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Cadmium uptake and accumulation ability of *Rosmarinus officinalis* and its growth changes

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Abstract

To investigate the effect of cadmium and its accumulation rate on growth of *Rosmarinus officinalis*, this project was performed in Padina Research and Development greenhouse, Mashhad, Iran. Experiment was conducted by using clay pots in completely randomized design with 4 replications and 5 cadmium concentrations (0, 50, 100, 200 and 400 μM). Some traits such as root and stem growth, cadmium accumulation rate in root and shoot, biological and transfer factors were measured. During project period no toxicity symptoms such as necrotic were found in treated plants with cadmium. Analysis of variance indicated that root and stem growth of treated plant significantly decreased. The highest cadmium accumulation rate in root (207 mg/kg) and stem (352.5 mg/kg) belonged to 400 μM. According to the results, biological and transfer factors in all of treated plants were high and reached to 13.3 and 2.27, respectively. Given that a plant is cadmium hyper-accumulator, can save higher than 100mg/kg of it and has higher than 1 of biological and transfer factors. So,*Rosmarinus officinalis* is introduced as a cadmium hyper-accumulator plant.

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Introduction

Soil and water contaminated with metals pose a major environmental and human health problem that is still in need of an effective and affordable technological solution. As, Cd, Cu, Hg, Pb and Zn are the most environmentally important metallic pollutants.Excess concentrations of some heavy metals in soils such as Cd, Cr, Cu, Ni, and Zn have caused the disruption in natural aquatic and terrestrial ecosystems. The ideal plant species to remediate a heavy metal contaminated site would be a rapidly growing, high biomass crops with an extensive root system that can tolerate and accumulate the contaminants of interest. Several studies have been conducted to evaluate the effect of different heavy metals concentration on live plants (SaiKachout*et al*., 2009). Heavy metal pollution is mainly the result of human activities such as agriculture, mining, construction and industrial processes (Ahmadi-Afzadiand Mousavi-Bideli,2005;Parsadost*et al*., 2007). According to Kabata-Pendias, Pendias and Whitney improper waste disposal activities and overuse of pesticides were among the most significant sources of heavy metal pollution in the environment (Nourmohammadi and Rezvani 2005).

Cadmium (Cd) is one of highly bioactive and toxic elements, its presence at elevated levels in soils and drinking water is threatening food safety and human health (Sun *et al*., 2008). It adversely affects biological activities as a teratogen, carcinogen or mutagen and having detrimental effects on the digestive system, respiratory system and immune system (Liao *et al*., 2005). Cd that is a highly toxic metal pollutant of soils, inhibits root growth, shoot growth and yield production, affects nutrient uptake and homeostasis, and is frequently accumulated by agriculturally important crops and then enters the food chain with a significant potential to impair animal and human health. The application of sewage sludge, city waste and Cd-containing fertilizers causes the increase of Cd content in soils. Excessive amount of Cd may cause decrease in uptake of nutrient elements, inhibition of various enzyme activities, induction of oxidative stress including alterations in enzymes of the antioxidant defense system (John *et al*., 2009).

One of the current technologies for cleaning contaminated sites is phytoremediation, this is a promising approach to the difficult problem of remediating heavy metal-polluted soils(Khosrobeki*et al*., 2010). In this method plants are used to remove pollutants from the environment. The use of metal-accumulating plants for cleaning soil and water contaminated with toxic metals is the most rapidly developing component of this environmentally friendly and cost-effective technology (Wei *et al*., 2005).

Hyperaccumulators are plants that have an innate capacity to absorb metal at levels 100 times greater than otherplants(Sun *et al*., 2008). Hyper-accumulators are explained based on the following characteristics: (1) shoot metal concentrations (threshold values) are >10,000 mg/kg dry weight of shoots for Zn and Mn, 1000 mg/kg for Co, Cu, Ni, As and Se and 100 mg/kg for Cd; (2) biological factor (ratio of metal concentration in plant to soil) is greater than 1.0, sometimes reaching 50; (3) transfer factor (ratio of metal concentration in shoots to roots) is greater than 1.0 (Wei and Zhou, 2004).

As regards, soil pollution with Cadmium metal is one of the most important environmental problems in many countries, phytoremediation of heavy metals in contaminated areas with hyper-accumulator plants is one of effective and environmental friendly methods andon the other hand, due to medicinal importance of *Rosmarinus officinalis* and its using for urban green spaces, the effect of cadmium contamination on plant growth and metal absorption rate by the plant was studied.

Material and methods

Experiment location and preparation plantlets

Hydroponic pot experiment was conducted in completely randomized design with 4 replications and

5 cadmium concentrations (0, 50, 100, 200 and 400µM) in Padina Research and Development greenhouse, Mashhad, Iran. Its average temperature was 23 and 17°C at day and night, respectively. Earthen pots with diameter of 12cm and height of 8cm were selected. The annual rooted cuttings of *Rosemarinus officinalis* were selected. They were placed in soil and irrigated with distilled water (in order to avoid transmission stress effect).After a week, cuttings were transferred to the culture pots (sand peat and perlite with a ratio of 1:1)after root complete separation of soil and wash to the extent there was no soil around roots. At this time Shoot and root length was measured and Hoagland hydroponic nutrition was started during this stage.

