Pak. J. Bot., 45(S1): 481-485, January 2013.

EFFECTS OF LEAD RESISTANT BACTERIA ON THE EARLY GROWTH OF *VIGNA MUNGO* L. (HEPPER) UNDER LEAD STRESS

SYEDA SHAIMA MERYEM AND AZRA YASMIN*

Department of Environmental Sciences, Fatima Jinnah Women University, Rawalpindi, Pakistan,
**Corresponding author e-mail: azrayasmin@fjwu.edu.pk*

Abstract

Three lead resistant bacteria *Bacillus pumilus* (TE07), *Bacillus cerus* (TE12 & TE14), were examined for their plant growth promotion /remediation potential. Their ability to promote early growth and their effects on metal uptake by three varieties of *Vigna mungo* L. (Hepper) i.e., NARC-Mash-2, NARC-Mash-3 and NARC-Mash-97 was screened out under different concentration (0, 1, 2, 5, and 10Mm) of lead. Different growth parameters (seed germination, seedling root and shoot length, seedling fresh and dry biomass, dry matter accumulation per seedling) and accumulation of lead by inoculated and non inoculated seedlings were observed and recorded. Results revealed that lead drastically reduce the seed germination and seedling growth of all three verities of Mash, while bacterial inoculations improved germination and various growth parameters of Mash varieties. All bacteria for variety NARC-Mash-3 and strain TE-12 for Variety NARC-Mash-2 had a positive relationship to combat lead stress by improving the seedling growth. Study also revealed that genetic variation of both plant variety and bacterial strain is important in developing a successful remedial mechanism.

Introduction

Heavy metal contamination is a major environmental problem throughout the world. Due to their technological importance they are used in many industries. Waste water from these industries has permanent toxic effects on human beings and environment (Anon., 2004). Lead is one of the biologically non essential element and a major heavy metal which has a soil retention time of 150-5000 years (Lerda, 1992; Roane, 1999). It enters the food chain through water and adversely affects the flora & fauna (Gupta *et al*., 2006). Enhanced levels of lead in soil could significantly reduce plant height, root-shoot ratio, dry weight, nodule per plant and chlorophyll content etc (Weirzbicka & Obidzinska, 1998; Farooqi *et al*., 2009; Kabir *et al.*, 2010).

Remediation of lead is getting more attention but the routine methods for its removal (physical or chemical) are either ineffective or extremely expensive (Anon., 2004). Use of such methods may also produce secondary wastes which may create problems. These constraints require an innovative, low cost, eco-friendly method for the removal of lead contamination. Some of the alternative strategies for the remediation are microbial and plant based metal remediation (Gadd, 2006). This is because numerous microorganisms harbor metal resistant detoxification mechanism. Like bacteria can combat metals in the environment by detoxifying their adverse effects either by binding with them or by mineralizing them (Nies, 1999). Bacterial transformation of metals is dependent on physical, chemical and biological properties of cells, the environmental factors and the type of metal. Microbial populations are known to affect the trace metal mobility and availability to the plants through release of chelates, acidification and redox changes. So the effective phytoremediation could be accomplished by bacteria having the potential of solubilizing essential minerals and heavy metals and promoting plant growth in contaminated soil. Use of bacterial strains to counter the toxic effects of heavy metals on the growth of plants has been reported by several workers (Burd *et al.*, 2000; Abou-shanab *et al*., 2006). Bacteria can promote plant growth indirectly by reducing/preventing the plant pathogens or directly through nitrogen fixation, phosphorus solubilization, siderophore formation, increasing phytohormones/enzymes etc (Glick *et al.*, 1999, Khan *et al*., 2011, Mehboob *et al.*, 2011; Kang *et al*., 2012; Inam-ul-Haq *et al*., 2012).

Present work was aimed to see the effects of three lead resistant bacteria on the growth of three varieties of *Vigna mungo* L. (Hepper) seedlings exposed to different concentrations of lead.

Materials and Methods

Three lead tolerant bacteria (TE07, TE12 & TE14) were selected for present study. These were isolated from tannery effluents and could tolerate 1000 µg/ml of lead in the growth medium (Tahir, 2009). While 3 different varieties of *Vigna mungo* L. (Hepper), NARC-Mash2 and NARC-Mash3 and NARC-Mash-97 were obtained from NARC, Islamabad.

