

EFFECTS OF HEAVY METALS ON EARLY GROWTH OF AFRICAN MARIGOLD (*Tagetes erecta*), FRENCH MARIGOLD (*Tagetes patula*) and SIGNET MARIGOLD (*Tagetes tenuifolia*)

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Abstract

Heavy metals in soils cause human health and environmental risks, therefore remediation of heavy metal contaminated sites is an important issue. Marigolds have been proposed as potential plants for phytoremediation of this type of contamination. In our experiment a seed germination test was carried out to investigate the toxic effects of heavy metals (Cu, Zn and Pb) on early growth of three different marigold species (African marigold, French marigold and Signet marigold). According to our results all tested heavy metals had significant ($p < 0.05$) toxic effects on seed germination and root/shoot elongation of the three plants. Signet marigold was the most sensitive plant to heavy metals, while African marigold and French marigold were able to tolerate low concentration of metals (below 400 mg l^{-1}) without considerable decline in growth parameters. These results indicate that African marigold and French marigold could be suitable for remediating heavy metal (Cu, Zn and Pb) contaminated soils.

Introduction

Heavy metals can accumulate in the topsoil at relatively high concentrations [1,2]. Previous studies have shown that heavy metals in soil pose potential threats to the environment, because in excessive concentrations they can be toxic to living organisms and endanger the health of humans and animals through the food chain [2,3,4]. For this reason, increasing attention has been paid in recent years to the phytoremediation of heavy metal contaminated soils, which is a cost-effective, environmental friendly and sustainable technique for restoration of these sites [1,4,5,6]. Applying ornamental plants for phytoremediation could be useful, especially in contaminated urban areas, since they are apart from the food chain, and they can beautify the environment [4,6,7]. Marigolds are commonly used as ornamental plants, and have been proposed as potential plants for phytoremediation of heavy metal contaminated soil [5,7,8,9,10]. Although much research has been conducted to investigate the bioaccumulation ability of marigolds, little information is available on the toxicity of metals on these plants. However high levels of metals can reduce plants biomass, which decrease its remediation potential [4]. The aim of our study was to compare the effects of selected metals (Cu, Zn and Pb) on seed germination and shoot/root elongation of marigold species.

Experimental

Three different marigold species were used in our experiment: African marigold (*Tagetes erecta*), French marigold (*Tagetes patula*) and Signet marigold (*Tagetes tenuifolia*). Seeds were obtained from Rédei Kertimag Ltd. Heavy metal concentrations in the test solution were 0, 50, 100, 200, 400, 800, 1600, 3200, 6400 mg l^{-1} , and they were added as $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$, $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$ and $\text{Pb}(\text{NO}_3)_2$. Chemicals were obtained from Reanal Laboratory Chemicals LLC.

The experimental procedure consisted of the following parts: 3 g cotton-wool was placed in plastic pots and twenty-five seeds were laid on each cotton-wool pads. The pads were moistened with approx. 50 ml test solution with a specific heavy metal concentration, and each pots were sealed with cellophane and set under a photoperiod of 12 h light and 12 h dark, and 25 ± 1 °C temperature. After six days, the number of germinated seeds was recorded, and root/shoot elongation were measured.

