Effects of Different Selenium Concentrations on Eco-Physiology of *Lactuca sativa*

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Abstract. The Effects of six selenium concentrations (0, 5, 10, 25, 50, 75, and 100 mg/kg) on eco-physiology of *Lactuca sativa* were conducted by a pot experiment. The result showed that with the Se concentration increased, the biomass, photosynthetic pigment contents and antioxidant enzyme activities of *L. sativa* seedlings all increased first and then decreased, and reached maximum at 5 mg/kg. Compared with control, shoot fresh weight, SOD activity and POD activity showed significant difference. At 100 mg/kg of Se concentration, the biomass, photosynthetic pigment contents and antioxidant enzyme activities of *L. sativa* seedlings all reached minimum and except SOD activity all showed significant difference.

1. Introduction
Selenium deficiency has been connected with several human diseases [1]. Plants can transform inorganic selenium and then meet human need of selenium nutrition through food chain [2]. Under stress, appropriate dose of Se can regulate plants Reactive oxygen species accumulation, Photosynthetic metabolism [3]. Different ways of Se application have different influence on plants physiological property. Compared with foliar application of Se, root application is more favorable to the enrichment of Se by *Astragalus membranaceus* [4], whereas blueberry is opposite [5]. Therefore, in order to improve physiology and ecology index of *Lactuca sativa* seedlings, the effects of different Se concentrations on eco-physiology of *L. sativa* was studied.

2. Materials and methods

2.1 Materials collection
*L. sativa* seeds were purchased from Guangdong Konong vegetable seed Industry Company. Sodium Selenite purchased from Chengdu Kelong Chemical Reagent Factory. The soil was sandy loam soil and taken from farmland around Sichuan agricultural university (Chengdu campus) and Se was not detected.

2.2 Experiment design
The soil samples were air-dried, crushed, passed through 5mm mesh, later, weighted 3.0 kg and placed into each 21 cm × 20 cm (high × diameter) plastic pot. Pure sodium selenite solution was added to the soil by concentrations of 0, 5, 10, 25, 50, 75, and 100 mg/kg, respectively, and fully mixed with the
soil. Keep the soil moist and place it for 30 days. Seeds of *L. sativa* were sterilized for 10 min with 10% hydrogen peroxide solution, rinsed with ultrapure water, placed uniformly into a pallet for germination. Later, seedlings planted into a 50-hole tray. When unfolded 2-3 real-leaf, uniform growth seedlings were selected and transplanted into prepared pots. Three seedlings planted into per pot. Each treatment was repeated five times. After 40 days cultivated, correlative indexes are harvested and determined. The biomass was determined by conventional method. The content of Photosynthetic pigments was determined by acetone-ethanol mixed (1:1) extraction method. The Antioxidant enzyme activity of superoxide dismutase (SOD), peroxidase (POD) and catalase (CAT) was analyzed by Nitrogen Blue Tetrazolium (NBT) photoreduction method, guaiacol method and ultraviolet spectrophotometric method, respectively.

2.3 Statistical analyses
The test data were sort out by software of EXCEL 2010 and statistical analyses were conducted by statistical software of SPSS 20.0. The obtained data were analyzed by univariate analysis of variance (ANOVA) with \( p < 0.05 \) as the standard and Duncan method was used to test the significance of the difference.

3. Results and discussion

3.1 Effects of selenium on the biomass of *L. sativa* seedlings
With the Se concentration increased, fresh weight of root and shoot both show a trend of increasing first and then decreasing. Fresh weight of root and shoot reached maximum at Se concentration of 5 mg/kg, compared with control, increased by 1.84% \( (p > 0.05) \) and 34.41% \( (p < 0.05) \). Above 5 mg/kg Se concentration, fresh weight of root and shoot were lower than control, and reached minimum at 100 mg/kg, decreased by 83.76% \( (p < 0.05) \) and 90.19% \( (p < 0.05) \). For root/shoot ratio, the minimum was 0.12 (5 mg/kg).

Table 1. Effect of selenium on the biomass of *L. sativa* seedlings.

| Treatments (mg/kg) | Root fresh weight (g/plant) | Shoot fresh weight (g/plant) | root/shoot ratio |
|-------------------|-----------------------------|-----------------------------|-----------------|
| 0                 | 7.08±0.04a                  | 43.85±1.82b                 | 0.16            |
| 5                 | 7.21±0.70a                  | 58.94±1.53a                 | 0.12            |
| 10                | 5.55±0.24b                  | 43.00±2.50b                 | 0.13            |
| 25                | 4.90±0.45b                  | 15.48±1.52c                 | 0.32            |
| 50                | 3.36±0.28c                  | 9.42±0.91d                  | 0.36            |
| 75                | 1.84±0.17d                  | 5.27±0.50e                  | 0.35            |
| 100               | 1.15±0.11d                  | 4.30±0.30e                  | 0.27            |

Values are means ± standard errors. Means with the same letter within each column are not significantly different at \( p < 0.05 \).

