J. Phytol. Res. **23**(2) : 289-293, 2010

EFFECTS OF HEAVY METALS ON SEED GERMINATION AND SEEDLING GROWTH IN *LENS ESCULENTA* MOENCH. CV. L4076

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Effects of heavy metals (Cu, Ni, Pb, Zn and Cd) on root length, shoot length, seed germination and fresh weight of *Lens esculenta* Moench.(lentil) were studied. For this purpose, lentil seeds were germinated at various concentrations (10, 50, 100, 200, 500 and 1000 ppm) of heavy metals under laboratory conditions for 10 days. It was determined that growth parameters studied were inhibited with increasing concentration of heavy metals with Cd the most toxic and Zn the least toxic among heavy metals.

Keywords : Germination; Heavy metals; *Lens esculenta;* Root length; Shoot length; Weight.

Introduction

Heavy metals are natural components of the Earth's crust. They cannot be degraded or destroyed. Heavy metal contamination affects the biosphere in many places worldwide. Excess concentrations of some heavy metals in soils have caused the disruption of natural aquatic and terrestrial ecosystems.

Abandoning cultivation in the contaminated soils is a mistake. Such soils turn into a wasteland posing hazards for the underground water and watercourses. In such conditions, a proper selection of plant species considering soil levels of heavy metals and the crop sensitivity to soil contamination is extremely important[1]. Studies are required to determine those varieties of agriculture plants which can tolerate heavy metal toxicity. The effects of heavy metals change according to grown plants species. Due to small soil requirements and high dietary value of lentil (*Lens esculenta* Moench.) seeds, expanding this plant cultivation seems justified. In this study, the effects of heavy metals (Cu, Ni, Pb, Zn and Cd) on root length, shoot length, seed germination and fresh weight of *Lens esculenta* Moench. cv. L4076, was determined.

Material and Methods

Certified seeds of *Lens esculenta* Moench. cv. L4076 were obtained from Durgapura Agricultural Research Station, Jaipur. Seeds were stored in glass stoppered bottles. After a preliminary selection for uniformity criteria (size and colour of seeds), the seeds were surface sterilized with 0.1 % $HgCl_2$ for two minutes[2], then washed with distilled water three times and then soaked for two hours in respective solutions of different concentrations (10, 50, 100, 200, 500 and 1000 ppm) of Copper sulphate, Nickel sulphate, Lead sulphate, Zinc sulphate and Cadmium sulphate. Seeds soaked in distilled water for two hours constituted the control. After the above treatments, seeds were removed and allowed to germinate in Petriplates on filter paper soaked in each of the above metallic solutions. Three replicates, each of 10 seeds, were kept for each concentration of each heavy metal. The experiments were carried out for ten days under laboratory conditions of temperature (25±2°C) and diffuse light. On the day of termination of experiment, percentage of germination, root length, shoot length and fresh weight of seedlings were recorded and statistically analyzed[3,4].

Result and Discussion

All data regarding the effects of heavy metals on root length, shoot length, seed germination percentage and seedlings fresh weight are recorded in Tables 1-4, respectively.

Effect of Heavy Metals on Root Length - Root length of seedlings of *L. esculenta* cv. L4076 slightly increased at low concentration (10 ppm) in Pb, (10 and 50 ppm) in Zn and(10 ppm) in Cd. Root length was adversely affected at higher concentration of heavy metals. A drastic reduction in root length at 1000 ppm of Cd was observed where it reduced to 0.3cm. Results were statistically not significant between concentration *versus* chemicals. Highly significant results were observed among chemicals and among concentrations (Table 1).

Effect of Heavy Metals on Shoot Length - Sharp decline in shoot length were observed with increase in

Table 1. Effect of heavy metals on Root Length (cm) of seedlings in *Lens esculenta* cv. L4076 (values are mean of three replicates each).

