Pak. J. Bot., 38(5): 1389-1396, 2006.

EFFECT OF LEAD AND CHROMIUM ON GROWTH, PHOTOSYNTHETIC PIGMENTS AND YIELD COMPONENTS IN MASH BEAN [*VIGNA MUNGO* (L.) HEPPER]

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Abstract

A pot experiment was carried out to study the effect of lead and chromium on growth, chlorophyll contents and yield components in two mash bean [*Vigna mungo* (L.) Hepper] cultivars i.e. Fs-1 and Mash-97. Fourteen-day old plants were exposed to 20 or 40 mg L^{-1} lead or chromium whereas control plants were treated with distilled water only. Application of both lead and chromium caused a significant reduction in all growth parameters as compared with that of control. The extent of decrease in growth due to chromium compared with lead. Although high concentration of both metals in the rooting media drastically reduced all photosynthetic pigments, chromium application caused more reducing effect as compared to chromium. In addition, all yield attributes of both cultivars of mash bean reduced due to both metals in rooting medium. The sensitivity of mash bean to chromium was greater as compared to lead. In conclusion, mash bean cultivar FS-1 proved to be tolerant as it showed less reduction in growth, photosynthetic pigments and yield as compared to Mash-97.
Keywords: lead, chromium, growth, photosynthetic pigments, yield

Introduction

Worldwide, metal contamination has extremely increased in the biosphere as a result of anthropogenic activities. This situation is alarming in the developing world where untreated waste water is extensively used for irrigation or is disposed off in water resources (UNIDO, 2002). These metals are deposited in different soil profiles leading to long-term metal contamination. These metals are mostly absorbed by plants easily and prove toxic to plants that can be observed as growth retardation as a result of alterations in biochemical process like inhibition of enzyme activity, protein penetration and impaired nutrition *etc.* (Poschenrieder, 1990; Arun *at al*., 2005).

In the recent era, lead (Pb) contamination has gained a considerable attention as a potent environmental pollutant. Significant increases in the Pb content of cultivated soils have been observed near urban and industrial areas where it tends to accumulate in the surface ground layer (de Abreu *et al*., 1998). Despite regulatory measures adopted in many countries to have a check on Pb input in the environment, it continues to be one of the most serious global environmental hazards in the developing world (Yang *et al*., 2000).

Chromium is a naturally occurring element found in rocks, animals, plants, soil, and in volcanic dust and gases. Chromium exists in the environment in several different forms with more common forms as chromium (III) and chromium (VI). Chromium (III) occurs naturally in the environment and is an essential nutrient whereas chromium (VI) is

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generally produced by industrial processes. The impact of Cr contamination in the physiology of plants depends on the metal speciation, which is responsible for its mobilization, subsequent uptake and resultant toxicity in the plant system (Shanker *et al*., 2005).

Mash bean [*Vigna mungo* (L.) Hepper] is among the most important pulse crops of the world. It has great value as food, fodder and green manure. In addition to improving the soil fertility, it is a cheap source of protein for direct human consumption. Chemical analysis of mash bean seed indicates that it contains protein (20-24%), oil (2.1%), Fats (1-2%), carbohydrates and a fair amount of vitamin A and B (James, 1981). Thus it has a great potential to improve protein deficiency in human beings by providing a low cost protein.

Keeping in view the increasing use of sewage water for irrigation and resultant excessive accumulation of lead and chromium in different soil profiles, it was of great importance to evaluate their toxic effects on growth, photosynthetic pigments and yield components in mash bean, a potential legume crop of Pakistan.

Materials and Methods

Ten seeds of the two cultivars [Fs-1 and Mash-97] were sown in plastic pots containing 10 kg sandy loam soil [ECe = 1.2 ds/m and pH = 7.95] and after germination thinned to leave four plants of similar size. Fourteen-day old plants were exposed to 20 and 40 mg L^{-1} of lead nitrate [$Pb(NO_3)_2$] or chromium chloride [$CrCl_3.6H_2O$] dissolved per liter distilled water, while control plants were treated with distilled water only. Plants were irrigated regularly with tap water during the course of study. The selected levels were used on the basis of lead (20-30 mg Kg^{-1} soil at 0-90 cm depth) and chromium (20-40 mg Kg^{-1} soil at 0-90 cm depth) content reported in Pakistani soils irrigated regularly by metal contaminated water in the vicinities of large cities like Faisalabad (Ensink *et al*., 2002; UNIDO 2002; van der Hoek *et al*., 2002; Ensink *et al*., 2007). The experiment was laid down in a completely randomized design (CRD) with four replicates.