Experiment was set following the method of Hasnain *et al*., (1993). Healthy seeds of each variety were selected and sterilized with 0.1% $HgCl_2$ solution. After that seeds were washed thoroughly to remove all the traces of mercury and soaked in autoclaved distilled water for 2-3 hours. Three lead resistant bacteria *Bacillus pumilus* (TE07), *Bacillus cerus* (TE12 & TE14) were grown on nutrient agar supplemented with lead (500µg/ml). Cultures were prepared by mixing bacterial growth in sterilized distilled water (10 ml) and OD was adjusted to 10^6 cells/ml. Presoaked sterilized seeds of each variety were divided into four groups and inoculated with respective bacterial culture (for inoculation seeds were co incubated with bacterial cultures at room temperature for 30 minutes), while leaving one group as un-inoculated control.

Control and Inoculated seeds (20 each) were placed in pre-labeled petri plates having filter papers. Ten ml of each treatment (lead concentration) was added in their respective plates and incubated in dark for 3 days. Seed germination was recorded for three consecutive days. On third day of germination, seedlings were supplied with 10 ml of Hewitt's nutrient solution (Hewitt, 1963) supplemented with respective lead concentrations and shifted to light (10

Klux) with a photoperiod of 16 h d^{-1} at 30±2°C in the growth chamber. Seedlings were harvested after 7 days of shifting and various growth parameters including germination, percentage root length, shoot length, seedling length (cm), fresh weight(g) dry weight(g) dry matter accumulation (mg/g) were recorded. Means of 8 replicates was used for compiling present results.

To analyze lead content in seedlings, wet digestion of dry biomass was performed in CEM microwave oven according to EPA protocol 3052 for microwave digestion of organic material. Digested material was filtered and diluted to get the absorption (Matusiewicz, 1997) on the atomic absorption spectrophotometer (spectra AA20). All data was subjected to statistical analysis.

Results and Discussion

Lead toxicity is important because of its constant increase in environment and adverse effects on various life forms. Though lead is non-biodegradable but can be transformed through various processes such as sorption, methylation, complexation or valence changes (Anon., 2004). This transformation can more easily be done with microorganisms, which posses many different resistant mechanisms. As these microorganisms especially bacteria have various interactions with other life forms specially plants so they may be helpful in combating the adverse effects. In present study the combined effects of lead and various lead tolerant bacteria were observed on three different varieties of Mash beans, results showed that there was a regular decrease (with a few exceptions) in the germination percentage of both inoculated and un-inoculated seeds with the increase in lead concentrations. When the effects of inoculations were compared, both decreases and increases were observed in case of Mash-2 and Mash-3, whereas all inoculations promoted the percentage germination of Mash-97 under all treatments of lead i.e., 0-10mM (Table 1). Many researchers have observed the inhibited germination and reduced plant growth due to lead toxicity (Iqbal & Shazia, 2004; Sharma & Durbey, 2005; Shafiq & Iqbal, 2005), while promotion in germination percentage due to bacterial inoculation under various heavy metal stresses is also reported (Hasnain *et al.,* 1993; Burd *et al.,* 2000; Gupta *et al.,* 2006). Improvement in germination and vigor index with bacterial inoculates is related with the increased activities of various antioxidant enzymes (Karthikeyan *et al.,* 2007) and / or increase in growth regulators (Ramamoorthy *et al*., 2000)

Seedling length comprised both of roots and shoot lengths and were uniformly decreased with the increase in the lead concentrations. Roots of both inoculated and un-inoculated seedlings were more severely affected as compared to shoots. Reduction in length parameters with increased concentrations of lead may be due to the reduction in mitotic activity or the lead ion incorporation into the cell wall components (Tomer *et al.,* 2000). Gupta *et al.,* (2006) reported a concentration dependent decrease in all growth parameters of Black gram under lead stress.

Mixed effects of bacterial inoculations were observed in different varieties of Mash. In case of Mash-3, all inoculations improved the seedling lengths and in many cases these were significant increases. In case of Mash-2, highly significant increase was observed in the absence of lead as compared to un-inoculated seedlings as well as under 10mM lead stress. For rest of the cases mixed response was observed. Strain TE-12 was the only one, which increased the seedling length from 0-10mM lead concentration as compared to non-inoculated Mash-2 seedlings (Table 2). Surprisingly all inoculations caused reductions under 0-2mM and then at 10mM as compared to un-inoculated respective treatments, while non-significant increase in this parameter was recorded under 5mM lead concentrations. Bacterial inoculations in many cases improved the germination, seedling length and weight parameters (Dobbelaere *et al*., 2003: Kang *et al.,* 2012).

