

Youngia erythrocarpa, a newly discovered cadmium hyperaccumulator plant

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Abstract The farmland weed *Youngia erythrocarpa* has been found to have the basic characteristics of a cadmium (Cd) hyperaccumulator. This study carried out preliminary and further Cd concentration gradient experiments and field experiment using *Y. erythrocarpa* to confirm this fact. The results showed that the biomass and resistance coefficient of *Y. erythrocarpa* decreased, but the root/shoot ratio and the Cd content in roots and shoots increased with the increase in soil Cd concentration. The Cd content in shoots of *Y. erythrocarpa* exceeded 100 mg/kg when the soil Cd concentration was 25 mg/kg in the two concentration gradient experiments, up to the maxima of 293.25 and 317.87 mg/kg at 100 mg/kg soil Cd. Both the bioconcentration factor of the shoots and the translocation factor exceeded 1 in all Cd treatments. In the field experiment, the total Cd extraction by shoots was 0.934–0.996 mg/m² at soil Cd levels of 2.04–2.89 mg/kg. Therefore, *Y. erythrocarpa* is a Cd hyperaccumulator that could be used to remediate Cd-contaminated farmland soil efficiently.

Keywords *Youngia erythrocarpa* · Cadmium · Resistance · Bioconcentration · Hyperaccumulator · Translocation

Introduction

The development of industry and agriculture has increased heavy metal contamination of soil and has severely damaged soil quality (Yang 2007; Nriagu and Pacyna 1988). Heavy metals can enter the human body through the food chain via edible crops, and this causes a potential threat to human health and raises a series of food quality and safety issues (Zhao and Lu 2010; Peralta-Videa et al. 2009). According to statistics, the heavy metal pollution of grain reaches 12,000,000 t every year in China (Wang 2005), and the “cadmium rice grain event” in China was the result of cadmium (Cd)-contaminated soil (Chang 2013). Cadmium enters soil through four main pathways: direct discharge of industrial waste without treatment, the discharge of solid waste (e.g., sludge, garbage) directly into soil, the misuse of heavy metal-containing pesticides and fertilizers, and atmospheric deposition (Zhang et al. 2010b). The problem of grain production safety needs to be solved urgently in Cd-contaminated soil because of an increase in urbanization, which simultaneously increases the population requiring grain and decreases the land available for grain production.

The main methods of heavy-metal-contaminated soil remediation are the physical method, chemical method, and bioremediation method (Xia and Chen 1997).

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Phytoremediation is a bioremediation method that is generally considered to be of low cost, non-destructive to soil structure, and have no secondary pollution. It is receiving much attention in heavy metal contamination remediation research (McGrath et al. 2002). Phytoremediation is a technology where a screened hyperaccumulator is planted on heavy-metal-contaminated soil; the plant roots then take up the metal and it is accumulated in the plant. Harvesting the plant then eliminates the heavy metal from the soil. Hundreds of hyperaccumulators have been found (Maestri et al. 2010), but most of them grow slowly and have a low shoot biomass and weak adaptive ability to farmland soil (Wu et al. 2004), especially those from mine areas (Reeves and Baker 2000), which have a very limited application to farmland soil. Therefore, screening new hyperaccumulators with good adaptation to the farmland environment, fast growth, and a strong reproductive capacity is becoming urgent. At present, the screened Cd hyperaccumulators for Cd-contaminated soil remediation cannot be applied to different regions or in different soil types; the available resources need further screening. Farmland weeds generally have a large biomass, fast growth, and strong resistance (Zhao 2004; Wei et al. 2005a); therefore, screening Cd hyperaccumulators from farmland weeds not only overcomes the problems of low biomass, slow growth, and a narrow growing area, but also avoids the potential problems of introduction to the farmland environment.

Youngia erythrocarpa is a farmland weed with a large biomass, fast growth, strong reproductive capacity, and wide distribution. A previous study showed that it had a strong capability for Cd accumulation and had the basic characteristics of a Cd hyperaccumulator. Therefore, this experiment was conducted to further identify the Cd hyperaccumulation characteristics of *Y. erythrocarpa* to discover a new Cd hyperaccumulator and provide new phytoremediation material for Cd-contaminated farmland soil.

