The effect of cadmium on growth and composition of essential oils of *Mentha piperita* L.

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Abstract

Cadmium is a non-essential element that induces various toxic responses in plants when accumulated above the threshold level. The aim of the present research was to study the effect of cadmium on growth factors, concentration of cadmium in the rhizomes and leaves, and quality and quantity of essential oils in the leaves of *Mentha piperita* L. The experiment was carried out in a completely randomized design with three replications. Rhizomes with uniform weight were planted in pots 30 × 50 × 35 cm. After foliation of all rhizomes, plants were irrigated every other day, for 2 months by different concentrations of $CdCl_2$ (0, 100, 500, and 1000 µM). Results demonstrated that the minimum stem length and fresh and dry weight of leaves were achieved in the plants treated with $CdCl_2$ (500 µM). With increasing the cadmium in treatments, the amount of cadmium in the rhizomes and leaves were also increased. In treatment of $CdCl_2$ (1000 µM) the accumulation of cadmium in the leaves was more than the rhizomes. There were not significant differences in the essential oils contents between treatments. Analysis of the chemical composition of essential oils indicated that the main constituents of all treated plants were 1, 8 Cineole, Dihydrocarvon, Pulegone, and Carvone. Limonene oxide was observed only in the leaves of control plants.

Key words: *Mentha piperita*; cadmium; essential oils; growth

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Introduction

The genus *mentha* includes 25-30 species that grow in tropical areas of Eurasia, Australia, and South Africa. There are 5 species and several varieties in Iran which are distributed in most parts of Iran especially in slopes of the Elborzs (Robson, 1987). *Mentha* species are widely used in conventional medicine for their antispasmodic, antiseptic effects (Edris et al., 2003). The essential oils in this plant are industrially important (Aflatuni, 2005; Kizil et al., 2010).

Current emphasis on soil, water, and air pollution, food quality, and food and energy shortages in certain parts of the world make it desirable to re-examine the role of excess metals in plant growth. Cadmium is an important environmental pollutant. Cd has no known function as nutrient and seem to be more or less toxic to the plants and microorganisms (Ekine and Agu, 2008; Nies, 1999). Several studies have suggested that an oxidative stress could be involved in Cd toxicity, by either inducing oxygen free radical production, or by decreasing enzymatic or non-enzymatic antioxidant

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(Benavides et al., 2005; Cho and Seo, 2005; Ramesh and Satakopan, 2010).

It was shown that Cd reduced the activity of nitrate reductase and reduced the absorption of nitrate (Hernandez et al., 1996). Cd also competes in the uptake, transport, and use of several essential minerals (Ca, Mg, P, and K) (Das et al., 1997) and causes desiccation stress. Cd toxicity can affect the plasma membrane permeability, causing a reduction in water content; in particular, Cd has been reported to interact with the water balance (Costa and Morel, 1994). However, Cd toxicity responses of different species vary greatly and are dependent on the interaction of the genotype and its concentration.

The main goal of this study was to investigate the effect of Cd on growth responses, Cd accumulation in different parts of *Mentha piperita* L. plants, and the chemical composition of their essential oils.

Materials and Methods

Plants culture

Rhizomes with uniform weight were planted in the pots 30 × 50 × 35 cm. After foliation of all rhizomes, plants were irrigated every other day, for 2 months by 0, 100, 500, and 1000 µM concentrations of $CdCl_2$. The experiment was carried out in a completely randomized design with three replications.

Cd accumulation in rhizome and leaf

The rhizomes and shoots of treated plants were dried in an oven. Cd concentration in tissues was estimated after digesting the samples in nitric acid:perchloric acid (3:1, v/v). Cd concentration was determined by an atomic absorption spectrophotometer(Irfan et al., 2014).

Extraction and analysis of essential oils

Essential oils were distilled from leaves of treated plants in a steam distillation apparatus for 90 min. The oils were dried over sodium sulfate. The chemical compositions of essential oils were determined by GC/MS. GC-MS analyses were carried out with the Agilent Technology 7890 GC. A fused silica column 5% phenyl-poly-dimethyl-siloxane (DB- 5MS 30 m x 0.25 mm i.d. and 0.25 µm film thickness) was used. The electron ionization energy was 70 eV. Ion-source temperature and the interface temperature were 200 °C and 270 °C, respectively. An injection at 260 °C injector temperature was employed. The oven temperature was programmed as follows: from 60 °C (4 min hold) raised at 3 °C/min to 100 °C (2 min hold), and raised from 3 °C/min to 225°C (4 min hold).