The data for relative increase in various growth attributes were started 10 days after treatment for three consecutive 7-day harvest intervals. After each harvest, roots and shoots were separated after harvesting from soil, and shoot and root lengths were measure using a meter rod. The fresh weight of shoots and roots were determined with the help of analytical balance, thereafter plants were oven dried at 70 ^{o}C to a constant dry weight and data for dry weights were recorded. The chlorophyll (Chl. a, b and total) contents were determined at third-harvest following the method of Arnon (1949). A 0.5 g fresh leaf sample was chopped into small pieces and extracted with 80% acetone. The absorbance was read at 645 nm and 663 nm using spectrophotometer (Hitachi, U-2001, Japan) and total amount of chlorophyll a, b and total chlorophyll were calculated. At maturity, data for number of pods per plant, seeds per pod, 100-seeds weight and yield per plant were recorded.

Statistical analysis: Analysis of variance (ANOVA) of the data was computed using a STATISTICA package for windows, Kernel Release 5.5A (StatSoft, Inc, Tulsa, OK 74104, USA). The Visual General Linear Model (GLM) was fitted using the same statistical package for assessing variation within treatments and interaction terms by splitting their degree of freedom into contrast comparisons.

Results

Application of metals in the rooting medium significantly reduced all growth attributes of both mash bean cultivars. The growth rate of both cultivars was found to be maximum at first interval thereafter decreased consistently. Metal stress reduced the growth rate of both cultivars as well. However, both levels of chromium were more toxic as compared to that of lead. Furthermore, highest level of both Cr and Pb maximally reduced the plant growth attributes. Moreover, this adverse effect of both metals was less on growth of FS-1 than that of Mash-97 (Fig. 1, 2 &3).

All photosynthetic pigments were drastically reduced by both metal treatments. Analysis of variance of the data for chlorophyll a, b and total chlorophyll contents revealed highly significant differences for genotypes and treatments. However genotype x treatment interaction differed non-significantly. Genotype FS-1 had higher photosynthetic pigments as compared to Mash-97 under both metal stresses. Overall, Fig. 5: Yield attributes of two mashbean cultivars when 14-day old plants were exposed to Pb or Cr in the root medium.

Table 1. Analyses of variance (ANOVA) and contrast comparisons for photosynthetic pigments of two mash bean genotypes under lead and chromium stresses.

General effect	d.f.	Chl a	Chl b	Total Chl
Intercept	1	34.63 ***	14.90 ***	136.34 ***
Genotypes (G)	1	0.213 ***	0.224 ***	0.513 ***
Treatment (T)	4	0.380 ***	0.186 **	0.313 ***
C vs All	1	0.927 ***	0.322 ***	0.777 ***
C vs Pb	1	0.441 ***	0.132 **	0.414 ***
C vs Cr	1	1.197 ***	0.452 ***	0.932 ***
Pb vs Cr	1	0.277 ***	0.143 ***	0.155 **
V_1C vs V_1 Pb	1	0.169 **	0.060 **	0.160 *
V_1C vs V_1 Cr	1	0.470 ***	0.260 ***	0.555 ***
V_2C vs V_2 Pb	1	0.288 ***	0.073 ***	0.260 **
V_2C vs V_2 Cr	1	0.742 ***	0.194 **	0.385 ***
V_1 Pb_1 vs V_1 Pb_2	1	0.040 *	0.097 **	0.146 *
V_1 Cr_1 vs V_1 Cr_2	1	0.088 **	0.054 *	0.063 *
V_2 Pb_1 vs V_2 Pb_2	1	0.053 *	0.045 *	0.054 *
V_2 Cr_1 vs V_2 Cr_2	1	0.132 **	0.064 *	0.068 *
G x T interaction	4	0.004 ns	0.003 ns	0.013 ns
V_1 Pb vs V_2 Pb	1	0.093 **	0.122 **	0.354 ***
V_1 Cr vs V_2 Cr	1	0.126 ***	0.055 **	0.094 *
V_1 Pb vs V_2 Cr	1	0.049 ***	0314 ***	0.533 ***
V_1 Cr vs V_2 Pb	1	0.002 ns	0.0006 ns	0.030 ns
Error	30	0.008	0.0065	0.015

*, **, ** = significant at 0.05, 0.01 and 0.001 levels, respectively. ns = non-significant, V_1 = FS-1, V_2 = Mash-97, C = Control, Pb_1 = Lead nitrate (20 mg/L), Pb_2 = Lead nitrate (40 mg/L), Cr_1 = Chromium chloride (20 mg/L), Cr_2 = Chromium chloride (40 mg/L), The 4 degree of freedom have been split into contrasts to check the variation within treatments (T) or variety x treatment (V x T) interaction.

