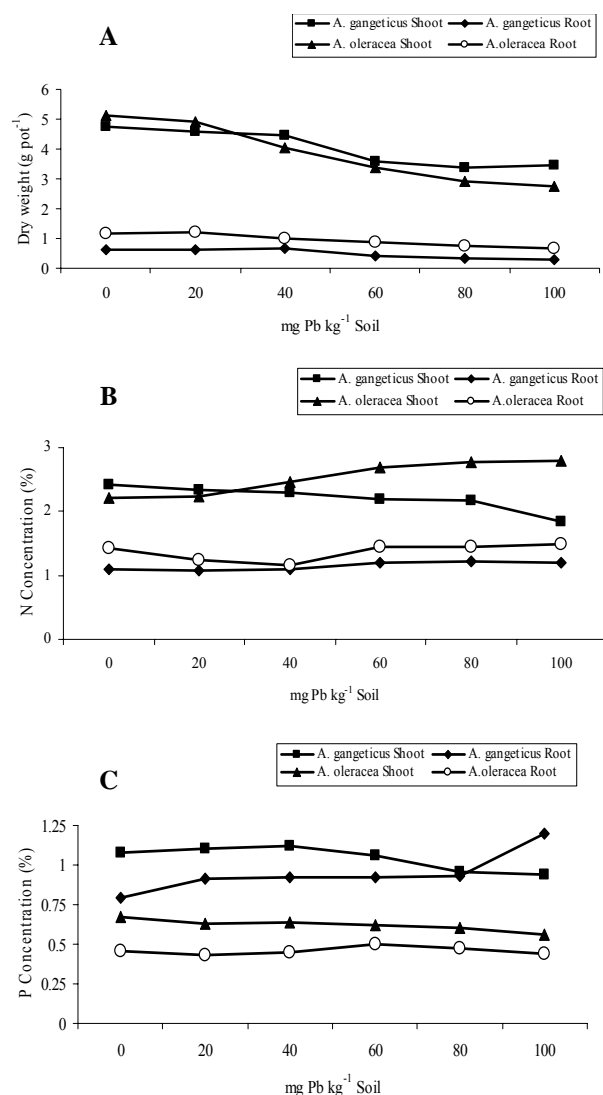
### Growth of *A. gangeticus* and *A. oleracea*

Dry weight of shoot and root of *A. gangeticus* and *A. oleracea* were significantly affected by Pb application. In general, Pb application caused a decrease in the shoot and root weight of *A. gangeticus* and *A. oleracea* (Figure 1). However, shoot and root weight of *Amaranthus gangeticus* L. significantly decreased when Pb was applied above 40 and 60 mg kg<sup>-1</sup>, respectively, compared with control. Similar results were found with *Amaranthus oleracea* L. At the highest dose of Pb (100 mg kg<sup>-1</sup>), shoot and root weight of *Amaranthus gangeticus* L decreased by 28 and 53 % when compared with control. The corresponding reductions for *Amaranthus oleracea* L. were 46 and 37 %, respectively. In agreement with the present study, Huang and Cunningham (1996) reported that increasing Pb concentration significantly decreased both shoot and root dry weight of corn and ragweed after 2 weeks of Pb exposure at 0, 5, 20, 50 and 100 μM. They also found that the shoot and root yield decreased linearly with increasing Pb concentration up to 50 μM. Above this Pb concentration, there was no further reduction in root growth. Kopittke *et al.* (2007a) reported that relative fresh mass of cowpea (*Vigna unguiculata*) decreased by 10% at a Pb<sup>2+</sup> activity of 0.2 μM for the shoots and at a Pb<sup>2+</sup> activity of 0.06 μM for the roots. A decrease of dry weight of two sunflower varieties cultivated in a hydroponic system spiked with Pb at 7.5 and 10 μM was observed by Nehnevajova (2005). Kosobrukhov *et al.* (2004) also reported a considerable decrease in dry weights of different plant parts under Pb treatments.

The inhibition of shoot growth may be due to a decrease in photosynthesis; it upsets mineral nutrition and water balance, changes hormonal status and affects membrane structure and permeability (Sharma and Dubey, 2005). The inhibition of root growth may be due to a decrease in calcium in root tips, leading to a decrease in cell division or cell elongation (Haussling *et al.*, 1988; Eun *et al.*, 2000).

