The effect of cadmium on the growth, tolerance and cadmium-accumulation of some ornamental plants in Vietnam

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Abstract
Cadmium (Cd) is a non-essential heavy metal that is toxic and causes negative effects on plant growth. In this study, five ornamental plants in Vietnam (Catharanthus roseus, Helianthus annuus, Tagetes erecta, Medicago sativa and Celosia argentea) were researched for the effect of Cd on their growth and tolerance. We collected data on shoot height, biomass, chlorophyll content, tolerance index and Cd accumulation of these plants after 30 days of exposure to Cd. The results suggested that Cd caused the inhibition of shoot growth for all species. Particularly, the shoot growth of C. argentea was promoted by 25 mg kg^{-1} Cd concentration in the soil after 30 days of treatment.

Similarly, the biomass and chlorophyll content of C. argentea were also increased at some concentrations. The tolerance index of C. argentea and the Cd accumulation of C. roseus are the highest values. Hence, C. argentea and C. roseus show that they have many special characteristics than the other species while living in the Cd polluted-environment. Therefore, they should be conducted for deep studies on heavy metal stress fields, especially Cd.

Keywords: Plant growth, cadmium stress, tolerance, ornamental plant, accumulation.

Introduction

Heavy metal pollution is a widespread problem in the world that is caused mainly by human activities and has an impact on the health of living organisms[25]. Agricultural activities, industry and urban development release large amounts of heavy metals into water and soil such as Cd, Pb, As etc.[9,20]. Cd is classified as a dangerous and toxic heavy metal according to the classification of the Agency for Toxic Substances and Disease Registry[15]. Like many other heavy metals, Cd is not necessary metal for the growth of organisms, high toxicity and cannot be decomposed in nature, so it has serious effects on the health of animals and plants when exposed[23]. Clinical studies show that when humans are exposed to Cd, they are more likely to develop kidney disease, liver failure, reproductive disorders, cancer and other health problems[15]. In plants, the toxicity of Cd can affect growth and development such as inhibiting shoot growth, accumulating of biomass and damaging the photosynthetic apparatus[6]. Plants develop a variety of defense systems to tolerante heavy metal stress including both enzymatic and non-enzymatic systems[2,7]. These defense systems enable plants to reduce Cd-related toxic symptoms and then continue to survive and grow under Cd stress[12]. Therefore, the research on Cd-tolerant plant species and Cd accumulation is the first step in selecting plant species with potential for remediating Cd-pollution. Vietnam is a tropical country with many commonly ornamental plants that are considered a rich source of plants for studies. In this study, we focused on understanding the impact of Cd on five ornamental plants in order to discover ornamental plants with Cd tolerance potential and accumulation for pollution treatment purposes in the future.

Material and Methods

Chemicals and plant seeds: $CdCl_2.½H_2O$ and ethylenediamine-tetraacetic acid disodium (Na_2EDTA) were provided by the Xilong Chemical Company (China). Ethanol absolute and HNO_3 were purcharsed from Vina Chemistry Solutions Company (Vietnam) and Merck (Germany) respectively. Soil was collected from the Green Saigon Company (Vietnam). Seeds of *Catharanthus roseus, Helianthus annuus, Tagetes erecta, Medicago sativa* and *Celosia argentea* were provided by the Vietnam Floating Company (Vietnam).

Induction of Cd stress: The soils were mixed with Cd^{2+} solution to reach the required concentrations (0, 25, 50, 100, 150 and 200 mg kg^{-1}) and incubated in the net house for 30 days. The 30-day-old plants were transferred into the pot containing the incubated soils and watered every day with an equal volume to maintain 60 – 70% humidity.

Plant collection and recording of the relevant parameters: After 30 days, the plants were sampled and washed with the water at the roots to clean the soils. Then, Cd attached to the root surface was washed with 15 mM Na_2EDTA for 20 mins. The shoot height and fresh weight were recorded. The plant samples were dried at 80^{o}C for 96 hours to get their dry weight.

Tolerance index (TI) was calculated by the average of root, shoot, leave biomass and height of the control versus treatments[8].

