

Effects of diverse doses of Lead (Pb) on different growth attributes of *Zea-Mays* L.

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ABSTRACT

The aim of the investigation was to determine the potential effect of lead on maize growth. Lead is considered as important potent environmental contaminant. Various ecological, environmental and evolutionary processes in the microsphere are disrupted because of lead toxicity to the microbial community. The effects of Lead Nitrate ($Pb(NO_3)_2$) as heavy metal on germination, early growth seedling, root-shoot length, root-shoot fresh and dry weights, total protein content and the uptake of lead by roots and shoots of *Zea-mays* were investigated. All of the parameters were reduced by the increased lead concentrations. Such growth retardation was due to metals toxicity that resulted in damages to various physiological and biochemical processes.

Keywords: Lead Uptake; *Zea-Mays* L.; Early Seedling Growth

1. INTRODUCTION

Heavy metals (HM) act as essential plant micronutrients however, excess amount of these contents become toxic pollutants [1]. Beyond threshold levels, heavy metals (HM) are transformed into contaminants. An inhibition of plant growth was also reported through the use of HM like Pd, Zn, Cu, and Cd [2]. Their release creates pollution into the atmosphere [3] and hence these are potential hazard for plant growth and development [2]. They produce toxic affects upon plants, animals as well as human health [4,5]. Toxic heavy metals concentrated

in the plant tissues comes from the common practices of sewage and industrial discharge on agricultural land.

Lead is considered as important potent environmental contaminant [6]. Various ecological, environmental and evolutionary processes in the microsphere are disrupted because of lead toxicity to the microbial community [7, 8]. It is commonly used in fertilizers, batteries, chemicals and ceramics, in different products like pottery, gasoline, lead glass, pesticides, paints, hair dyes, rubber toys and newsprint. Major lead content in the soil comes from weathering of geological rock formations, lead mine's discharge, automobile exhausts, industrial applications, smelting operations, fertilizer impurities, use of lead arsenate in metal plating and finishing operations, tetra-methyl lead applications as anti knocking agent in petrol [9] and plants obtain lead from such agencies [1]. Increase in lead concentration in cultivated soils is detected in close proximity to industrial sites.

Zea-mays L. as one of the most important cereal crops in the world and ranks third most significant cash crop of Pakistan [10]. It is an important aspect to understand the potential capacity in metals uptake and distribution and its effects on maize growth. Leaves of *Zea-mays* indicate significant quantity of accumulation and further translocation of lead occurring in concentration dependent manner. Accumulation of lead in leaves depends upon its absorption from the aerial sources. Leaf morphology also contributes a lot for its intake. The *Zea-mays* seedlings showed strong inhibition of primary root growth with the applications of lead [11]. The lead toxicity causes cessation of root growth which was accompanied by the inhibition of growth at root tips [12]. Such observations were confirmed by histochemical and electron microscopic

studies. Heavy metals like lead may form complex with the synthetic chelate EDTA, thus are enhancing their absorption capability by soil particles. Through this process, heavy metals can be removed from contaminated water. K^+ ions leakage from root cells of *Zea-mays* is also attributed to lead toxicity [13].

Lead inhibits water imbalance, disturbed mineral nutrition, enzyme activities, change in hormonal status and membrane permeability alteration. Lead at high concentrations inhibits cellular activities thus causing cell death [14]. Increased lead concentration hampers the synthesis of chlorophyll because of impaired uptake of Iron and Magnesium through plants. The photosynthetic apparatus is damaged due to its affinity for protein N- and S-ligands. At higher concentrations of lead, inhibition of respiration is observed. The remediation of lead affected sites is carried out by relatively narrow range of engineering based technologies [15]. The rhizofiltration and phytoremediation provide better horizons for the utilization of such technique for the clean-up of lead contaminated soils.

The main objective of the present study was to investigate Lead accumulation in *Zea-mays* to evaluate the effects of lead toxicity on protein contents and to determine its effects on different growth parameters like seed germination, root and shoot length, root and shoot fresh and dry weight. The study also throws light in understanding the capacity of *Zea-mays* in metal uptake and distribution and its effects on the growth of maize.

2. MATERIAL AND METHODS

The certified seeds of *Zea-mays* were obtained from Punjab Seed Corporation, Lahore. Seeds were washed thoroughly under running tap water and finally with distilled water before sowing. Six concentrations of metal salt (Lead Nitrate $Pb(NO_3)_2$) *i.e.*, Control (0.0 mM), 01, 25, 50, 100, 200, 500 mM were prepared, respectively. Pots (15 × 3 cm) were sterilized and filled with soil. Ten sterilized seeds were sown in each pot and 10 ml of varied Lead nitrate concentrations was added to each pot at the time of sowing. Pots were kept in dark for 72 hours for germination at normal atmospheric temperature and pressure. After three days of germination, pots were then shifted to light (8000 - 10,000 Lux). The seedlings growth was observed for 14 days. Three replicates were used per treatment per experiment and experiment was repeated thrice. Data of seed germination root and shoot length, fresh and dry weight, total protein contents, lead uptake by root and shoot was recorded of 14-day old seedlings. Analysis of data and statistics was carried out as reported in an earlier study. Total Proteins was estimated through Kjeldahl's method in accordance with "official methods of analysis".

