



**Full Length Article**

## Evaluation of Phytoremediation Potential of Barbados Nut Grown in Cadmium-Contaminated Soil under Different Irrigation Regimes

Jie Li<sup>1,2</sup>, Qiliang Yang<sup>2\*</sup>, Zhengtao Shi<sup>1</sup>, Xiaogang Liu<sup>2</sup> and Zhennan Zang<sup>2</sup>

<sup>1</sup>School of Tourism and Geography sciences, Yunnan Normal University, Kunming 650500, China

<sup>2</sup>Faculty of Agricultural and Food, Kunming University of Science and Technology, Kunming 650500, China

\*For correspondence: 929631470@qq.com

Received 31 August 2019; Accepted 11 December 2019; Published 13 February 2020

### Abstract

Cadmium (Cd) is a widespread and toxic heavy metal that is a threat to plant, animal and human health. Phytoremediation, the use of tolerant species for the remediation of polluted areas, is a green technology that can be used for environmental pollution control. This pot study was conducted to evaluate the effect of soil Cd on growth, irrigation-water use efficiency, and Cd uptake, translocation and accumulation in Barbados nut under different irrigation regimes. Barbados nut was grown at low (50 mg kg<sup>-1</sup>), medium (100 mg kg<sup>-1</sup>) and high (200 mg kg<sup>-1</sup>) Cd levels as well as with no Cd under four irrigation regimes: 30, 50, 70 and 90% of soil water-holding capacity (SWC). Moreover 50% soil saturation percentage was considered as 100% SWC. The results revealed that the growth and dry matter accumulation of Barbados nut decreased with increasing soil Cd levels; however, this decrease declined with increasing irrigation amounts. In addition, Cd accumulation in different tissues was found to be in the order of root > stem > leaf, and excess soil Cd supply might inhibit Cd transportation from root to canopy. The higher dry matter accumulation, Cd accumulation, bioconcentration factor and irrigation-water use efficiency were observed at 50~70% SWC and low Cd levels (50 mg kg<sup>-1</sup>). Moreover, Barbados nut survived under severe drought (30% SWC) and the highest soil Cd concentrations (200 mg kg<sup>-1</sup>), exhibiting a strong capacity to resist drought and Cd stress. In conclusion, Barbados nut plants can be used for the phytoremediation of Cd-polluted soils in arid regions © 2020 Friends Science Publishers

**Keywords:** Barbados nut; Bioconcentration factor; Cd contents; Irrigation water-use efficiency; Phytoremediation

### Introduction

Heavy metals contamination is the leading worldwide environmental threat which seriously affects humans and animals health, and normal growth and yield of plants (Agbogidi *et al.* 2014; Bolan *et al.* 2014; Zahoor *et al.* 2018). Phytoremediation technology is a type of in situ technology that is considered the most effective method for the remediation of heavy metal-contaminated soils (Rascio and Navari-izzo, 2011). As most plants do not satisfy the restoration requirements for heavy metal contaminated soil; researchers worldwide are searching for new suitable plant species to be used in phytoremediation. When selecting a species for phytoremediation, there are several factors that must be taken into account. For example, the species should have rapid growth, high biomass production capacity, a profuse root system, tolerance to adverse environmental conditions, and a strong ability to tolerate and accumulate pollutants and should be inedible, economically beneficial and easy to harvest (Alkorta *et al.* 2004).

Barbados nut (*Jatropha curcas* L.), which belongs to the Euphorbiaceae family, is a deciduous perennial stem-succulent shrub tree species. It is widely distributed in

tropical and subtropical Asia, Africa, and America (Pandey *et al.* 2012). Many previous studies have demonstrated that Barbados nut has obvious advantages for remediating soils contaminated with heavy metals (Du *et al.* 2011; Liang *et al.* 2011; Agbogidi *et al.* 2014; Chang *et al.* 2014). In recent years, some studies have shown that the root system of Barbados nut can immobilize heavy metal pollutants, reduce pollution in the surrounding environment *via* the migration of pollutants, and beautify the environment and generate higher economic benefits (Tordo *et al.* 2000; Wong 2003). Other studies have shown that their roots, stems or leaves have the ability to effectively remove heavy metals such as zinc (Zn), lead (Pb), chromium (Cr), Cd and copper (Cu). The highest levels of Zn (29.5 mg kg<sup>-1</sup>), Cu (0.44 mg kg<sup>-1</sup>) and Cd (8.35 mg kg<sup>-1</sup>) accumulation were found in roots, whereas the highest Pb (4.63 mg kg<sup>-1</sup>) and Cr (0.33 mg kg<sup>-1</sup>) concentrations were observed in leaves and stems, respectively (Ahmadpour *et al.* 2010). The existing studies indicated that the remediation effect of Barbados nut on soil contaminated with heavy metals could be effectively improved through soil conditioning and fertilization (Du *et al.* 2011); but the method of adjusting soil water content to remediate soil contaminated with various heavy metals has

not yet been studied.

Drought stress is a dominating limiting factor in agricultural production in arid and semiarid regions. Studies have shown that hydraulic conductance, net photosynthetic rate, transpiration and biomass production decreased with drought stress, and thus the water use efficiency of Barbados nut was reduced (Santana *et al.* 2015). Yang *et al.* (2013a) found that Barbados nut improved its water transmission efficiency from the root to the canopy by enhancing the Huber value; therefore, Barbados nut showed improved resistance to drought stress when irrigated at intervals of 12 days. However, there are few studies on the optimal moisture modulation mode for enhancing the remediation of Cd-contaminated soil by the plant. The objectives of this study, therefore, were to investigate the growth, irrigation-water use efficiency, Cd uptake, translocation, and accumulation in Barbados nut under different soil Cd concentrations and irrigation regimes. In addition, findings of this study will determine that can Barbados nut be used as a phytoremediation species to remediate Cd-contaminated soil in arid and semiarid areas?

