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EFFECTS OF LEAD ON SEED GERMINATION AND SEEDLING GROWTH OF WHEAT (TRITICUM AESTIVUM) L.

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Abstract

Lead is one of the highly toxic heavy metal and being a dangerous environmental pollutant and exerts harmful effects on plant growth. The effects of different concentrations (0, 20, 40, 60, 80, 100 ppm) of lead on seed germination and seedling growth performance of wheat (*Triticum aestivum*) as compared to control was studied. Lead treatment in the form of lead acetate at 100 ppm completely inhibited the seed germination and seedling growth of *T. aestivum* as compared to control. Lead treatment at 40 ppm produce significant ($p<0.05$) reduction in seed germination while lead treatment at 20 ppm concentration produced significant reduction in shoot length as compared to control. Root growth is an important growth variable and found greatly reduced at different concentrations of lead treatment. The treatment of lead at 20 ppm produced significant ($p<0.05$) reduction in seedling dry weight of *T. aestivum* as compared to control. However, increase in concentration of lead treatment at 80 ppm was found sufficient to cause significant reductions in seedling dry weight of *T. aestivum* as compared with control.

The seedlings of *T. aestivum* were also tested for percentage of tolerance to lead. The results showed that *T. aestivum* has high tolerance to lead at 20 ppm (92.50%) and lowest at 80 ppm (64.50%) of lead. *T. aestivum* seedlings showed better percentage of tolerance (73.25 %) to lead at 60 ppm. The seedlings of *T. aestivum* were tested for the establishment of seedling vigor index (SVI) to different level of lead treatment. The results showed that *T. aestivum* has greater seedling vigor index was (1893) to lead at 0 ppm and lowest (302) at 80 ppm of lead. *T. aestivum* seedlings showed moderate SVI (1586) at 20 ppm lead treatment. *T. aestivum* showed better SVI (847) to lead treatment at 60 ppm.

Key words: Crop, germination, growth, lead, tolerance, toxicity.

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Introduction

Plants are an integral part of life in many indigenous communities (Bhatia *et al.*, 2014). Human communities have now modified the plant cover almost beyond recognition over wide areas (Eyre, 1984). Rapid increase in the industrial and anthropogenic activities and discharge of untreated chemicals in the environment are responsible for spreading of different types of chemical compounds in the air, soil and water, affecting the environment and growth of plants. Among the toxic elements release in the environment, lead is highly toxic to the growth of plants. Lead (Pb) is an environmental pollutant extremely toxic to plants and other living organisms including humans (Lamhamdi et al., 2011). During the past couple of decades, the concentrations of heavy metals in the environment of city have appeared at dangerous level. Metals are toxic to both plants and fungi, and elevated soil metal concentrations have been documented to change the structure of ectomycorrhizal communities (Crane *et al.* 2012). The toxicity and tolerance to lead depends on the concentration, type of salt, soil properties and plant species.

Seed health plays an important role for successful cultivation and yield exploitation of a crop species (Rajput *et al.*, 2005). Given the ever-increasing environmental pollution with metal(loid)s, the impacts of metals on seed metabolism, viability and germination in comparison to the numerous publications on the effects of metals in vegetative tissues, particularly roots and shoots (Kranner and Colville, 2011). Attention has been given, in developed countries, about the effects of metal toxicities on germination and growth of plants. Lead is a global environmental pollutant that is present in soil, water, air and biota. The increase in concentration of heavy metal decreased plant growth and responsible to death (Kumar *et al.*, 2011). Studies on differential tolerance of mung bean cultivars to metalliferous mine wastes and tannery effluents were observed (Samantaray and Rout, 1999; Bera and Kanta-Bokria, 1999). Lead is the heaviest of the non-radioactive meals that also naturally occur in substantial quantities in the earth's surface. Pb easily accumulates in the surface of the soil (De Abrreu et al., 1998). Lead is an important industrial heavy metal, which contaminates environment and ultimately, food, water urban soil and air (Haq et al., 2013). Lead stress causes multiple direct and indirect effects on plant growth and metabolism and alters some physiological processes (Diaz et al., 2001).

In Pakistan, substantial quantities of agricultural chemical are used annually to enhance yield (Nuzhat, *et al.*, 2005). Residues and addition of lead containing compounds in the environment have been reported to persist in the food grains from many parts of the world. The ever increase in lead concentration over the wide areas of Karachi and rural areas raises serious questions as to its effects on the growth and vigor of plants. A decline in the agriculture areas play an important role, which can lead to certain restriction in the availability of crop for human beings. Although the data on the effects of lead on mung bean crop is presently seems scanty. The increase in concentration of metals likewise lead can produce toxic effects on plants growth. The response of plant growth to toxic effects of heavy metals has become the subject of great interest in recent years because of their nature of high toxicity to plants. Attention has been given in developed countries, about the effects of metal toxicities on plants growth. Therefore, a study was carried out with the aim to determine the effect of lead on seed germination and seedling growth of an important country crop wheat *T. aestivum*.

