

## The effect of nano-silicone on biochemical characteristics of European borage (*Borago officinalis* L.) under the cadmium stress

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### Extended abstract

#### Introduction

Environmental pollution with heavy metals has spread in the world. The impact these pollutants on the human health and food chain, as one of the factors of economic and health concerns, needs more attention. Cadmium (cd) as one of the most important heavy metals compared to other metals is rapidly absorbed and accumulated in plant tissues, and its transfer to the food chain is the result of widespread contamination of the soil with cadmium. Nano-silicon (Si) is more effective than usual fertilizers in protecting plants against biotic and abiotic stresses. Medicinal plants are one of the most important sources of drug production that humans have used for many years, and their importance is expanding day by day. Many secondary metabolites of *Borago officinalis* and its products are consumed in the world pharmaceutical markets, so it requires special attention to improve the quality of culture, production efficiency, and health. This attention requires increasing knowledge about the physiological mechanisms of this plant affected by environmental factors such as cadmium stress. This study aimed to investigate the effect of nano-silicon on alleviating the detrimental effects of cadmium stress on physiological properties such as antioxidant enzyme activity in *Borago officinalis*.

#### Materials and methods

Treatment was performed by adding cadmium in the form of cadmium nitrate ( $\text{Cd}(\text{NO}_3)_2$ ) and nano-silicon in hydroponic cultures of borage plants at the 7-8 leaf stage. This study was conducted as a factorial experiment based on a completely randomized design with three replications. Experimental factors were cadmium at three levels (0 (control), 25 mg L<sup>-1</sup> and 75 mg L<sup>-1</sup>) and nano silicon at two different levels (0 and 1.5 mM). One day, one week, and two weeks after treatment, plant leaves were sampled and the amount of cadmium accumulation and biochemical and physiological properties were measured.

#### Results and discussion

The results showed that the accumulation of cadmium in the aerial parts of the borage plant was significantly increased with increasing the cadmium concentration in the hydroponic medium. The utilization of nano-silicon significantly reduced the amount of cadmium absorption and accumulation in borage plants.

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Cadmium nitrate increased the amount of hydrogen peroxide compared to the control. The use of nano-silicon significantly reduced the amount of hydrogen peroxide under cadmium stress at one and two weeks after treatment. One of the important consequences of increasing the concentration of heavy metals in plants is increasing in the production and accumulation of reactive oxygen species (ROS) such as superoxide, hydroxyl and hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) radicals in the cells. This reactive oxygen species leads to the oxidation and destruction of macromolecules such as protein, DNA, cell membrane damage and ion leakage.

With increasing the concentration of cadmium, the amount of malondialdehyde (MDA) and proline in the cells were significantly increased. Malondialdehyde (MDA) and proline levels were significantly reduced by nano-silicon treatment. Heavy metals such as cadmium lead to the production of hydroxyl radicals, followed by lipid peroxidation. With increasing the lipid peroxidation, the cell membrane is destroyed and malondialdehyde levels increase. In the present study, the amount of malondialdehyde was significantly reduced under the influence of nano-silicon treatment. Proline accumulation in plants may also be a biomarker of cadmium stress, because proline can scavenge free radicals and protect the cells from their damaging effects.

With increasing the concentration of cadmium, the activity of polyphenol oxidase, peroxidase, and catalase enzymes was increased. In addition, the activity of these enzymes was reduced with the utilization of nano-silicon. The highest activities of these enzymes were recorded under cadmium concentration at 75 mg l<sup>-1</sup> and the lowest activities of these enzymes were related to the treatment of nano-silicon and control. Increasing the level of antioxidant activity can sweep ROS produced by heavy metals, and protects the cell from the damaging effects of oxidative stress, which increases the plant's tolerance to environmental stresses, including heavy metals. The use of nano-silicon positively reduces cadmium accumulation in the plant maybe by maintaining the photosynthetic capacity and regulating the uptake and transfer of cadmium under cadmium stress conditions. On the other hand, nano-silicon reduces the amount of malondialdehyde (MDA) and reactive oxygen species (H<sub>2</sub>O<sub>2</sub>) may be through the reduction of cadmium uptake and accumulation in the plant cells and tissues as well as increase in the efficiency of enzymatic and non-enzymatic antioxidant systems.