3.2 Effects of selenium on the photosynthetic pigment contents of *L. sativa* seedlings
The contents of chlorophyll *a*, chlorophyll *b*, total chlorophyll and carotenoid increased first and then decreased with the increase of selenium concentration. When Se concentration was 5 mg/kg, the contents of chlorophyll *a*, chlorophyll *b*, total chlorophyll and carotenoid reached the maximum value, and increased by 11.63% \( (p > 0.05) \), 8.11% \( (p > 0.05) \), 10.83% \( (p > 0.05) \), 3.32% \( (p > 0.05) \) respectively, compared with the control. When Se concentration was 100 mg/kg, the contents of chlorophyll *a*, chlorophyll *b*, total chlorophyll and carotenoid reached the minimum value, and decreased by 65.10% \( (p < 0.05) \), 40.20% \( (p < 0.05) \), 59.40% \( (p < 0.05) \), 19.45% \( (p < 0.05) \) respectively, compared with the control.
Table 2. Effect of selenium on photosynthetic pigment contents of *L. sativa* seedlings.

| Treatment (mg/kg) | Chlorophyll *a* (mg/g) | Chlorophyll *b* (mg/g) | Total chlorophyll (mg/g) | Chlorophyll *a/b* | Carotenoids (mg/g) |
|------------------|------------------------|------------------------|-------------------------|-------------------|-------------------|
| 0                | 0.997±0.023a           | 0.296±0.011ab          | 1.293±0.035a            | 3.366             | 3.281±0.059ab     |
| 5                | 1.113±0.054a           | 0.320±0.031a           | 1.433±0.020a            | 3.507             | 3.390±0.069a     |
| 10               | 1.100±0.075a           | 0.315±0.030ab          | 1.415±0.107a            | 3.498             | 3.358±0.082ab    |
| 25               | 0.797±0.0787b          | 0.258±0.024abc         | 1.055±0.104b            | 3.108             | 3.103±0.175bc    |
| 50               | 0.566±0.011c           | 0.242±0.001bcd         | 0.808±0.013c            | 2.334             | 2.937±0.023cd    |
| 75               | 0.399±0.017d           | 0.198±0.016cd          | 0.597±0.033d            | 2.016             | 2.750±0.171de    |
| 100              | 0.348±0.033d           | 0.177±0.015d           | 0.525±0.051d            | 1.990             | 2.643±0.098e     |

Values are means ± standard errors. Means with the same letter within each column are not significantly different at *p* < 0.05.

3.3 Effects of selenium on antioxidant enzyme activities of *L. sativa* seedlings

The same as biomass and photosynthetic pigment content, with the increase of Se concentrations, antioxidant enzyme activities of *L. sativa* seedlings increased first and then decreased. When Se concentration of 5 mg/kg, the SOD, POD and CAT activity reached maximum and increased than control by 15.95% (*p* < 0.05), 18.01% (*p* < 0.05) and 8.43% (*p* > 0.05) respectively. Rather, at 100 mg/kg, the SOD, POD, CAT activity reached minimum and was lower than control by 4.29% (*p* > 0.05), 36.60% (*p* < 0.05) and 60.28% (*p* < 0.05) respectively.

Table 3. Effects of selenium on antioxidant enzyme activities of *L. sativa* seedlings.

| Treatment (mg/kg) | SOD activity (U/g) | POD activity (U/g/min) | CAT activity (U/g/min) |
|------------------|--------------------|------------------------|------------------------|
| 0                | 443.08±43.71b      | 540.53±14.71b          | 17.80±1.67a            |
| 5                | 513.74±31.59a      | 637.87±27.91a          | 30.0±1.09a             |
| 10               | 510.54±37.39a      | 530.40±28.00b          | 16.60±1.19ab           |
| 25               | 436.36±15.23b      | 514.14±21.87b          | 14.80±1.35ab           |
| 50               | 430.43±13.19b      | 472.80±46.30b          | 9.3±0.92ab             |
| 75               | 427.24±14.67b      | 395.20±24.14c          | 7.14±0.70b             |
| 100              | 424.05±17.89b      | 342.67±19.99c          | 7.07±0.19b             |

Values are means ± standard errors. Means with the same letter within each column are not significantly different at *p* < 0.05.

4.Conclusions

The low Se concentration (5 mg/kg) promotes *L. sativa* biomass, photosynthetic pigment contents and antioxidant enzyme activities increase, while high Se (>5 mg/kg) inhibits *L. sativa* growth and physiological and ecological response. When Se concentration was 100 mg/kg, the biomass, photosynthetic pigment contents and antioxidant enzyme activities of *L. sativa* all reached minimum and POD and CAT activities showed significant difference. Therefore, 5 mg/kg of Se concentration stimulating *L. sativa* resistance to stress, however, 100 mg/kg has toxic effect on *L. sativa*. 
Acknowledgement
This work was financially supported by the Application Infrastructure Project of Science and Technology Department of Sichuan Province (2016JY0258).

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