Materials and methods

Y. erythrocarpa screening experiment

Y. erythrocarpa had been found to grow well in soil polluted with 60 mg/kg Cd in March 2012. The roots and shoots of these plants were harvested at the blooming stage and washed with tap water and then deionized

water. The roots and shoots were then dried at 110 °C for 15 min to de-enzyme, oven dried at 75 °C to constant weight, and sieved to 0.149 mm. Samples of 0.5 g were then digested in HNO₃/HClO₄ (4:1 v/v) until transparent and filtered into a volumetric flask to 50 mL by deionized water. The Cd content was determined by an iCAP 6300 ICP spectrometer (Thermo Scientific, USA). The bioconcentration factor (BCF) was calculated as the Cd content in shoot (root)/Cd concentration in soil (Zhang et al. 2011), and the transfer factor (TF) was calculated as the Cd content in shoot/Cd content in roots (Rastmanesh et al. 2010). The Cd contents in roots and shoots of *Y. erythrocarpa* reached 151.52 and 188.64 mg/kg, respectively, and the BCFs of roots and shoots reached 2.52 and 3.14, respectively, and the IF reached 1.24. These results indicated that *Y. erythrocarpa* might be a Cd hyperaccumulator (Brooks et al. 1977; Brooks 1998).

Preliminary concentration gradient experiment

The preliminary concentration gradient experiment was conducted at the Ya'an Campus Farm of Sichuan Agricultural University (29° 59' N, 102° 59' E) from September to December 2012, using the inceptisol soil (the purple soil in the Genetic Soil Classification of China) samples from the same farm. The basic properties of the soil were as follows: pH, 7.02; organic matter, 41.38 g/kg; total nitrogen (N), 3.05 g/kg; total phosphorus (P), 0.31 g/kg; total potassium (K), 15.22 g/kg; alkali solution N, 165.30 mg/kg; available P, 5.87 mg/kg; and available K, 187.03 mg/kg. The total Cd content was 0.101 mg/kg, and the available Cd content was 0.021 mg/kg. The physical and chemical properties of soil and its heavy metal content were determined according to Bao (2000).

The soil samples were air dried and sieved to 5 mm in September 2012, then 3.0 kg of the air-dried soil was weighed into polyethylene pots (15 cm tall, 18 cm diameter), and Cd was added to the soils as CdCl₂·2.5H₂O at 0, 25, 50, 75, and 100 mg/kg. The pots were soaked in the Cd solutions for 4 weeks, and then the soil in each pot was mixed. Plant samples were collected from the Ya'an Campus Farm of Sichuan Agricultural University. Five uniform seedlings with two euphyllas were transplanted into each pot in October 2012. Each treatment was repeated three times, making a total of 15 pots, and put in a completely randomized design with 10-cm spacing between pots. The soil moisture content was

maintained at 80 % of field capacity until the plants were harvested.

After 60 days of cultivation, all plants were dug up and processed as described above. The biomasses of roots and leaves were measured, and their Cd contents were determined. The BCF and TF were calculated, and the root/shoot ratio was calculated as the root biomass/shoot biomass (Lukačová Kuliková and Lux 2010), the resistance coefficient was calculated as the total biomass in the experimental group/the total biomass in the control group (Zhao et al. 2012), and the amount of extracted Cd was calculated as the Cd content in plant×the biomass of plant (Zhang et al. 2010a).

Further concentration gradient experiment

The further concentration gradient experiment was conducted in the same place as the preliminary concentration gradient experiment from January to April 2013. The soil was the same as in the screening experiment. Dried soil (12.0 kg) was put into 20-cm-tall, 27-cm-diameter plastic pots, and Cd was added to the soils as CdCl₂·2.5H₂O at 0, 25, 50, 75, and 100 mg/kg. The pots were soaked in the Cd solutions for 4 weeks, and then the soil in each pot was mixed. Seedlings were transplanted into each pot in February 2013, and each treatment was repeated three times. The source of plant seedlings and their cultivation and management were as described in the preliminary concentration gradient experiment.

After 60 days of cultivation, all plants were dug up and processed as described above. The biomasses of roots and leaves were measured and their Cd contents were determined. The BCF, TF, root/shoot ratio, resistance coefficient, and amount of Cd extracted were calculated.

