



Phytoremediation of Heavy Metals (Cr and Pb) Contaminated Soils by Pot Marigold (*Calendula officinalis* L.) with the Use of Salicylic Acid

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

The pot experiment was conducted to estimate the phytoremediation potential of pot marigold (*Calendula officinalis* L.) an ornamental plant for the chromium and lead contaminated soils. The grown as a test plant Cr and Pb was applied as @ 0, 30 and 60 mg/kg and salicylic acid (SA) were applied as @ 0, 2 and 4 mmol/kg. The results demonstrated that the applied of SA (4 mmol/kg) significantly decreased the dry biomass yield of root and shoot i.e. Cr (3.18±0.23 and 15.84±1.28

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g/pot) and Pb (3.89 ± 0.28 and 16.14 ± 1.28 g/pot) respectively, control pot were compared to (T₆) 60 mg/kg Cr and Pb contaminated soil. The applied of SA (4 mmol/kg) significantly increased the uptake by root and shoot of pot marigold plants i.e. Cr (20.93 ± 2.11 and 21.43 ± 2.03 mg/kg) and Pb (22.87 ± 1.03 and 23.09 ± 1.14 mg/kg) respectively, in comparison to the control pot (T₉) 60 mg/kg Cr and Pb contaminated soil. When SA applied, then the highest translocation factor (TF), bioaccumulation factor (BAF) and remediation factor (RF) values were recorded i.e. Cr (1.277 ± 0.121 , 0.442 ± 0.014 mg/kg and $0.863 \pm 0.061\%$) and Pb (1.263 ± 0.201 , 0.500 ± 0.041 mg/kg and $0.755 \pm 0.061\%$) in that order. The application of SA in Cr and Pb polluted soils considerably enhanced the uptake of these metals (Cr and Pb) though, reduced the growth and dry biomass yield of pot marigold. Therefore, SA 4 mmol/kg application may be recommended to reduce dry biomass of *Calendula officinalis* L. and enhance the Cr and Pb concentration in root and shoot of *Calendula officinalis* L. Thus, it may be concluded that salicylic acid (SA) can play a significant role in reducing the uptake of Chromium and Lead through *Calendula officinalis* L.

Keywords: Bioaccumulation; *Calendula officinalis* L. heavy metals; phytoremediation; salicylic acid (SA).

1. INTRODUCTION

In recent years, the escalating concern over environmental pollution and its detrimental effects on plant health has led to innovative approaches aimed at optimizing soil conditions (Papadkar et al., 2024). Heavy metal pollution is a significant environmental concern (Ganvir & Guhey, 2021). Soil containing these metals presents environmental issues that impact plants, animals, and ultimately human health because of their persistent and extremely poisonous nature (Atta et al., 2024; Ganvir & Papadkar, 2022). Research has shown that plants growing in polluted environments exhibit altered metabolism (Hussnain et al., 2023), growth reduction (Sarwar et al., 2023), lower biomass production (Komal et al., 2024), and oxidative damage along with the accumulation of pollutants (Khan et al., 2024). As a result, managing harmful materials is becoming more and more necessary to safeguard agricultural land. Compared to plants and animals, soil enzymes are more susceptible to heavy metal stress in the making those useful markers of the quality and conditions of soil (Abeed et al., 2022).

Lead is one of the most well-known trace heavy metals with a lengthy history of toxicity. Because of its long biological life, tendency to spread, and poisonous nature, exposure to lead is becoming a major concern (Talukdar, 2011). As Pb levels in the growing media rise, it has been observed to accumulate in the stem, leaves, roots, and seeds of plants (Komal et al., 2024). Lead buildup in leaves is dependent on its absorption from aerial sources (Obroucheva et al., 1998). The type of salts, concentration, and plant species all affect its effect. In certain situations,

smaller doses may accelerate metabolic processes, even though effects are more noticeable at larger concentrations and durations. Enzymatic activities, plant water status, mineral nutrition, seed germination, seedling growth, and photosynthesis are the main processes impacted (Patra et al., 2004). The application of waste-water treatment sludge, the use of fertilizers containing metals, the deposition of mining waste, and other comparable human activities have been the main causes of heavy metal pollution in agricultural soils (Unsal et al., 2013; Ganvir & Guhey, 2020). In addition to changing the biochemical and physiological cycles of plants, heavy metals like lead (Pb), cadmium (Cd), chromium (Cr), and nickel (Ni) can readily infiltrate the food chain and progressively build up in living things (Raj & Maiti, 2020). Because lead toxicity disrupts some cellular enzymes, it changes the usual metabolic routes in plants, including respiration, photosynthesis, and other important metabolic functions (Ravikumar & Thamizhuni, 2014; Ruley et al., 2004).