Materials and methods

The healthy seeds of *Triticum aestivum* L. (Poaceae) were obtained from local market and surface sterilized with 0.20% solution of sodium hypochlorite (NaOCl) for one minute to avoid any fungal contamination. The seeds were washed with distilled water and transferred in Petri dishes

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(90 mm diameter) on Whatman No. 42 filter paper at room temperature 23 – 30 ºC and relative humidity 74 – 80%. There were three replicates. Initially, 5 ml solution of lead acetate $Pb(CH_3COO)_2$ in different ranges 0, 20, 40, 60, 80 and 100 ppm were applied. All solutions were daily changed. In control, no treatment was given except distilled water. Experiment was completely randomized. After ten days, seed germination percentage, shoot, root and seedling length (cm) were noted for *T. aestivum.* The seedling dry weight was determined by drying the plant materials in an oven at 80°C for 24 hours.

Analysis of variance and Duncan's Multiple Range Test using personal computer software packages SPSS version 14.0 statistically analyzed the data obtained.

A tolerance index was determined by the following formulae as described by Iqbal and Rahmati (1992):

Mean root length in metal solution/Mean root length in distilled water X 100

Seedling vigor index (S.V.I.) was determined as per the formula given by Bewly and Black (1982).

Results

Lead treatment was found toxic to the seed germination and all seedling growth parameter of *T. aestivum*. The seed germination and seedling growth performance of *T. aestivum* were tested in different concentrations (0, 20, 40, 60, 80 and 100 ppm) of lead as compared to control (Table 1-2, Fig.1 - 2). Lead treatment was found highly toxic to the seed germination and all seedling growth parameter of *T. aestivum*. Lead treatment at 100 ppm completely inhibited the seed germination while, lead treatment at 20 ppm did not produce any significant effects on seed germination of *T. aestivum* as compared to control (Table 1). Lead treatment at 40 ppm produced significant ($p<0.05$) effects on seed germination of *T. aestivum* as compared to control. Lead treatment at 20 ppm concentration produced toxic effects on shoot length. Root growth is an important growth variable and found negatively affected by different concentration of lead treatment. The results also showed that lead treatment in the substrate at 40 ppm produced significant effect on seedling growth of *T. aestivum* as compared to control. The treatment of lead at 20 ppm produced significant ($p<0.05$) effect on seedling dry weight of *T. aestivum* as compared to control.

Table 1. Effects of lead on seed germination and seedling growth of wheat (*Triticum aestivum*)

Treatments (ppm)	Seed germination (%)	Shoot length (cm)	Root length (cm)	Seedling length (cm)	Seedling dry weight (mg)
0	100.00 ±0.00 d	10.93 ±0.23d	8.00 ±0.51c	18.93 ±0.75d	26.33 ±1.45e
20	100.00 ±0.00 d	8.46 ±0.46c	7.40 ±0.26c	15.86 ±0.73c	19.66 ±1.45d
40	73.33 ±3.33 c	8.13 ±0.18c	7.20 ±0.06c	15.40 ±0.25c	18.66 ±0.66cd
60	66.66 ±3.33 c	6.83 ±0.54b	5.86 ±0.59b	12.70 ±1.13b	15.33 ±1.45bc
80	26.66 ±3.33 b	6.16 ±0.17b	5.16 ±0.26b	11.33 ±0.43b	12.66 ±1.45b
100	00.00 ±0.00 a	0.00 ±0.00a	0.00 ±0.00a	00.00 ±0.00a	00.00 ±0.00a

Number followed by the same letters in the same bar are not significantly different ($p<0.05$) according to Duncan's Multiple Range Test.
± standard error

Lead treatment at all concentration decreased high percentage of seed germination, shoot, and root length and seedling dry weight of *T. aestivum* (Table 2). Lead treatment at 20 ppm concentration was less toxic for decrease in the rate of seed germination (0%), shoot length (22.59

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%), root length (7.50%), seedling length (16.21 %) and seedling dry weight (25.33%) as compared to control. Lead treatment at 40 ppm concentration showed more decrease in seed germination (30%), shoot length (19%), root length (23.72%), seedling length (20.62 %) and seedling dry weight (21.99%) of *T. aestivum*. Lead concentration of 60 ppm decrease seed germination (36.67 %), shoot length (23.23%), root length (27.79 %), seedling length (24.37%) and seedling dry weight (22.40 %) of *T. aestivum* as compared to control. Lead concentration of 80 ppm decrease seed germination (50.00 %), shoot length (41.96 %), root length (41.01 %), seedling length (41.25 %) and seedling dry weight (37.59 %) of *T. aestivum* as compared to control. Lead concentration of 100 ppm showed highest percentage of decease in seed germination (73.34 %), shoot length (72.05 %), root length (65.25 %), seedling length (69.31 %) and seedling dry weight (72.80 %) of *T. aestivum* as compared to control.

