Study on Cadmium Accumulation Characteristics of Grape Seedlings

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Abstract: The cadmium (Cd) accumulation characteristics of grape seedlings were investigated through a pot experiment. The results showed that with the increase of Cd concentrations, the root, stem, leaf and shoot biomasses of grape seedlings decreased, and the Cd contents in roots, stems, leaves and shoots of grape seedlings had the increase trend. The antioxidant enzyme (SOD and CAT) activity and soluble protein content of grape seedlings reduced when the dose of soil Cd was not more than 10 mg/kg, and improved when the dose of soil Cd was higher than 10 mg/kg. Therefore, Cd treatment could inhibit the growth of grape seedlings.

Introduction

With the improvement of people's living standards, the fruit has more and more proportion in the human diet [1]. Grape is one of the world's most important fruit tree species [2-3], and China's fresh grape planting area is the second of world for five consecutive years [4-5]. However, due to the long-term use of pesticide and fertilizer, the heavy metal accumulation in soil, and the heavy metal content in fruit excessive [6-7]. So, it is very important to study the Cd accumulation characteristics of grape. Under Cd stress, the Cd contents in different varieties of grape seedlings are significant different, and grafting could reduce the Cd content in fruits of grape [8]. There are few studies on the Cd accumulation characteristics of grape or other fruit trees [9-11]. The aim of this study was to study the Cd accumulation characteristics of grape seedlings, and provided a reference for grape production in Cd-contaminated soil areas.

Materials and Methods

Materials. The soil samples used in the experiment were collected from the Chengdu campus of Sichuan Agricultural University (30° 42′N, 103° 51′E) in May 2016, which were uncontaminated by heavy metals. The grape seedlings were 'Jufeng' and 'Xiahei', which were the annual cutting seedlings with 25 cm height of young sprout.

Experimental Design. The soil samples were air-dried and passed through a 5-mm sieve. Four kilograms of the air-dried soil was weighed into each polyethylene pot (18 cm high, 21 cm in diameter). Cd was added to soils as $CdCl_2 \cdot 2.5H_2O$ at 0, 1, 5, 10, 15 and 20 mg/kg in April 2016, and the soil moisture was maintained at 80% of field capacity for one month. The uniform grape seedlings were transplanted into each pot in May 2016, and watered every day to keep the soil moisture content maintaining at 80% of field capacity. One pot planted one seedling, and each treatment was 6 replicates, respectively. After grape seedlings grew two months (July 2016), the whole plants were harvested. Then, the roots, stems, and leaves were washed with tap water, further washed with deionized water three times, and then dried at 80°C to constant weight for dry weight determination. The plant samples were finely ground and sieved through a 0.149-mm mesh nylon sieve before digestion. The Cd

concentrations were determined using an iCAP 6300 ICP spectrometer (Thermo Scientific, Waltham, MA, USA) [12]. The measured Cd values were checked against certified standard reference material (GBW-07602, bush branches and leaves) obtained from the China National Center for Standard Reference Materials.

Statistical Analyses. Statistical analyses were conducted using SPSS 13.0 statistical software (IBM, Chicago, IL, USA). Data were analyzed by one-way analysis of variance with least significant difference (LSD) at the p = 0.05 confidence level.

Results and Discussion

Biomass of Grape Seedlings. Under Cd stress, the biomass of grape seedlings decreased, but there were no obvious toxic symptoms in any treatments during the observation period (Table 1). With the increase of soil Cd concentrations, the root, stem, leaf and shoot biomasses of grape seedlings had the decreasing trend. At 1, 5, 10, 15 and 20 mg/kg soil Cd treatments, the root biomass of 'Jufeng' seedlings decreased by 8.73% ($p > 0.05$), 9.61% ($p > 0.05$), 20.96% ($p < 0.05$), 29.69% ($p < 0.05$), and 44.54% ($p < 0.05$), respectively, compared with the control, and the shoot biomass decreased by 1.32% ($p > 0.05$), 7.02% ($p > 0.05$), 18.41% ($p < 0.05$), 32.98% ($p < 0.05$) and 52.45% ($p < 0.05$), respectively, compared with the control. For 'Xiahei' seedlings, the root biomass decreased by 1.07% ($p > 0.05$), 3.33% ($p > 0.05$), 6.78% ($p > 0.05$), 20.21% ($p < 0.05$), and 27.82% ($p < 0.05$), respectively, compared with the control, at 1, 5, 10, 15 and 20 mg/kg soil Cd treatments, and the shoot biomass decreased by 1.56% ($p > 0.05$), 5.63% ($p > 0.05$), 14.17% ($p < 0.05$), 24.42% ($p < 0.05$) and 37.99% ($p < 0.05$), respectively, compared with the control. So, Cd stress inhibited the growth of grape seedlings.