Data analysis

Experiments followed a randomized complete block design with three replications. Analysis of variance was performed by the General Linear Model procedure (SPSS ver. 16) and differences among treatments were evaluated by Duncan Test ($p \leq 0.05$).

Results

Growth parameters

There was a significant difference between means of shoot length in 500 µM treatment and the others. Maximum and minimum shoot lengths were observed in control (39.49 cm) and in the 500 µM treatment (33.00 cm), respectively (Table 1).

Table 1
Means of shoot length, internode length, number of leaves, and fresh and dry weights of the leaves/shoots in response to different concentrations of cadmium chloride; grouped by Duncan test ($p \leq 0.05$); The same letter show no significant difference.

$CdCl_2$ (µM)	Shoot Length (cm)	Internode Length (cm)	Number of Leaves	FW of Leaves per Shoot (g)	DW of Leaves per Shoot (g)
0	39.41 a	3.83 a	15.75 a	2.84 ab	0.76 a
100	38.33 ab	4.75 a	13.41 a	1.95bc	0.47 ab
500	33.00 b	3.93 a	14.08 a	1.50 c	0.36 b
1000	38.72 ab	4.65 a	19.90 a	3.09 a	0.56 ab

Table 2
Means of Cd levels in the leaves and rhizomes in response to different concentrations of cadmium chloride; grouped by Duncan test ($p \leq 0.05$); means with same letter have no significant differences.

$CdCl_2$ (μM)	Cd in leaves (mg/gDW)	Cd in rhizome (mg/gDW)	% Essential oil
0	0.30d	0.21d	0.796a
100	1.24c	1.47c	0.330ab
500	3.28b	3.53ab	0.450ab
1000	4.34a	3.87a	0.623a

Different Cd concentrations had too little effect on internode length and leaf numbers. Maximum levels of both factors were gained in the 1000 μM treatment (Table 1).

There were no significant differences between means of fresh and dry weights of leaves in 500 μM treatment and the others. The Minimum of these factors was observed in the 500 μM treatment (Table 1).

Cadmium accumulation in rhizome and leaf

The present study showed that the Cd level in rhizome and leaf significantly increased when the concentration of Cd in the solution

Table 3
Percentage of essential oils composition of different $CdCl_2$ treated plants

Compound (arranged basd on retention time)	%			
	0 μM $CdCl_2$	100 μM $CdCl_2$	500 μM $CdCl_2$	1000μM $CdCl_2$
2-hexenal	0.16	-	0.14	-
alpha.-thujene	0.05	-	-	-
alpha.-pinene	1.85	0.84	1.45	1.09
camphene	0.55	-	0.45	0.40
sabinene	1.08	-	0.71	0.63
2-.beta.-pinene	1.33	0.76	0.56	0.45
beta.-myrcene	1.11	0.74	0.91	0.97
pseudolimonene	0.05	-	-	-
1,8 cineole	28.03	16.47	23.59	24.38
cis-sabinenehydrate	0.15	-	0.11	0.12
limonene oxide	0.12	-	-	-
pinocarvone	0.16	-	0.15	0.16
borneol	0.14	0.90	0.95	1.08
isopulegone	0.24	0.99	0.82	0.72
dihydrocarvone	18.62	17.71	17.78	17.37
pulegone	19.81	24.22	27.48	24.56
carvone	21.04	25.86	18.66	21.07
endobornyl acetate	1.08	1.75	1.10	1.34
p-mentha-3,8-diene	0.90	1.35	1.84	1.34
4,8-o-menthatriene	0.07	-		0.11
cis-2,6-dimethyl-2,6-octadiene	0.12	-	0.12	0.12
cis-l-carvyl acetate	0.33	-	0.32	0.41
.alpha.-bourbonene	0.09	-	-	0.10
acetic acid, decyl ester	0.10	-	0.09	0.10
trans-caryophyllene	1.18	3.75	1.22	1.50
alpha.-caryophyllene	0.08	1.05	1.22	0.11
beta.-farnesene	0.12	-	0.13	0.17
bicyclogermacrene	0.09	-	-	0.16
caryophyllene oxide	0.48	0.72	0.66	0.61
delta.-cadinene	0.21	0.60	0.19	0.25
phytol	0.09	-	-	0.22
benzene-dicarboxylic acid	-	2.30	-	-

increased. In the treatment of 1000 µM $CdCl_2$ rhizome accumulated relatively less quantities of Cd than leaves (Table 2).