Chlorophyll content: The extraction method and parameters of chlorophyll were followed by Su et al[22]. Absorbance values of 645 and 663 nm were read by the Multiskan SkyHigh Spectrophotometer (Thermo Scientific). The concentration of chlorophyll was calculated according to the following formula:

Chlorophyll a (µg/g) = $12.72\ OD_{663\ nm} - 2.59\ OD_{645\ nm}$
Chlorophyll b (µg/g) = $22.9\ OD_{645\ nm} - 4.67\ OD_{663\ nm}$
Total chlorophyll (µg/g) = ((Chlorophyll a + Chlorophyll b).$V_{ethanol\ (mL)}$)/(Fresh weight $_{(g)}$ .1000)

Determination of Cd content: Dry plant samples were treated following the method of Feng et al[8]. The concentration of Cd in samples was determined by an Atomic absorption spectrophotometer (ASS).

Statistical analyses: Each of the experiments was replaced three times. The data was analyzed by the one-way ANOVA (p value < 0.05) of SPSS software (version 29.0.0). The graphs were drawn using Microsoft Excel 2016.

Results and Discussion

Effect of Cd-concentration on shoot height: As a toxin, Cd inhibits plant growth and development[10]. Five ornamental plants were used in our study. The growth of shoot, which was observed by shoot height, was inhibited under Cd stress (Fig. 1). Following a 30-day of treatment with various Cd concentrations, the shoot height was reduced to about 25.13 – 49.25% in *C. roseus* that makes this specie as the most affected by Cd-toxin compared to the other four species (*T. erecta*, *H. annuus* and *M. sativa* were decreased 27.74 – 34.86%, 25.20 – 33.67% and 10.90 – 19.89% respectively). In contrast, *C. argentea* improved 13.08% of the shoot height at 25 mg kg^{-1} Cd.

The decrease of shoot length (10.34 – 39.12% compared to the control) in *C. argentea* is the result of treatment at higher concentrations. The stimulation of shoot growth at low Cd concentrations was also observed in *Zea mays*[14] that suggests low Cd concentrations acting as a hormetic factor for the shoot growth of plants[5]. Thus, our results indicated that Cd toxicity-affected ornamental plants led to inhibition of shoot growth which is supported by many previous studies[1, 3].

Effect of Cd-concentration on biomass: The Cd-treatment on five ornamental plants in this study illustrated that there was a drop in whole plant biomass including fresh and dry weight (Fig. 2). The higher is Cd concentration treatment, the lower is the biomass. As a result, the plant groups exposed to soil containing 200 mg kg^{-1} Cd produced the least biomass. Although there is improvement of biomass of plants living in the low-heavy metal environment, a large amount of heavy metal in the soil environment inhibits biomass of plants. Hormetic effect of heavy metals[5,13] can occur in *C. argentea* in our experiments. After 30 days-treatment, the evidence indicates that corresponding to the increase in shoot height of *C. argentea*, the biomass also increased in fresh weight (37.26% and 30.78%) and dry weight (58.40% and 56.93%) at 25 and 50 mg kg^{-1} Cd respectively. This feature is not found in the other four species of our study.

Based on the hormetic effect of heavy metals, *C. argentea* has similar mechanisms to *Brassica juncea*[21], *Lonicera japonica*[11] and *Sesuvium portulacastrum.*[8] The presence of heavy metal concentrations of soil tends to increase permeability, changes membrane potential, damages the enzyme system and affects physiological functions in cells, which are the reasons for growth prevention in plants[26]. The slow development of plants undergoing Cd stress is due to Cd affecting nutrient uptake, photosynthesis and other growth-related processes in the cell[19]. Consequently, as biomass index, *C. argentea* is a potential specie at low-Cd contaminant area while people should consider *H. annuus* planted in long-term under heavy metal stress.