Name of Chemical	Control (cm)	Concentration (ppm)					
		10	50	100	200	500	1000
Copper sulphate	9.6	6.1	5.8	5.6	4.7	2.93	2.5
Nickel sulphate	9.6	7.9	7.1	7.1	6.7	3.5	2.5
Lead sulphate	9.6	10.3	9.5	8.1	8.0	8.0	7.6
Zinc sulphate	9.6	11.0	10.7	9.5	8.1	8.0	8.0
Cadmium sulphate	9.6	10.3	9.2	6.8	3.6	1.6	0.3

Source	D:F.	S.S.	M.S.S.	F Ratio
Chemicals	4	264.652	66.163	7.312**
Concentrations	6	400.715	66.785	7.381**
Interaction	24	155.827	6.492	0.717NS
Error	70	633.326	9.047	

NS=Not Significant, **= Highly Significant.

Table 2. Effect of heavy metals on Shoot Length (cm) of seedling in *Lens esculenta* cv. L4076 (values are mean of three replicates each).

Name of Chemical	Control (cm)	Concentration (ppm)					
		10	50	100	200	500	1000
Copper sulphate	7.2	5.1	4.4	4.5	4.3	3.3	4.4
Nickel sulphate	7.2	4.6	4.6	6.1	5.3	4.7	4.7
Lead sulphate	7.2	5.1	4.1	5	5.3	3.2	4.1
Zinc sulphate	7.2	7.1	6.1	6	5.8	5.1	4.6
Cadmium sulphate	7.2	5.4	4.6	3.6	3.2	1.7	0.3

Source	D.F.	S.S.	M.S.S.	F Ratio
Chemicals	4	57.827	14.456	50.155***
Concentrations	6	132.888	22.148	76.839***
Interaction	24	50.887	2.120	7.356**
Error	70	20.176	0.288	

= Highly Significant, *=Very highly Significant.

Table 3. Effect of heavy metals on Seed Germination(%) of seedling in *Lens esculenta* cv. L4076 (values are mean of three replicates each).

Name of Chemical	Control (%)	Concentration (ppm)					
		10	50	100	200	500	1000
Copper sulphate	100	90	100	100	96	93.3	93.3
Nickel sulphate	100	96	96	100	100	93.3	96
Lead sulphate	100	100	90	100	96	100	96
Zinc sulphate	100	100	100	100	100	100	96
Cadmium sulphate	100	100	100	93.3	90	16	1

Source	D.F.	S.S.	M.S.S.	F Ratio
Chemicals	4	105.181	26.295	36.813***
Concentrations	6	73.390	12.231	17.124***
Interaction	24	234.419	9.767	13.674***
Error	70	50	0.714	

***=Very highly Significant.

Table 4. Effect of heavy metals on Fresh Weight (g) of seedlings in *Lens esculenta* cv. L4076 (values are mean of three replicates each).

Name of Chemical	Control (g)	Concentration (ppm)					
		10	50	100	200	500	1000
Copper sulphate	0.19	0.17	0.13	0.13	0.13	0.10	0.11
Nickel sulphate	0.19	0.17	0.11	0.15	0.12	0.13	0.09
Lead sulphate	0.19	0.18	0.15	0.14	0.14	0.12	0.14
Zinc sulphate	0.19	0.20	0.17	0.15	0.14	0.11	0.11
Cadmium sulphate	0.19	0.08	0.06	0.07	0.04	0.04	0.02

Source	D.F.	S.S.	M.S.S.	F Ratio
Chemicals	4	0.0949	0.0237	82.713***
Concentrations	6	0.1065	0.0177	61.860***
Interaction	24	0.0253	0.00105	3.681**
Error	70	0.0200	0.00028	

= Highly Significant, *=Very highly Significant.

concentration of heavy metals. At maximum concentration *i.e.* 1000 ppm the shoot length was 0.3 cm in Cd, 4.1 cm in Pb, 4.4 cm in Cu, 4.6 cm in Zn and 4.7 cm in Ni when compared with control *i.e.* 7.2 cm. Statistically, very highly significant results were observed among chemicals and among concentrations but highly significant between concentration *versus* chemicals (Table 2).