### Effects of Pb on nutrient uptake

Plants cultivated in soil contaminated with heavy metals are subject to modification of the chemical composition of not only the content of heavy metals but



**Figure 1. Effects of lead on growth (A), nitrogen (B) and phosphorus (C) concentrations of *A. gangeticus* and *A. oleracea*.**

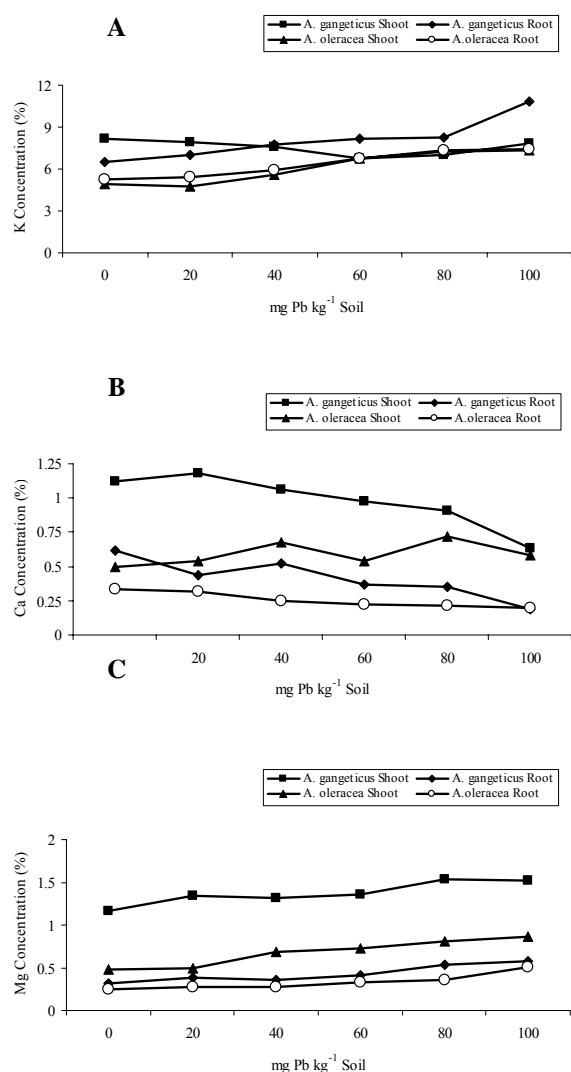
also macronutrients (Ciecko *et al.*, 2004). Nitrogen concentration in the present experimental plants was dependent on both species and organ of the plant. Nitrogen concentration in shoot of *A. gangeticus* was declined by increasing amounts of Pb application in soil (Figure 1). However, a significant decrease in nitrogen concentration in shoots of *A. gangeticus* was observed only with 100 mg Pb kg<sup>-1</sup> soil treatment as compared to the control. The reduction in nitrogen concentration in shoots of *A. gangeticus* was 24 % with 100 mg kg<sup>-1</sup> Pb application compared with control. Lead application in soil neither affected nor showed any definite trend of variation in nitrogen concentration in roots of *A. gangeticus*. Contrary to the results found for

*A. gangeticus*, Pb application in soil significantly increased nitrogen concentration in shoots of *A. oleracea*. However, the values of nitrogen concentration in shoots of *A. oleracea* with 40-100 mg kg<sup>-1</sup> Pb application were not significantly different from each other. At the highest rate of Pb application (100 mg kg<sup>-1</sup>), nitrogen concentration in shoots of *A. oleracea* increased by 26% compared with control. Nitrogen concentrations in roots of *A. oleracea* were significantly different among the treatments. The values of nitrogen concentration decreased with increasing Pb application up to 40 mg kg<sup>-1</sup> and then increased.

Phosphorus concentrations in shoots of *A. gangeticus* and *A. oleracea* significantly decreased with higher rates of Pb application in soil (Figure 1). This is in agreement with Walker *et al.* (1997) who reported that Pb decreased the uptake of phosphorus in *Zea mays*. However, Huang and Cunningham (1996) found that phosphorus concentrations in shoots of both corn and ragweed were not significantly affected by Pb. In contrast to shoot phosphorus concentrations, root phosphorus concentrations in *A. gangeticus* increased with higher rates of Pb application in soil in the present study. However, Pb did not significantly affect root phosphorus concentrations in *A. oleracea*. Similar results were found by Huang and Cunningham (1996) with corn.