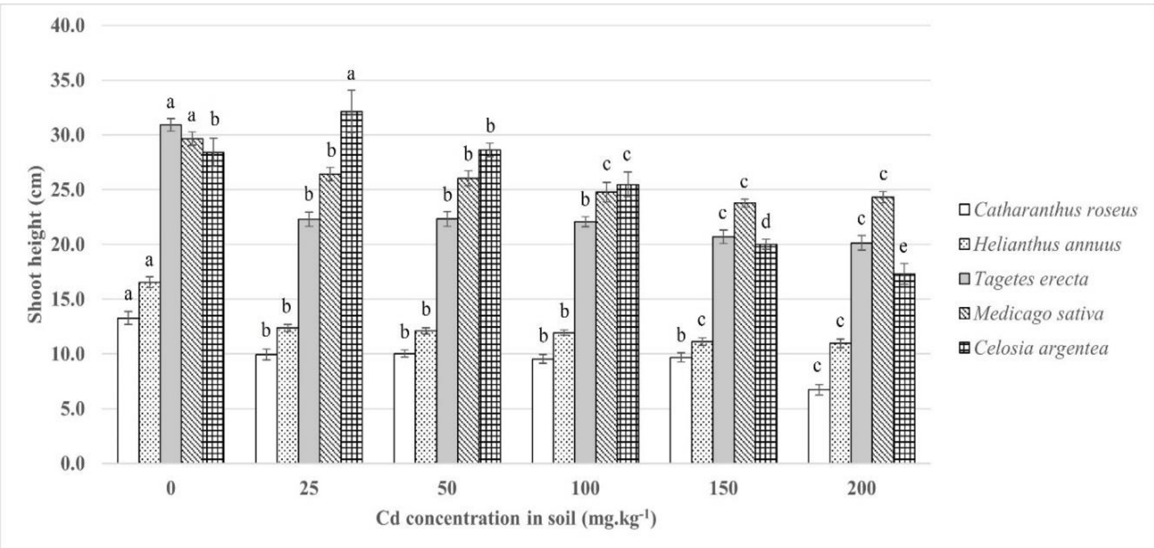

Fig. 1: The height of five plants' shoot was treated with Cd-concentrations after 30 days. The letters (a, b, c, d and e) show the significant differences (p < 0.05).