## Materials and Methods

### Experimental outline

The experiment was conducted in a greenhouse of the faculty of Agricultural and Food, Kunming University of Science and Technology, Kunming, Yunnan, China (25°1'N, 102°8'E, 1862 m) from November 2013 to August 2014. During the experimental period, the mean temperature was 20 to 38°C from 8:00 to 19:00, and the relative humidity was in the range of 30 to 55%.

### Experimental method

In total 280 Barbados nut seedlings were selected and transferred (one seedling in each pot) to plastic pots (30 cm in diameter at the top edge, 22.5 cm in diameter at the bottom, and 30 cm in depth) with 14 kg soil. The experimental soil was dry red soil (Rhodoxeralfs), and its basic physical and chemical properties are summarized in Table 1. All pots were irrigated with tap water to reach (upper limit) 90% SWC (soil water-holding capacity) every 10 days, and 0.64 g kg<sup>-1</sup> CO(NH<sub>2</sub>)<sub>2</sub> and 3.43 g kg<sup>-1</sup> KH<sub>2</sub>PO<sub>4</sub> were supplied as basal fertilizers.

Different amounts of Cd in the form of cadmium chloride hydrate (CdCl<sub>2</sub>·2.5 H<sub>2</sub>O) were mixed with 14 kg soil and used to fill the pots before transplanting the seedlings. One hundred days later, we selected 48 experimental seedlings based on their health and similarity in height and leaf number and transplanted them to plastic pots with Cd-contaminated soil before the first irrigation treatments. They were randomly divided into sixteen groups with three replications (4×4×3). Over a 10-day period after transplanting, different amounts of irrigation water were

applied to all treatments. Barbados nut was grown at low (50 mg kg<sup>-1</sup>), medium (100 mg kg<sup>-1</sup>) and high (200 mg kg<sup>-1</sup>) Cd levels as well as with no Cd under four irrigation regimes: 30, 50, 70 and 90% of soil water-holding capacity (SWC); a 50% soil saturation percentage was considered 100% SWC.

### Measurements

Plant height and basic diameter were measured by a ruler and an electronic screw micrometer every two weeks. Variation of plant height and stem diameter was the measured value on August 20 minus that on May 21. Roots, stem with branches and leaf blades were respectively harvested on 20 August. We thoroughly rinsed fresh tissues with distilled water to remove surface adhering Cd and put them in different paper bags. Paper bags with different plant tissues were first dried in an oven at 105°C for 30 min and then dried in a desiccator at 80°C for about 72 h until a constant weight was reached, and weighed these tissues by an electronic balance with an accuracy of 0.01 g. Leaf area was determined using Auto CAD (2007) method (Rico-Garcia *et al.* 2009), and total leaf area equals to per leaf area multiplied by total leaf dry mass and dividing per leaf dry mass, and total leaf dry mass include the fallen leaves during the experimental period. In addition, the dry plant tissues were pulverized by a plant pulverizer and sent to the analytical test center of Kunming, Yunnan Province, China to measure the Cd concentrations. The Cd contents (C') in the plant, Translocation factor (TF), Bioconcentration factor (BF) and Irrigation water-use efficiency (IUE) were respectively calculated using the following equations (Santana *et al.* 2015; Zhu *et al.* 2018):

$$C' = C_{\text{leaf}} \times M_{\text{leaf}} + C_{\text{stem}} \times M_{\text{stem}} + C_{\text{root}} \times M_{\text{root}} \quad (1)$$

$$TF = \frac{C_{\text{shoot}}}{C_{\text{root}}} = \frac{C_{\text{leaf}} \times M_{\text{leaf}} + C_{\text{stem}} \times M_{\text{stem}}}{M_{\text{leaf}} + M_{\text{stem}}} \times C_{\text{root}} \quad (2)$$

$$BF = \frac{C_{\text{whole plant}}}{C_{\text{soil}}} = \frac{C_{\text{leaf}} \times M_{\text{leaf}} + C_{\text{stem}} \times M_{\text{stem}} + C_{\text{root}} \times M_{\text{root}}}{M_{\text{leaf}} + M_{\text{stem}} + M_{\text{root}}} \times C_{\text{soil}} \quad (3)$$

$$IUE = \frac{I_{\text{total}}}{M_{\text{leaf}} + M_{\text{stem}} + M_{\text{root}}} \quad (4)$$

Where, C is Cd concentration and M is dry mass.

### Statistical analysis

Data were analyzed for statistical significance using Excel 2003 (Microsoft, Redmond, WA, USA) and SPSS version 20 software (IBM, Chicago, IL, USA). Analysis of variance was used to compare the statistical difference and least significant difference (LSD) test was used to compare treatments means  $P < 0.05$ .