Most interesting observation was recorded for Mash-97 seedlings, where bacterial inoculations reduced the length parameter as compared to control (Table 2). The three bacterial strains caused a non-significant increase only under 5mM lead stress. Though the germination of un-inoculated seeds of Mash-3 was good as compared to inoculated seedlings and reverse was the case for Mash-97, where all the inoculations under all lead concentrations improved the germination (Table 1), but the growth of seedlings was other way round. This showed that after germination bacteria developed a positive (synergistic) relationship towards the growing roots of Mash-3 seedlings, while they interacted negatively in case of Mash-97. Root exudates might play an important role to determine the nature of relationship between bacteria and the growing root tips.

As for as the weight parameters are concerned, there was a regular decrease in fresh weight per seedling and an increase in the dry weight per seedling, with the increase in lead concentration. Bacterial inoculations caused both increase and decreases in this parameter but there was no specific strain based pattern or concentration based pattern. For dry matter accumulation, almost a uniform trend was observed. For majority of the treatments, a gradual decrease (0-2mM) and then a gradual increase (5 & 10mM) in accumulated dry matter were observed (Table 3).

Lead accumulation was studied through atomic absorption spectrophotometer after digestion. It showed a regular increase from 1-10mM lead concentration. In most of the cases the lead contents measured in the presence of bacteria were less as compared to un-inoculated ones and in most cases these reductions were significant. While in few treatments enhancement in lead accumulation (as compared to control treatments) was also observed (Table 4). These increases/decreases in lead accumulation were neither regular nor specific with reference to bacterial strains or lead treatments. For example increased accumulation was observed under 1mM (Mash-97), 2mM (Mash-2) and 5mM (Mash-3), while for rest of the treatments for these varieties, inoculations caused a reduction in this parameter. Bacteria can improve the plant growth by increasing the nutrients availability or by increasing the resistance against toxic metal or by reducing the metal availability to the roots (Reed & Glick, 2005; Vivas *et al.,* 2006; Li *et al*., 2007; Rajkumar & Freitas, 2008).

Table 1. Effect of Lead resistant bacteria (TE-07, TE-12, TE-14) on the germination (%age) of Mash varieties under Lead stress (mM).

Var	Mash-2					Mash-3					Mash-97				
	0mM	1mM	2mM	5mM	10mM	0M	1mM	2mM	5mM	10mM	0mM	1mM	2mM	5mM	10mM
Control	95.00 ±2.88	88.33 ±1.67	83.33 ±3.33	90.00 ±2.88	85.00 ±2,88	85.00 ±2.88	88.33 ±1.66	86.66 ±1.66	83.33 ±3.33	83.33 ± 4.40	85.00 ± 5.0	80.00 ±2.88	76.66 ±1.66	75.00 ±2.88	70.00 ± 2.88
TE-07	93.33 ±3.33	91.67 ±4.40	88.33 ±4.40	83.33 ±4.40	76.66 ±1.66	83.33 ±4.40	78.33 ±4.40	86.66 ±1.66	83.33 ±4.40	83.33 ± 1.66	96.66 ±1.66	91.66 ±1.66	83.33 ±1.66	83.33 ±3.33	86.66 ± 4.40
TE=12	96.66 ±3.33	93.33 ±1.66	91.66 ±1.66	88.33 ±1.66	80.00 ± 4.40	98.33 ±1.66	78.33 ±3.33	75.00 ±5.00	85.00 ± 5.0	86.66 ±1.66	95.00 ±2.88	91.66 ±3.33	90.00 ±2.88	91.66 ±1.66	81.66 ± 4.40
TE-14	88.30 ±3.33	86.67 ±4.40	85.00 ±2.88	83.30 ±1.66	78.33 ± 4.40	88.33 ±3.33	86.6 ±1.66	81.67 ±1.66	80.00 ±2.88	80.66 ± 3.33	93.30 ±1.66	91.66 ±6.00	90.00 ±7.63	85.00 ±2.88	75.00 ± 2.88

Table 2. Effect of Lead resistant bacteria (TE-07, TE-12, TE-14) on the seedling lengths (cm) of Mash varieties under Lead stress (mM).

