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Effects of Cadmium Stress on Photosynthesis of *Cyphomandra betacea* Seedlings

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Abstract: A pot experiment was conducted to study the effects of cadmium (Cd) stress on photosynthetic characteristics of *Cyphomandra betace* seedlings. The annual *C. betacea* seedlings were treated with Cd of different concentration gradients (0 - 20 mg/kg). The results showed that with the increase of soil Cd concentration, the chlorophyll *a*, chlorophyll *b*, total chlorophyll and carotenoid content of *C. betacea* decreased, the net photosynthetic rate, stomatal conductance, CO_2 concentration of intercellular, transpiration rate and light use efficiency decreased, the vapor pressure deficit of leaf and water use efficiency increased, compared with the control. Therefore, Cd stess could effect the photosynthesis of *C. betacea* seedlings significiently, and had a bad influnence on their growth.

Introduction

Cyphomandra betacea is a kind of fruit tree with commercial development value [1], it belongs to the Solanaceae family, native to Peru, South America and is now mostly cultivated in southwest China [2]. *C. betacea* can be both ornamental and edible, and is thought to has the potential to develop as an effective drug for cancer [3]. Previous studies have shown that cadmium (Cd) might strongly inhibit plant growth by reducing chlorophyll content, damaging plant cell structure, destroying physiological metabolic processes such as photosynthesis., and even led to plant death [4,5]. For Photosynthesis, Cd stress could lead to the increase of relative electrical conductivity and the injury of *Medicago sativa* leaves [6],could also reduce the photosynthetic capacity and fluorescent parameters of *Nicotiana tabacum* [7], and could effect the electron transfer in the process of photosynthesis, and might have a hormesis in *Lonicera japonica* Thunb[8]. But there are significant differences in the sensitivity and tolerance of different crops to Cd stress[9].Therefore, a pot experiment was conduct to study the photosynthetic Characteristics of *C. betacea* seedlings in soil with Cd of different concentration gradients (0 - 20 mg/kg). The objectives of this study were to provide guidance for the cultivation, application and safe production of *C. betacea*.

Materials and Methods

Materials. The seeds of *C. betacea* were collected from a perennial *C. betacea* of Chengdu Academy of Agriculture and Forestry(30° 42′ N, 103° 51′ E) in August, 2016, air-dried and stored at 4 °C respectively.

Experimental Design. The experiment was conducted at Chengdu Campus of Sichuan Agricultural University (30° 42′ N, 103° 51′ E) from May to August 2017. The soil samples were air-dried and passed through a 5-mm mesh in May 2017, and then 3.0 kg of soil was weighed into each polyethylene pot (15 cm tall, 18 cm diameter). Cd was added to make a final soil Cd concentrations of 0, 1, 5, 10, 15, 20 mg/kg with a saturated heavy metal solution in the form of $CdCl_2 \cdot 2.5H_2O$. The soils were mixed aperiodically during the next 4 weeks, and the soil moisture was kept at 80%. The seeds of *C. betacea* were sown in the farmland of the Chengdu Campus in June 2017. Two weeks later, three seedlings of each treatment were transplanted into each pot, and each treatment was repeated five times with a 10-cm spacing between pots. In order to reduce the

marginal effect, the position of the pots was completely randomly arranged and exchanged during the whole growth process. When *C. betacea* seedlings grow two month (August 2017) under Cd stress, the upper mature leaves of *C. betacea* were collected to determine the photosynthetic pigment (chlorophyll *a*, chlorophyll *b*, total chlorophyll and carotenoid) content [10]. The photosynthesis of each plant was determined by using LI-6400 portable photosynthesis meter (LI-COR Inc, USA). The photosynthetic parameters of the photosynthesis meter were manual controled, with CO_2 concentration 400 μmol/mol, temperature 25 ºC, light intensity 1000 $\mu mol/m^2/s^1$) . The determination of photosynthetic parameters were net photosynthetic rate (Pn), transpiration rate (Tr), vapor pressure (VpdL), stomatal conductance (Gs) and CO_2 concentration of intercellular (Ci), and each treatment was repeated three times. Water use efficiency (WUE) = net photosynthetic rate (Pn) / transpiration rate (Tr), Light use efficiency (LUE) = net photosynthetic rate (Pn) / light intensity [11].