Potassium concentration in shoots of *A. gangeticus* was not affected by Pb application (Figure 2). However, the highest level of Pb application significantly increased K concentration in roots of *A. gangeticus*. Although K concentration in roots of *A. gangeticus* gradually increased with increasing rate of Pb application, the values for K concentration were not significantly different up to 80 mg Pb kg<sup>-1</sup> soil (Figure 2). Lead application significantly increased K concentrations in both shoots and roots of *A. oleracea*. The highest concentrations of K in shoots and roots were found with the highest rate of Pb application. Potassium concentration in shoot and root of *A. oleracea* increased by 50 and 41 % respectively with the highest rate of Pb application as compared to control. The results of the present study are in contrast to the findings of Walker *et al.* (1997) with *Cucumis sativus* seedlings and *Zea mays*. Kopittke *et al.* (2007a) reported that above critical Pb<sup>2+</sup> activity, an increase in Pb<sup>2+</sup> decreased K concentration in shoot of cowpea. Kibria (2008) also reported that Pb application significantly decreased potassium concentration in straw and roots of rice, shoot and root of radish and leaf, stem and root of Indian spinach.

Calcium concentration in shoots and roots of *A. gangeticus* and roots of *A. oleracea* were significantly decreased by Pb application (Figure 2). However, Ca concentrations in shoots of *A. gangeticus* with Pb



**Figure 2. Effects of lead on potassium (A), calcium (B) and magnesium (C) concentrations of *A. gangeticus* and *A. oleracea*.**

application up to 80 mg kg<sup>-1</sup> were statistically similar to that with control. The results of the present study are in agreement with that of other workers. For example, Kibria (2008) found Ca concentrations to be decreased in grain, straw and roots of rice, shoot and root of radish and leaf, stem and root of Indian spinach due to Pb application. Similar results were found by Huang and Cuningham (1996) with corn. Calcium concentration in corn shoots decreased by more than 40% after 2 weeks of 20µM Pb treatment. However, in their study, the same Pb treatment did not significantly affect Ca concentration in shoots of ragweed. Lead application neither affected nor showed any definite trend of variation in Ca concentration in shoots of *A. oleracea* in the present study.

Lead application at higher rates significantly increased magnesium concentration in shoots and roots of *A. gangeticus* and roots of *A. oleracea* (Figure 2). Magnesium concentrations in shoot of *A. gangeticus* and root of *A. oleracea* were not affected by Pb application up to 60 mg kg<sup>-1</sup>. Lead application up to 80 mg kg<sup>-1</sup> did not affect Mg concentration in roots of *A. gangeticus* and shoots of *A. oleracea*. On the other hand, Kibria (2008) reported that Pb application significantly decreased Mg concentration in grain, straw and roots of rice and leaf, stem and root of Indian spinach. However, in his study Pb did not affect Mg concentration in both shoots and roots of radish. Huang and Cuningham (1996) reported that Mg concentrations in shoots for both corn and ragweed significantly decreased after 2 week of 20µM Pb treatment. Lead decreased the uptake of magnesium by *Cucumis sativus* seedlings and *Zea mays* (Walker *et al.*, 1997). Similar results were found by Kopittke *et al.* (2007a) with cowpea. In Pb treated rice plants there was no definite trend in the concentration of magnesium in various parts (Chatterjee *et al.*, 2004).

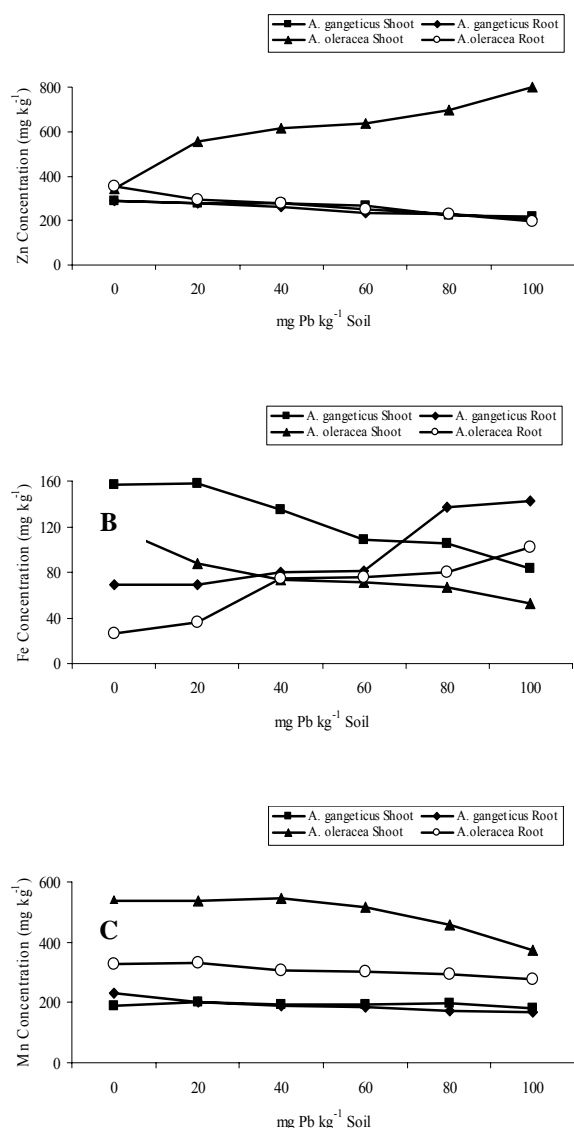
Zinc content of shoots and roots of *A. gangeticus* and roots of *A. oleracea* decreased significantly with increasing rate of Pb application showing a negative relation between Pb and Zn (Figure 3). On the contrary, shoots of *A. oleracea* increased with increasing Pb application. Kopittke *et al.* (2007a and b) showed negative effects between Pb and Zn in shoots and roots of cowpea and shoots of signal grass and Rhodes grass.