## Results

### Growth

Different soil Cd concentrations and irrigation regimes and

the interactions among them had significant effects on all results indicate that soil Cd concentration  $\times$  irrigation

**Table 1:** Physicochemical properties of the soil

Soil	Total nitrogen (mg kg <sup>-1</sup> )	Total potassium (mg kg <sup>-1</sup> )	Total phosphorus (mg kg <sup>-1</sup> )	pH	Pb (mg kg <sup>-1</sup> )	Cu (mg kg <sup>-1</sup> )
Rhodoxerafals	0.87	13.92	0.68	5.5	4.33	2.75
Bulk density (g cm <sup>-3</sup> )	Organic matter (mg kg <sup>-1</sup> )	Cation exchange capacity (mg kg <sup>-1</sup> )	Soil water-holding capacity (%)	Zn (mg kg <sup>-1</sup> )	As (mg kg <sup>-1</sup> )	Cd (mg kg <sup>-1</sup> )
1.20	13.20	12.6	25.2	7.62	2.54	1.67

Here Pb: Lead; Cu: Copper; Zn: Zinc; As: Arsenic; Cd: Cadmium

**Table 2:** Effects of different soil Cd concentrations and irrigation regimes on growth and dry matter of Barbados nut

Treat-ments	Growth variables			Dry matter (g plant <sup>-1</sup> )					
	Plant height (cm plant <sup>-1</sup> )	Stem diameter (cm plant <sup>-1</sup> )	Leaf area (m <sup>2</sup> plant <sup>-1</sup> )	M <sub>l</sub>	M <sub>s</sub>	M <sub>r</sub>	M <sub>c</sub>	M <sub>t</sub>	
	Mean $\pm$ SE	Mean $\pm$ SE	Mean $\pm$ SE	Mean $\pm$ SE	Mean $\pm$ SE	Mean $\pm$ SE	Mean $\pm$ SE	Mean $\pm$ SE	
I <sub>1</sub>	Cd <sub>1</sub>	4.61 $\pm$ 0.21i	2.10 $\pm$ 0.13h	0.05 $\pm$ 0.00h	1.59 $\pm$ 0.06f	9.29 $\pm$ 0.81gh	6.63 $\pm$ 1.19def	10.88 $\pm$ 0.87g	17.52 $\pm$ 1.88i
	Cd <sub>2</sub>	3.32 $\pm$ 0.46ij	1.93 $\pm$ 0.32h	0.05 $\pm$ 0.00h	1.53 $\pm$ 0.13f	8.84 $\pm$ 1.16gh	5.64 $\pm$ 0.4def	10.37 $\pm$ 1.28g	16.01 $\pm$ 1.5i
	Cd <sub>3</sub>	2.23 $\pm$ 0.11jk	1.83 $\pm$ 0.08h	0.04 $\pm$ 0.00h	1.49 $\pm$ 0.18f	7.72 $\pm$ 0.78h	5.41 $\pm$ 0.84ef	9.21 $\pm$ 0.89g	14.62 $\pm$ 1.63i
	Cd <sub>4</sub>	1.05 $\pm$ 0.18k	1.81 $\pm$ 0.00h	0.04 $\pm$ 0.00h	1.42 $\pm$ 0.13f	7.07 $\pm$ 0.65h	4.48 $\pm$ 0.60f	8.49 $\pm$ 0.73g	12.97 $\pm$ 1.08i
I <sub>2</sub>	Cd <sub>1</sub>	28.99 $\pm$ 0.38b	6.05 $\pm$ 0.37cd	0.25 $\pm$ 0.01c	8.72 $\pm$ 0.62bc	22.58 $\pm$ 1.06cd	16.99 $\pm$ 1.29ab	31.30 $\pm$ 1.28cd	48.29 $\pm$ 2.38bc
	Cd <sub>2</sub>	27.78 $\pm$ 0.30b	5.23 $\pm$ 0.21def	0.23 $\pm$ 0.01cd	8.03 $\pm$ 0.4cd	20.87 $\pm$ 1.18cd	15.5 $\pm$ 0.68b	28.90 $\pm$ 1.48d	44.39 $\pm$ 1.84cd
	Cd <sub>3</sub>	18.21 $\pm$ 0.37f	5.07 $\pm$ 0.27ef	0.19 $\pm$ 0.02fg	6.47 $\pm$ 0.48de	16.18 $\pm$ 1.26e	12.18 $\pm$ 0.75c	22.65 $\pm$ 1.72ef	34.82 $\pm$ 1.64ef
	Cd <sub>4</sub>	9.41 $\pm$ 0.33h	3.89 $\pm$ 0.00g	0.17 $\pm$ 0.01g	5.65 $\pm$ 0.83de	12.2 $\pm$ 0.99fg	8.54 $\pm$ 0.54d	17.85 $\pm$ 1.79f	26.39 $\pm$ 2.22gh
I <sub>3</sub>	Cd <sub>1</sub>	32.76 $\pm$ 0.76a	7.35 $\pm$ 0.09a	0.34 $\pm$ 0.00a	11.58 $\pm$ 0.64a	28.48 $\pm$ 1.28a	18.88 $\pm$ 1.06a	40.07 $\pm$ 1.91a	58.94 $\pm$ 2.95a
	Cd <sub>2</sub>	28.84 $\pm$ 0.84b	7.07 $\pm$ 0.48a	0.32 $\pm$ 0.00a	10.86 $\pm$ 0.52a	26.19 $\pm$ 0.78ab	16.92 $\pm$ 0.8ab	37.05 $\pm$ 1.28ab	53.97 $\pm$ 1.86ab
	Cd <sub>3</sub>	21.10 $\pm$ 0.83e	5.76 $\pm$ 0.33cde	0.25 $\pm$ 0.01c	8.58 $\pm$ 0.60bc	20.10 $\pm$ 1.30d	12.06 $\pm$ 1.18c	28.68 $\pm$ 1.80d	40.74 $\pm$ 2.05de
	Cd <sub>4</sub>	10.11 $\pm$ 0.65h	5.62 $\pm$ 0.34cde	0.21 $\pm$ 0.01ef	7.34 $\pm$ 0.58	14.34 $\pm$ 1.05ef	8.09 $\pm$ 1.00de	21.68 $\pm$ 1.61ef	29.78 $\pm$ 2.6fgh
I <sub>4</sub>	Cd <sub>1</sub>	25.41 $\pm$ 1.08c	6.91 $\pm$ 0.54ab	0.31 $\pm$ 0.01a	10.81 $\pm$ 1.12a	24.21 $\pm$ 1.45bc	16.17 $\pm$ 1.36ab	35.02 $\pm$ 2.52bc	51.19 $\pm$ 3.56bc
	Cd <sub>2</sub>	23.33 $\pm$ 0.26d	6.15 $\pm$ 0.26bc	0.28 $\pm$ 0.01b	9.83 $\pm$ 0.71ab	21.58 $\pm$ 1.46cd	14.34 $\pm$ 0.61bc	31.41 $\pm$ 2.08cd	45.74 $\pm$ 2.66cd
	Cd <sub>3</sub>	14.80 $\pm$ 0.42g	5.20 $\pm$ 0.16def	0.22 $\pm$ 0.00de	7.66 $\pm$ 0.54cde	15.61 $\pm$ 0.89e	8.38 $\pm$ 1.20de	23.27 $\pm$ 1.42e	31.65 $\pm$ 2.21fg
	Cd <sub>4</sub>	8.61 $\pm$ 1.02h	4.73 $\pm$ 0.15f	0.18 $\pm$ 0.01g	6.69 $\pm$ 0.53de	11.83 $\pm$ 0.65fg	6.18 $\pm$ 0.54def	18.52 $\pm$ 1.14ef	24.7 $\pm$ 1.68h
Significant test (F value)									
F (I)	572.19**	33.47**	105.02**	162.59**	122.86**	68.63**	150.84**	146.65**	
F (Cd)	972.39**	222.38**	700.15**	21.87**	67.30**	58.42**	54.45**	71.76**	
F (I $\times$ Cd)	43.53**	25.42**	12.59**	2.39*	4.90**	4.31**	4.37**	5.54**	
LSD value	15.28	6.65	10.59	5.02	5.36	4.38	5.52	5.75	