Field experiment

The field experiment was conducted at the Ya'an Campus Farm of Sichuan Agricultural University from October 2013 to February 2014. The inceptisol soil samples came from the Cd-contaminated soils of the earlier experiment on the Ya'an Campus Farm (Cd-contaminated area). The basic properties of the soil were as follows (Lin et al. 2014): pH, 6.98; organic matter, 35.01 g/kg; total N, 1.19 g/kg; total P, 0.63 g/kg; total K, 20.64 g/kg; alkali solution N, 80.63 mg/kg; available P, 31.78 mg/kg; and available K, 115.97 mg/kg. The

plots were divided into three treatments based on their distances from an irrigation canal—near (I), middle (II), and far (III)—and the distances from the canal were 1, 2, and 3 m, respectively. The total Cd concentrations in the soils of the three treatments were 2.89, 2.55, and 2.04 mg/kg, respectively. Each plot was 1.0 m² (1.0×1.0 m), and seedlings of *Y. erythrocarpa* were planted directly in the soil at a density of 100 plants/m² (in a 10×10 cm grid) in October 2013. Each treatment was repeated three times (three plots). The source of *Y. erythrocarpa* seedlings and their cultivation and management were as in the pot experiment. After *Y. erythrocarpa* had matured (after 120 days), the shoots were harvested to determine biomasses and Cd content as in the pot experiment.

Results and discussion

Biomass of *Y. erythrocarpa*

Cadmium is a non-essential plant element, and high Cd contents are toxic to plants (Zhang et al. 2010b). The biomass and the resistance coefficient of Cd hyperaccumulators decreased with an increase in soil Cd concentration, suggesting that Cd hyperaccumulators could be harmed by high concentrations of Cd as can common plants (Wei et al. 2005b; Zhang et al. 2013). In the preliminary concentration gradient experiment, because of the high soil Cd concentration, the roots, shoots, and total biomasses of *Y. erythrocarpa* decreased with increasing Cd concentration in soil ($P<0.05$, Table 1), but there were no obvious toxic symptoms in any treatments during the observation period. These results were the same as other Cd hyperaccumulators (Wei et al. 2013; Adki et al. 2013) and was found to be because the root system of the hyperaccumulator was damaged by heavy metal stress, which decreased the uptake of water and nutrients, eventually leading to biomass decrease (Šottníková et al. 2003; Zhou and Qiu 2005). The total biomass of *Y. erythrocarpa* decreased by 9.51, 28.89, 54.66, and 81.84 % at 25, 50, 75, and 100 mg/kg Cd in soil, respectively, compared with the control in the preliminary concentration gradient experiment. The resistance coefficient of *Y. erythrocarpa* also decreased with the increase in soil Cd concentration, suggesting the growth of *Y. erythrocarpa* was inhibited by a high concentration of Cd. The root/shoot ratio increased with increasing Cd concentration in soil; *Y. erythrocarpa*

Table 1 Biomass of *Y. erythrocarpa* in the two concentration gradient experiments

Treatments (mg/kg)	Roots (g/pot)	Shoot (g/pot)	Total biomass (g/pot)	Resistance coefficient	Root/shoot ratio
Preliminary concentration gradient experiment					
0	0.504±0.007e	2.535±0.085e	3.039±0.092e	1.000	0.199
25	0.472±0.011d	2.278±0.122d	2.750±0.133d	0.905	0.207
50	0.372±0.006c	1.789±0.139c	2.161±0.134c	0.711	0.208
75	0.248±0.005b	1.130±0.120b	1.378±0.115b	0.453	0.219
100	0.142±0.008a	0.410±0.048a	0.552±0.056a	0.182	0.346
Further concentration gradient experiment					
0	0.614±0.009e	3.135±0.076e	3.749±0.085e	1.000	0.196
25	0.528±0.007d	2.665±0.096d	3.193±0.088d	0.852	0.198
50	0.443±0.008c	2.165±0.110c	2.608±0.102c	0.696	0.205
75	0.280±0.042b	1.328±0.010b	1.608±0.052b	0.429	0.211
100	0.150±0.009a	0.645±0.020a	0.795±0.011a	0.212	0.233

appeared to enhance its resistance by increasing the root/shoot ratio (Table 1). The biomass and resistance coefficient decreased while the root/shoot ratio increased with increasing Cd concentration in the further concentration gradient experiment, which provided further reference of resistance and tolerance to *Y. erythrocarpa*. The total biomass of *Y. erythrocarpa* decreased by 14.83, 30.43, 57.11, and 78.79 % at 25, 50, 75, and 100 mg/kg Cd in soil, respectively, compared with the control in the further concentration gradient experiment (Table 1).