Table 1 Biomass of grape seedlings

Treatments	Roots (mg/plant)	Stems (mg/plant)	Leaves (mg/plant)	Shoots (mg/plant)
Jufeng				
0	2.29±0.16a	2.53±0.24a	5.02±0.33a	7.55±0.57a
1	2.09±0.17ab	2.49±0.21a	4.96±0.10a	7.45±0.31a
5	2.07±0.13ab	2.44±0.16a	4.58±0.17a	7.02±0.33ab
10	1.81±0.20bc	2.26±0.13ab	3.90±0.14b	6.16±0.27b
15	1.61±0.17cd	1.75±0.30b	3.31±0.18c	5.06±0.48c
20	1.27±0.18d	1.12±0.18c	2.47±0.11d	3.59±0.30d
Xiahei				
0	8.41±0.20a	9.93±0.24a	9.97±0.20a	19.90±0.44a
1	8.32±0.31a	9.78±0.17a	9.81±0.27a	19.59±0.45a
5	8.13±0.18a	9.32±0.45ab	9.46±0.76ab	18.78±0.31a
10	7.84±0.23a	8.46±0.48b	8.62±0.25bc	17.08±0.74b
15	6.71±0.27b	7.21±0.30c	7.83±0.18c	15.04±0.11c
20	6.07±0.34c	6.04±0.37d	6.30±0.28d	12.34±0.65d

Cd Content in Grape Seedlings. With the increase of soil Cd concentrations, the Cd contents in roots, stems, leaves and shoots of grape seedlings had the increase trend (Table 2). The order of Cd contents in roots, stems, leaves and shoots were ranked as roots > stems > shoots > leaves. So, the Cd was main accumulated in the roots of grape seedligs. At 1, 5, 10, 15 and 20 mg/kg soil Cd treatments, the Cd contents in roots of 'Jufeng' seedlings were 2.59, 24.63, 65.43, 97.78 and 128.74 times of the control, and the Cd contents in shoots were 1.21, 1.27, 1.35, 1.43 and 1.52 times of the control. For 'Xiahei' seedlings, the Cd contents in roots were 1.81, 26.77, 41.12, 52.38 and 79.34 times of the control, and the Cd contents in shoots were 1.70, 1.79, 1.91, 2.03 and 2.22 times of the control. So, different varieties of grape had different Cd accumulation characterics, and 'Jufeng' seedlings

accumulated more Cd in roots and shoots than that in ‘Xiahei’ seedlings. The roots of ‘Jufeng’ seedlings were more sensitive than ‘Xiahei’ seedlings to Cd stress.

Table 2 Cd content in grape seedlings

Treatments	Roots (mg/kg)	Stems (mg/kg)	Leaves (mg/kg)	Shoots (mg/kg)
Jufeng				
0	0.737±0.027e	0.169±0.008e	0.162±0.011d	0.165±0.010d
1	1.908±0.147e	0.209±0.007d	0.196±0.006c	0.200±0.006c
5	18.150±2.617d	0.214±0.008cd	0.206±0.008bc	0.209±0.008c
10	48.222±2.514c	0.228±0.002bc	0.220±0.016bc	0.223±0.011bc
15	72.062±4.155b	0.238±0.004ab	0.235±0.013ab	0.236±0.010ab
20	94.885±2.991a	0.247±0.007a	0.252±0.012a	0.250±0.010a
Xiahei				
0	0.485±0.022e	0.120±0.002e	0.114±0.004d	0.117±0.003e
1	0.879±0.172e	0.212±0.004d	0.186±0.005c	0.199±0.004d
5	12.985±1.152d	0.228±0.003c	0.192±0.008c	0.210±0.004cd
10	19.941±2.205c	0.236±0.006bc	0.210±0.015bc	0.223±0.011bc
15	25.403±2.259b	0.246±0.006b	0.231±0.013ab	0.238±0.010b
20	38.481±3.562a	0.270±0.010a	0.250±0.015a	0.260±0.013a