Var	Mash-2					Mash-3					Mash-97				
	0mM	1mM	2mM	5mM	10mM	0M	1mM	2mM	5mM	10mM	0mM	1mM	2mM	5mM	10mM
Control	14.62 ± 0.22	12.80 ± 0.91	11.50 ± 0.14	6.70 ± 0.76	0.52 ± 0.15	19.92 ± 1.67	12.51 ± 1.54	12.42 ± 1.47	5.93 ± 0.28	2.43 ± 0.07	23.41 ± 0.98	13.46 ± 1.26	13.08 ± 0.94	5.61 ± 1.25	2.42 ± 0.75
TE-07	19.94 ± 1.52	12.59 ± 1.22	11.69 ± 1.12	6.40 ± 0.51	2.44 ± 0.45	20.30 ± 1.43	13.20 ± 1.18	13.06 ± 1.25	7.30 ± 1.22	2.70 ± 0.55	21.10 ± 0.89	13.80 ± 1.04	12.25 ± 1.31	6.32 ± 0.28	1.98 ± 0.06
TE=12	17.31 ±0.63	13.26 ± 1.00	12.33 ± 1.66	8.07 ±0.29	2.50 ± 1.18	23.14 ± 2.66	13.02 ± 1.28	13.43 ± 0.89	7.30 ± 0.73	2.45 ± 0.82	20.92 ± 1.19	12.85 ± 1.49	12.42 ± 1.07	6.36 ± 1.46	2.49 ± 0.61
TE-14	18.90 ± 1.31	15.07 ± 1.40	9.82 ± 1.15	5.35 ± 0.65	2.05 ± 0.29	20.89 ± 0.56	14.01 ± 1.02	13.17 ± 0.89	6.40 ± 0.97	2.33 ± 0.66	21.70 ± 0.97	12.11 ± 1.35	11.28 ± 0.94	5.81 ±0.31	2.65 ± 0.19

Table 3. Effect of Lead resistant bacteria (TE-07, TE-12, TE-14) on the dry matter accumulation (mg/gm) of Mash varieties under Lead stress (mM).

Var	Mash-2					Mash-3					Mash-97				
	0mM	1mM	2mM	5mM	10mM	0M	1mM	2mM	5mM	10mM	0mM	1mM	2mM	5mM	10mM
Control	24.33 ± 2.85	17.04 ± 2.00	15.92 ± 2.65	20.77 ± 2.25	39.69 ± 1.88	24.73 ± 1.23	27.03 ± 3.50	25.93 ± 3.87	25.17 ± 3.55	37.25 ± 5.32	28.35 ± 1.46	14.24 ± 0.44	13.81 ± 1.25	17.36 ± 1.04	35.10 ± 5.26
TE-07	18.15 ± 1.16	22.75 ± 2.23	22.90 ± 2.66	20.05 ± 1.03	39.05 ± 3.42	27.27 ± 1.29	14.66 ± 1.39	14.60 ± 0.37	21.55 ±2.47	39.82 ± 0.71	22.62 ± 3.13	19.55 ± 2.80	12.61 ± 0.70	18.00 ± 1.76	45.22 ± 4.40
TE=12	18.20 ± 1.45	18.73 ± 1.78	20.87 ± 1.64	21.66 ± 2.04	38.02 ± 2.71	27.27± 1.29	15.66 ± 1.39	14.60 ± 0.37	20.55 ± 2.47	40.82 ± 0.71	27.03 ± 1.93	14.17 ± 1.27	15.34 ± 1.00	15.51 ± 1.88	42.98 ± 0.94
TE-14	24.29 ± 2.66	21.24 ± 0.97	20.46 ± 1.76	24.24 ± 1.54	50.31 ± 4.13	24.36 ± 1.77	12.78 ± 0.84	14.51 ± 2.51	17.43 ± 1.04	27.44 ± 2.66	24.23 ± 1.76	14.88 ± 1.55	15.26 ± 1.27	26.20 ± 2.11	43.67 ± 3.35

Table 4. Effect of Lead resistant bacteria (TE-07, TE-12, TE-14) on the lead accumulation (mg/g dry wt.) of Mash varieties under Lead stress (mM).