Here M<sub>l</sub>: leaf dry matter; M<sub>s</sub>: stem dry matter; M<sub>r</sub>: root dry matter; M<sub>c</sub>: canopy dry matter; M<sub>t</sub>: total dry matter

Irrigation regimes I<sub>1</sub>, I<sub>2</sub>, I<sub>3</sub> and I<sub>4</sub> were 30, 50, 70 and 90% soil water-holding capacity (SWC), respectively and Cd concentrations of soil Cd<sub>1</sub>, Cd<sub>2</sub>, Cd<sub>3</sub> and Cd<sub>4</sub> were 0, 50, 100, 200 mg kg<sup>-1</sup>

Values are means  $\pm$  standard errors (n = 3)

LSD, least significant test; ANOVA, analysis of variance tests

\* and \*\* indicate significant difference ( $P < 0.05$  and  $P < 0.01$ )

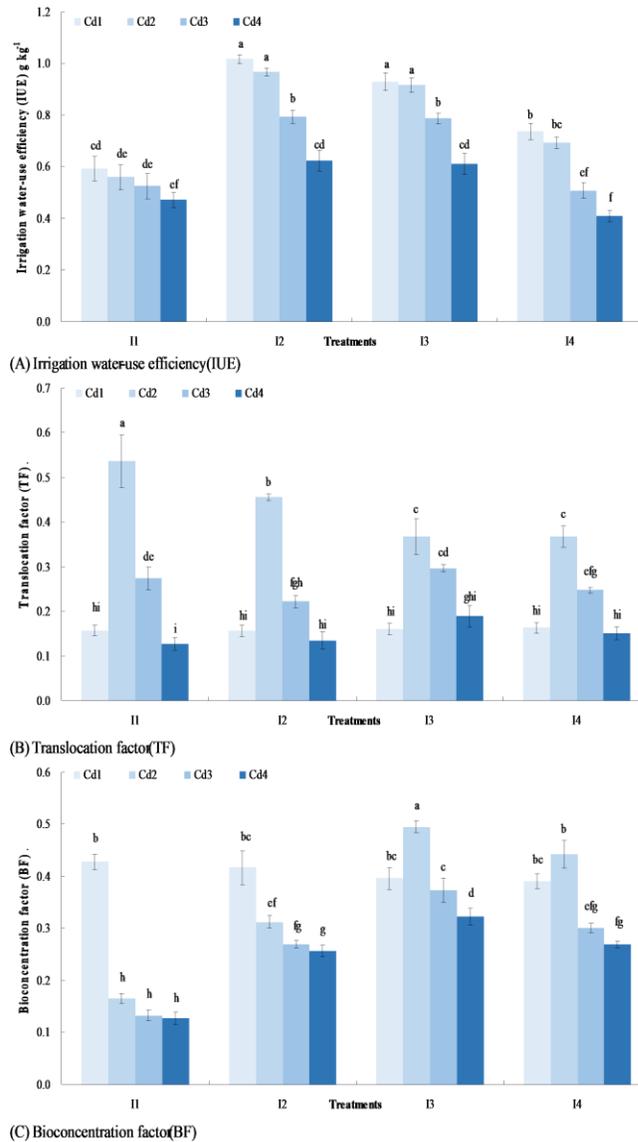
I  $\times$  Cd means the interactions among irrigation regimes (I) and soil Cd concentrations (Cd)