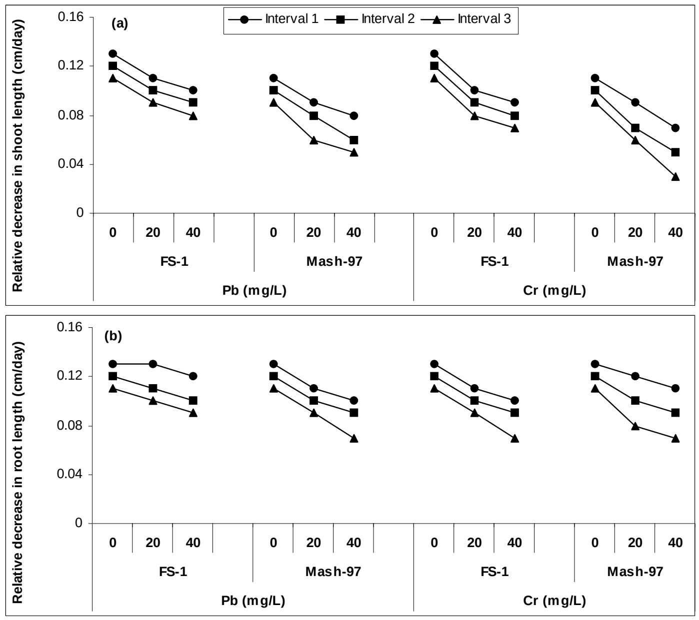

Fig. 1. Relative decrease in shoot and root lengths of two mashbean cultivars when 14- day old plants were exposed to Pb or Cr in the root medium.

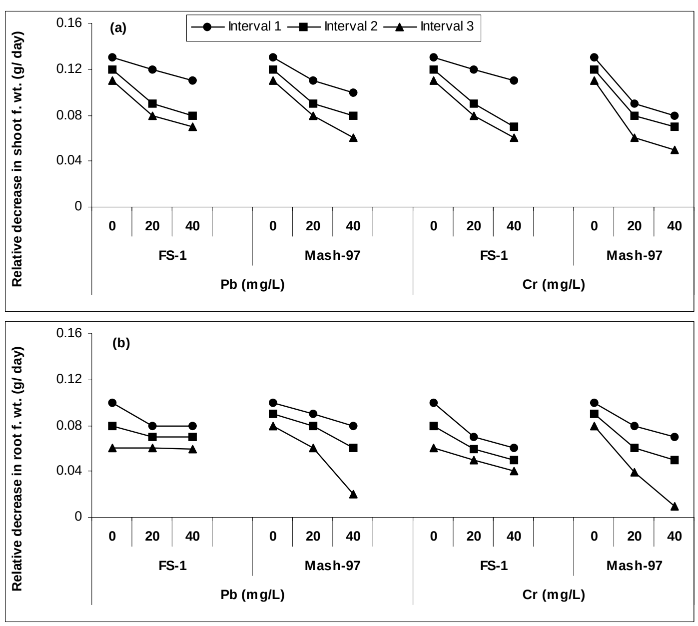

Fig. 2. Relative decrease in shoot and root fresh weights of two mashbean cultivars when 14-day old plants were exposed to Pb or Cr in the root medium.

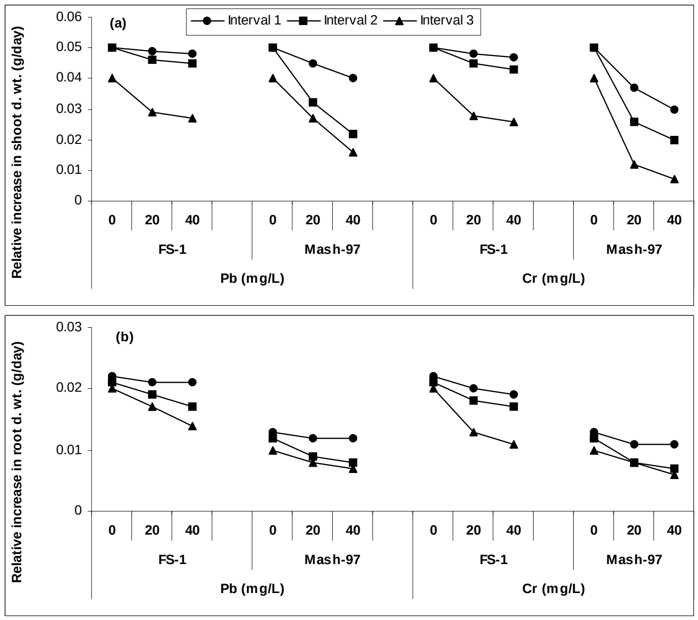

Fig. 3. Relative decrease in shoot and root dry weights of two mash bean cultivars when 14-day old plants were exposed to Pb or Cr in the root medium.