Chemical composition of essential oils

In different treatments, no significant differences were verified in essential oil yield. However, mean yield was slightly higher in control plants (Table 2).

Analysis of the chemical composition of essential oils indicated that the main constituents of all treated plants were 1,8 Cineole, Diydrocarvon, Pulegone, and Carvone. The high level of 1,8 Cineole and the minimum level of Pulegone were achieved in the control. Limonene oxide was observed just in the control and also benzene-dicarboxylic acid was found just in the plants of 100 µM $CdCl_2$ (Table 3).

Discussion

The results demonstrated that the growth factors slightly were affected by Cd. It seems *Mentha piperita* is relatively cadmium resistant. This means that peppermint can be grown in soils containing Cd.

Cd reduces Photosynthesis (Dalla Vecchia et al., 2005) and it can change the water balance and decrease the size and number of Xylem (Barceló and Poschenrieder, 1990). Also, this metal decreases the absorption of necessary nutritional elements such as Ca, Mg, and Fe (Gussarsson et al., 1996). So Cd uptake at toxic level causes mineral deficiency, desiccation, and cellular metabolic disturbances in plants (Gomes and Soares, 2013; Marshner, 2012).

With increasing the cadmium in treatments, the amount of cadmium in the rhizomes and leaves also increased. In treatment of $CdCl_2$ (1000 µM) the accumulation of cadmium in the leaves was more than that in the rhizomes. Metals can move from the soil to plant roots. Accumulation of cadmium in root is a tolerance processes in some species. In these plants, much part of absorbed cadmium is remained linked to wall or storage in root cells vacuoles. In cucumber, cadmium accumulation in root is more than leaf (Moreno-Caselles et al., 2000). Translocation of metals from root to shoots has been the subject of numerous studies. Jarvis et al. (1976) found that the roots of some crops (lettuce, cress, etc) released much more of their absorbed Cd for translocation to the shoots than other crops. In *Brassica juncea*, *Silence vulgaris*, and *Arabidopsis halleri*, leaf epidermis is the main site of cadmium accumulation whereas in *Brassica napus*, the most concentration of cadmium is in mesophyll (Dixit et al., 2001). Zheljazkov et al. (2006) indicated that at elevated Cd concentrations in the growth medium, Cd transport from roots to shoots of the three species (peppermint, basil, and dill) was impaired (Zheljazkov et al., 2006). These findings correspond with those of the present study.

Our results indicated no detectable amount of Cd in the oils of treated plants. There were no significant differences in essential oils contents between treatments. Chemical composition of essential oils indicated that the main constituents of all treated plants were 1,8 Cineole, Dihydrocarvon, Pulegone, and Carvone. The study by Kizil et al. (2010) revealed that menthol (38/06%) and cineol (3/62%) were the main components of mint (*Mentha piperita*) essential oils and carvone (50/33%) and D. Limonene (16/47%) were dominant compounds of essential oils in *M. spicata.*

The effect of heavy metals on essential oils has been reported by many researchers (Nasiri et al., 2010; Prasad et al., 2010; Zheljazkov et al., 2006). Zheljazkon et al. (2006) evaluated the effect of Cd, Pb, and Cu on yields and essential oils of peppermint, basil, and dill. Their results showed the tested treatments slightly altered chemical composition of the essential oils of basil and dill, and reduced the menthol content in the peppermint oil.

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References

Aflatuni, A., 2005. The yield and essential oil content of mint *(Mentha ssp.)* in Northern Ostrobothnia.(http://herkules.oulu.fi/isbn9514277465/). Oulu University, Finland, p 52.

Barceló, J. and **C. Poschenrieder. 1990**. P'lant water relations as affected by heavy metal stress: a review'. *Journal of Plant Nutrition*, 13(1): 1-37.

Benavides, M.P., S. M. Gallego, S.M. and **M.L. Tomaro.2005**. 'Cadmium toxicity in plants'. *Brazilian Journal of Plant Physiology*, 17(1: 21-34.

Cho, U.-H. and **N.-H. Seo. 2005**. 'Oxidative stress in *Arabidopsis thaliana* exposed to cadmium is due to hydrogen peroxide accumulation'. *Plant Science*, 168(1): 113-120.

Costa, G. and **J. L. Morel. 1994**. 'Water relations, gas exchange and amino acid content in Cd-treated lettuce'. *Plant Physiology and Biochemistry*, 32(4): 561-570.