growth related-traits of Barbados nut ( $P < 0.05$ ) (Table 2). With increasing soil Cd concentrations, the plant height, stem diameter, leaf area and total dry matter of Barbados nut were decreased at 50–90% SWC. Moreover, Barbados nut grown at 70% SWC at low Cd concentrations, *i.e.*, 0–50 mg kg<sup>-1</sup> of soil, showed the maximum plant height, stem diameter, leaf area and total dry matter (Table 2). In addition, the minimum values of these traits were recorded at 30% SWC, and there was no significant difference among them at the different soil Cd concentrations. In addition, the leaf, stem, root and canopy dry matter showed the same tendency as total dry matter. Therefore, increasing Cd doses and water stress were associated with a strong reduction in the biomass yield, and there was an especially large reduction under the high Cd level (200 mg kg<sup>-1</sup>) and 30% SWC.

### Cadmium concentrations and Cd contents in different organs of barbados nut

The effects of different soil Cd concentrations and irrigation regimes on Cd concentrations and Cd contents in different organs of Barbados nut are shown in Fig. 1. The

regime had significant effects on Cd accumulation in Barbados nut ( $p < 0.05$ ). Cd enrichment exhibited a positive correlation with Cd concentration in soil, so that with the increasing soil Cd concentration, Cd accumulation in the organs increased significantly (Table 3). The total Cd concentrations in the plants at 70 and 90% SWC at the high Cd level (200 mg kg<sup>-1</sup>), 69.97 mg kg<sup>-1</sup> and 63.03 mg kg<sup>-1</sup>, respectively, were higher than those in the other treatments. However, considering the total biomass yield, the Cd contents per plant at 70% SWC with the high (200 mg kg<sup>-1</sup>) and medium (100 mg kg<sup>-1</sup>) Cd level, 69.97 mg plant<sup>-1</sup> and 63.03 mg plant<sup>-1</sup>, were higher than those in the other treatments. In addition, there were no significant differences among root, stem and leaf Cd concentrations or Cd contents at 30% SWC with different soil Cd concentrations, but they were in the order of root > stem > leaf at low, medium and high Cd levels. Furthermore, the percentages of Cd in the canopy and root were 46.17% and 53.83%, respectively, at the low Cd level (50 mg kg<sup>-1</sup>); those values were higher in the canopy (35.74% and 25.94%) and lower in the root (64.26% and 74.06%) than those of them medium Cd level (100 mg kg<sup>-1</sup>) and high Cd level (200 mg kg<sup>-1</sup>).



**Fig. 1:** Effects of different soil Cd concentrations and irrigation regimes on irrigation water-use efficiency (IUE), translocation factor (TF) and bioconcentration factor (BF) of Barbados nut  
Here Irrigation regimes: I<sub>1</sub>, I<sub>2</sub>, I<sub>3</sub> and I<sub>4</sub> were 30, 50, 70 and 90% soil water-holding capacity (SWC), respectively; and Cd concentrations of added to the soil Cd<sub>1</sub>, Cd<sub>2</sub>, Cd<sub>3</sub> and Cd<sub>4</sub> were 0, 50, 100, 200 mg kg<sup>-1</sup> Bars and points with different letters are significantly different (least significant test (LSD),  $P < 0.05$ ). Error bars represent the standard deviation ( $n = 3$ )

**Irrigation water-use efficiency (IUE), translocation factor (TF) and bioconcentration factor (BF) of barbados nut**

The different soil Cd concentrations and irrigation regimes and the interactions among them had significant effects on IUE ( $p < 0.05$ ) (Fig. 1A) and BF ( $p < 0.05$ ) (Fig. 1C). The IUE decreased with the increasing soil Cd concentration and increased first and then decreased with the increasing irrigation amount (Fig. 1A). The maximum IUE was observed at 50–70% SWC and lower Cd concentrations, *i.e.*, 0–50 mg kg<sup>-1</sup> soil, whereas the minimum value was observed at 90% SWC and the high Cd level (200 mg kg<sup>-1</sup>). However,

unlike IUE, the BF decreased with the increasing soil Cd concentration at lower irrigation regimes *i.e.*, 30% SWC and 50% SWC, but increased first and then decreased at higher irrigation regimes, *i.e.*, 70% SWC and 90% SWC. Based on the interactions among irrigation regime and soil Cd concentration, the 70% SWC irrigation regime can be used to effectively remediate soil containing lower or moderate amounts of Cd, *i.e.*, 50–100 mg kg<sup>-1</sup>. In addition, the TF was significantly affected by the soil Cd concentration ( $P < 0.05$ ) but not by irrigation regimes or the interactions among the two treatments ( $P > 0.05$ ) (Fig. 1B). The higher values of TF were 0.54 and 0.46 for 30% SWC and 50% SWC, respectively, at the low Cd level (50 mg kg<sup>-1</sup>).