Var	Mash-2					Mash-3					Mash-97				
	0mM	1mM	2mM	5mM	10mM	0M	1mM	2mM	5mM	10mM	0mM	1mM	2mM	5mM	10mM
Control	-	11.67 ± 1.76	12.99 ± 0.41	71.53 ± 4.05	187.8 ± 31.2	-	20.62 ± 2.09	22.15 ± 4.18	66.94 ± 4.12	173.9 ± 18.1	-	7.91 ± 0.76	30.86 ± 1.50	225.0 ± 2.28	240.0 ± 13.4
TE-07	-	6.19 ± 1.39	13.24 ± 2.94	50.84 ± 2.26	206.0 ± 19.7	-	4.15 ± 0.12	18.44 ± 0.21	91.50 ± 1.04	151.3 ± 1.04	-	18.05 ± 5.32	25.84 ± 6.35	91.16 ± 1.19	275.9 ±31.2
TE=12	-	37.0 ± 6.55	19.28 ± 0.58	110.1 ± 11.5	168.2 ± 36.0	-	5.53 ± 0.36	19.73 ± 1.71	121.7 ± 5.80	142.6 ± 4.64	-	10.06 ±0.76	29.04 ± 2.56	90.42 ± 2.75	185.6 ± 22.8
TE-14	-	6.15 ± 2.13	23.97 ± 6.93	81.20 ± 4.88	184.5 ±25.1	-	8.02 ± 2.49	17.80 ± 10.6	173.8 ± 1.61	156.7 ± 9.83	-	8.20 ± 1.01	32.41 ±3.06	57.85 ± 14.7	186.5 ± 3.91

Variations in the accumulation trend might be due to the genetic variability found in three varieties of Mash as well as the three bacterial inoculations. Another reason might be the specific relationship of the bacteria with the plant varieties, which was evident in case of Mash 3. In that variety, all inoculations enhanced the seedling growth (Table 2), but 2 strategies were adopted by the seedling-microbe-interaction to combat the stress; one by keeping the toxic metal ions away from the growing seedlings whereas at higher concentrations by accumulating more lead. In few cases a positive while for others a negative correlation was observed between the dry matter accumulation and lead accumulation by the seedlings.

Present work revealed that lead tolerant bacteria may be effectively used to improve the growth of Mash under lead stress. This study also showed that apart from other factors, effective plant-microbe interaction is dependent upon genetic variability of both the bacterial strain and the plant variety.

References

Abou-Shanab, R.A., J.S. Angle and R.L. Chaney. 2006. Bacterial inoculants affecting nickel uptake by *Alyssum murale* from low, moderate and high Ni soils. *Soil. Biol. Biochem*., 38: 2882-2889.

Anonymous. 2004. *Bioremediation of Arsenic, Chromium, Lead and Mercury*. United states Environment Protection Agency, Office of solid waste and emergency response technology innovation office Washington DC.

Burd, G.I., D.G. Dixon, B.R. Glick. 2000. Plant growth promoting bacteria that decrease heavy metal toxicity in plants. *Can. J. Microbiol.*, 46: 237-245.

Dobbelaere, S., J. Vanderleyden and Y. Okon. 2003. Plant growth promoting effects of diazotrophs in the rhizosphere. *Critical Rev. Plant Sci.*, 22: 107-149.

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. *Pak. J. Bot.*, 41: 27-33.

Gadd, G.M. 2006. *Fungi in Biogeochemical Cycles*. Cambridge University Press. Cambridge.

Glick, B.R., C.L. Patten, G. Holguin and D.M. Penrose. 1999. *Biochemical and genetic mechanism used by plant growth promoting bacteria.* Imperial College press, London, UK.

Gupta, D.K., A. Srivastava and V.P. Singh. 2006. Phytoremediation of induced lead toxicity in *Vigna mungo* (L.) hepper by vetiver grass. Rohilkhand University.

Hasnain, S., S. Yasmin and A. Yasmin. 1993. The effect of lead-resistant Pseudomonads on the growth of *Triticum aestivum* seedling under the lead stress. *Environ. Pollu.*, 81: 179-184.

Hewitt, E.J. 1963. Minerals nutrition of plants in culture media, In: (Steward, F.C. ed.) *Plant Physiology*. Academic Press, New York, pp. 99-137.

Inam-ul-Haq, M., S. Mehmood, H.M. Rehman, Z. Ali and M.I. Tahir. 2012. Incidence of root rot diseases of soyabean in Multan, Pakistan and its management by the use of plant growth promoting rhizobacteria. *Pak. J. Bot.*, 44 (6): 2077-2080.

Iqbal, M.Z. and Y. Shazia. 2004. Reduction of germination and seedling growth of Leucaena leucocephala caused by lead and cadmium individually and combination. *Ekologia (Bratislava)*, 23(2): 162-168.