Plants were cultured in soil containing up to 100 mg/kg Cd for 60 days in two pot experiments.

Table 2 Cadmium accumulation characteristics of *Y. erythrocarpa*

Treatments (mg/kg)	Root (mg/kg)	Shoot (mg/kg)	BCF of root	BCF of shoot	TF
Preliminary concentration gradient experiment					
0	5.21±0.21e	0.99±0.24e	–	–	0.19
25	59.90±3.90d	119.37±1.93d	2.40	4.77	1.99
50	123.87±5.17c	157.76±7.96c	2.48	3.16	1.27
75	189.27±3.23b	234.54±11.04b	2.52	3.13	1.24
100	255.99±6.51a	293.25±9.35a	2.56	2.93	1.15
Further concentration gradient experiment					
0	11.88±0.47e	1.59±0.09e	–	–	0.13
25	65.51±2.94d	121.90±11.30d	2.62	4.88	1.86
50	135.30±7.90c	178.93±6.47c	2.71	3.58	1.32
75	215.75±4.55b	261.36±11.26b	2.88	3.48	1.21
100	292.31±6.91a	317.87±16.37a	2.92	3.18	1.09

Values are means (±SE) of three replicate pots. Different lowercase letters within a column indicate significant differences ($P<0.05$). The resistance coefficient was calculated as the total biomass in the experimental group/the total biomass in the control group.

Cadmium accumulation characteristics of *Y. erythrocarpa*

According to the identification of hyperaccumulators by Brooks (Brooks et al. 1977; Brooks 1998), plants that can grow well in Cd-contaminated soil and the Cd content in shoots reached the critical value (100 mg/kg), and the BCF and the TF were >1 at the same time, are Cd hyperaccumulators. In the preliminary concentration gradient experiment, *Y. erythrocarpa* plants grew normally and the Cd content in shoots reached 119.37 mg/kg at the dose of 25 mg/kg soil Cd, while the BCF was 4.77 and the TF was 1.99 (Table 2). All of these match the basic characteristics of a Cd hyperaccumulator. Cadmium contents in roots and shoots of *Y. erythrocarpa* and the BCF of roots (2.40–2.56) increased with the increase in soil Cd concentration, while the BCF of shoots (4.77–2.93) and the TF (1.99–1.15) decreased (Table 2). This experiment found that the ratio of root Cd content in *Y. erythrocarpa* increased as the soil Cd concentration increased, which weakened the toxicity of the rapidly increased shoot Cd content.

Plants were cultured in soil containing up to 100 mg/kg Cd for 60 days in two pot experiments. Values are means (±SE) of three replicate pots. Different

Table 3 Extraction amount of cadmium of *Y. erythrocarpa*

Treatments (mg/kg)	Preliminary concentration gradient experiment			Further concentration gradient experiment		
	Cd extraction of root (µg/pot)	Cd extraction of shoot (µg/pot)	Amount extraction of Cd (µg/pot)	Cd extraction of root (µg/pot)	Cd extraction of shoot (µg/pot)	Amount extraction of Cd (µg/pot)
0	2.63±0.07d	2.51±0.69c	5.14±0.63c	7.29±0.39d	4.98±0.40a	12.27±0.79e
25	28.27±2.50c	271.92±18.96a	300.19±21.46a	34.59±2.07c	324.86±18.42b	359.45±20.49d
50	46.08±1.18a	282.23±36.17a	328.31±37.35a	59.94±4.58a	387.38±5.69a	447.32±1.16c
75	46.94±1.74a	265.03±40.62a	311.97±38.88a	60.41±7.79a	347.09±17.57b	407.50±25.36b
100	36.35±1.12b	120.23±10.25b	156.58±11.36b	43.85±1.59b	205.03±16.92c	248.88±15.32a

lowercase letters within a column represent significant differences ($P < 0.05$). The bioconcentration factor (BCF) is defined as Cd content in shoot or root/Cd concentration in soil. The translocation factor (TF) is defined as Cd content in shoot/Cd content in roots.