The second most prevalent inorganic metal pollutant in agricultural soil is chromium (Cr), which is mostly caused by its extensive industrial use in textile colors, leather tanning, and electroplating (Singh et al., 2013). The non-essential element Cr, on the other hand, can be absorbed by plants through sulfate transporters together with other essential elements, and it can lead to a number of metabolic abnormalities in plants. Plant death can result from Cr poisoning, which also disrupts respiration and photosynthesis, causes oxidative damage, and inhibits critical enzyme activity (Shanker et al., 2005; Singh et al., 2013). In the Salicylic acid,

sometimes referred to be 2-hydroxy benzoic acid, is a varied group of phenolic chemicals that are synthesized by plants and consist of an aromatic ring with a hydroxyl group or its functional derivative. This is salicylic acid a chelator the complex compound to simple compound converted of heavy metals. These are salicylic acid increased of solubility molecular from contaminated soil, because highly accumulate of Cr and Pb (Hayat et al., 2024). Plant development, thermo-genesis, flower induction, and ion uptake are all exclusively influenced by salicylic acid. It also reverses the effects of ABA on leaf abscission and affects stomata mobility and ethylene biosynthesis. Moreover, it increases the amount of photosynthetic pigments, speeds up photosynthetic reactions, and alters the activity of some crucial enzymes in both stress-free and stress-induced conditions (Ma et al., 2001).

Phytoremediation techniques remove of heavy metals from the environment by using a specific class of plants called hyper-accumulators (Raj & Maiti, 2020). Plant-based remediation is considered a green technology that is easy to apply, economically viable, and environmentally beneficial, but it still has a number of drawbacks, such as slow plant development, time consumption, and sensitivity to heavy metal accumulation (Razmi et al., 2021). Plants exposed to varying quantities of heavy metals may develop phytotoxicity, which could negatively impact the plants' ability to perform phytoremediation (Cameselle & Gouveia, 2019). Furthermore, the phytoremediation of heavy metals might also seem like an extremely difficult undertaking because the effectiveness of this biological therapy may be influenced by a number of factors, including the environment, the heavy metals' bioavailability, the choice and growth of plants, and others (Razmi et al., 2021). The translocation factor (TF) of hyper-accumulator plant species should be more than one, and they can accumulate metals 100 times more than other plant species (Soylak et al., 2013).

The family of Asteraceae includes the annual, short-lived perennial ornamental plant known as pot marigold (*Calendula officinensis* L.). *Calendula officinensis* is a quickly expanding plant that can be used to clean up places contaminated by heavy metals (Karimi & Koksai, 2022). This plant's ability to withstand elevated levels of Cr and Pb suggests that it could be utilized for phytoremediation of soils contaminated by metals (Liu et al., 2010). Although ornamental

plants don't alter the food chain while still improving the environment, their application for phytoremediation of metal-polluted soil has been seen as a very suitable economical, environmentally benign, and aesthetically viable preference (Khan et al., 2024).

These are the fundamental objectives of the recent study i.e., to evaluate the phytoremediation potential of pot marigold (*Calendula officinensis* L.) plants grown in Cr & Pb contaminated soils., to study the effects of SA on physiological characteristics (Dry biomass) of pot marigold plants., to study the effects of SA on the accumulation of heavy metals (Cr & Pb) by pot marigold plants grown in contaminated soils.

