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The Toxicity Effects of Lead and Zinc on Seed Germination and Plant Growth of Common Leguminous Yellow Flame Roadside Tree species *Peltophorum pterocarpum* (DC.) Baker ex Heyne

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Abstract: Environmental pollution by heavy metals due to anthropogenic, industrial, automobile activities and its impact on living organisms is a worldwide problem. The addition of the heavy metals in the environment found responsible for brought up significant changes in the nature, structure and composition of the plant communities, plant growth and even involved in the extinction of some plant species to some extent from the universe. In present study, the toxicity test was performed to record the effects of lead and zinc on seed germination and plant growth and biomass production performance of yellow flame [*Peltophorum pterocarpum* (DC) K Heyne] invitro studies. The treatment of different concentrations of Pb and Zn at 100-900 ppm showed variable effects on rate of seed germination percentage and seedling growth of yellow flame as compared to control. Lead and zinc treatment at 100 ppm produced significant ($p<0.05$) effect on root growth of *P. pterocarpum*. The lead treatment at 500 ppm showed maximum rate of seed germination percentage and root elongation of *P. pterocarpum*. Zn treatment at 500 ppm showed lowest and at 700 ppm showed maximum rate of seed germination of *P. pterocarpum*. Lead and zinc treatment at all level did not produce any significant effect on shoot, seedling growth and seedling dry weight of *P. pterocarpum*. It was concluded that the order of inhibitory effects of metals on germination and growth performance for *P. pterocarpum* were recorded in order of Pb>Zn.
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1. Introduction

Heavy metals are a group of elements which have a density greater than 5g/cm^3 (Stobrawa and Lorenc-Plucinska, 2008). Heavy metals toxicity in plants studied by different researchers (Cuypers et al., 2009; Yadev, 2010; Shafiq and Iqbal, 2012; Singh et al., 2015; Wang et al., 2015; Shafiq et al., 2019). Global industrial growth and automobiles activities have polluted the air, water and soil with the introduction of many hazardous compounds, including heavy metals likewise Pb, Cd, Cu, Cr, Co, Ni, Fe and Zn. These heavy metals at higher level are not only toxic to plants growth but also cause severe human health hazards when leach out into food chain (Kumar et al., 2013). Heavy metals are essential to life only in trace amount while their excess amount causes cellular damage and Pb element has ability to displace essential metal ions or blocking functional groups (Siddhi et al., 2018).

Lead (Pb) is a toxic heavy metal and attracted considerable attention keeping in view of its potential risk to the living organisms. The environmental conditions of Karachi city are constantly deteriorating due to industrial, auto vehicular and anthropogenic activities. Lead availability and uptake induced toxicity effects on seed germination and plant growth (Xiong, 1998; Shafiq and Iqbal, 2005; Farooqi et al., 2009; Zhejiang et al., 2009; Mohtadi et al., 2012; He et al., 2014). The studies of different researchers have reported about the ever increase concentrations of different types of toxic pollutants and heavy metals in city environment of Karachi. Lead is a widely distributed metal in the environment and contamination in soils resulted in soil fertility deterioration (Majer et al., 2002; Toplan et al., 2004) but also resulted in yield decline. High level of lead and heavy metals in the street dust, road side plants, vegetation and soil of Metropolitan city of Karachi reported (Ara et al., 1996; Khalid et al., 1996; Iqbal et al., 1998).

Zinc (Zn) is trace element and required in small amount for normal plants growth and increase in its concentration decreased plant growth. There are many reports are available on essential role, toxicity and accumulation of zinc in the plants (MacFarlane and Burchett, 2002; Auda and Ali, 2010; Taniguchi et al., 2013). The farmers usually use organic or mineral substances to improve plant growth and increase yield and crop quality and the presence of extensive mineral substances causes high concentrations of heavy metals like zinc. The treatment with 0, 3 of $Na_2HAsO_4 \cdot 7H_2O$ and 5 mM of $ZnSO_4$ for 72 h caused a significant reduction of radicle and cotyledon growth, chlorophyll and carotenoid contents, the accumulation of mineral nutrients (Mabrouk et al., 2019).

Peltophorum pterocarpum (DC) K Heyne is a popular ornamental perennial shade tree and commonly grown in street and roadside. *P. pterocarpum* is fast growing deciduous tree and propagated by seeds where, flowers are pollinated by insects. *P. pterocarpum* belongs to family Fabaceae, widely grown in tropical south east Asian countries, Sri Lanka, Malaysia, India, Indonesia, Philippines. *P. pterocarpum* is a large handsome tree and widely grown in greater part of Pakistan (eflora, 2021). The phytochemical screening of flower extract showed the presence of phenolic compounds, flavonoids, saponins, steroids, tannins, xanthoproteins, carboxylic acids, coumarins, carbohydrates and the presence of significant antibacterial activity against human pathogens in the flowers of *Peltophorum pterocarpum* (DC.) Baker ex Heyne (Sukumaran et al., 2011). *P. pterocarpum* is a deciduous tree with a girth of up to 1 m and grows to 35 m tall. *P. pterocarpum* is Native to Singapore and local conservative status in Singapore declared as (CR) Critically Endangered (FFW, 2021) and wood is used in making cabinet work.