In the further concentration gradient experiment, the shoot Cd content in *Y. erythrocarpa* also reached the critical value of a Cd hyperaccumulator at the same dose of Cd in soil, and the BCF of shoots was 4.77 while the TF was 1.99. The study proved that *Y. erythrocarpa* is a hyperaccumulator. With increasing soil Cd concentration, the Cd contents in shoots and roots of *Y. erythrocarpa* and the BCF (2.62–2.92) of roots increased while the BCF of shoots (4.88–3.18) and TF (1.86–1.09) decreased. The shoot Cd content of *Y. erythrocarpa* reached 317.87 mg/kg at 100 mg/kg soil Cd content (Table 2), which was close to the shoot Cd content of *Solanum photeinocarpum* (Zhang et al. 2011).

Amount extraction of cadmium of *Y. erythrocarpa*

The efficiency of hyperaccumulators for phytoremediation is related to their heavy metal extraction amount on contaminated soil. If the extraction amount is bigger, the phytoremediation effect is better. In this study, both the preliminary concentration gradient and the further

concentration gradient experiments showed that the Cd extraction amount of *Y. erythrocarpa* was maximum at 50 mg/kg Cd in soil, which reached 328.31 and 447.32 µg/pot, respectively, and the Cd extraction amount at 75 mg/kg Cd in soil was second (Table 3). Therefore, *Y. erythrocarpa* would perform better at phytoremediation below the dose of 75 mg/kg Cd in soil, especially 0–75 mg/kg Cd stress.

Plants were cultured in soil containing up to 100 mg/kg Cd for 60 days in two pot experiments. Values are means (±SE) of three replicate pots. Different lowercase letters within a column represent significant differences ($P < 0.05$). The amount of extracted Cd is defined as the Cd content in plant × plant biomass.

Field experiment

The biomass of shoots after 60 days was 223.5–228.9 g/m², and the Cd content in shoots was 4.11–4.35 mg/kg (Table 4). The total extraction of Cd in shoots was 0.934–0.996 mg/m², indicating that *Y. erythrocarpa* had a very good remedial effect at low concentrations of Cd contamination. Additionally, the plant used for phytoremediation should have the characteristics of fast growth, a deep rooting system, and easy propagation (Ghosh and Singh 2005). *Y. erythrocarpa* is an annual herb that grows to heights of 15–35 cm and lives on

Table 4 Biomass and Cd accumulation of *Y. erythrocarpa* in the field experiment

Treatment	Cd concentration in soil (mg/kg)	Biomass of shoot (g/m ²)	Cd content in shoot (mg/kg)	Total extraction of Cd in shoot (mg/m ²)
I	2.89	228.9±4.14a	4.35±0.02a	0.996±0.03a
II	2.55	223.5±1.52a	4.24±0.07ab	0.948±0.08b
III	2.04	227.2±2.81a	4.11±0.08b	0.934±0.04b

grass slopes, ditches, and plain wasteland between 460- and 1850-m elevations. It is widely distributed in China (Shi 1997). At the same time, *Y. erythrocarpa* is a weed species with a deep root system, rapid growth, and strong adaptation; it can grow almost all year round. Therefore, *Y. erythrocarpa*, as a Cd hyperaccumulator, can effectively remediate Cd-contaminated farmland soil.

Plants were cultured in soil containing 2.04–2.89 mg/kg Cd for 60 days in a field experiment. Values are means (\pm SE) of three replicate plots. The plots were divided into three treatments based on their distances from an irrigation canal: near (I), middle (II), and far (III). Different lowercase letters in a column represent significant differences ($P < 0.05$).

Conclusions

Y. erythrocarpa is a weed with a wide distribution, rapid growth, and strong adaptation. It has a strong tolerance to Cd stress and is a Cd hyperaccumulator that will effectively remediate Cd-contaminated farmland soil. In future work, investigations will focus on Cd uptake and tolerance mechanisms of *Y. erythrocarpa*.

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