2. MATERIALS AND METHODS

2.1 Site and Design of Experiment

The pot experiment was carried out in Cr and Pb contaminated soils at Sheila Dhar Institute (SDI) experimental farm Mumfordganj at longitudes 81°52'E and latitudes 20°20'N, situated with elevation slope 101m at SDISS, University of Allahabad, Prayagraj. The pot experiment was carried out as an overall completely randomized design (CRD) at SDI experimental farm in the Rabi season during the year of 2023-24. Samples of soil were collected from SDI farm and then pots were filled with 5 kg soil samples. After the pots had been filled with soil and pot marigold (*Calendula officinalis* L.) seeds were sown as a test plant with 90% germination power of seeds and after 20 days of germination of seeds, the some seedling were thinned. The work of pot experiment was consisted of 09 treatments with a control pot. The design of experiment is given in Table 1.

2.2 Sampling of Soil and Analysis

Soil samples were taken at a depth of 0–20 cm from the SDI experimental farm in order to analyze the physico-chemical characteristics and heavy metal levels of the soil. After the soil samples were collected, they were allowed to air dry at ambient temperature, crushed into fine particles, and then sieved through a 2 mm sieve. The di-acid digestion method was used to measure the concentration of total Cr and Pb in a mixture of concentrated HNO₃ and HClO₄ (1:4 by volume). An Atomic Absorption Spectrophotometer at Banaras Hindu University in Varanasi, Uttar Pradesh, India, was used to measure the amounts of Cr and Pb in digested final soil samples (Mani et al., 2015).

Table 1. Treatments combinations of Chromium and Lead with SA

Treatments	Combinations of Cr	Combinations of Pb
T ₁	Control	Control
T ₂	Cr 0 mg/kg + SA 2 mmol/kg	Pb 0 mg/kg + SA 2 mmol/kg
T ₃	Cr 0 mg/kg + SA 4 mmol/kg	Pb 0 mg/kg + SA 4 mmol/kg
T ₄	Cr 30 mg/kg + SA 0 mmol/kg	Pb 30 mg/kg + SA 0 mmol/kg
T ₅	Cr 30 mg/kg + SA 2 mmol/kg	Pb 30 mg/kg + SA 2 mmol/kg
T ₆	Cr 30 mg/kg + SA 4 mmol/kg	Pb 30 mg/kg + SA 4 mmol/kg
T ₇	Cr 60 mg/kg + SA 0 mmol/kg	Pb 60 mg/kg + SA 0 mmol/kg
T ₈	Cr 60 mg/kg + SA 2 mmol/kg	Pb 60 mg/kg + SA 2 mmol/kg
T ₉	Cr 60 mg/kg + SA 4 mmol/kg	Pb 60 mg/kg + SA 4 mmol/kg

Note- Cr: chromium, Pb: lead, SA: salicylic acid

2.3 Sampling and Analysis of Plants

After carefully washing the plants with tap water, they were given 0.1 N HCl, detergent solution (0.2%), de-ionized water, and displacement drum washer (DDW). The dry biomass and length/height measurements of the roots and shoots were taken independently. The plant samples were oven-dried at 60 °C for biochemical analysis and at 75 °C in a hot-air oven for the analysis of heavy metals. Dried plant samples were finally ground to a fine powder. One gram of plant material was digested using a 15 ml tri-acid mixture that contained concentrated HNO₃ (16 M, 71%), H₂SO₄ (18 M, 96%), and HClO₄ (11 M, 71%) in a 5:1:2 ratio. The mixture was then cooked on a hot plate at 60 °C for 30 minutes in order to analyze the heavy metals in plants. Total content of Cr and Pb in the extracts of tri-acid mixture were determined by the Atomic Absorption Spectrophotometer at Motilal Nehru National Institute of Technology, Prayagraj (U.P.), India (Kumar & Mani, 2010).