Table 2. Percent reduction in seed germination and seedling growth of wheat (*T. aestivum*) using different concentration of lead (20, 40, 60, 80, 100 ppm) as compared to control (0 ppm).

Treatments (ppm)	Seed germination	Shoot length	Root length	Seedling length	Seedling dry weight
20	0	22.59	7.50	16.21	25.33
40	26.67	25.61	9.25	18.64	29.13
60	33.34	37.51	26.75	32.90	41.77
80	73.34	43.64	35.50	41.14	51.90
100	100.00	100.00	100.00	100.00	100.00

The seedlings of *T. aestivum* were also tested for percentage of tolerance to lead. The results showed that *T. aestivum* has high tolerance to lead at 20 ppm (92.50%) and lowest at 80 ppm (64.50%) of lead. *T. aestivum* seedlings showed better percentage of tolerance (73.25 %) to lead at 60 ppm (Fig. 1). The seedlings of *T. aestivum* were tested for the establishment of seedling vigor index (SVI) to different level of lead treatment. The results showed that *T. aestivum* has greater seedling vigor index was (1893) to lead at 0 ppm and lowest (302) at 80 ppm of lead. *T. aestivum* seedlings showed moderate SVI (1586) at 20 ppm lead treatment. *T. aestivum* showed better SVI (847) to lead treatment at 60 ppm (Fig. 2).

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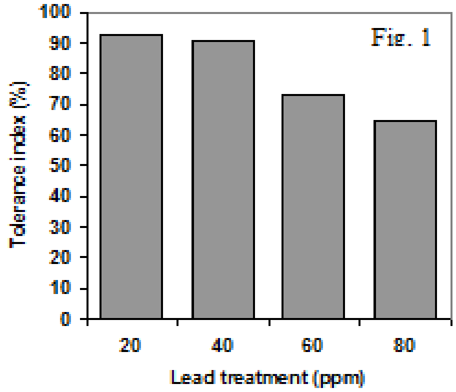

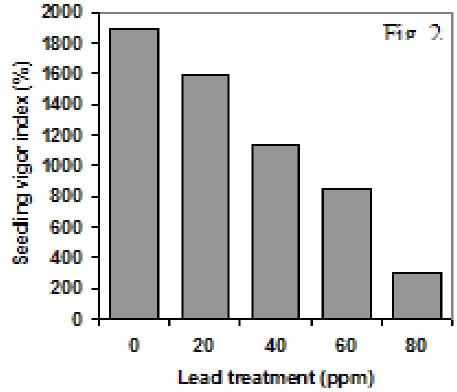

Fig. 1 - 2. Percentage tolerance index and seedling vigor index in seedlings of wheat (*T. aestivum*) to different concentration of lead treatment.

Discussion

World vegetation is an important component of our planet. Plants have a unique role for the existence of all heterotrophic organisms including human population (Kralova and Masarovieova, 2006). Few studies have been studied on the influence of abiotic stress on plants. Due to their direct or indirect presence, they may affect the development, growth, basic metabolisms in plants or any other living organisms and each kind of these, such as heavy metals, salt stress, chilling, drought or UV-B radiation (Szollosi, 2014). The plants under stress condition are most likely to be adversely affected by high concentrations of pollutants. Lead is considered a toxic and dangerous heavy metal. The lead treatment at 150 mM showed significant physiological, photosynthetic and ultra-structural changes in seedlings of *Vigna unguiculata* (Kasim *et al.*, 2014). In the present experiments, the effect of lead acetate on seed germination, root, shoot, seedling growth and seedling dry weight of an important crop wheat (*T. aestivum*) were recorded. The seed germination and seedling growth of wheat responded differently to lead treatment at higher concentration as compared to control. High percentage of decrease in seed germination of wheat at 40-60 ppm lead treatment provided evidence that the treatment of lead in excess may be inhibitory to plant growth and development. In this study, an inhibition in seed germination of wheat at 100 ppm lead treatment was observed. Inhibition due to heavy metal such as mercury in seed germination of another crop *Phaseolus aureus* was observed (Sharma, 1982). Metal toxicity is also an important factor governing germination and growth of plants. The permeability of metals can decreased the growth of plants. In present studies, the reduction in seedling growth of *T. aestivum* was observed when treated with different concentration of lead. The reduction in the seedling and root growth of *T. aestivum* with the increase in concentration of lead in the immediate environment provides further evidence that the lead in excess may be inhibitory to plant growth and development. Excessive amount of toxic element usually caused reduction in plant growth (Kubota and Allaway, 1972). Toxic metal ions enter cells by means of the same uptake processes as essential micronutrient metal ions. The amounts of metal absorbed by a plant depend on the concentrations and speciation of the metal in the soil solution, its movement successively from the bulk soils to the root surface, then into the root and finally into the shoot. Excessive concentrations of metals result in phytotoxicity through: (i) changes in the permeability of the cell membrane; (ii) reactions of sulphydryl (–SH) groups with cations; (iii) affinity for reacting with phosphate groups and active groups of ADP or ATP; and (iv) replacement of essential ions (Patra *et al.* 2004). The plant