Antioxidant Enzyme Activity of Grape Seedlings. Different from biomass, with the increase of soil Cd concentrations, the antioxidant enzyme (SOD and CAT) activity reduced when the dose of soil Cd was not more than 10 mg/kg, and improved when the dose of soil Cd was higher than 10 mg/kg (Table 3). At 1, 5, 10, 15 and 20 mg/kg soil Cd treatments, the SOD activity of ‘Jufeng’ seedlings reduced by 30.19% ($p < 0.05$), 32.54% ($p < 0.05$), 50.47% ($p < 0.05$), 32.93% ($p < 0.05$), and 29.07% ($p < 0.05$), respectively, compared with the control. The CAT activity of ‘Jufeng’ seedlings reduced by 11.18% ($p > 0.05$), 18.64% ($p < 0.05$) and 33.83% ($p < 0.05$) at 1, 5 and 10 mg/kg soil Cd treatments, and improved by 9.08% ($p > 0.05$) and 56.96% ($p < 0.05$) at 15 and 20 mg/kg soil Cd treatments, respectively, compared with the control. For ‘Xiahei’ seedlings, the SOD activity reduced by 4.80% ($p < 0.05$), 6.05% ($p < 0.05$) and 6.75% ($p < 0.05$) at 1, 5 and 10 mg/kg soil Cd treatments, and improved by 1.72% ($p > 0.05$) and 7.82% ($p < 0.05$) at 15 and 20 mg/kg soil Cd treatments, respectively, compared with the control. The CAT activity of ‘Xiahei’ seedlings reduced by 12.67% ($p < 0.05$), 33.94% ($p < 0.05$), 39.17% ($p < 0.05$), 34.00% ($p < 0.05$) and 7.20% ($p < 0.05$) at 1, 5, 10, 15 and 20 mg/kg soil Cd treatments, respectively, compared with the control. The soluble protein content of grape seedlings had the same trend as the antioxidant enzyme activity (Table 3). The soluble protein content of ‘Jufeng’ seedlings increased by 5.86% ($p > 0.05$) and 21.38% ($p > 0.05$) at 1 and 20 mg/kg soil Cd treatments, and decreased by 18.40% ($p < 0.05$), 30.02% ($p < 0.05$) and 39.41% ($p < 0.05$) at 5, 10 and 15 mg/kg soil Cd treatments, respectively, compared with the control. For ‘Xiahei’ seedlings, The soluble protein content increased by 13.57% ($p < 0.05$), 10.26% ($p < 0.05$), 27.75% ($p < 0.05$) and 56.56% ($p < 0.05$) at 1, 5, 15 and 20 mg/kg soil Cd treatments, and decreased by 16.89% ($p < 0.05$) at 10 mg/kg soil Cd treatments, respectively, compared with the control. The SOD specific activity of ‘Jufeng’ seedlings was ranked as 15 mg/kg > 0 mg/kg > 5 mg/kg > 10 mg/kg > 1 mg/kg > 20 mg/kg, and the order of SOD specific activity of ‘Xiahei’ seedlings was 10 mg/kg > 0 mg/kg > 5 mg/kg > 1 mg/kg > 15 mg/kg > 20 mg/kg.

Conclusions

Under Cd stress, with the increase of Cd concentrations, the root, stem, leaf and shoot biomass of grape seedlings decreased, and the Cd contents in roots, stems, leaves and shoots of grape seedlings had the increase trend. The antioxidant enzyme (SOD and CAT) activity and soluble protein content of

grape seedlings reduced when the dose of soil Cd was not more than 10 mg/kg, and improved when the dose of soil Cd was higher than 10 mg/kg.

Table 3 Antioxidant enzyme activity of grape seedlings

Treatments	SOD activity (U/g)	CAT activity (U/g)	Soluble protein content (mg/g)	SOD specific activity (U/mg)
Jufeng				
0	249.88±5.67a	28.97±1.41bc	10.76±0.32b	23.22±1.21a
1	174.43±1.49b	25.73±0.41cd	11.39±0.29b	15.31±0.52bc
5	168.56±4.32b	23.57±1.18d	8.78±0.19c	19.20±0.07b
10	123.77±3.92c	19.17±1.17e	7.53±0.22d	16.44±1.02bc
15	167.59±3.32b	31.60±3.44b	6.52±0.98d	25.70±3.40a
20	177.25±3.42b	45.47±0.66a	13.06±0.31a	13.57±0.58c
Xiahei				
0	175.82±1.18b	33.62±1.57a	6.63±0.43d	26.52±1.91b
1	167.38±1.87c	29.36±0.50b	7.53±0.35c	22.23±1.27c
5	165.18±3.58c	22.21±0.41c	7.31±0.19c	22.60±0.13c
10	163.95±0.21c	20.45±0.46c	5.51±0.12e	29.75±0.61a
15	178.85±1.69b	22.19±1.32c	8.47±0.09b	21.12±0.02c
20	189.57±0.63a	31.20±0.60b	10.38±0.25a	18.26±0.39d

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