2.4 Translocation Factor

Translocation factor (TF) was used to find out the efficiency of phytoremediation. TF is a suggestion of the ability of particular plant to movement of metals from root to shoot of plants (Marchiol et al., 2004). The formula it represented:

$$\text{Translocation Factor} = \frac{\text{Metal concentration in shoot}}{\text{Metal concentration in root}}$$

2.5 Bioaccumulation Factor

The bioaccumulation factor (BAF), which combines the concentration in plants and soil, can be used to assess how well heavy metals accumulate in plants (Talukdar, 2011).

$$\text{Bioaccumulation Factor} = \frac{\text{Metal concentration in shoot}}{\text{Metal concentration in soil}}$$

2.6 Remediation Factor

The remediation factor is the proportion of heavy metals absorbed by the stem and shoot from the contaminated soil (Meers et al., 2005), calculated based on the following equations,

$$\text{RF}\% = \frac{M_{\text{shoot}} \times W_{\text{shoot}}}{M_{\text{soil}} \times W_{\text{soil}}} \times 100$$

Where, the metal content in M_{shoot} is expressed as mg kg⁻¹, W_{shoot} is the yield of dry biomass plant shoot in gram, M_{soil} was calculated by total metal content in contaminated soils and W_{soil} every pot soil weight in grams.

2.7 Statistical Analysis

The pot experiment was conducted as a complete randomized design with each treatment replicated thrice. Statistical analysis of data was done following analysis of variance (ANOVA), when the ANOVA was significant that 0.05 ≤ mean were separated using critical difference (CD), at P level of significance. The diagrams were plotted using the Graph Pad prism-8.01 version.

3. RESULTS AND DISCUSSION

3.1 Physical and Chemical Properties of Soil

The calculated sand, silt and clay percentage is varied from 56.26±2.24, 22.54±1.56 and 21.18±1.18% respectively. The pH value of the soil is 7.6±0.07, which indicates that the soil is moderately saline. In soil samples' electrical conductivity, cation exchange capacity, and organic carbon ranged from 0.36±0.03 dSm⁻¹, 20.76±1.34 cmol (p+)/kg and 0.57±0.06%, respectively. The soil samples' total nitrogen and

phosphorus contents varied from 0.13 ± 0.02 to $0.12\pm 0.02\%$, respectively. In soil samples' amounts of Pb and Cr ranged from 0.52 ± 0.05 & 0.48 ± 0.04 mg kg⁻¹, respectively (Table 2). The heavy metals solubility capacity may be influenced by physico-chemical properties of soil, like pH, EC, and organic matter (OM). These are

most important harmful of contaminated soils were related to the decrease in the level of physiological properties from contaminated soils as a nutrients collection and improved nutrient cycling. In this way similar results have also been found by (Annu & Garg, 2015; Edogbo et al., 2020).

Table 2. Physical and chemical properties of soil

Parameters	Unit	Value
Sand	%	56.26±2.24
Silt	%	22.54±1.56
Clay	%	21.18±1.18
pH	—	7.6±0.07
EC at 25 °C	dS/m	0.36±0.03
CEC	Cmol(p ⁺)/kg	20.76±1.34
Organic carbon	%	0.57±0.06
Total N ₂	%	0.13±0.02
Total P ₂ O ₅	%	0.12±0.02
Total Chromium	mg/kg	0.52±0.05
Total Lead	mg/kg	0.48±0.04

Note- EC (electrical conductivity), CEC (cation exchange capacity), OC (organic carbon), Pb (lead) & Cr (chromium), each value of the three replicates (n=3, mean±SD)

Root dry biomass of *Calendula officinalis* L. plant

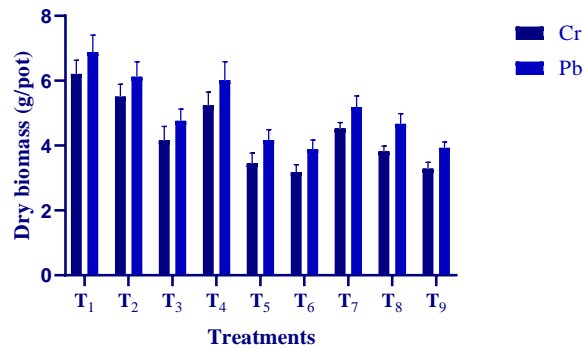


Fig. 1. The effect on dry biomass of pot marigold (*Calendula officinalis* L.) in root
For every value in the three replicates (n = 3, mean±SD), there is significantly difference at P<0.05