Another batch of 14 days old seedlings grown in various concentration of lead were used for estimation of metals content in root and shoot parts. Lead contents were estimated through ashing of plant material. Oven dried samples of roots and shoots were taken in porcelain crucibles (in duplicates). Samples were heated slowly at 450°C - 500°C in muffle furnace and held at the same temperature for 5 - 7 hours till white ash was obtained. Samples showing any black residue were wetted with a few drops of conc. nitric acid, heated on a burner to eliminate nitric acid fumes and kept in the furnace for another two hours till the ash turned white. The ash was dissolved in 5 ml of 6 N HCl, heated to dissolve any other remains. Acid washed filter paper was used for filtering solution followed by washing with hot distilled water. Final volume was made 100 ml by adding distilled water. Determinations of Lead were carried out on a Hitachi Model. 170-10 atomic absorption spectrophotometer equipped with hollow cathode lamp of magnesium, operated at 217 nm. Lead contents were calculated by comparison with the standard treatments with same amount of reagent *i.e.*, 5 ml of 6 N HCl as for sample.

3. RESULTS

Lead had inhibitory effect on percentage germination. There was a continuous decrease in the percentage germination with the increase of Lead concentration. At 1.0 mM concentration, it was decreased by 10% while it reached upto 100% at 500 mM. All decreases were significant in relation to control. Lead produced deleterious effects on seedling growth. A steady decline in the seedling growth was observed with the increase in lead concentration. Shoot length was decreased at all concentrations, even the lowest treatment of 1.0 mM reduced shoot growth by 12.07% while at 200 mM the percentage decrease over control was up to 95%. A reduction in root length was also prevalent comparable to the shoot length. A significant sharp decrease (97.74%) was observed at 200 mM compared to control (Table 1, Figure 1).

The results were similar as that of drastic effects of lead upon the germination and growth of *Zea-mays*. Both fresh weight and dry weight of the seedling demonstrated

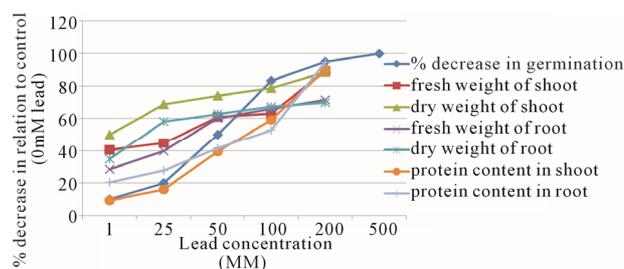


Figure 1. % decrease in growth parameters in different concentrations of lead.

the continuous reduction in all concentrations as compared to the control. The relative decrease at 200 mM was 89.37% for fresh weight of shoot and 71.6% for fresh weight of root. Dry weight of shoot and root showed decrease at 1.0 mM treatment, with the reduction in shoot dry weight (50%) compared to root (34.89%) in relation to control (**Table 2, Figure 1**).

The effect of lead on total protein content of shoot and root varied. In shoots, the total proteins were increased at 1.0 mM while decreased at 25 - 500 mM. At 1.0 mM concentration, the reduction in protein content in shoot was 9.13% while 20.34% in root in relation to control (**Table 3, Figure 1**). **Table 3** reveals that the total uptake

of Pb was directly proportional to the amount of lead provided to the seedlings.

4. DISCUSSION

The present study is confined to determine the effect of Lead Nitrate on germination, total protein content and the uptake of lead by roots and shoots of *Zea-mays*. The reduction in plant growth is also attributed to excessive accumulation of lead in the soils. The decrease in seed germination is attributed by the heavy metal treatment [16,17]. Reduced seed germination is observed in corn treated with 20, 50, 100 and 200 µg/ml lead acetate [18].

Table 1. Effects of Pb(NO₃)₂ on seed germination, length and fresh weight of shoot and root of 14-days old seedlings of *Zea-mays* L.