Kabir, M., M.Z. Iqbal, M. Shafiq and Z.R. Farooqi. 2010. Effects of lead on seedling growth of *Thespesia poulnea* L. *Plant Soil Environ.*, 56(4): 194-199.

Kang, S.M., A.L. Khan, M. Hamayun, Z.K. Shinwari, Yoon-Ha Kim, Gil-Jae Joo and In-Jung Lee. 2012. *Acinetobacter calcoaceticus* ameliorated plant growth and influenced gibberellins and functional biochemicals. *Pak. J. Bot.*, 44(1): 365-372.

Karthikeyan, B., V.A. Jaleel and M. Deiveekasundaram. 2007. Alterations in seedling vigor and antioxidant enzyme activation in *Catharanthum roseus* under seed priming with native diazotrophs. *J. Zhejiang Univer. Sci.*, 8: 453-457.

Khan, A.L., M. Hamayun, S.A. Khan, Z.K. Shinwari, M. Kamaran, S.M. Kang, J.G. Kim and I.J. Lee. 2011. Pure culture of *Metarhizium anisopliae* LHL07 reporgrams soybean to higher growth and mitigates salt stress. *World J. Microb Biotech*., 28(4): 1483-94.

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

Li, W.C., Z.H. Ye and M.H. Wong. 2007. Effects of bacteria on enhanced metal uptake of the Cd/Zn hyperaccumulating plant, *Sedum alferdii. J. Exp. Bot.*, 58: 4173-4182.

Matusiewicz, H. 1997. Developmenat of high pressure closed vessel system for Microwave-assisted sample digestion. In: *Microwave Enhanced Chemistry, fundamental, Sample Preparation and Applications*, (Eds.): H.M. Skip, Kingston and S.J. Haswel. *Am. Chem. Soc*., Washington, pp. 353.

Mehboob, I., Z.A. Zahir, M. Ashraf, A. Tanveer and Farooq-e-Azam. 2011. Growth promoting activities of different *Rhizobium spp.* in wheat. *Pak. J. Bot.*, 43(3): 1643-1650.

Nies, D.H. 1999. Microbial heavy metal resistance. *Appl. Microbiol. Biotechnol*., 51: 730-50.

Rajkumar, M. and H. Freitas. 2008. Effects of inoculation of plant growth promoting bacteria on Ni uptake by Indian mustard. *Bioresour. Technol.*, 99: 3491-3498.

Ramamoorthy, K., N. Natarajan and A. Lakshaman. 2000. Seed biofortification with *Azospirillum* spp. For improvement of seedling vigor and productivity in rice (*Oryza sativa* L.). *Seed. Sci. Technol*., 28: 809-815.

Reed, M.L.E. and B.R. Glick. 2005. Growth of canola (*Brassica napus*) in the presence of plant growth-promoting bacteria and either copper or polycyclic aromatic hydrocarbons. *Can. J. Microbiol.*, 51: 1061-1069.

Roane, T.M. 1999. Lead resistance in two bacterial isolates from heavy metal contaminated soils. *Microbiol. Ecol*., 37: 218-224.

Shafiq, M. and M.Z. Iqbal. 2005. The toxicity effects of heavy metals on germination and seedling growth of *Cassia siamea* Lamark. *J. New Seeds*, 7: 95-105.

Sharma, P. and R.S. Dubey. 2005. Lead toxicity in plants, *Braz. J. Plant Physiol.*, 17(1): 35-52.

Tahir, U. 2009. *Heavy metal resistant bacteria from tannery effluents.* MSc. Thesis. Department of Environmental Sciences, Fatima Jinnah Women University, Rawalpindi, Pakistan.

Tomar, M., I. Kaur, Neelu and A.K. Bhatnagar. 2000. Effect of enhanced lead in soil on growth and development of *Vigna radiata* (L.) Wilezek. *Ind. J. Pant Physiol*., 5: 13-18.

Vivas, A., B. Biro, J.M. Ruiz-Lozano, J.M. Barea and R. Azcon. 2006. Two bacterial strains isolated from a Zn-polluted soil enhance plant growth and mycorrhizal efficiency under Zn-toxicity. *Chemosphere*, 62: 1523-1533.

Wierzbicka, M. and J. Obidzinska. 1998. The effect of lead on seed imbibitions and germination in different plant species. *Science*, 137: 155-171.

(Received for publication 1 September 2012)