Pb/Zn reported primary contaminant in Pb/Zn smelting site soil (Go et al. 2023). The contamination of soil, water and air by heavy metals is an important issue for environmental concern. Pb and Zn are considered an important heavy metal for ecotoxicology concern and little is known about the effects of heavy metals (Pb and Zn) on the germination and growth performance of an important street plant species, *P. pterocarpum*. The present studies were carried out with the aim to evaluate the effects of selected heavy metals viz. Pb and Zn on seed germination and seedling growth performance of *P. pterocarpum*.

2. Material and Methods

The healthy and same size seeds of *Peltophorum pterocarpum* (DC) K Heyne were randomly collected from city area (Gurumandir) of Karachi-Pakistan, brough in laboratory and stored for experiment. All the seeds were checked for viability by putting 10 seeds in each petri dish with a filter paper lined of same size and provided 10 ml of distilled water for 10 days. The seeds were surface sterilized with 0.50% of mercuric chloride for one minute to prevent any fungal growth. The seeds were selected for experiment, if they showed at least 50% germination. The rate of seed germination percentage at different time interval was checked. The first trials were taken in month of July (summer) when the temperature was good and favorable for seed germination of *P. pterocarpum.* Six different concentrations 1, 5, 10, 20, 30 and 60 ppm of Zinc nitrate and Lead nitrate were prepared. Ten seeds were placed in Petri dishes of 400 mm diameter on filter paper (Whatman No. 42) of equal size. Five ml solutions of lead nitrate and zinc nitrate concentrations 1, 5, 10, 20, 30 and 60 ppm were provided to respective petri dishes for ten days and the total number of seed germinated was recorded. The second trial were taken in December (winter) with different concentrations 10, 100, 200, 300, 500 and 700 ppm of Zinc nitrate and Lead nitrate prepared from stock solution of 1000 ppm. In winter the temperature was low and seeds of *P. pterocarpum* did not show good germination. These seeds may be dormant and the temperature might be not suitable for germination. Finally, third trial of experimental set up was performed in February using six different concentrations 0, 100, 300, 500, 700 and 900 ppm of Zinc nitrate and Lead nitrate which were prepared from 1000 ppm stock solution as follows.

0 ppm = 0 ml solution + 1000 ml distilled water
100 ppm = 100 ml solution + 900 ml distilled water
300 ppm = 300 ml solution + 700 ml distilled water
500 ppm = 500 ml solution + 500 ml distilled water
700 ppm = 700 ml solution + 300 ml distilled water
900 ppm = 900 ml solution + 100 ml distilled water

Ten seeds were placed in Petri dishes of 400 mm diameter on filter paper (Whatman No. 42) of equal size. Five ml solutions of lead nitrate and zinc nitrate concentrations 100, 300, 500, 700 and 900 ppm were provided to respective petri dishes for ten days and the total number of seed germinated was recorded. The control set received only distilled water. The experiments were designed on the basis of three replicates and the Petri dishes were kept at room temperature (32±2°C) with 240 Lux light intensity and the experiment lasted for 10 days. The rate of seed germination was recorded daily. The experiment was completely randomized and seed germination, root, shoot and seedling lengths were recorded. The seedling dry biomass was determined by placing the seedling in an oven at 80°C for 24 hours and dry biomass was measured with electrical balance. Mean percent seed germination, root, shoot, seedling growth and seedling dry weight data were statistically analyzed by Analysis of Variance (ANOVA) and Duncan's Multiple Range Test (DMRT) to determine the level of significance at $p < 0.05$ on personnel computer using COSTAT version 3.

3. Results and Discussion

The Plants are now being widely used for heavy metals toxicity test. However, studies in response of roadside plants to metals such as Pb and Zn stress and toxicity are very scant. Pb and Zn both pose great threat to plant growth when grown in excess environment. This work reports the differential impact on seed germination and early seedling growth of *P. pterocarpum.* The effects of different concentrations (0, 100, 300, 500, 700 and 900 ppm) of selected heavy metals (Pb and Zn) on seed germination, seedling growth and biomass performance of *P. pterocarpum* was recorded (Table 1-2). The metal takes long time after their introduction for chemical degradation (Bolan et al., 2014). The exposure of heavy metals stress to plants leads to damaging effects on germination and growth characteristics. The germination and growth of plants in presence of metals can be adversely affected. In present study the seed germination percentage of *P. pterocarpum* was found quite low in all concentrations of both heavy metals as compared to control treatment. There was no significant difference in seed germination of *P. pterocarpum* was found when treated with both metal solutions and control. The lowest percentage of seed germination of *P. pterocarpum* was found when treated with lead (Pb) by 300 ppm.

An increase in the concentrations of lead at 1000 mg/kg reported significantly inhibitory effect on seed germination percentage, root length, shoot length, tolerance index, fresh weight and dry weight of soybean (*Glycine max* (L.) Merr in seven days old seedlings as compared to control treatment (Gupta *et al.,* 2016). Shoot and seedling height of *P. pterocarpum* was strongly affected by Pb treatments as compared to control.