**Table 3:** Effects of different soil Cd concentrations and irrigation regimes on Cd concentrations and Cd contents in different organs of Barbados nut

Treatments	Cd concentration (mg kg <sup>-1</sup> )					Cd content (mgkg <sup>-1</sup> )					
	leaf	stem	root	canopy	total	leaf	stem	Root	canopy	total	
I <sub>1</sub>	Cd <sub>1</sub>	0.24 ±0.04g	0.24±0.02f	1.54±0.15i	0.24±0.01j	0.67±0.05i	0.37±0.05h	2.22±0.36f	9.85±0.82e	2.59±0.31g	12.44±1.03g
	Cd <sub>2</sub>	1.39±0.09f	7.41±0.66e	12.31±1.19hi	4.4±0.33i	7.04±0.51h	2.11±0.13g	67.06±13.62e	69±6.16g	69.17±13.74e	138.17±19.73f
	Cd <sub>3</sub>	3.01±0.13e	7.50±0.64e	25.05±1.59gh	5.26±0.26hi	11.85±0.57g	4.52±0.67g	58.26±9.13e	137.43±27.08fg	62.78±9.01e	200.21±36.00f
	Cd <sub>4</sub>	4.52±0.41d	8.04±0.43e	60.02±4.06cde	6.28±0.32h	24.19±1.14f	6.47±1.04g	56.27±2.48e	272.62±51.54fg	62.74±2.64e	335.36±52.85f
I <sub>2</sub>	Cd <sub>1</sub>	0.24±0.02g	0.24±0.01f	1.54±0.10i	0.24±0.02j	0.67±0.04i	2.12±0.33g	5.35±0.42e	26.16±2.77g	7.46±0.68e	33.62±3.27f
	Cd <sub>2</sub>	2.87±0.11e	14.67±0.76d	25.02±1.09gh	8.77±0.34f	14.19±0.59g	22.95±0.29f	307.97±32.19cd	388.05±25.82ef	330.93±32.46cd	718.98±56.74e
	Cd <sub>3</sub>	5.77±0.23c	14.86±0.96d	55.56±3.03def	10.32±0.44fg	25.4±1.12f	37.14±1.37cd	242.42±33.89d	676.85±57.45cd	279.56±35.13d	956.4±66.71de
	Cd <sub>4</sub>	7.59±0.29b	20.56±1.21c	125.7±10.71b	14.07±0.5cd	51.28±3.34c	42.83±6.12c	250.27±22.6d	1083.23±149.3ab	293.1±24.94d	1376.33±164.36b
I <sub>3</sub>	Cd <sub>1</sub>	0.27±0.00g	0.24±0.01f	1.55±0.12i	0.25±0.01j	0.68±0.04i	3.09±0.21g	6.76±0.54e	29.28±2.88g	9.86±0.74e	39.13±3.43f
	Cd <sub>2</sub>	3.14±0.21e	22.06±1.27c	45.66±3.26ef	12.6±0.72de	23.62±0.90f	34.26±3.63cd	578.86±45.16a	770.01±47.73cd	613.12±48.10a	1383.13±79.58b
	Cd <sub>3</sub>	7.60±0.61b	28.7±1.97b	75.38±3.38c	18.15±1.04b	37.23±1.70d	64.9±5.29a	578.43±60.84a	912.38±113.33bc	643.34±62.53a	1555.71±159.76b
	Cd <sub>4</sub>	8.86±0.49a	40.52±1.94a	160.53±14.19a	24.69±1.15a	69.97±3.98a	64.5±2.02a	578.53±35.47a	1309.66±215.38a	643.02±37.48a	1952.68±239.17a
I <sub>4</sub>	Cd <sub>1</sub>	0.26±0.02g	0.24±0.01f	1.54±0.11i	0.25±0.01j	0.68±0.04i	2.79±0.28g	5.85±0.12e	24.61±1.18g	8.64±0.24e	33.26±1.40f
	Cd <sub>2</sub>	2.92±0.25e	20.17±0.89c	40.66±3.79fg	11.55±0.38ef	21.25±1.48f	28.37±0.22ef	437.82±49.58b	587.2±77.57de	466.19±49.48b	1053.39±125.56cd
	Cd <sub>3</sub>	7.06±0.37b	21.94±1.73c	68.64±2.21cd	14.5±0.68c	32.55±1.19e	53.86±2.69b	344.34±43.47c	572.76±74.84de	398.19±44.34bc	970.95±97.18de
	Cd <sub>4</sub>	8.84±0.27a	30.08±1.56c	150.18±7.09a	19.46±0.69b	63.03±1.92b	59.09±4.83ab	355.87±26.84bc	932.03±106.55bc	414.96±26.6bc	1346.99±122.73bc
Significant test (F values)											
F(I)	74.64**	177.52**	61.62**	239.94**	143.65**	156.17**	112.80**	39.24**	128.73**	73.18**	
F(Cd)	526.37**	356.34**	399.09**	600.32**	718.55**	197.19**	107.97**	73.74**	122.87**	102.06**	
F(I×Cd)	12.82**	27.78**	13.12**	36.96**	27.21**	19.79**	12.67**	5.32**	14.20**	8.61**	
LSD value	9.33	9.10	7.89	11.40	11.03	7.40	5.87	4.18	6.27	5.20	

Here Irrigation regimes I<sub>1</sub>, I<sub>2</sub>, I<sub>3</sub> and I<sub>4</sub> were 30, 50, 70 and 90% soil water-holding capacity (SWC), respectively; and Cd concentrations of soil Cd<sub>1</sub>, Cd<sub>2</sub>, Cd<sub>3</sub> and Cd<sub>4</sub> were 0, 50, 100, 200 mg kg<sup>-1</sup>