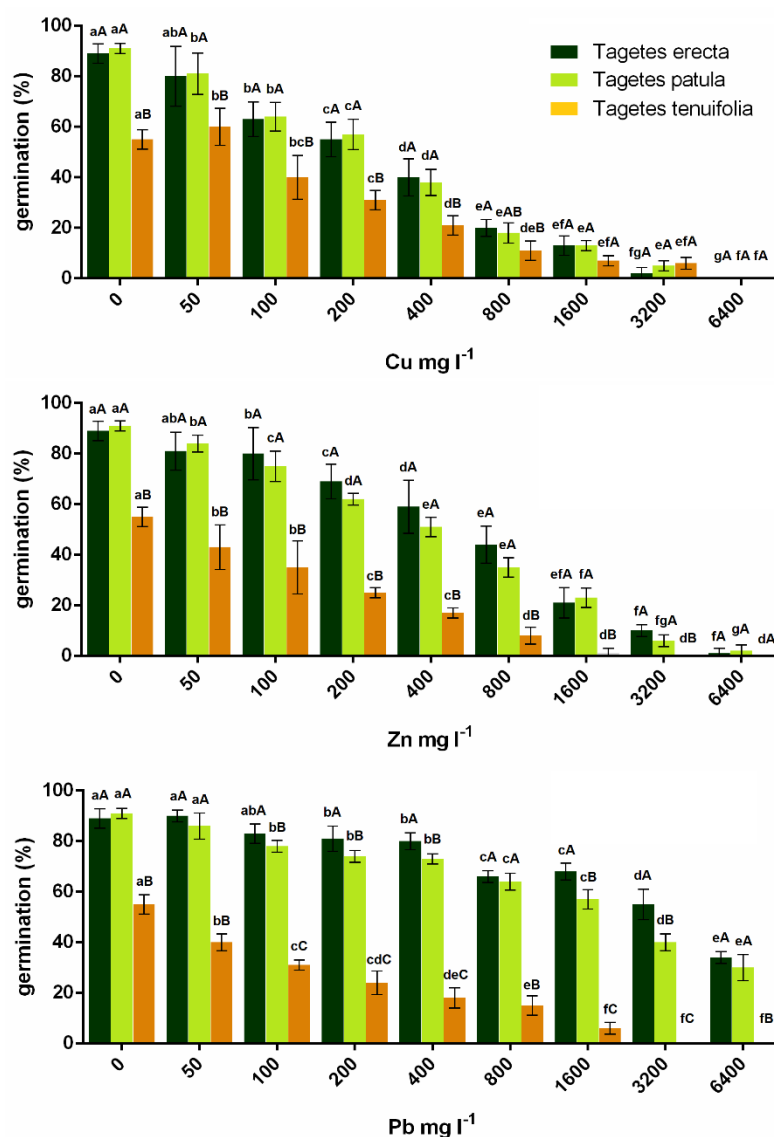
The experiment was conducted in a completely randomized design with four replications. The data were recorded as means \pm standard deviations and analyzed by Graphpad Prism 6. Two-way analysis of variance (ANOVA) and Tukey multiple comparisons were carried out to test for any significant differences (at 95% significance level) between the means. In figures the same small letter above the column means there is no significant difference at different heavy metal concentration, and the same capital letter means there is no significant difference among plants.

Results and discussion

Increasing concentration of heavy metals in the test solution significantly ($p < 0.05$) decreased the germination rates of all tested species (Fig. 1.). Signet marigold had significantly ($p < 0.05$) lower germination rates than African marigold and French marigold in all heavy metal treatments except for above 800 mg Cu l⁻¹ concentration. At 50 mg Cu l⁻¹ concentration there was a slightly increase in germination rate of Signet marigold, however all three plants had less germination rate at 100 Cu mg l⁻¹ compared with control. At 6400 mg Cu l⁻¹ germination was not observable. Zn also significantly decreased germination rates firstly at 100 mg l⁻¹ concentration, however the decline in germination rates of African marigold and French marigold was less than Cu in all concentrations. On the contrary, Zn was the most toxic heavy metal to Signet marigold. Pb had the least toxic effects on germination of African marigold and French marigold. Signet marigold was more sensitive to Pb than the other plants, and it was not germinated above 1600 mg Pb l⁻¹.