## Conclusions

The results of the present study showed that the accumulation of cadmium in the borage tissues under cadmium stress was reduced due to the application of nano-silicon. In other words, the use of nano-silicon improved the physiological characteristics of the borage plant by reducing the uptake and transfer of cadmium nitrate. This reduction in the amount of uptake and accumulation of cadmium nitrate in plant tissues is important for the health of agricultural products and human communities.

**Keywords:** Antioxidant enzymes, *Borago officinalis*, Heavy metals, Reactive oxygen species (ROS)

**Table 1. Analysis of variance of biochemical traits under cadmium stress and nano-silicon treatment one day after stress application**

S.O.V	df	H <sub>2</sub> O <sub>2</sub> <sup>†</sup>	MDA <sup>†</sup>	Proline	Protein	Polyphenol oxidase	Peroxidas	Catalase
Cadmium(C)	2	0.005**	117.863**	0.548**	0.362**	1.395**	0.004**	0.053**
Nanosilicon(N)	1	0.001**	36.652**	0.038**	0.564**	0.098**	0.004**	0.092**
C × N	2	0.001**	14.672**	0.042**	0.193**	0.039*	0.002**	0.034**
Error	12	3.26010 <sup>-5</sup>	0.560	0.003	0.004	0.006	4.20919 <sup>-5</sup>	0.001
C.V%	-	14.190	16.043	27.351	13.566	12.615	15.457	26.429

ns, \* and \*\* are non-significant, significant at the probability level of 0.05 and 0.01, respectively

<sup>†</sup> MDA: Malondialdehyde, H<sub>2</sub>O<sub>2</sub>: Hydrogen peroxide

**Table 2. Analysis of variance of biochemical traits and cadmium concentration under cadmium stress and nano-silicon treatment one week after stress**

S.O.V	df	Cadmium Concentration	Hydrogen peroxide	Malondialdehyde	Proline
Cadmium (C)	2	21.853**	0.001**	174.594**	2.010**
Nanosilicon (N)	1	2.494**	0.006**	93.321 <sup>ns</sup>	0.050*
C × N	2	6.677**	0.001**	5.709**	0.124**
Error	12	0.178	6.5901 <sup>-5</sup>	1.036	0.010
C.V%	-	17.37	17.004	19.616	28.245

**Table 2. Continued**

S.O.V	df	Protein	Polyphenol oxidase	Peroxidase	Catalase
Cadmium (C)	2	0.636**	1.955**	0.044**	0.089**
Nanosilicon (N)	1	0.226**	0.195**	0.041**	0.099**
C × N	2	0.259**	0.065**	0.035**	0.068**
Error	12	0.004	0.009	6.82710 <sup>-5</sup>	0.001
C.V%	-	13.171	16.555	16.232	20.553

ns, \* and \*\* are non-significant, significant at the probability level of 0.05 and 0.01, respectively

**Table 3. Analysis of variance of biochemical traits under cadmium stress and nano-silicon treatment two weeks after stress**

S.O.V	df	Hydrogen peroxide	Malondialdehyde	Proline	Protein	Polyphenol oxidase	Peroxidase	Catalase
Cadmium (C)	1	0.001**	143.59**	2.399**	0.428**	2.072**	0.0001**	0.004**
Nanosilicon (N)	1	0.0003*	1.062 <sup>ns</sup>	0.001 <sup>ns</sup>	0.82**	0.002 <sup>ns</sup>	7.484 <sup>-5**</sup>	8.071 <sup>-6ns</sup>
C × N	1	2.212 <sup>-5ns</sup>	8.653**	0.091**	0.003**	0.004 <sup>ns</sup>	0.0001**	0.0002**
Error	8	1.4511 <sup>-5</sup>	0.492	0.002	0.005	0.002	1.36910 <sup>-6</sup>	1.2361 <sup>-5</sup>
C.V%	-	16.985	15.850	28.079	11.686	15.211	17.858	23.688

ns, \* and \*\* are non-significant, significant at the probability level of 0.05 and 0.01, respectively

**Table 4. Average comparison of the biochemical traits of European borage related to the effect of nano silicon and cadmium in three time periods**