Shoot dry biomass of *Calendula officinalis* L. plant

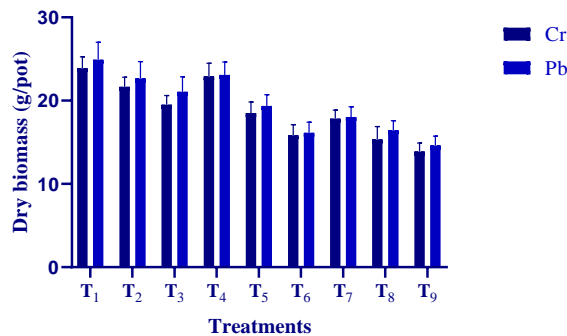


Fig. 2. The effect on dry biomass of pot marigold (*Calendula officinalis* L.) in shoot
For every value in the three replicates (n = 3, mean±SD), there is significantly difference at P<0.05

3.2 The Effect on Dry Biomass of Pot Marigold (*Calendula officinalis* L.) in Root

The experimental results showing the effect of SA chelator on dry biomass of pot marigold are evident (Fig 1). In figure illustrates the effects of Cr and Pb concentration and chelators on root dry biomass of pot marigold. The dry biomass of root was gradually and significantly decreased with the increase in Cr and Pb concentration in soil in the presence and absence of chelator, although no toxicity symptoms such as chlorosis or necrosis were observed in any treated plant. The applied in lower dose of SA (2 mmol/kg) significantly less reduced dry biomass yield of pot marigold in root i.e. Cr (3.46±0.31 g/pot) and Pb (4.17±0.32 g/pot) respectively, when compared the control pot to (T₅) 30 mg/kg of Cr and Pb contaminated soils. While the under applied higher dose of SA (4 mmol/kg) significantly reduced dry biomass yield of pot marigold plants in root i.e. Cr (3.29±0.19 g/pot) and Pb (3.93±0.18 g/pot) respectively, when compared the control pot to (T₉) 60 mg kg⁻¹ of Cr and Pb contaminated soils. The maximum dry biomass yield in the control pot, while the minimum dry biomass yield in the (T₉) treatment. Because SA concentration increased, the pot marigold plants dry biomass yield of the root decreased. The applied of SA showed the deleterious effects on pot marigold plants vitality which is evidenced by dry biomass yield of root and shoot of pot marigold plants. It also demonstrates that pot marigold is more affected than pot marigold by the total and dissolved soil Cr & Pb. However, the regression model indicates that pot marigold shows a less pronounced decrease in dry matter. Therefore, it may be said that this plant is more resilient to pollution from Cr and Pb. Found similar results (Ravikumar & Thamizhunivzn, 2014).

3.3 The Effect on Dry Biomass of Pot Marigold (*Calendula officinalis* L.) in Shoot

The experimental results showing the effect of SA chelator on dry biomass of pot marigold are present in (Fig. 2). In figure illustrates the effects of Cr and Pb concentration and chelators on shoot dry biomass of pot marigold. The dry biomass of shoot was gradually and significantly decreased with the increase in Cr and Pb concentration in soil in the presence and absence of chelator, although no toxicity

symptoms such as chlorosis or necrosis were observed in any treated plant. The applied in lower dose of SA (2 mmol/kg) significantly less reduced dry biomass yield of pot marigold in shoot i.e. Cr (18.51±1.33 g/pot) and Pb (19.37±1.33 g/pot) respectively, when compared the control pot to (T₅) 30 mg/kg of Cr and Pb contaminated soils. While the under applied higher dose of SA (4 mmol/kg) significantly reduced dry biomass yield of pot marigold plants in shoot i.e. Cr (13.89±1.01 g/pot) and Pb (14.61±1.12 g/pot) respectively, when compared the control pot to (T₉) 60 mg kg⁻¹ of Cr and Pb contaminated soils. The maximum dry biomass yield in the control pot, while the minimum dry biomass yield in the (T₉) treatment. Because SA concentration increased, the pot marigold plants dry biomass yield of the shoot decreased. The applied of SA showed the deleterious effects on pot marigold plants vitality which is evidenced by dry biomass yield of root and shoot of pot marigold plants. It also demonstrates that pot marigold is more affected than pot marigold by the total and dissolved soil Cr & Pb. However, the regression model indicates that pot marigold shows a less pronounced decrease in dry matter. Therefore, it may be said that this plant is more resilient to pollution from Cr and Pb. This type of experiment was done found similar results (Ravikumar & Thamizhunivzn, 2014).