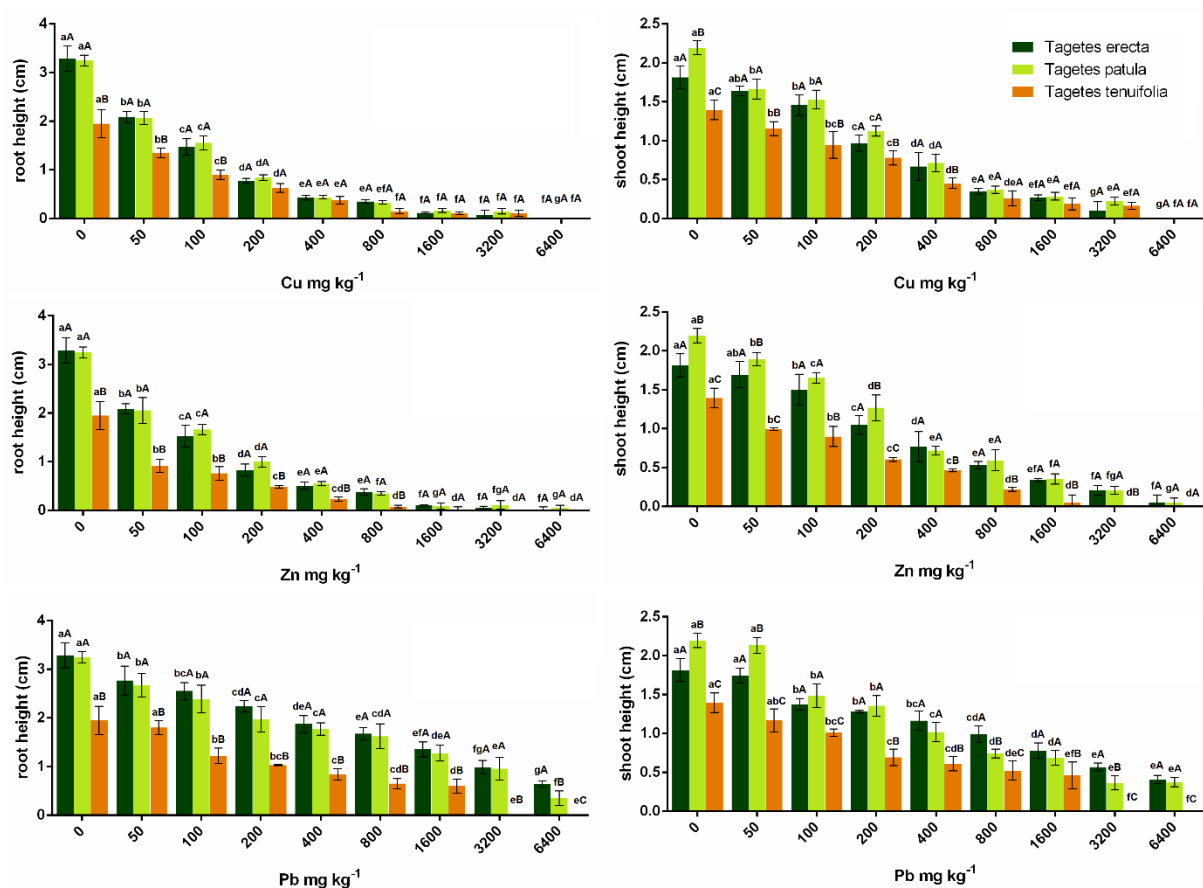
All tested heavy metals had a significant toxic effect ($p < 0.05$) on root lengths and shoot heights of the three plants (Fig. 2.). Signet marigold had significantly ($p < 0.05$) lower root lengths and shoot heights than African marigold and French marigold except for above 100 mg Cu l⁻¹ and 1600 mg Zn l⁻¹ concentrations. Toxic effects on root elongation was firstly observed at the least concentration (50 mg l⁻¹) of all tested metals. Cu was the most toxic heavy metal to root lengths, since 100 mg Cu l⁻¹ caused more than 50% decline in this growth parameter. Zn decreased root lengths by 50% at 200 mg Zn l⁻¹, while Pb decreased root lengths by 50% only at 800 mg Pb l⁻¹. Less toxic effects were observable on shoot elongation compared with the results of root elongation. Cu and Zn decreased shoot heights by 50% firstly at 400 mg l⁻¹ concentration. Shoot heights of French marigold and Signet marigold is decreased by 50% at 400 mg Pb l⁻¹, while only 1600 mg Pb l⁻¹ reduced shoot heights of African marigold at such a rate. Interestingly at 50 mg Pb l⁻¹ French marigold had significantly higher shoot heights than African marigold, however at 800 and 3200 mg Pb l⁻¹ African marigold had higher shoot heights.

Figure 1. Germination rates of marigolds.



The results showed that heavy metals caused significant ($p < 0.05$) decline in the growth parameters of the three plants. It is expected, because plant seeds were in direct contact with the toxicity of heavy metals in the test solution, and inhibition effects of heavy metals on growth parameters of African marigold has been also observed in previous studies [5,8,9,11]. Heavy metals had greater adverse effects on plant root lengths than on shoot heights. Inhibition of root elongation is known to be a more sensitive indicator to metal toxicity, since roots are the responsible for absorption and accumulation of metals [3,4,12]. Based on our results, Signet marigold is the most sensitive plant to the tested heavy metals among the three species. Between the effects of metals on African marigold and French marigold, no considerable differences were observed, however African marigold was more tolerant to high levels of Pb. Between the tested heavy metals the following series of phytotoxicity was observed in our experiment: Cu>Zn>Pb for African marigold and French marigold, and Zn>Cu>Pb for Signet marigold.

Figure 2. Root lengths and shoot heights of marigolds.



Conclusion

It was concluded that Signet marigold was very sensitive to the tested heavy metals, therefore only African marigold and French marigold can be used to remediate heavy metal (Cu, Zn and Pb) contaminated soils. These two plants can tolerate low concentration of metals (below 400 mg l^{-1}) without considerable decline in growth parameters. Marigolds may tolerate higher levels of heavy metals in natural environment, since the seeds were germinated in hydroponic solution during the experiment, which is quite different from natural soils. In soils metals could be tied up in insoluble forms, and they are less available to plants [13]. Although hydroponic experiments have very limited relevance to the natural environment, these research can be useful in demonstrating the tolerance of marigolds to Cu, Zn and Pb [14].

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