Conc. of Pb(NO ₃) ₂ in mM	Mean % Germination	Mean length of shoot (cm/seedling)	Mean length of root (cm/seedling)	Mean fresh weight of shoot (g/seedling)	Mean fresh weight of root (g/seedling)
0.0	100 ^a ± 0	32.90 ^a ± 1.168	22.10 ^a ± 1.861	1.41 ^a ± 0.060	0.88 ^a ± 0.0441
1.0	90 ^b ± 0	29.40 ^b ± 0.306	19.20 ^a ± 0.493	0.84 ^b ± 0.029	0.63 ^b ± 0.0115
25	80 ^c ± 0	20.63 ^c ± 1.646	12.96 ^b ± 1.241	0.78 ^c ± 0.018	0.53 ^c ± 0.1763
50	50 ^d ± 0	18.27 ^d ± 0.498	9.06 ^c ± 0.384	0.55 ^d ± 0.076	0.35 ^d ± 0.0233
100	16.66 ^e ± 3.333	6.50 ^e ± 1.656	3.53 ^d ± 0.284	0.52 ^d ± 0.015	0.30 ^d ± 0.0288
200	5.00 ^f ± 2.511	1.13 ^f ± 0.612	0.50 ^e ± 0.288	0.15 ^e ± 0.026	0.25 ^e ± 0.0202
500			No Germination		

^{a, b, c, d, e, f} = Values having different alphabets are significantly different from each other at 5% level of significance (Duncan's multiple range test of composite mean).

Table 2. Effect of Pb(NO₃)₂ on dry weight and protein content of shoot and root of 14-days old seedlings of *Zea-mays* L.

Conc. of Pb(NO ₃) ₂ mM	Mean dry weight shoot (g/seedling)	Mean dry weight of root/seedling (g)	Protein in g/dry weight of shoots	Protein in g/dry weight of roots
0.0	1.12 ^a ± 0.015	0.43 ^a ± 0.01667	29.80 ^a ± 0.057	18.83 ^a ± 0.0333
1.0	0.56 ^b ± 0.088	0.28 ^b ± 0.01667	27.08 ^b ± 0.005	15.00 ^b ± 0.1154
25	0.35 ^c ± 0.028	0.18 ^c ± 0.01333	25.05 ^c ± 0.011	13.63 ^c ± 0.1905
50	0.29 ^d ± 0.026	0.16 ^d ± 0.0333	18.11 ^d ± 0.034	11.00 ^d ± 0.1732
100	0.24 ^d ± 0.023	0.14 ^e ± 0.00667	12.13 ^e ± 0.028	8.90 ^e ± 0.2886
200	0.13 ^e ± 0.015	0.13 ^f ± 0.01000	3.00 ^f ± 0.057	1.02 ^f ± 0.0057

^{a, b, c, d, e, f} = Values having different alphabets are significantly different from each other at 5% level of significance (Duncan's multiple range test of composite mean).

Table 3. Uptake of Pb (NO₃)₂ through shoots of 14-day old seedlings of *Zea-mays* L.

Conc. of Pb(NO ₃) ₂ mM	Uptake of Pb by shoots in ppm	Absorption	Uptake of Pb by roots in ppm	Absorption
0.0	0.3942 ppm	0.001	0.3942 ppm	0.0013
1.0	0.5710 ppm	0.0056	0.5710 ppm	0.0076
25	1.0523 ppm	0.0266	1.0523 ppm	0.0376
50	1.1051 ppm	0.3208	1.1051 ppm	0.5908
100	1.2250 ppm	0.5108	1.2250 ppm	0.6108
200	1.4569 ppm	0.7108	1.4569 ppm	0.8108
500	0.0	0.0	0.0	0.0

Overall germination observed was greatly suppressed at all lead treatments. A sharp decrease in crop productivity in soils contaminated with lead poses a serious problem for agriculture. Lead toxicity indicates decreased dry mass of shoots and root and percentage germination [19]. Present study, revealed that the root-shoot length showed inhibition in seed germination at all lead concentrations. Fresh and dry weights of both roots and shoots were also reduced at all treatments. Lead contents decrease germination index, tolerance index, germination percent, root/shoot length and dry mass of roots and shoots [20]. Reduction in dry matter yield of *Zea-mays* was also reported. The effects of lead depend on concentration, soil properties, type of soil and plant species. The total protein content of shoots increased at lower concentration of lead followed by reduction at higher concentrations. An increase in concentration of lead cause decrease of protein contents in roots (**Figure 1**). Many of the physiological processes, such as chlorophyll synthesis, photosynthetic rate, respiration, and protein level are inhibited due to heavy metals as evaluated by Iqbal *et al.* [21]. An increase in uptake of lead occurs up to certain concentration *i.e.*, 200 mM. Beyond this limit the lead concentration decreased in both roots and shoots. The growth inhibition on a priori basis results from damage to physiological and biochemical processes.

5. CONCLUSION

The aim of the investigation was to determine the potential of lead to effect on maize growth. Lead is considered as important potent environmental contaminant. Various ecological, environmental and evolutionary processes in the microsphere are disrupted because of lead toxicity to the microbial community. It is concluded that, the root-shoot length showed inhibition in seed germination at all lead concentrations. Fresh and dry weights of both roots and shoots were also reduced at all treatments.

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