Treatment	Concentration	Time <sup>†</sup>	Hydrogen peroxide	Malondialdehyde	Proline	Protein	Polyphenol oxidase
			μ mol g <sup>-1</sup> F. Wt	μ mol g <sup>-1</sup> F. Wt	μ mol g <sup>-1</sup> F. Wt	mg g <sup>-1</sup>	U mg <sup>-1</sup> Protein
Control	0	24 h	0.0250 <sup>e</sup>	3.2030 <sup>c</sup>	0.0414 <sup>d</sup>	1.131 <sup>a</sup>	0.0672 <sup>d</sup>
Cadmium(C)	81 μM	24 h	0.0929 <sup>b</sup>	6.5467 <sup>c</sup>	0.3592 <sup>c</sup>	0.653 <sup>c</sup>	0.5680 <sup>c</sup>
Cadmium	243 μM	24 h	0.1036 <sup>a</sup>	14.828 <sup>a</sup>	0.7990 <sup>a</sup>	0.286 <sup>d</sup>	1.1844 <sup>a</sup>
Nanosilicon(N)	1.5 mM	24 h	0.0405 <sup>d</sup>	2.686 <sup>e</sup>	0.0636 <sup>d</sup>	1.128 <sup>a</sup>	0.0424 <sup>d</sup>
C + N	81 μM+1.5 mM	24 h	0.0643 <sup>c</sup>	4.9083 <sup>d</sup>	0.3456 <sup>c</sup>	1.004 <sup>b</sup>	0.4800 <sup>c</sup>
C + N	243 μM+1.5 mM	24 h	0.0714 <sup>c</sup>	8.42 <sup>b</sup>	0.5144 <sup>b</sup>	0.999 <sup>b</sup>	0.8535 <sup>b</sup>
Control	0	1 W	0.0286 <sup>c</sup>	3.957 <sup>d</sup>	0.0562 <sup>d</sup>	1.461 <sup>a</sup>	0.0733 <sup>d</sup>
Cadmium	81 μM	1 W	0.0643 <sup>b</sup>	11.682 <sup>b</sup>	1.0871 <sup>b</sup>	1.000 <sup>c</sup>	0.8602 <sup>b</sup>
Cadmium	243 μM	1 W	0.1179 <sup>a</sup>	16.368 <sup>a</sup>	1.4383 <sup>a</sup>	0.407 <sup>d</sup>	1.4159 <sup>a</sup>
Nanosilicon	1.5	1 W	0.0131 <sup>d</sup>	3.203 <sup>e</sup>	0.2483 <sup>c</sup>	1.248 <sup>b</sup>	0.0889 <sup>d</sup>
C + Nn	81 μM+1.5 mM	1 W	0.0226 <sup>cd</sup>	5.918 <sup>c</sup>	0.9604 <sup>b</sup>	1.272 <sup>b</sup>	0.6151 <sup>c</sup>
C + N	243 μM +1.5 mM	1 W	0.0619 <sup>b</sup>	10.773 <sup>b</sup>	1.0573 <sup>b</sup>	1.021 <sup>c</sup>	1.0208 <sup>b</sup>
Control	0	2 W	0.0254 <sup>bc</sup>	3.9633 <sup>c</sup>	0.0783 <sup>d</sup>	1.447 <sup>b</sup>	0.0759 <sup>b</sup>
Cadmium	81 μM	2 W	0.401 <sup>a</sup>	12.580 <sup>a</sup>	1.1463 <sup>a</sup>	1.037 <sup>d</sup>	0.9425 <sup>a</sup>
Nanosilicon	1.5	2 W	0.0222 <sup>c</sup>	5.067 <sup>c</sup>	0.2742 <sup>c</sup>	1.579 <sup>a</sup>	0.0887 <sup>b</sup>
C + N	81 μM+1.5 mM	2 W	0.0315 <sup>b</sup>	10.287 <sup>b</sup>	0.9948 <sup>b</sup>	1.234 <sup>c</sup>	0.8845 <sup>a</sup>

The means with common letters in each column are not significantly different based on Duncan's multiple range tests at the 5% probability level.

<sup>†</sup> In units of hour (h) or week (W)