Effect of Heavy Metals on Seed Germination - 100 % germination was seen in control and remained 100% upto 500 ppm concentration in Zn, upto 50 ppm in Cd. Germination was 100% at 50 ppm and 100 ppm in Cu, at 100 and 200 ppm in Ni and at 10 ppm, 100 ppm and 500 ppm in Pb. A significant reduction occurred in seed germination from 200 ppm to 1000 ppm in Cu and 100 ppm to 1000 ppm in Cd. Statistically, very highly significant results were obtained among chemicals, among concentrations and between concentration *versus* chemicals (Table 3).

Effect of Heavy Metals on Fresh Weight - The heavy metals showed inhibitory effect on fresh weight of the seedling at higher concentrations. Only Zn at 10ppm showed an increase in fresh weight. It declined to 0.02g/ seedling in Cd when compared with control where it was 0.19g/seedling. Results were very highly significant among chemicals and among concentrations. Highly significant results were seen between concentration *versus* chemicals (Table 4).

Observation of data tables concludes that all seedling growth parameters decreased significantly in *Lens esculenta* cv. L4076 with application of heavy metals. In general, higher concentrations were inhibitory to the growth parameters measured in *L. esculenta* cv. L4076. Wani *et al.*[5] reported that metal accumulation in roots and shoots of green plants decreased in the order Cd> Cu>Cr. Beri and Setia[6] reported that with increasing concentration of heavy metals growth and yield of lentil raised in sand cultures decreased, however, low levels of Ni, Cd and Pb were slightly promotory. Heavy metal toxicity in legume microsymbiont system was studied by Athar and Ahmad[7]. They described that the phytotoxicity was apparently due to the susceptibility of the symbolic apparatus against toxic doses of heavy metals. Faizan and Khan[8] studied the effect of coal ash application on growth, productivity and biochemical characteristics of lentil. They reported that higher coal ash levels affected the crop adversely resulting in loss of growth and yield. Physiological effect of organic mercury on the growth of *Arachis hypogea* seedlings was studied by Bhanumathi *et al.*[9] Higher concentration of paper mill effluent inhibits both seed germination and seedling growth[10]. Suthar *et al.*[11] studied the impact of distillery effluent on seed germination and seedling growth of some plants and reported that increased concentration of distillery effluent significantly inhibited plant development.

Inhibition of seed germination was reported with Cu and Ni in lentil[12], chickpea[13] and *Pisum sativum*[14]. Accumulation of heavy metals in leguminous crops (bean, soybean, peas, lentils and gram) was studied by Angelova *et al.*[15]. They concluded that the accumulation and absorption of the heavy metals in the studied crops from soil was highest in soybean followed by beans, lentils gram and peas. Yamur *et al.*[16] reported the effects of sewage biosolid on seed protein ratios, seed NP contents, some morphological and yield characters in lentil. According to them the application of sewage biosolid to lentil increased plant seed yield significantly. Zeidan[17] studied the effect of organic manure and phosphorous fertilizers on lentil and reported that increasing rates of applied organic manure and phosphorous markedly increased the growth, yield and quality of lentil plants in sandy soil.

The present results were similar with the earlier results that reduction in seed germination and seedling growth was reported with increasing application of heavy metals on *Cyamopsis tetragonoloba* by Jain *et al.*[18] and on *Raphanus sativus* by Gupta *et al.*[19]. Similar results were obtained for *Phaseolus aconitifolius*[20]. Observations also agree with Ayaz and Kadioglu[21], Khurana *et al.*[22] and Yasar and Ahmet[23,24] who in their studies on lentil reported that higher concentrations of Zn, Pb, Cu and Cd inhibited the yield, seed germination, root growth and mitotic index . Ergun and Oncel[25] also reported that after 250 ppm concentration, both Zn and Cd were inhibitory. Cd was found to be more toxic than Zn on growth and some biochemical parameters of lentil seedlings.

The results of this study concluded that among all the heavy metals studied, effect of Zn was found to be the least toxic and Cd the most toxic for all the growth parameters (root length, shoot length, seed germination and fresh weight) in *Lens esculenta* cv.L4076.

Acknowledgement

Authors are thankful to Prof. S. Kshetrapal for her valuable guidance and Head, Department of Botany, University of Rajasthan, Jaipur for providing laboratory and library facilities.

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