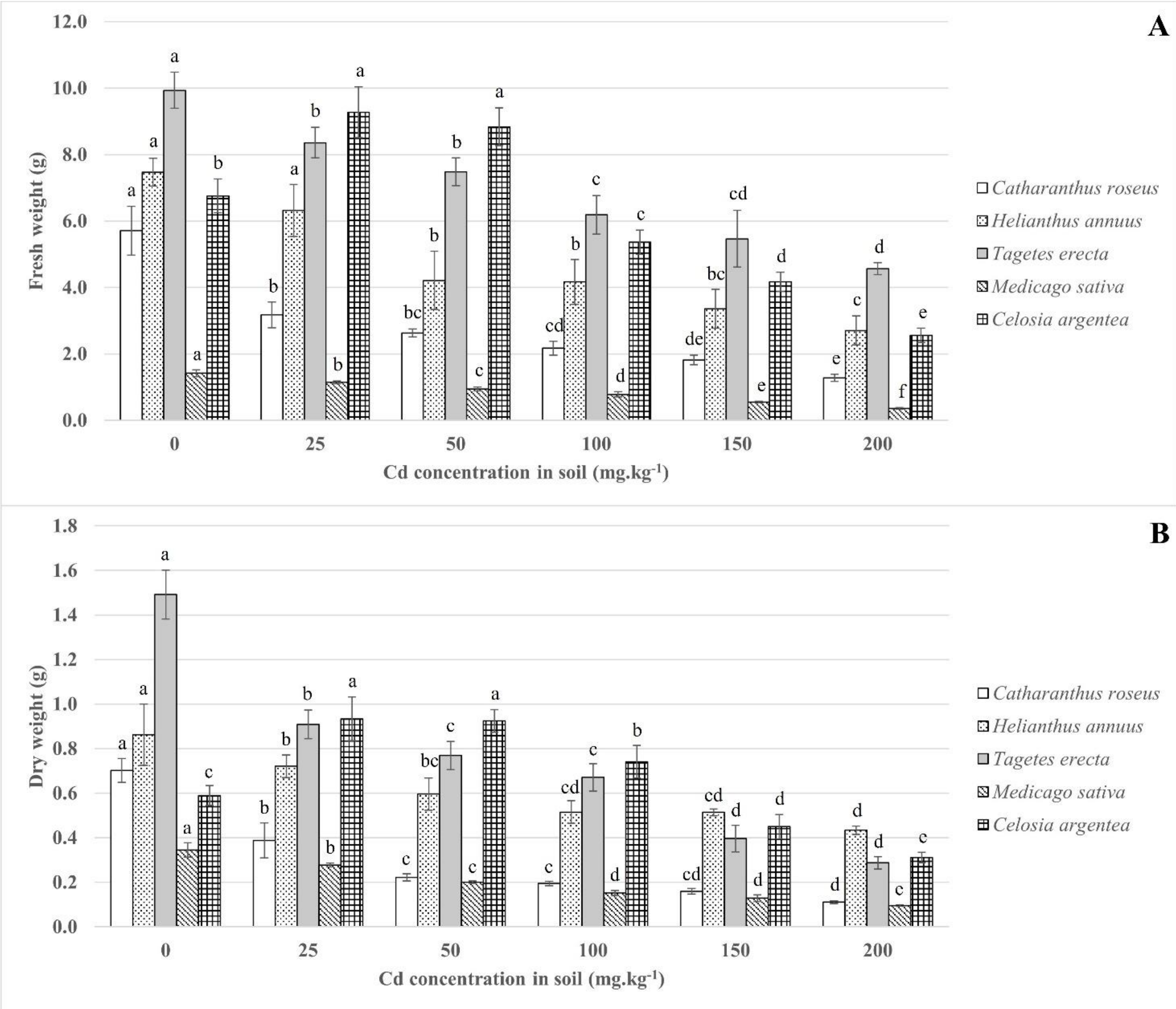

Fig. 2: (A) The fresh weight and (B) dry weight of five ornamental plants were treated with different Cd-concentrations after 30 days. The letters (a, b, c, d and e) show the significant differences ($p < 0.05$).

Effect of Cd-concentration on chlorophyll: Exposure to Cd caused a fall in the chlorophyll of leaves in ornamental plants (Fig. 3). In *C. roseus, H. annuus, T. erecta* and *M. sativa* experiments, the content of chlorophyll witnessed a decrease from 12.81% to 52.82% after 30 days. These results demonstrate clearly that photosynthetic apparatus of the survey plants was affected negatively by Cd treatment. This toxicity of Cd was also illustrated by other researchers around the world[17, 27]. For *C. argentea*, it increased at all of Cd-concentractions. This trend is also observed in species such as *Dianthus carthusianorum*[16] and *Z. mays*[14]. Brzóska et al[4] suggested that Cd and Zn have some similar activity properties in the cell. Simultaneously, Torabian et al[24] indicated that Zn relates to increasing chlorophyll in plants.

The tolerance of ornamental plants to Cd: Figure 4 shows the TI values of five survey plants undergone various levels of Cd stress after 30 days, the TI value declines when Cd concentration rises. *C. argentea* had the highest TI value (around 1.27) whereas the figure for *C. roseus* is the lowest (around 0.60) compared to *H. annuus, T. erecta* and *M. sativa* (versus 0.75, 0.82 and 0.82 respectively) at 25 mg kg^{-1} Cd. It can be seen that there are similar results from the other Cd concentration treatments. According to Rascio and Navari-Izzo,[18] the tolerance of plants is a characteristic associated with heavy metal accumulation. Consequently, *C. argentea* suggests the potential plant in the tolerance to Cd stress than the other four ornamental plants. Simultaneously, all five ornamental plants of this research should be encouraged to conduct extensive research in the phytoremediation fields.

Cd accumulation: After 30 days treatment, the Cd-accumulation was observed in whole plant of five survey species in which roots tended to accumulate more Cd than shoots at the concentrations investigated (Fig. 5). Roots of *C. roseus* showed significantly higher accumulation (up to 242.00 mg kg^{-1} Cd of dry biomass) compared to the other four species (Fig. 5B) from 2 to 14-times at all Cd concentrations in soil. In this study, the more Cd is in the soil, the more Cd accumulates in roots of plants.