Iron concentration in roots of *A. gangeticus* and *A. oleracea* showed gradual increase over control with all rates of Pb and the lowest values were obtained with the highest rate of Pb in both the species (Figure 3). An opposite trend was observed for shoots of both species. In agreement with the present study, an increase in Fe concentration in the roots of cowpea (greater than three folds) with increase in Pb<sup>2+</sup> activity was reported by Kopittke *et al.* (2007a).

Shoots and roots of *A. oleracea* and roots of *A. gangeticus* showed to decrease significantly in Mn concentration with the increase of Pb application with the lowest values being obtained with the highest rate (Figure 3). Similar results were found by Kopittke *et al.* (2007b) in shoots of signal grass and Rhodes grass. However, Mn concentration in shoots of *A. gangeticus* was not affected by Pb treatments in the present study.

## Conclusions

Lead application significantly decreased shoot and root weight of *A. gangeticus* and *A. oleracea*. The reductions of shoot and root weight of *A. gangeticus* were 28 and 53% and of *A. oleracea* 46 and 37%, respectively, when compared with control. A decrease in N, P, Ca and Zn



**Figure 3. Effects of lead on zinc (A), iron (B) and manganese (C) concentrations of *A. gangeticus* and *A. oleracea*.**

concentrations in shoots of *A. gangeticus* and an increase of K and Fe concentrations in roots were observed by Pb application. Potassium concentration in both shoots and roots of *A. oleracea* was also increased. Lead application at higher rates significantly increased Mg concentration in both shoots and roots of *A. gangeticus* and *A. oleracea*. Manganese concentration was found to decrease in shoots and roots of *A. oleracea* and roots of *A. gangeticus* due to Pb application.