3.4 The Uptake of Pot Marigold (*Calendula officinalis* L.) in Root

The uptake of Cr & Pb through pot marigold plants grown in contaminated soil. The pot investigations have presented (Fig. 3) in comparison with the control pot containing the polluted soils; the use of SA significantly increased the Cr and Pb contents in pot marigold plants. The applied lower dose of SA (2 mmol/kg) significantly enhanced the lower concentration of root i.e. Cr (6.84±0.51 mg/kg) and Pb (6.99±0.51 mg/kg) respectively, when compared with control pot to (T₅) 30 mg/kg of Cr and Pb contaminated soils. While the higher dose of SA (4 mmol/kg) is applied significantly enhanced the higher concentration of shoot i.e. Cr (20.93±2.11 mg/kg) and Pb (22.87±1.03 mg/kg) respectively, when compared with control pot to (T₉) 60 mg/kg of Cd and Pb contaminated soils. Whereas the maximum concentration found in T₆ of pot marigold plants in root i.e. Cr (10.42±0.76 mg/kg) and Pb (10.64±0.0.76 mg/kg) respectively, when compared to all treatments. The results are also in conformity with that of where they evaluated the effect of SA on heavy metals (Cu, Pb, Zn and

Cd) uptake by pot marigold and found that SA, both have the capacity to increase the metal accumulation, but more increase in metal accumulation was noticed with SA treatments, than citric acid treatments (Yusuf et al., 2013). The increasing heavy metal concentration in soil the potential of all chelator is reduced due to the translocation of heavy metals in the plants. The higher potential of SA to accumulate Cr and Pb in pot marigold may be due to difference in chemical structure and higher stability of Cr and Pb with SA in soil. The hydrophobicity of SA complex with heavy metals in the soil as a result of which more hydrophobic compounds passed through the apoplastic pathway, and thus are less resistance against their entry into the cell. The results of present study clearly indicate that pot marigold potential accumulated the Cr and Pb from treated soils and addition of SA had increased the accumulation of Cr than control treatments. Similar results have also been found by (Li et al., 2016).

3.5 The Uptake of Pot Marigold (*Calendula officinalis* L.) in Shoot

The uptake of Cr & Pb through pot marigold plants grown in contaminated soil. The pot investigations have presented (Fig. 4) in comparison with the control pot containing the polluted soils; the use of SA significantly increased the Cr and Pb contents in pot marigold plants. The applied lower dose of SA (2 mmol/kg) significantly enhanced the lower concentration of shoot i.e. Cr (8.09±0.56 mg/kg) and Pb (8.11±0.63 mg/kg) respectively, when compared

with control pot to (T₅) 30 mg/kg of Cr and Pb contaminated soils. While the higher dose of SA (4 mmol/kg) is applied significantly enhanced the higher concentration of shoot i.e. Cr (21.43±2.03 mg/kg) and Pb (23.09±1.14 mg/kg) respectively, when compared with control pot to (T₉) 60 mg/kg of Cd and Pb contaminated soils. Whereas the maximum concentration found in T₆ of pot marigold plants in root i.e. Cr (11.23±1.01 mg/kg) and Pb (11.56±0.0.76 mg/kg) respectively, when compared to all treatments. The results are also in conformity with that of where they evaluated the effect of SA on heavy metals (Cu, Pb, Zn and Cd) uptake by pot marigold and found that SA, both have the capacity to increase the metal accumulation, but more increase in metal accumulation was noticed with SA treatments, than citric acid treatments (Yusuf et al., 2013). The increasing heavy metal concentration in soil the potential of all chelator is reduced due to the translocation of heavy metals in the plants. The higher potential of SA to accumulate Cr and Pb in pot marigold may be due to difference in chemical structure and higher stability of Cr and Pb with SA in soil. The hydrophobicity of SA complex with heavy metals in the soil as a result of which more hydrophobic compounds passed through the apoplastic pathway, and thus is less resistance against their entry into the cell (Grcman et al., 2001). The results of present study clearly indicate that pot marigold potential accumulated the Cr and Pb from treated soils and addition of SA had increased the accumulation of Cr than control treatments. Similar results have also been reported by others (Wu et al., 2011; Thakkar, 2014).