The shoots of *T. erecta* had higher Cd accumulation (maximum up to 51.78 mg kg^{-1} Cd of dry biomass) than the other four (Fig. 5A).

At different levels of Cd stress, the amount of Cd in shoots of plants also changed. For instance, the shoots of *C. argentea* were best able to accumulate 25 mg kg^{-1} Cd, the figure for *C. roseus* is 47.83 mg kg^{-1} Cd of dry biomass at 50 mg kg^{-1} Cd, while the best accumulation is *T. erecta* (51.78 mg kg^{-1} Cd of dry biomass) at 150 mg kg^{-1} Cd. Therefore, it is necessary to select suitable species for the target of Cd accumulation in shoots.

At the Cd concentrations of 25 and 50 mg kg^{-1}, total Cd accumulation of whole plants decreases in the order of *C. roseus*, *C. argentea*, *M. sativa*, *H. annuus* and *T. erecta*. At high Cd concentrations of the soil, *H. annuus* showed more Cd accumulation than *M. sativa* and *T. erecta*. These findings illustrate that *C. roseus* and *C. argentea* were two species for further phytoremediation.

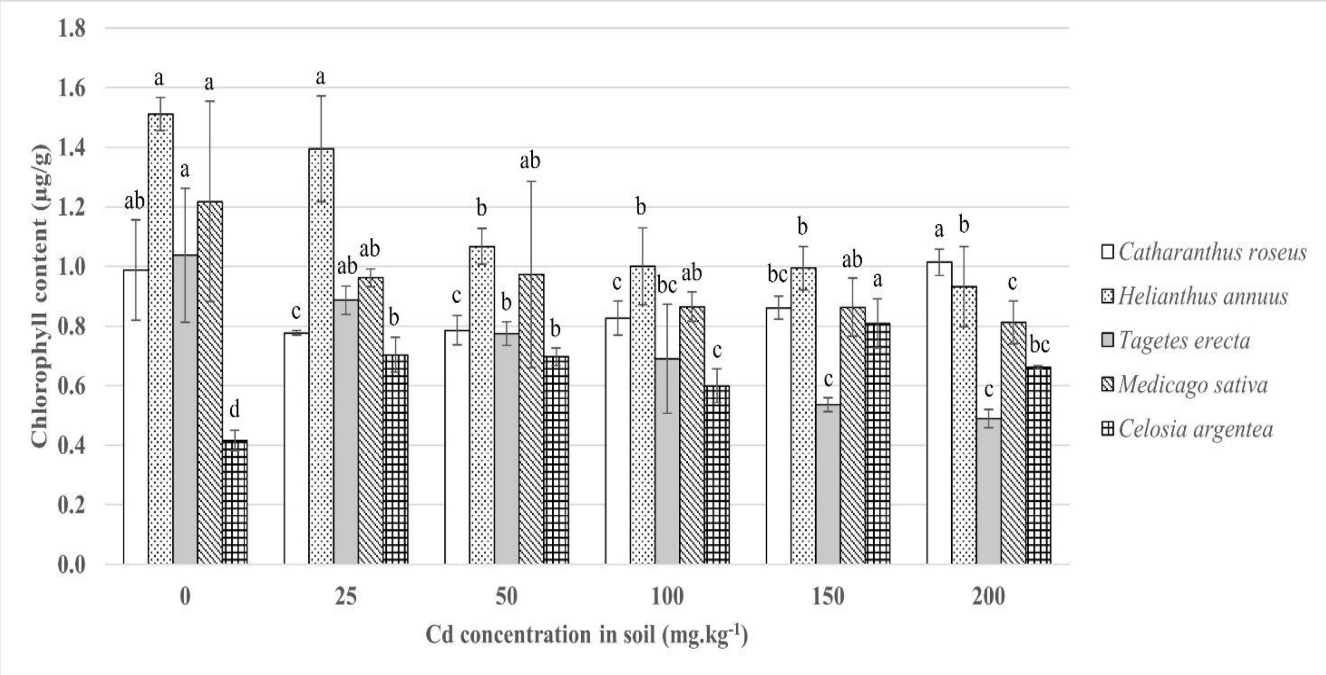

Fig. 3: The chlorophyll content of five ornamental plants was treated with Cd-concentrations after 30 days. The letters (a, b and d) show the significant differences ($p < 0.05$).