Values are means ± standard errors (n = 3); LSD, least significant test; ANOVA, analysis of variance tests; \* and \*\* indicate significant difference ( $P < 0.05$  and  $P < 0.01$ ); I×Cd means the interactions among irrigation regimes (I) and soil Cd concentrations (Cd)

## Discussion

The characteristics of phytoremediation species are high biomass production, a tolerance for heavy metals and the ability to absorb heavy metals (Ahmadpour *et al.* 2010; Mahar *et al.* 2016; Rostami and Azhdarpoor 2019). The results of this study demonstrated that Barbados nut can be used as the phytoremediation species because of its high tolerance for Cd and water stress. The increase in Cd doses and water stress led to a significant drop in leaf area, growth and the amount of dry matter accumulation (Table 2). These parameters decreased because the plant reduced water transpiration to cope with the negative impacts of the environment and to reduce damage to the plant (Santana *et al.* 2015). However, Barbados nut could survive at 30% SWC with the high Cd level (200 mg kg<sup>-1</sup>) without visual signs of phytotoxicity; Barbados nut showed a great ability to resist severe drought and Cd stress. Barbados nuts are rich in endophytic bacteria that can produce organic acids to adjust the pH in Cd-contaminated soils and alleviate the toxicity of Cd ions to plant growth (Guo *et al.* 2014); in addition, an antioxidant protection mechanism is activated when Cd enters the plant organs (Iannelli *et al.* 2002) that prevents too many Cd ions from entering Barbados nut tissues in a short period and reduces the damage to plant growth under Cd stress.

The results of this study demonstrated that Barbados nut can be used as the phytoremediation species because of its strong ability to absorb Cd from the soil into its tissues. In this pot experiment study, the Cd accumulation content in the different tissues was found to be in the order of root > stem > leaf (Table 3), which meant that most of the Cd absorbed from the soil was retained in roots and only small

amounts were transported to the stem and leaf. Therefore, the root system is the main organ enriching the soil Cd for most plants (Wójcick and Tukendorf 1999; Ranieri *et al.* 2005). This conclusion is different from the finding of Chang *et al.* (2014) (stem > leaf > root), because the soil Cd concentrations and the ages of the trees were different between these studies. In addition, the percent of Cd contents in the canopy decreased with the soil Cd concentrations from 50 mg kg<sup>-1</sup> to 200 mg kg<sup>-1</sup>, which implied that the relatively low concentration of heavy metals in the soil led to a higher transfer coefficient. The roots are affected first by oxidative stress and Cd ions toxicity because the roots directly touch the soil Cd; thus, the water absorption capability of the roots and the quantity of water transported from the roots to the canopy decline (Benavides *et al.* 2005; Yang *et al.* 2013b). Additionally, the Cd accumulation and the bioconcentration factor (BF) in the 70% SWC treatment were the highest (Table 3 and Fig. 1), which could be explained in two ways. First, the conditions of soil moisture (water vapor) and heat were optimized at 70% SWC, which meant that a more favorable root-zone microenvironment for plant growth was created (Yang *et al.* 2013b). Sec, root activity and soil microbial activity were increased, and the ability of microbes to activate heavy metals in the soil was improved by a favorable root-zone microenvironment that promoted the absorption of more Cd by the plant root (Cicatelli *et al.* 2014; Mani *et al.* 2015). Therefore, soil moisture that is too high or too low is not conducive to plant growth and Cd absorption by the root system.

Water stress with soil Cd contamination leads to a significant drop in biomass production; thus, IUE is decreased (Zhu *et al.* 2018). Hence, the lowest IUE was

noted from 30% SWC with a high Cd level (200 mg kg<sup>-1</sup>). In this experiment, the average IUE values at 50% SWC and 70% SWC with the no-Cd level and low Cd (50 mg kg<sup>-1</sup>) treatments were significantly higher than those in the other treatments (Fig. 1A). The reason for the increase in IUE at the 50% SWC level may be the lower irrigation amount, but at the 70% SWC level, the higher yield may be the reason. In fact, the results show that the best irrigation treatment for Barbados nut remediation of Cd-contaminated soil is irrigation based on 70% SWC. Under these conditions, the plant will have a higher biomass yield, BF, and IUE and hence will optimally accumulate soil Cd. In addition, further validation experiments based on the concentration gradient are needed to obtain the maximum capacity of this plant species for absorbing Cd.

## Conclusion

Barbados nut can be used for the phytoremediation species of Cd-contaminated soil as it exhibited a strong capacity to resist severe drought (30% SWC) and high Cd stress (200 mg kg<sup>-1</sup>). The optimal irrigation regime under which Barbados nut achieves high biological yield and irrigation water-use efficiency and takes up and accumulates large amounts of Cd in its tissues at low (50 mg kg<sup>-1</sup>) and medium (100 mg kg<sup>-1</sup>) Cd level is 70% SWC.

## Acknowledgements

This research work was supported by the National Natural Foundation of China (No. 51379004), the General Program of Applied Basic Research of Yunnan Province (No. 2013FB024) and Innovation Fund Designated for Graduate Students of Yunnan Normal University (No. Yjs2018147).