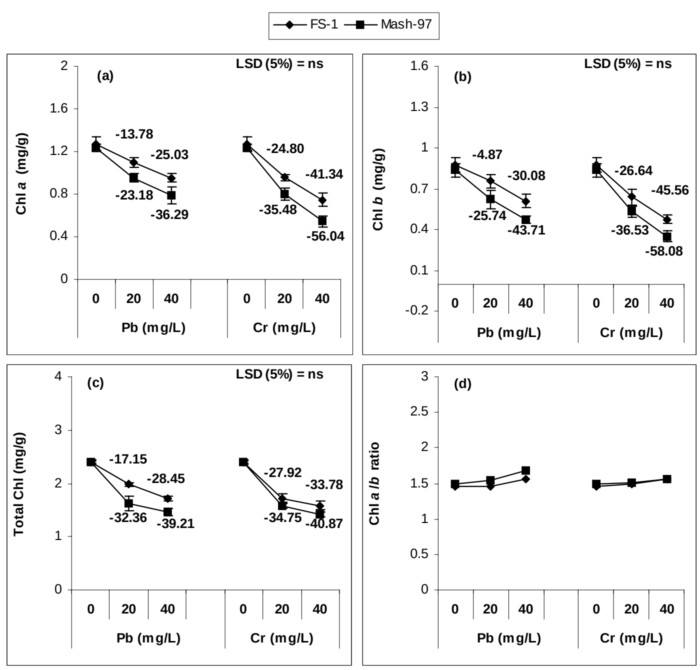

Fig. 4. Photosynthetic pigments of two mash bean cultivars when 14-day old plants were exposed to Pb or Cr in the root medium.

Table 2. Analyses of variance (ANOVA) and contrast comparisons for yield attributes of two mash bean genotypes under lead and chromium stresses.

General effect	d.f.	Pods/plant	Seeds/pod	Yield/plant	100 seed weight
Intercept	1	230.40 ***	455.62 ***	6.14 ***	3.55 ***
Genotypes (G)	1	6.40 ***	3.02 *	0.0023 ***	0.123 ***
Treatment (T)	4	4.83 ***	8.18 ***	0.0027 ***	0.085 ***
C vs All	1	9.50 ***	18.90 ***	0.0051 ***	0.194 ***
C vs Pb	1	6.02 ***	10.02 ***	0.0034 ***	0.120 ***
C vs Cr	1	10.08 ***	21.33 ***	0.0061 ***	0.209 ***
Pb vs Cr	1	0.78ns	2.53 *	0.0007 **	0.018 *
V_1C vs V_1 Pb	1	2.04 *	4.16 *	0.0012 ***	0.065 **
V_1C vs V_1 Cr	1	6.00 **	12.04 ***	0.0056 ***	0.106 **
V_2C vs V_2 Pb	1	4.16 **	7.04 **	0.0020 ***	0.055 **
V_2C vs V_2 Cr	1	4.17 **	9.37 ***	0.0025 ***	0.103 ***
V_1 Pb_1 vs V_1 Pb_2	1	3.13 *	2.00 *	0.0014 ***	0.049 **
V_1 Cr_1 vs V_1 Cr_2	1	2.00 *	3.12 *	0.0005 *	0.010 *
V_2 Pb_1 vs V_2 Pb_2	1	2.00 *	3.13 *	0.0006*	0.015 *
V_2 Cr_1 vs V_2 Cr_2	1	2.00 *	3.13 *	0.0005 *	0.068 ***
G x T interaction	4	0.21 ns	0.21 ns	0.0001 ns	0.004 ns
V_1 Pb vs V_2 Pb	1	5.06 **	3.06 *	0.0015 ***	0.044 **
V_1 Cr vs V_2 Cr	1	1.00 ns	0.25 ns	0.0002 *	0.052 **
V_1 Pb vs V_2 Cr	1	5.06 **	5.06 **	0.0020 ***	0.099 ***
V_1 Cr vs V_2 Pb	1	1.00 ns	0.0001 ns	0.0000 ns	0.015 *
Error	30	0.300	0.425	0.0004	0.0023