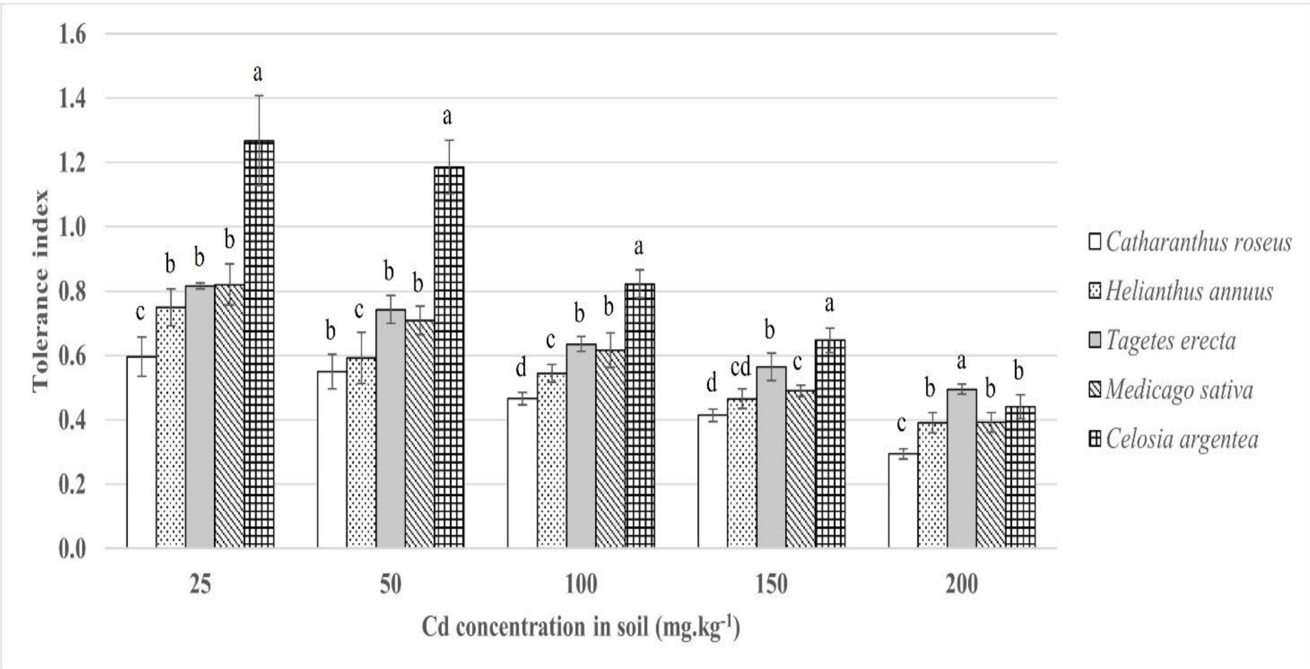

Fig. 4: The TI values of plants was treated with the various levels of Cd after 30 days. The letters (a, b and d) show the significant differences ($p < 0.05$).