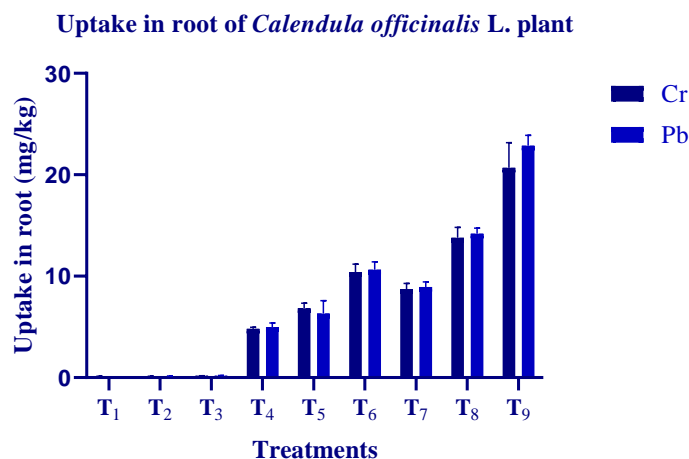


Fig. 3. The uptake of pot marigold (*Calendula officinalis* L.) in root
For every value in the three replicates ($n = 3$, mean±SD), there is significantly difference at $P < 0.05$.

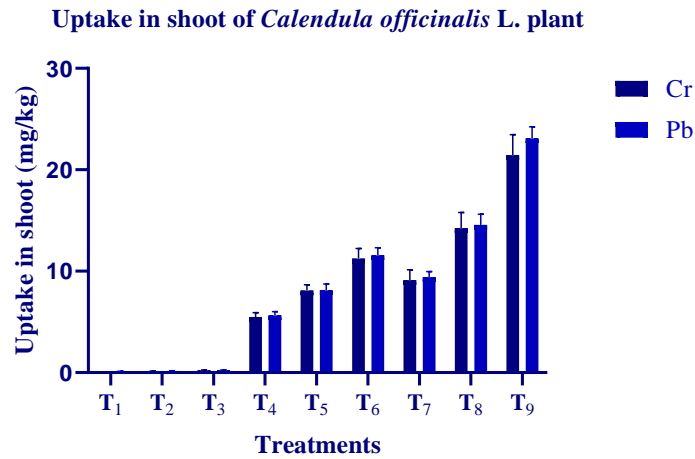


Fig. 4. The uptake of pot marigold (*Calendula officinalis* L.) in shoot
 For every value in the three replicates ($n = 3$, mean \pm SD), there is significantly difference at $P < 0.05$