Statistical Analyses. Statistical analyses were conducted using SPSS 17.0 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

Photosynthetic Pigment Content in *C. betacea* seedlings. Cd decreased the content of chlorophyll *a*, chlorophyll *b*, total chlorophyll and carotenoid in *C. betacea* seedlings (Table 1). The content of chlorophyll *a*, chlorophyll *b*, total chlorophyll and carotenoid were similar to the contorl when the soil Cd concentration was less than 5 mg/kg. But with the increase of soil Cd concentration, they decreased evidently. Under soil Cd concentration of 1, 5, 10, 15 and 20 mg/kg, the chlorophyll *a* content decreased by -2.84% ($P > 0.05$), 7.75% ($P > 0.05$), 16.03% ($P < 0.05$), 21.22% ($P < 0.05$) and 21.29% ($P < 0.05$) respectively, the chlorophyll *b* content decreased by -6.02% ($P > 0.05$), 4.87% ($P > 0.05$), 14.39% ($P < 0.05$), 23.19% ($P < 0.05$) and 16.08% ($P < 0.05$) respectively, the total chlorophyll content decreased by -2.910 ($P > 0.05$), 7.01% ($P > 0.05$), 15.59% ($P < 0.05$), 21.73% ($P < 0.05$) and 19.95% ($P < 0.05$) respectively, the carotenoid content decreased by -0.39 ($P > 0.05$), 6.60% ($P > 0.05$), 9.21% ($P < 0.05$), 20.66% ($P < 0.05$) and 15.91% ($P < 0.05$) respectively, compared with the control. Cd stress decreased the chlorophyll *a*/*b* of *C. betacea* seedlings.

Table 1 Photosynthetic pigment content in *C. betacea* seedlings

Cd concentration (mg/kg)	Chlorophyll *a* (mg/g)	Chlorophyll *b* (mg/g)	Total chlorophyll (mg/g)	Chlorophyll *a*/*b*	Carotenoid (mg/g)
0	5.279±0.098ab	1.828±0.047ab	7.107±0.145ab	2.887±0.021ab	1.031±0.020a
1	5.376±0.023a	1.938±0.010a	7.314±0.013a	2.775±0.026cd	1.035±0.010a
5	4.870±0.237bc	1.739±0.081b	6.609±0.318bc	2.801±0.006c	0.963±0.065ab
10	4.433±0.336cd	1.565±0.106c	5.999±0.442cd	2.831±0.024bc	0.936±0.029bc
15	4.159±0.123d	1.404±0.056d	5.563±0.179 d	2.964±0.031a	0.818±0.023d
20	4.155±0.225d	1.534±0.044cd	5.689±0.269 d	2.708±0.069d	0.867±0.022cd

Different lowercase letters indicate significant differences based on one-way analysis of variance in SPSS 17.0 followed by the least significant difference test ($P < 0.05$). 0 = Cd free , 1 = 1 mg/kg soil Cd concentration, 5 = 5 mg/kg soil Cd concentration, 10 = 10 mg/kg soil Cd concentration, 15 = 15 mg/kg soil Cd concentration, 20 = 20 mg/kg soil Cd concentration. The same as below.

Photosynthetic Characteristics of *C. betacea seedlings*. Cd stress had a significant effect on the photosynthesis of *C. betacea* seedlings (Table 2). Compared with the respective control, Cd stress decreased the Pn of *C. betacea* seedlings, and increased the Vpdl of that. With the increase of soil Cd concentration, the Gs, Ci and Tr increased when the of Cd concentration was less than 1 mg/kg, and decreased when the Cd concentration was more than 1 mg/kg. Under soil Cd concentration of 1

mg/kg, *C. betacea* seedlings decreased Pn and VpdL by 5.72% ($P < 0.05$) and 23.96% ($P < 0.05$), respectively, increased Gs, Ci and Tr by 65.99% ($P < 0.05$), 27.99% ($P < 0.05$) and 21.89% ($P < 0.05$). Under soil Cd concentration of 5, 10, 15 and 20 mg/kg, the Pn decreased by 33.78% ($P < 0.05$), 41.22% ($P < 0.05$), 38.16% ($P < 0.05$) and 49.20% ($P < 0.05$) respectively, the Gs dereeased by 54.89% ($P < 0.05$), 50.48% ($P < 0.05$), 36.28% ($P < 0.05$) and 47.97% ($P < 0.05$) respectively, the Ci hardly changed respectively, the Tr decreased by 58.81% ($P < 0.05$), 48.90% ($P < 0.05$), 53.29% ($P < 0.05$) and 47.65% ($P < 0.05$) respectively. the VpdL increased by 0.51% ($P > 0.05$), 7.51% ($P < 0.05$), 5.63% ($P < 0.05$) and 12.86% ($P < 0.05$) respectively. For WUE, Fig. 1 shows that Cd stress treatment increased the water use efficiency of *C. Betacea.* Compared with the control, the WUE was 29.87% ($P < 0.05$) lower under soil Cd concentration of 1 mg/kg, and increased by 60.59% ($P < 0.05$) and 32.84% ($P < 0.05$) under soil Cd concentration of 5 and 15 mg/kg respectively. The same as the Pn, Cd stress decreased the LUE of *C.betacea* seedlings (Fig. 2). The light use efficiency of *C.betacea* seedlings decreased by 5.33% ($P < 0.05$), 32% ($P < 0.05$), 41.33% ($P < 0.05$), 37.33% ($P < 0.05$), and 49.33% ($P < 0.05$) respectively under soil Cd concentration of 1, 5, 10, 15 and 20 mg/kg.