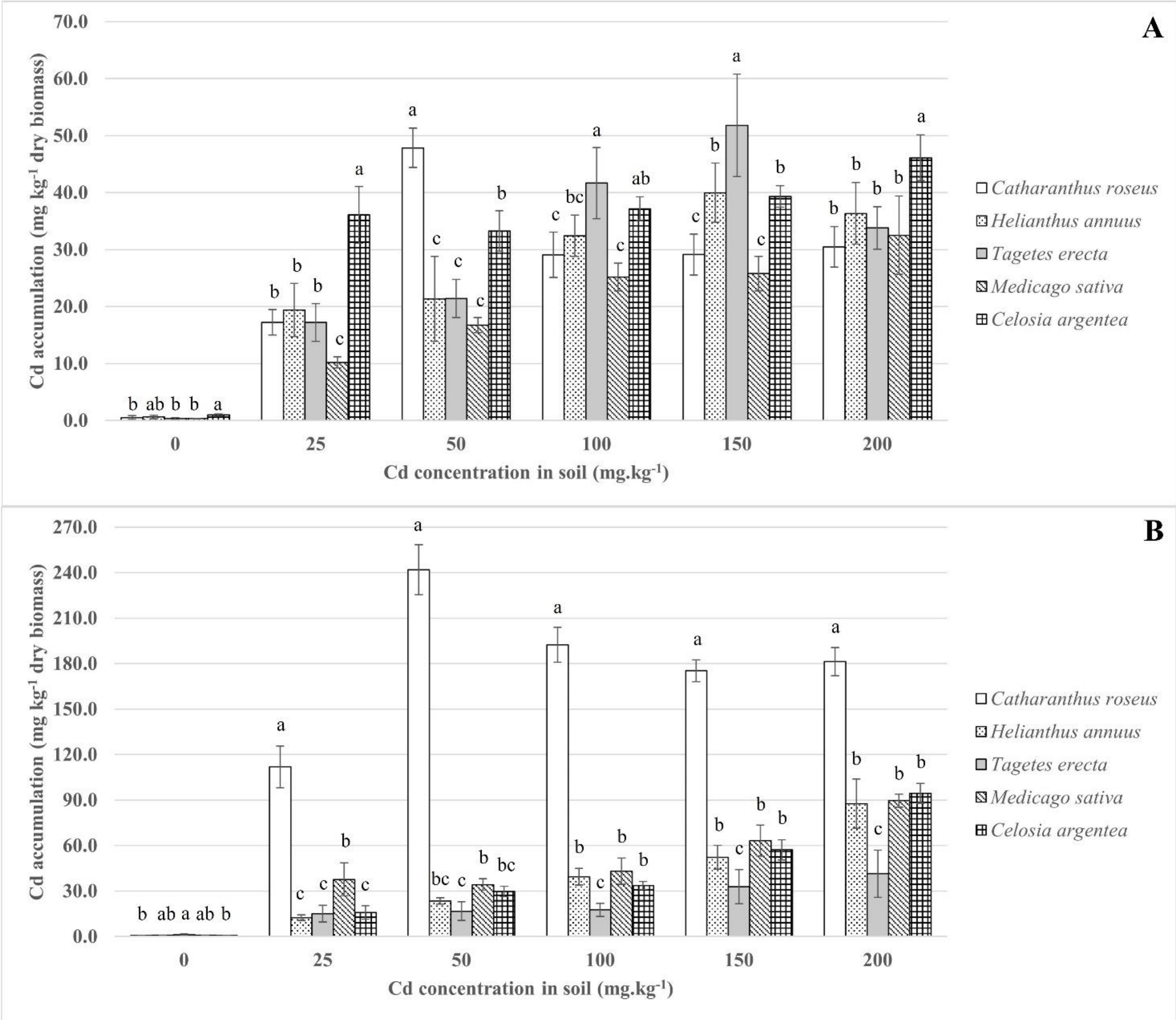

Fig. 5: The Cd accumulation of (A) shoot and (B) root of five ornamental species was treated with different Cd-concentrations after 30 days. The letters (a, b, c) show the significant differences ($p < 0.05$).

Conclusion

In short, all five ornamental plants can live in Cd polluted-soil. However, Cd had an inhibitory effect on shoot growth. Cd stimulates the growth of *C. argentea* at several levels of stress. These results not only show the impact of Cd on the development of servey species, but also illustrate as the base for conducting further studies in the phytoremediation field, because one species (*C. roseus*) is the best Cd accumulator of plants and another (*C. argentea*) is highly tolerant to Cd.

Acknowledgement

This research was funded by Vietnam National University, Ho Chi Minh City (VNU-HCM) under grant number B2021-18-04.

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(Received 15th March 2023, accepted 12th May 2023)