

Selenium Accumulation Characteristics of *Perilla frutescens*

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Abstract: Pot experiments were conducted to study the effects of different concentrations of selenium (Se) on growth and Se-rich characteristics of *Perilla frutescens*. The results showed that Se treatment had significant effect on growth and Se content. With Se concentrations increased, the biomass of *P. frutescens* significantly decreased. The root to shoot ratio increased firstly and then decreased, which reached the peak when Se concentration was 10 mg/kg. With Se concentrations increased, Se content increased and the difference between each treatment was extremely significant. In addition, the content of Se in leaf > root > stem. Translocation factor was on the rise as a whole, compared with control. Also, Se treatment had significant effect on the amount of Se accumulation in *P. frutescens*. With Se concentrations increased, the accumulation of Se showed different trends. However, as a whole, the accumulation of Se tends to increase. In addition, translocation accumulation factor showed a rising trend.

1. Introduction

Perilla frutescens is the traditional Asian crop, which has multiple uses, such as used for seed oil and for medicinal uses [1]. Selenium (Se) is an essential micronutrient for human health and plays an extremely important role in human body's function [2-3]. Se can not be synthesized in human body. Plants can realize the transformation from inorganic Se to organic Se by adding exogenous Se, which can obviously improve the content of Se in plants [4-5], which can satisfy the Se supplement in human body. Organic Se has higher absorption and utilization rate, and has little toxicity to human body [6]. Therefore, the application of sodium selenite in *P. frutescens* is an economical and feasible measure to obtain Se containing health herbs-*P. frutescens*. However, excessive intake of trace element Se by human body will have toxic effect, which is beneficial to higher plants at low concentration of Se, but high levels of elements will cause toxic effect, such as slow growth, curly leaves and even death [7]. So, how to use safer biological enrichment carriers and suitable Se concentration in the process of Se enrichment is very important. Therefore, in this experiment, the Se enrichment characteristics and growth of *P. frutescens* were studied by using different concentrations of Se.

2. Materials and methods

2.1 Materials

The experiments were conducted at Sichuan Agricultural University (30° 42' N, 103° 51' E), Wenjiang, Chengdu, Sichuan, China. *P. frutescens* seeds are purchased from the market. Se is purchased from Chengdu Kelong chemical reagent factory. All chemicals used in experiments were of analytical grade.



2.2 Experimental design

The soil samples were air-dried and passed through a 5-mm mesh in April 2017, and then 3.0 kg of soil was weighed into each polyethylene pot 18 cm × 26 cm (height × diameter). Na₂SeO₃ solution was added to the soil to divide the Se content into 0 (control), 5, 10, 25, 50, 75 and 100 mg/kg. The soils were mixed immediately and again after 4 weeks, during this period soil moisture was kept at 80%.

Seeds were sterilized in 10% Na₃PO₄ solution for 5 min, rinsed in distilled water five times, and were placed on 9-cm-diameter petri dishes with three layers of filter paper moistened with distilled water and germinated at 25 °C in the darkness. Seeds were considered germinated when the seed coat was broken and a radical was visible. After germination, seeds were planted in seedling tray filled with nursery substrate. When the third leaf expanded, uniform seedlings were transplanted into the previously prepared polyethylene pots, four plants seedlings per pot, each treatment repeated three times, randomly placed, and routinely administered.

After one month of plants growing, the plants were harvested and washed in tap water to remove soils from the root surface, and then rinsed in deionized water. Plants were separated into roots, stem, and leaves, killed out at 15 min at 105 °C and dried to constant weight at 70 °C and comminuted. The total Se concentration was determined by Hydride Generation Atomic Fluorescence Spectrometry (HG-AFS 9230) [8]. Calculate above ground part bioconcentration factor, translocation factor and translocation accumulation factor. The bioconcentration factor (BCF) = the Se content of plant above ground part / soil Se concentration, translocation Factor (TF) = the Se content of plant above ground part / root Se content, translocation accumulation factor (TAF) = Se accumulation in the aerial parts of plants / accumulation of Se in roots [9].

2.3 Statistical analyses

Statistical analysis was carried out by with SPSS 18.0 statistical software. The data were analyzed by one-way ANOVA, with the least significant difference at the 5% confidence level.

3. Results and discussion

3.1 Biomass of *P. frutescens*

As shown in Table 1, biomass of *P. frutescens* gradually decreased with increasing Se concentration. Compared with control, when Se concentrations were 5, 10, 25, 50, 75, 100 mg/kg, roots biomass decreased by 4.65%, 21.46%, 72.60%, 78.96%, 90.57% and 93.59%, and shoots biomass respectively decreased by 15.17%, 34.19%, 69.83%, 73.79%, 80.91% and 83.56%, respectively, compared with control. Except that the difference of stem biomass was not significant when Se concentration was 75, 100 mg/kg, the difference between the treatments was statistically significant ($P < 0.05$). Besides, compared with control, the root to shoot ratio increased firstly and then decreased with the increase of soil Se concentration, when Se concentrations were 5, 10 mg/kg, the root to shoot ratio increased and other Se concentrations treatments, the root to shoot ratio decreased.

Table 1. Biomass of *P. frutescens*.