Sesbania drummondii seedling growth was found significantly inhibited with lead and zinc treatment. The uptake of metals followed the order Pb > Zn in roots and Pb > Zn in shoots (Israr al., 2011). Seedling height of *P. pterocarpum* showed higher mean root length by Zn treatments as compared to Pb. Maximum seedling size (4.166 cm) which includes the length of root (0.866 cm), shoot (3.30 cm) of *P. pterocarpum* was recorded for control which was decreased to 2.25 cm, 0.10 cm and 2.15 cm when treated with 500 ppm solution of lead metal. Seedling dry weight of *P. pterocarpum* was affected by Pb treatments as compared to Zn. Seedling dry weight of *P. pterocarpum* showed higher value in control treatments as compared to Pb and Zn. In another studies, treatments of Zn (0, 2.5, 5.0, 7.5 and 10 mM) doses reduced seedling dry mass in mustard varieties (Chowardhara, *et al.,* 2019).

Overall, results showed that Zinc (Zn) treatment showed less toxic effects than lead (Pb) on seed germination, seedling growth and seedling dry weight of *P. pterocarpum* as compared to control treatment. Plants species are able to develop the capability of adaptation to grow in contaminated site and the mechanism of action of toxicity depends on the availability of chemical compounds (Ren, 2003) in the surrounding environment.

5. Conclusion

It was concluded that lead and cadmium treatment produced toxic effects on seed germination and seedling growth of *P. pterocarpum* as compared to control. The tolerance indices and seedling vigor index was found less at higher concentration of both metal treatment. It is suggested that the value of difference in tolerance indices and seedling vigor index should be consider while planting *P. pterocarpum* in metal contaminated areas.

Table 1. Effects of different concentrations of lead on rate of seed germination (%), root, shoot, seedling length and seedling dry weight of *Peltophorum pterocarpum* (DC) K Heyne]

Treatments (ppm)	**SG (%)**	Root length (cm)	Shoot length (cm)	Seedling size (cm)	Seedling dry weight (g)
00	16.66 a	0.866 a	3.300 a	4.166 a	0.020 a
100	13.33 a	0.180 b	2.700 a	2.880 a	0.012 a
300	10.00 a	0.200 b	3.330 a	3.530 a	0.010 a
500	23.00 a	0.100 b	2.150 a	2.250 a	0.010 a
700	20.00 a	0.200 b	3.000 a	3.200 a	0.010 a
900	20.00 a	0.200 b	3.250 a	3.450 a	0.010 a
L.S.D.	19.21	0.549	3.16	2.94	0.015

Number followed by the same letters in the same column are not significantly different according to Duncan Multiple Range Test at $p<0.05$ level.

Zinc treatment with 500 ppm showed lowest percentage of seed germination of *P. pterocarpum*. Increased in concentration of lead at 500 ppm and Zn treatment at 700 ppm improved and showed maximum rate of seed germination percentage of the seed germination of *P. pterocarpum* as compared to control. The different physiological responses to Pb stress by 50, 150, 300, 600, 800, 1000 mg/L for the seed germination of *Salsola passerina* Bunge and *Chenopodium album* L. were reported (Hu *et al.,* 2012). Lead and zinc treatment at 100 ppm significantly ($p<0.05$) decreased root length of *P. pterocarpum* as compared to control.

Table 2. Effects of different concentration of Zinc on rate of seed germination (%), root, shoot, seedling size and seedling dry weight of *Peltophorum pterocarpum* (DC) K Heyne]

MT (ppm)	SG (%)	Root length (cm)	Shoot length (cm)	SZ (cm)	SDW (g)
00	20.00 a	1.000 a	3.830 a	4.830 a	0.020 a
100	13.33 a	0.300 b	1.600 a	1.900 a	0.120 a
300	20.00 a	0.380 b	3.300 a	3.680 a	0.010 a
500	6.60 a	0.216 b	1.080 a	1.296 a	0.010 a
700	23.33 a	0.143 b	1.990 a	2.133 a	0.010 a
900	20.00 a	0.160 b	2.160 a	2.320 a	0.006 a
L.S.D.	20.10	0.456	2.96	3.30	0.115

MT= Metal treatment; SDW= Seedling dry weight; SZ= Seedling size; Number followed by the same letters in the same row are not significantly different according to Duncan Multiple Range Test at $p<0.05$ level

Root growth of *P. pterocarpum* was strongly affected by Pb treatments as compared to Zn. Root growth of *P. pterocarpum* showed lowest mean root length by Pb treatments at 500 ppm as compared to control. The effect of the lead and zinc on root growth of *P. pterocarpum* was also observed and agreed with the findings of Ismail *et al.,* (2013). The growth of tobacco in Cd^{2+} and $Cd^{2+}+Pb^{2+}$ polluted soil for 50, 100, and 150 d resulted in some abnormal external morphological and anatomical changes in ripe region of lateral roots (Yuan *et al.,* 2011).

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