Evaluation of cadmium accumulation

According to SaiKachout and *et al.* (2009) Cd concentrations of 0, 50, 100, 200 and 400µM were applied after 3 weeks.$CdCl_2$. 2.5 H_2O was used as Cd source. Plants were harvested after 35 days, then root length and stem height were measured. The plant samples were washed twice with tap water and once with distilled water, and dried separately at 100°C for 10 minutes, then at 70°C in an oven until completely dry. 1gr of plant sample was become to ashes by graphics furnace and digested by nitric acid and hydrochloric acid. cadmium concentration was determined by using atomic absorption spectrophotometer (AAS) method (Sun *et al.*, 2008).

Statistical analysis

Data were placed in Microsoft Office Excel 2010. Biological (metal concentration in plant/metal concentration in soil) and transfer (metal concentration in shoot/ metal concentration in root) factors were calculated by means of formula for data. Analysis of variance and means comparison were performed by SPSS software (the version of 15) and the Duncan's multiple range tests at the level of 5%, respectively.

Results

Analysis of variance indicated (Table 1) that cadmium significantly affected all measured traits ($\alpha \leq 0.05$).

Table 1. Analysis of variance of the cadmium effect on growth of *Rosmarinus officinalis*and the metal accumulation rate in it.

				Mean	Squares (MS)		
SOV	Df	Stem growth	Root growth	Root Cd concentration	Shoot Cd concentration	Cd biological factor	Cd Transfer factor
Cd	4	4.575*	3.34*	19973.32*	63382.32*	33.504*	0.989*
Error	15	0.167	0.038	51.167	151.03	0.482	0.099
CV(%)	-	0.65	0.62	0.68	0.83	0.32	0.35

ns, non significant; *, significant at P≤0.05; **, significant at P≤0.01.

Table 2. Effect of cadmium concentrations on measured traits of *Rosmarinus officinalis*.

Cd concentrations	Stem growth (cm)	Root growth (cm)	Root Cd concentration (mg/kg)	Shoot Cd concentration (mg/kg)	Cd biological factor	Cd transfer factor
0µM	3.5a	3.00a	15.5e	34.00e	6.04e	1.04c
50µM	1.25b	1.00b	65.25d	74.75d	13.3a	1.15c
100µM	1.00b	0.88b	84.5c	88.25c	7.85b	2.27a
200µM	1.25b	1.13b	105.75b	147.5b	6.56d	1.4bc
400µM	1.00b	0.88b	207a	352.5a	7.84c	1.71b

Means in a column followed by the same letter are not significantly different at P≤0.05.

According to the results, the highest stem (3.50cm) and root (3.00 cm) growth belonged to the control treatment. No significant difference was observed between root and stem growth in treated plants with cadmium toxicity concentrations (Fig. 1). The lowest shoot (34 mg/kg) and root Cadmium accumulation (15.5 mg/kg) belonged tothe control treatment. Treatment of 400 µM showed the highest shoot and root Cadmium accumulation with 352.5 and 207 mg/kg, respectively. The lowest transfer(1.04) and biological (6.04) factor belonged to 50 µM treatment. The highest cadmium transfer (2.27) and biological (13.3) factor were observed in 100 and 50 µM, respectively (Table 2).

Discussion

Root and stem growth was decreased after cadmium application. In fact, cadmium prevents the cell division and growth in meristematiczone. On the other hand, premature differentiation and lignifying cell walls in elongation zone may be the other reason for reducing root growth (Fusconi*et al.*, 2007; Shariyat*et al.*, 2010).

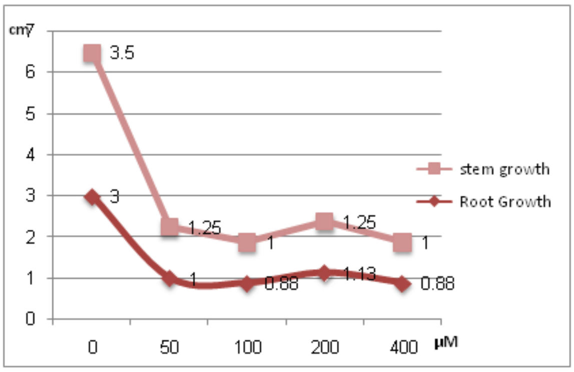

Fig. 1. The effect of cadmium different concentrations on root and stem growth of *Rosmarinus officinalis*.

Generally, cadmium accumulation in plants root and shoot was increased by adding high cadmium concentrations to nutrient solution. These results were matched with theresults of other investigators (Prabha*et al.*, 2007; Taji and Golchin, 2010).Based on the results of the project, no symptoms of contamination and disease such as leaves discoloration and wilting, stem hogging and root fragility observed in plant samples under stress of heavy metal. The results were consistent with Shariyat*et al.* on *Eucaliptusoccidentalis* and opposed with Ghasemi and Shahabi results on *Solanumlycopersicum*(Shariyat*et al.*, 2010; Ghasemi and Shahabi, 2010). So,*Rosmarinus officinalis* is introduced as a cadmium hyper-accumulator plant and can be planted in urban green spaces for pollution reduction.

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