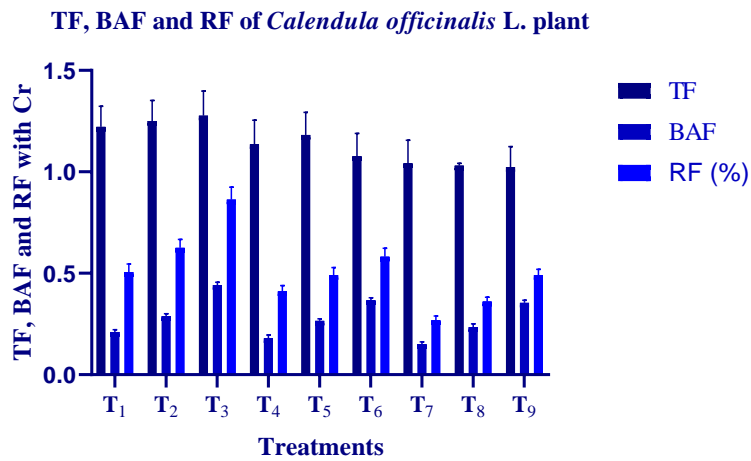


Fig. 5. The BAF, TF and RF of Cr of pot marigold

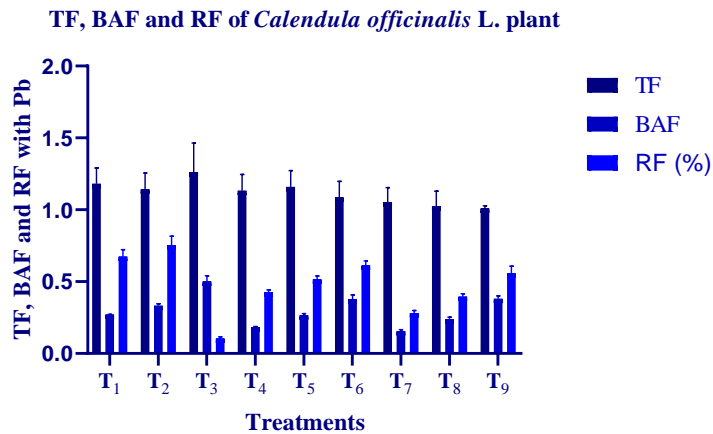


Fig.6. The BAF, TF and RF of Pb of pot marigold

3.6 The BAF, TF and RF of Cr and Pb of Pot Marigold (*Calendula officinalis* L.) Plant

The translocation factor, bioaccumulation factor and remediation factor are three vital indexes to determine the phytoremediation capability of plants. Significant effect of Cr and Pb level and chelator application on phytoremediation indices, TF, BAF and RF was observed. TF indicates the capability of plant to translocate heavy metals from the root to the shoot and BAF represent the accumulation of heavy metal from soil to aerial portion of plant and RF represents the efficiency of remediate in contaminated soils. The values of the TF, BAF, and RF factors of Cr and Pb are shown in the data in (Figs. 5&6). The TF and BAF of maximum concentration i.e. Cr (1.277 ± 0.102 and 0.442 ± 0.014 mg/kg) and Pb (1.263 ± 0.201 and 0.500 ± 0.041 mg/kg) respectively, whereas the TF and BAF of minimum concentration i.e. Cr (1.023 ± 0.101 and 0.150 ± 0.012 mg/kg) and Pb (1.009 ± 0.019 and 1.55 ± 0.011 mg/kg) in that order. It is significantly uptake of Cr & Pb in value was greater than 1 of TF concentration and less than 1 of BAF. However, the RF maximum value of Cr ($0.863\pm 0.061\%$) and Pb ($0.755\pm 0.061\%$) and minimum value of Cr ($0.268\pm 0.0021\%$) and Pb ($0.105\pm 0.011\%$), Whereas RF values is less than 1. The capacity of various plant species to absorb and transport heavy metals, such as Cr and Pb, in contaminated soils may also result from their genetic variety. The chelating agent significantly increased the RF values for Cr compared to Pb. This increases their solubility and bioavailability in the soils allowing the plant to absorb these metals more efficiently influencing TF, BAF and RF. The application of chelator increased TF and BAF, which can improve the Cr and Pb phytoremediation by pot marigold plant. This type of experiment also conducted by (Hussnain et al., 2023) and found similar results.

4. CONCLUSION

The phytoremediation potential of pot marigold (*Calendula officinalis* L.) was studied by growing in Cr and Pb treated soil with the help of chelator. According to the results of this study, pot marigold (*Calendula officinalis* L.) as an ornamental plant, showed a significant potential against stress, toxicity, and uptake of Cr and Pb. The use of chelator SA effectively increase Cr and Pb concentration in root and shoot of plants, TF, BAF and RF compared to the non-chelators

applied treatments. SA applications improve the metals accumulation through eliminating the physiological barriers in the root by removing of contaminated soils. The pot marigold (*Calendula officinalis* L.) is a flowering ornamental plant and does not a component of human and animal food chain, has an appropriate mechanism to overcome the Cr and Pb stress in plan and has BAF less than 1 for Cr and Pb; therefore, it can be used as a Cr and Pb hyper-accumulator plant in Cr and Pb polluted soils. In addition, due to the biodegradable nature of SA and increased efficiency of the TF of Cr and Pb than the non-biodegradable SA synthetic chelate, finally it is recommended that SA can be selected as the good chelator in order to increase the efficiency of Cr and Pb phytoremediation. Therefore the current study recommends that phytoremediation is the best and most eco-friendly approach to mitigate the metal pollutant found in contaminated soils.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during writing or editing of this manuscript.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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