*, **, ** = significant at 0.05, 0.01 and 0.001 levels, respectively. ns = non-significant, V_1 = FS-1, V_2 = Mash-97, C = Control, Pb_1 = Lead nitrate (20 mg/L), Pb_2 = Lead nitrate (40 mg/L), Cr_1 = Chromium chloride (20 mg/L), Cr_2 = Chromium chloride (40 mg/L), The 4 degree of freedom have been split into contrasts to check the variation within treatments (T) or variety x treatment (V x T) interaction.

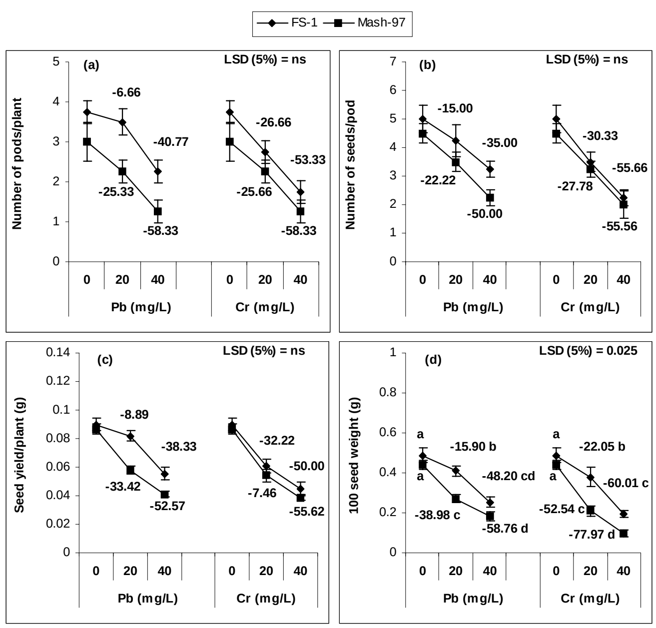

Fig. 5. Yield attributes of two mash bean cultivars when 14-day old plants were exposed to Pb or Cr in the root medium.

Discussion

In this study, metal treatments drastically reduced growth of both mash bean cultivars. It was observed that both metals were more toxic at higher levels as compared to their lower ones. This decrease in growth might be attributed to decrease in photosynthetic pigments that were drastically reduced in this study (Sharma & Sharma, 2003) that lead to decreased photosynthesis and hence decreases supply of photosynthesis products to the actively growing organs that suppressed growth (Fargašová, 1998). Moreover disturbance in nutrient uptake and metabolism as a result of increased metal content in the growth environment (data not reported) might explain this reduction in growth (Panda & Choudhary, 2005).

Chlorophyll a, b and total chlorophyll were drastically reduced under both metal treatments especially at higher level. In this study, severe chlorosis on older leaves and scarce appearance on younger leaves suggested that decline in chlorophyll content in shoots of metal treated plants result mostly from its enhanced degradation or reduced synthesis (Stobart *et al*., 1985; Somashekaraiah *et al*., 1992). Moreover, chlorophyll a/b ratio was not affected under metal treatments (Figure 4). This suggested that both chlorophyll a and b were equally sensitive to metal stress in both mash bean cultivars (Gajewska *et al.*, 2006).

All yield parameters were drastically reduced under both metal stresses. This reduction in yield might be due to decreased photosynthesis as a consequence of reduction in photosynthetic pigments under both metal stresses (Fargašová, 1998). In addition, decrease vegetative growth under metal stress lead to suppressed reproductive growth (Arun *et al*., 2005). Moreover, flower and pod senescence as a consequence of metal toxicity lead to production of less number of viable pods and seeds that reduced yield under metal stress (Sharma & Dubey, 2005).

Overall, highly toxic effects of chromium and lead metals on growth, photosynthetic pigments and yield components in mash bean were observed in this study. Moreover, mash bean cultivar FS-1 proved to be more tolerant as compared to Mash-97.

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(Received for publication 18 September, 2006)

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Published by the Pakistan Botanical Society.
Printed at Karachi University Press, Karachi-75270, Pakistan. Phone: 9243186, 4969237.
Copies available from the Chief Editor, Pakistan Journal of Botany, Department of Botany, University of Karachi, Karachi-75270, Pakistan. Phone: 4387867.
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