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reaction with respect to it stresses are complex and involves in many kinds of physiological and biochemical processes. Similarly, the inhibition in 5 d old seedling growth of *Vigna radiata* (L.) Wilczek cv. Pusa Baisakhi in the presence of 1.0 mM lead acetate increased drastically, if NaCl (6 and 12 EC) was also present in the nutrient media along with the metal salt (Singh *et al.*, 2003).

The root elongation tests have been used to evaluate the damage caused by toxic compounds present in various composts (USEPA, 1982). Many species including cabbage, lettuce, carrot, cucumber, tomato and oats have been recommended for the phytotoxicity test (F.D.A., 1982). The roots are normally considered in relation to their ability to supply water and nutrients to the plants. They are also required to produce hormones, which may regulate the growth and performance of both root and shoot (Blackman and Davies, 1985). The root growth of *T. aestivum* was found decreased more than 75 percent at 80 ppm lead concentration. The results of this investigation have shown that lead treatment is more toxic for *T. aestivum* for root development. These findings are also agree with the work of (Kopittke *et al.,* 2007) and found a low concentration of 1 µM lead (Pub) reduced the shoot and root of cowpea (*Vigna unguiculata*). The primary site of Pb^{2+} toxicity was the root, caused severe reductions in root growth, loss of apical dominance, the formation of localized swellings behind the root tips (due to the initiation of lateral roots), and the bending of some root tips (Kopittke *et al.,* 2007).

The significant decrease in seedling dry weight of *T. aestivum* due to metal toxicity of lead was also recorded. The treatment of lead in *T. aestivum* provided evidence that the trace element in nutrient medium if present in excess may be inhibitory to plant growth and development especially at more than 60 ppm. Toxicants accumulate in the plant when soluble forms are present in high quantities. The exact amount of accumulation depends upon the solubility of the pollutants in soil (Treshow, 2010). The biomass production of *T. aestivum* was initially nonsignificant and decreased with increasing the lead concentrations.

According to tolerance test it could be seen that tolerance to lead was higher at low concentration of lead in the seedlings of *T. aestivum*. These results showed that the reason of tolerance against heavy metals might be a physiological association of the tolerance mechanism to these metals. The seedling growth of *T. aestivum* showed high percentage of tolerance to lead at 20 ppm concentration. The treatment of 100 ppm concentration of lead produced lowest percentage of tolerance in seedling of mungbean.

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Conclusion

It is concluded that during the past decade, the concentration of heavy metals like lead and other metals in the environment has been increased due to automobiles, industries, agro chemicals and anthropogenic activities and responsible for limiting the crop yield. Plants experience oxidative stress upon exposure to heavy metals that leads to cellular damage (Yadav, 2010). Lead is highly toxic pollutant for plants and the environment because of their toxicity towards all living organisms. The results showed that treatment of different concentrations of lead to seedlings of *T. aestivum* responded differently. The response of *T. aestivum* seedlings in the form of tolerance indices to lead treatment was found suitable pollutant indicator to study the deleterious effects of the lead. The present findings proved the deleterious effects of lead at higher concentration to the seedlings of *T. aestivum.* The development of novel phytoremediation strategies using plants to lessen the burden of heavy metals contamination from the environment is an increasingly important demand worldwide. The information from the present studies would be helpful in understanding the level of lead tolerance in seedlings of *T. aestivum* while growing in lead polluted areas. The findings may contribute to ecological fragility, the potential of crop in coordinating in land management programmes. The cultivation management of wheat in lead polluted areas would help in reducing the burden of pollution to some extent. Heavy reliance on metals containing agrochemicals such as fungicides, nematicide, and pesticides (Lead arsenate) should be discouraged. The continuous release of lead into the immediate environment may endanger the growth performance of other crops. Current research shows that lead treatment at different concentration has produced an important effect on seed germination and seedling growth of *T. aestivum*. Increase in the concentration of lead in the medium, brought up certain toxic changes in germination of *T. aestivum*. Special care should be taken to monitor the toxic pollutants available in the immediate environment. The accumulation of such types of toxic pollutants in larger concentrations by crop can produce harmful effects to crops and ecosystems. Special efforts need to be made to identify sources of lead toxicity and there is also a need to be carried out an ongoing effort to develop tolerance indices. Better sources of resistances to metal are badly needed.

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