Dalla Vecchia, F., N. La Rocca, I. Moro, S. De Faveri, C. Andreoli and **N. Rascio. 2005**. 'Morphogenetic, ultrastructural and physiological damages suffered by submerged leaves of *Elodea canadensis* exposed to cadmium'. *Plant Science*, 168(2): 329-338.

Das, P., S. Samantaray and **G. Rout. 1997**. 'Studies on cadmium toxicity in plants: a review'. *Environmental pollution*, 98(1): 29-36.

Dixit, V., V. Pandey and **R. Shyam. 2001**. 'Differential antioxidative responses to cadmium in roots and leaves of pea (*Pisum sativum* L. cv. Azad)'. *Journal of Experimental Botany,* 52(358): 1101-1109.

Edris, A., A. Shalaby, H. Fadel and **M. Abdel-Wahab. 2003**. 'Evaluation of a chemotype of spearmint (*Mentha spicata* L.) grown in Siwa Oasis, Egypt'. *European Food Research and Technology.* 218(1): 74-78.

Ekine, R. and **G. Agu. 2008**. 'Fetal contamination with cadmium following chronic exposure of rat dams during gestation'. *African Journal of Applied Zoology and Environmental Biology*, 7(1): 120-124.

Gomes, M.P. and **A. M. Soares. 2013**. 'Cadmium effects on mineral nutrition of the Cd-hyperaccumulator *Pfaffia glomerata*'. *Biologia*, 68(2): 223-230.

Gussarsson, M., H. Asp, S. Adalsteinsson and **P. Jensén. 1996**. 'Enhancement of cadmium effects on growth and nutrient composition of birch (*Betula pendula*) by buthionine sulphoximine (BSO)'. *Journal of Experimental Botany*, 47(2): 211-215.

Hernandez, L., R. Carpena-Ruiz and **A. Garate. 1996**. 'Alterations in the mineral nutrition of pea seedlings exposed to cadmium '. *Journal of Plant Nutrition*, 19(12): 1581-1598.

Irfan, M., A. Ahmad and **S. Hayat. 2014**. 'Effect of cadmium on the growth and antioxidant enzymes in two varieties of *Brassica juncea*'. *Saudi journal of biological sciences*, 21(2): 125-131.

Jarvis, S., L. Jones and **M. Hopper. 1976**. 'Cadmium uptake from solution by plants and its transport from roots to shoots'. *Plant and Soil,* 44(1): 179-191.

Kizil, S., N. Hasimi, V. Tolan, E. Kilinc and **U. Yuksel 2010**. 'Mineral content, essential oil components and biological activity of two mentha species (*M. piperita* L., *M. spicata* L.)'. *Turkish Journal of Field Crops*, 15(2): 148-153.

Marshner, P., 2012. Mineral Nutriton of Higher Plants, London, UK.

Moreno-Caselles, J., R. Moral, A. Pérez-Espinosa, and **M. Pérez-Murcia. 2000**. 'Cadmium accumulation and distribution in cucumber plant'. *Journal of Plant Nutrition*, 23(2): 243-250.

Nasiri, Y.,S. Zehtab-Salmasi, S. Nasrullahzadeh, N. Najafi and **K. Ghassemi-Golezani.2010**. 'Effects of foliar application of micronutrients (Fe and Zn) on flower yield and essential oil of chamomile (*Matricaria chamomilla* L.)'. J. *Med. Plants Res*, 4(17): 1733-1737.

Nies, D.H., 1999. 'Microbial heavy-metal resistance'. *Applied microbiology and biotechnology*, 51(6): 730-750.

Prasad, A., A. K. Singh, S. Chand, C. Chanotiya and **D. Patra. 2010**. 'Effect of chromium and lead on yield, chemical composition of essential oil, and accumulation of heavy metals of mint species'. *Communications in soil science and plant analysis*, 41(18): 2170-2186.

Ramesh, B. and **V. Satakopan. 2010**. 'Antioxidant activities of hydroalcoholic extract of *Ocimum sanctum* against cadmium induced toxicity in rats'. *Indian Journal of Clinical Biochemistry*, 25(3): 307-310.

Robson, N., 1987. 'Carminative property of peppermint in magnesium trisilicate mixture, BP'. *Anaesthesia*, 42(7): 776-777.

Zheljazkov, V.D., L. E. Craker and **B. Xing. 2006**. 'Effects of Cd, Pb, and Cu on growth and essential oil contents in dill, peppermint, and basil'. *Environmental and Experimental Botany*, 58(1): 9-16.