(mg/kg) Treatments	Roots (g/plant)	Stems (g/plant)	Leaves (g/plant)	Shoots (g/plant)	Root/ shoot ratio
0	0.903±0.011a	0.615±0.008a	1.001±0.006a	1.615±0.014a	0.559
5	0.850±0.005b	0.470±0.001b	0.900±0.014b	1.370±0.012b	0.620
10	0.701±0.004c	0.287±0.008c	0.776±0.005c	1.063±0.003c	0.659
25	0.240±0.007d	0.112±0.004d	0.375±0.008d	0.487±0.012d	0.493

50	0.180±0.012e	0.093±0.012e	0.330±0.009e	0.423±0.004e	0.426
75	0.083±0.002f	0.060±0.009f	0.248±0.010f	0.308±0.019f	0.268
100	0.054±0.005g	0.053±0.006f	0.213±0.006g	0.265±0.012g	0.202

Note: Different letters indicated significant differences among treatments at 0.05 levels.

3.2 Se content in *P. frutescens*

Different concentrations of Se treatment had significant effect on Se content in *P. frutescens* (Table 2). Se content increased and the difference between each treatment was extremely significant. In addition, the distribution of Se content in plants: leaf > stem > root. Within the range of 0-100 mg/kg, TF increased firstly and then decreased, increased lastly with the increase of soil Se concentration. When Se concentration was 100 mg/kg, Translocation Factor reached the peak, which was 0.469. However, the trend was on the rise as a whole, compared with control.

3.3 Se accumulation amount of *P. frutescens*.

Se treatment had significant effect on Se accumulation amount of *P. frutescens* (Table 3). With Se concentrations increased, the accumulation of Se in the roots and stems showed a twists and turns change. With Se concentrations increased, the accumulation of Se in the roots increased firstly and then decreased and increased, increased lastly. When Se concentration was 50 mg/kg, the accumulation of Se in the roots reached the peak, which increased by 41.04 times compared with the control. The accumulation of Se in the stems increased firstly and then decreased and increased, increased lastly. However, the trend was on the rise as a whole, compared with control. The accumulation of Se in the leaves and shoots increased. In addition, TAF showed a rising trend.

Table 2. Se content in *P. frutescens*.

Treatments (mg/kg)	Roots (mg/kg)	Stems (mg/kg)	Leaves (mg/kg)	Shoots (mg/kg)	TF
0	3.78±0.14g	1.17±0.085g	1.02±0.057g	1.08±0.068g	0.286
5	75.65±1.39f	16.91±1.739f	28.88±2.942f	24.77±2.480f	0.327
10	145.02±8.56e	35.51±2.800e	53.62±5.119e	48.72±4.613e	0.336
25	405.42±15.09d	55.81±3.946d	135.46±7.722d	117.11±7.024d	0.289
50	795.71±21.75c	162.43±5.869c	245.69±10.875c	227.34±12.029c	0.286
75	850.94±22.53b	252.37±4.610b	353.17±11.554b	333.54±8.612b	0.392
100	938.70±25.19a	329.19±8.033a	467.18±11.710a	439.83±12.958a	0.469

Note: Different letters indicated significant differences among treatments at 0.05 levels.

Table 3. Se accumulation amount in *P. frutescens*.

(mg/kg) Treatments	Roots (µg/plant)	Stems (µg/plant)	Leaves (µg/plant)	Shoots (µg/plant)	TAF
0	3.41±0.170e	0.72±0.062d	1.02±0.063f	1.74±0.125e	0.510
5	64.28±0.776cd	7.95±0.840bc	26.00±2.252e	33.95±3.092d	0.528
10	101.59±6.512b	10.21±0.526b	41.59±4.244d	51.80±4.770c	0.510
25	97.36±6.577b	6.26±0.226c	50.80±1.756c	57.06±1.982c	0.586
50	143.37±13.118a	15.15±1.451a	81.08±5.683b	96.23±4.232b	0.671
75	70.39±3.940c	15.15±2.436a	87.71±6.540b	102.86±8.977b	1.461

100	50.41±6.264d	17.32±1.614a	99.44±0.249a	116.76±1.864a	2.316
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Note: Different letters indicated significant differences among treatments at 0.05 levels.

4. Conclusions

Se treatment had significant effect on growth and Se content. With Se concentrations increased, biomass of *P. frutescens* significantly decreased, which showed that high concentration of Se inhibited the growth of *P. frutescens*, and the inhibitory effect increased with increasing concentration, showing a dose effect. Compared with control, when Se concentrations were 5, 10 mg/kg, the root to shoot ratio increased, showed the growth of the aerial portion had inhibited stronger than the growth of the roots, the growth of *P. frutescens* was less inhibited. When Se concentrations were 25, 50, 75, 100 mg/kg, the root to shoot ratio decreased, the roots were strongly suppressed, because the roots first touched a high concentration of Se, and the inhibition was the most significant. With Se concentrations increased, Se content increased and the difference between each treatment was extremely significant. Within the range of 0-100 mg/kg, TF was on the rise as a whole, compared with control. With Se concentrations increased, the accumulation of Se showed a different trend. However, as a whole, the accumulation of Se tends to increase. In addition, TAF showed a rising trend. Therefore, the *P. frutescens* absorbed Se was a dosage effect, and the Se content of the *P. frutescens* increased as the Se concentration was increased.

In conclusion, high concentrations of Se stress were not conducive to the growth of *P. frutescens*. Only the concentration of 5 - 10 mg/kg of Se was in inhibiting root growth. Therefore, *P. frutescens* could grow normally and improve Se content and Se accumulation amount of *P. frutescens* under Se stress of 5-10 mg/kg.

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