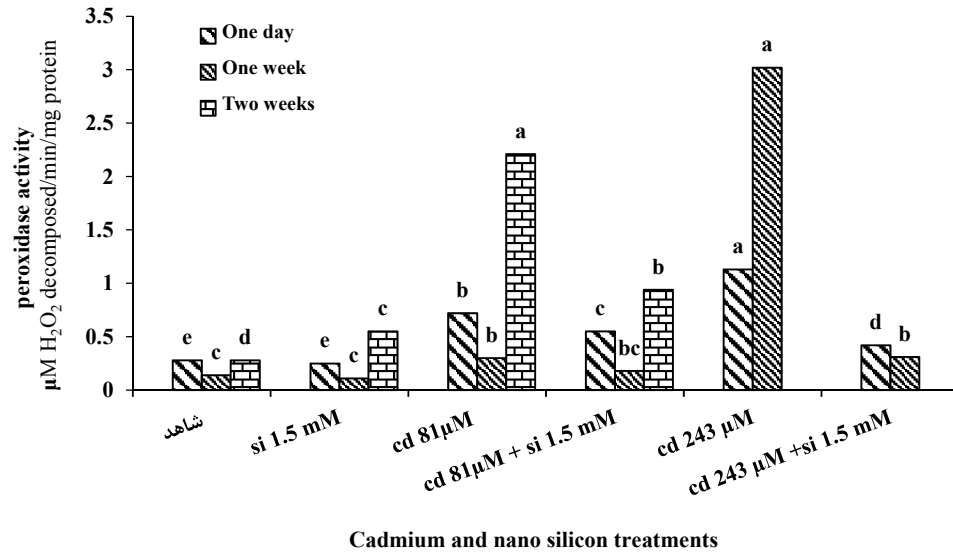


Fig. 1. Comparison of activity of peroxidase enzyme in European borage under the influence of nano-silicon and cadmium

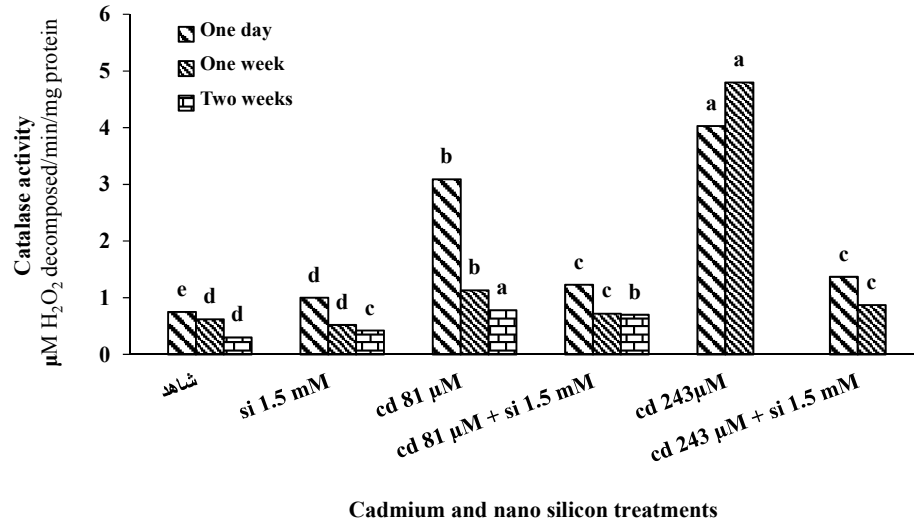
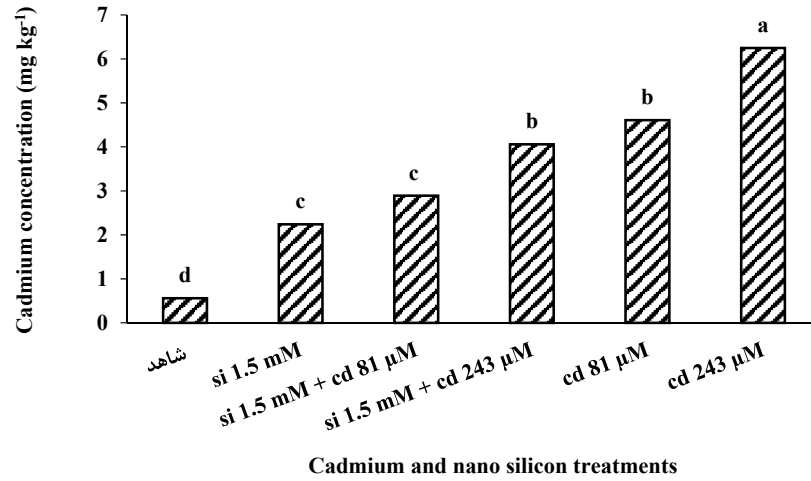


Fig. 2. Comparison of activity of catalase enzyme in European borage under the influence of nano silicon and cadmium.



**Fig. 3.** Comparison of the concentration mean of cadmium in borage leaves under the influence of nano-silicon and cadmium in one week after application of stress.