Table 2 Photosynthetic characteristics of *C. betacea* seedlings

Cd concentration (mg/kg)	Pn (μmol $CO_2/m^2/s$)	Gs (mol $H_2O/m^2/s$)	Ci (μmol CO_2/mol)	Tr (mmol $H_2O/m^2/s$)	VpdL (kPa)
0	7.52±0.41a	0.0838±0.0003b	241.59±20.00b	1.595±0.006b	1.757±0.129c
1	7.09±0.03b	0.1391±0.0008a	309.20±9.89a	2.152±0.159a	1.336±0.065d
5	4.98±0.01c	0.0378±0.0009d	174.12±6.21d	0.657±0.014d	1.766±0.003c
10	4.42±0.06d	0.0415±0.0005d	204.30±15.04c	0.815±0.008c	1.889±0.012b
15	4.65±0.05d	0.0534±0.0026c	243.46±22.24b	0.745±0.058cd	1.856±0.107b
20	3.82±0.06e	0.0436±0.0020cd	245.67±19.26b	0.835±0.075c	1.983±0.015a

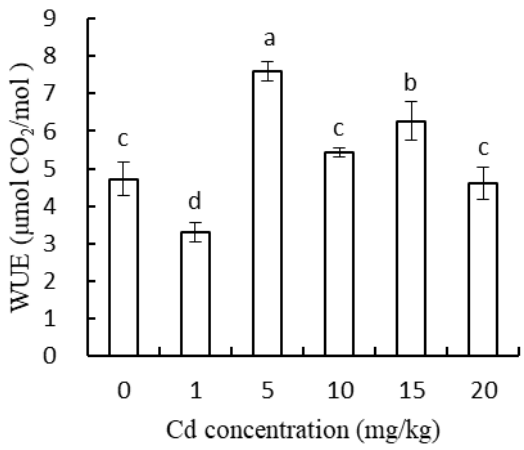

Fig. 1 WUE of *C. betacea* seedlings. Values are means of three replicate pots. Different lowercase letters indicate significant differences based on one-way analysis of variance in SPSS 17.0 followed by the least significant difference test ($P < 0.05$). 0 = Cd free , 1 = 1 mg/kg soil Cd concentration, 5 = 5 mg/kg soil Cd concentration, 10 = 10 mg/kg soil Cd concentration, 15 = 15 mg/kg soil Cd concentration, 20 = 20 mg/kg soil Cd concentration.

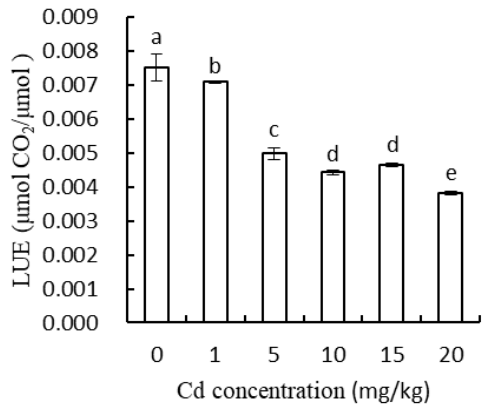

Fig. 2 LUE of *C. betacea* seedlings. Values are means of three replicate pots. Different lowercase letters indicate significant differences based on one-way analysis of variance in SPSS 17.0 followed by the least significant difference test ($P < 0.05$). 0 = Cd free , 1 = 1 mg/kg soil Cd concentration, 5 = 5 mg/kg soil Cd concentration, 10 = 10 mg/kg soil Cd concentration, 15 = 15 mg/kg soil Cd concentration, 20 = 20 mg/kg soil Cd concentration.

Conclusions

Under Cd stess, the chlorophyll *a*, chlorophyll *b*, total chlorophyll and carotenoid content in *C. betacea* decreased, the Pn, Gs, Ci, Tr and LUE decreased, the Vpdl and WUE increased compared with the control. The results showed that with the increase of soil Cd concentration, the photosynthesis of *C. betacea* seedlings increased when the concentration of Cd was less than 1 mg/kg and decreased when the concentration was more that. Therefore, Cd stess could effect the photosynthesis of *C. betacea* seedlings by decreasing the photosynthetic pigment content, the Pn, Gs, Ci, Tr and LUE.

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