## References

- Allen, S.E., H.M. Grimshaw and A.P. Rowland. 1986. Chemical Analysis. p. 285-344. In: Methods in Plant Ecology. 2<sup>nd</sup> Ed. P.D. Moore and S.B. Chapman (eds.). Blackwell Scientific Publications, Oxford, UK.
- Antosiewicz, D.M. 1993. Mineral status of dicotyledonous crop plants in relation to their constitutional tolerance to lead. *Environment and Experimental Botany* 33: 575-589.
- BARC (Bangladesh Agricultural Research Council), 1997. Fertilizer Recommendation Guide. BARC Soils Publications No 41, Dhaka, Bangladesh.
- Chatterjee, C., B.K. Dube, P. Sinha and P. Srivastava. 2004. Detrimental effects of lead phytotoxicity on growth, yield and metabolism of rice. *Communications in Soil Science and Plant Analysis* 35: 255-265.
- Ciećko, Z., S. Kalembsa, M. Wyszowski and E. Rolka. 2004. The effect of elevated cadmium content in soil on the uptake of nitrogen by plants. *Plant and Soil* 50: 283-294.
- Day, P.R. 1965. Particle fractionation and particle size analysis. p. 545-567. In: Methods of Soil Analysis. Part I. C.A. Black (ed.). Agronomy Monograph, Academic Press, New York.
- Eun, S.O., H.S. Youn and Y. Lee. 2000. Lead disturbs microtubule organization in the root meristem of *Zea mays*. *Physiologia Plantarum* 110: 357-365.
- Excel Inc., 2003. Microsoft Excel for Windows, Microsoft Corporation, USA.
- Hausling, M., C.A. Jorns, G. Lehmbecker, C. Hercht-Bucholz and H. Marschner. 1988. Ion and water uptake in relation to root development of Norway spruce [*Picea abies* (L.) Karst]. *Journal of Plant Physiology* 133: 486-491.
- Huang, J.W. and S.D. Cunningham. 1996. Lead phytoextraction: species variation in lead uptake and translocation. *New Phytology* 134: 75-84.
- Jackson, M.L. 1973. Soil Chemical Analysis. Prentice Hall of India Private Limited, New Delhi, India.
- Kabata-Pendias, A. 2001. Trace elements in soils and plants. 3<sup>rd</sup> Ed. CRC Press, Boca Raton, USA. 413 p.
- Kabata-Pendias, A. and H. Pendias. 1999. Biogeochemistry of Trace Elements. PWN, Warsaw, Poland, 397 p.
- Kibria, M.G. 2008. Dynamics of cadmium and lead in some soils of Chittagong and their influx in some edible crops. Ph. D. Thesis, University of Chittagong, Bangladesh.
- Kibria, M.G., K.T. Osman and M.J. Ahmed. 2006. Cadmium and lead uptake by rice (*Oryza sativa* L.) grown in three different textured soils. *Soil & Environment* 25 (2): 70 -77.

- Kibria, M.G., K.T. Osman and M.J. Ahmed. 2007. Cadmium and lead uptake by radish (*Raphanus sativus* L.) grown in three different textured soils. *Soil & Environment* 26(2): 106 -114.
- Kopittke, P.M., C.J. Asher, R.A. Kopittke and N.W. Menzies. 2007a. Toxic effects of  $Pb^{2+}$  on growth of cowpea (*Vigna unguiculata*). *Environmental Pollution* 150: 280-287.
- Kopittke, P.M., C.J. Asher, F.P.C. Blamey and N.W. Menzies. 2007b. Toxic effects of  $Pb^{2+}$  on the growth and mineral nutrition of signal grass (*Brachiaria decumbens*) and Rhodes grass (*Chloris gayana*). *Plant and Soil* 300: 127-136.
- Kosobrukhov, A., I. Knyazeva and V. Mudrik. 2004. Plantago major plants responses to increase content of lead in soil: growth and photosynthesis. *Plant Growth Regulator* 42: 145-151.
- Malkowski, E., A. Kita, W. Galas, W. Karez and J.M. Kuperburg. 2002. Lead distribution in corn seedlings (*Zea mays* L.) and its effect on growth and the concentrations of potassium and calcium. *Plant Growth Regulator* 37: 69-76.
- Nehnevajova, E. 2005. Non-GMO approach for the improvement of heavy metal accumulation and extraction of high yielding crop species for efficient phytoextraction of contaminated soil. Ph. D. Thesis. School of Architecture, Civil and Environmental Engineering Swiss Federal Institute of Technology Lausanne (EPFL).
- Salim, R., M.M. Al-Subu and A. Attallah. 1993. Effects of root and foliar treatments with lead, cadmium and copper on the uptake, distribution and growth of radish plants. *Environment International* 19: 393-404.
- Sharma, P. and R.S. Dubey. 2005. Lead toxicity in plants. *Brazilian Journal of Plant Physiology* 17: 35-52.
- SPSS Inc. 2003. Statistics. SPSS Inc. Chicago.
- Trivedi, S. and L. Erdei. 1992. Effects of cadmium and lead on the accumulation of  $Ca^{2+}$  and  $K^{+}$  and on the influx and translocation of  $K^{+}$  in wheat of low and high  $K^{+}$  status. *Physiologia Plantarum* 84: 94-100.
- Vesk, P.A. and W.G. Allaway. 1997. Spatial variation of copper and lead concentrations of water hyacinth plants in a wetland receiving urban run-off. *Aquatic Botany* 59: 33-44.
- Walker, W.M., J.E. Miller and J.J. Hasset. 1997. Effect of lead and cadmium upon the calcium, magnesium, potassium and phosphorus concentration in young corn plants. *Soil Science* 124: 145-151.
- Wozny, A. and E. Jerezynska. 1991. The effect of lead on early stages of *Phaseolus vulgaris* L. growth in vitro conditions. *Biologia Plantarum* 33: 32-39.