## References

- Agbogidi OM, AE Mariere, OA Ohwo (2014). Metal concentration in Plant tissues of *Jatropha curcas* L. grown in crude oil contaminated soil. *J Sustain For* 1:9–17
- Ahmadpour P, AM Nawi, A Abdu, H Abdul-hamid, DK Singh, A Hassan, S Jusop (2010). Uptake of heavy metals by *Jatropha curcas* L. planted in soils containing sewage sludge. *Amer J Appl Sci*, 7:1291–1299
- Alkorta I, J Hernández-Allica, JM Becerril, I Amezaga, I Albizu, C Garbisu (2004). Recent findings on the phytoremediation of soils contaminated with environmentally toxic heavy metals and metalloids such as zinc, cadmium, lead, and arsenic. *Rev Environ Sci Biotechnol* 3:71–90
- Benavides MP, SM Gallego, ML Tomaro (2005). Cadmium toxicity in plants. *Braz J Plant Physiol* 17:21–34
- Bolan N, A Kunhikrishnan, R Thangarajan, J Kumpiene, J Park, T Makino, K Scheckel (2014). Remediation of heavy metal (loid) s contaminated soils - to mobilize or to immobilize? *J Hazard Mater* 266:141–166
- Chang FC, CH Ko, MJ Tsai, YN Wan, CY Chung (2014). Phytoremediation of heavy metal contaminated soil by *Jatropha curcas*. *Ecotoxicology* 23:1969–1978
- Cicatelli A, P Torrigiani, V Todeschini (2014). Arbuscular mycorrhizal fungi as a tool to ameliorate the phytoremediation potential of poplar: biochemical and molecular aspects. *iFor-Biogeoosci For* 7:333–341
- Du R, J Bai, S Wang, Q Wu, H Zheng, Q Li, R Chou (2011). Response of soil microbial community function to chemical-aided remediation of multi-metal contaminated soils using *Jatropha curcas*. *Acta Sci Circumst* 31:575–582
- Guo J, X Lv, J Yang, H Yang, Y Gao, C Li (2014). Screening of endophytic bacteria inhibiting canker of *Jatropha curcas*. *J Yunnan Agric Univ* 29:610–613
- Iannelli MA, F Pietrini, L Fiore, L Pettrilli, A Massacci (2002). Antioxidant response to cadmium in *Phragmites australis* plants. *Plant Physiol Biochem* 40:977–982
- Liang J, X Liu, L Tang, Y Xu, S Wang, F Chen (2011). Biological characteristics of *Jatropha curcas* root border cells and toxic effect of cadmium on the cell viability. *Chin J Ecol* 30:1423–1428
- Mahar A, P Wang, A Ali, Z Guo, MK Awasthi, AH Lahori, Q Wang, F Shen, R Li, Z Zhang (2016). Impact of CaO, fly ash, sulfur and Na<sub>2</sub>S on the (im)mobilization and phytoavailability of Cd, Cu and Pb in contaminated soil. *Ecotoxicol Environ Saf* 134:116–123
- Mani D, C Kumar, NK Patel (2015). Integrated micro-biochemical approach for phytoremediation of cadmium and zinc contaminated soils. *Ecotoxicol Environ Saf* 111:86–95
- Pandey VC, K Singh, JS Singh, A Kumar, B Singh, RP Singh (2012). *Jatropha curcas*: A potential biofuel plant for sustainable environmental development. *Renew Sustain Ener Rev* 16:2870–2883
- Ranieri A, A Castagna, F Scebba, M Careri, I Zagnoni, G Predieri, LSD Toppi (2005). Oxidative stress and phytochelatin characterisation in bread wheat exposed to cadmium excess. *Plant Physiol Biochem* 43:45–54
- Rascio N, F Navari-izzo (2011). Heavy metal hyperaccumulating plants: How and why do they do it? And what makes them so interesting? *Plant Sci* 180:169–181
- Rico-Garcia E, F Hernandez-Hernandez, GM Soto-Zarazua (2009). Two new methods for the estimation of leaf area using digital photography. *Intl J Agric Biol* 11:397–400
- Rostami S, A Azhdarpoor (2019). The application of plant growth regulators to improve phytoremediation of contaminated soils: A review. *Chemosphere* 220:818–827
- Santana TAD, PS Oliveira, LD Silva, BG Laviola, AFD Almeida, FP Gomes (2015). Water use efficiency and consumption in different Brazilian genotypes of *Jatropha curcas* L. subjected to soil water deficit. *Biomass Bioener* 75:119–125
- Tordo GM, AJM Baker, AJ Willis (2000). Current approaches to the revegetation and reclamation of metalliferous mine wastes. *Chemosphere* 41:219–228
- Wójcik M, A Tukendorf (1999). Cd - tolerance of maize, rye and wheat seedlings. *Acta Physiol Plantarum* 21:99–107
- Wong MH (2003). Ecological restoration of mine degraded soils, with emphasis on metal contaminated soils. *Chemosphere* 50:775–780
- Yang Q, F Li, F Zhan, X Liu (2013a). Interactive effects of irrigation frequency and nitrogen addition on growth and water use of *Jatropha curcas*. *Biomass Bioener* 59:234–242
- Yang Q, F Zhang, F Li, X Liu, (2013b). Hydraulic conductivity and water-use efficiency of young pear tree under alternate drip irrigation. *Agric Water Manage* 119:80–88
- Zahoor A, F Ahmad, M Hameed, SMA Basra (2018). Contribution of structural and functional traits in turgor maintenance of *Pistia stratiotes* under cadmium toxicity. *Intl J Agric Biol* 20:1391–1396
- Zhu G, H Xiao, Q Guo, Z Zhang, J Zhao, D Yang (2018). Effects of cadmium stress on growth and amino acid metabolism in two *Compositae* plants. *Ecotoxicol Environ Saf* 158:300–308