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Characteristics of Cd uptake and accumulation in two Cd accumulator oilseed rape species

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Abstract: Two pot experiments were conducted under greenhouse conditions to investigate the characteristics of Cd uptake and accumulation by two Cd accumulator oilseed rape varieties and one Indian mustard grown on a loamy soil that had been artificially contaminated by different amounts of $CdSO_4$ (0, 20, 40, 60, 80, 100 mg/kg soil). The relationship between shoot Cd uptake of the two oilseed rape cultivars and the soil Cd concentrations could be simulated via quadratic equations. The curve showed that maximum shoot Cd uptake of Indian mustard was 314.7 μg/pot at soil Cd concentration of 87.8 mg/kg, while maximum uptake of the variety Xikou Huazi was 543.3 μg/pot at soil Cd concentration of 69.1 mg/kg and that of the variety Zhongyou Za-1hao was 576.7 μg/pot at soil Cd concentration of 84.0 mg/kg, suggesting that shoot Cd uptake ability of the two Cd accumulator oilseed rapes was significantly higher than that of the Indian mustard. Xikou Huazi had higher phytoremediation potential for Cd contaminated soil. Shoot Cd accumulation ability of the two Cd accumulator oilseed rapes was correspond and Cd was easier translocated to the shoot than hyperaccumulator Indian mustard as comparation plant. Shoot Cd distribution pattern showed consistent and significant reduction from older leaves to younger ones of two oilseed rapes and Indian mustard. Cd uptake by oilseed rapes in growth prophase was higher than that of growth anaphase.

Keywords: Cd accumulator oilseed rape; uptake; accumulate; characteristics

Introduction

Phytoremediation has emerged as a potentially cost-effective alternative to engineering-based and promising technologies. But accumulator or hyperaccumulator plant germplasm resources are the prerequisite of the technologies (Shen, 1998; Wang, 2001; Brown, 1994). It has been recently reported that several members of the Brassicaceae family from the genus Brassica displayed an enhanced ability to accumulate and tolerate Cd (Kumar, 1995; Salt, 1995a; 1995b; Ebbs, 1997a; 1997b; Brown, 1995). Indian mustard is a high biomass and Cd accumulating crop within the Brassicaceae family, which has been identified as potentially useful plant for phytoremediation (Salt, 1995a; 1995b). But Indian mustard has its geographical growth requirement and has not been extensively grown in China. China has a wide variety of oilseed rape species and oilseed rape is also a member of the Brassicaceae family. In previous studies two cultivars of oilseed rape—Xikou Huazi and Zhongyou Za-1hao were selected from many varieties of oilseed rapes (Su, 2002; Wang, 2003). Hydroponics and pot trail suggested that oilseed rape—Xikou Huazi and Zhongyou Za-1hao have phytoremediation potential for Cd contaminated soil. But at present the characteristics and mechanisms of Cd uptake and accumulation in Cd accumulator oilseed rapes have not been thoroughly understood. The objectives of the present study therefore were to ulteriorly lucubrate the characteristics and potential of Cd uptake and accumulation in Cd accumulator oilseed rapes with comparison to that of Indian mustard in pot trials.

1 Materials and methods

1.1 Materials

The soil used in the study was collected from experiment section of China Agricultural University. Selected soil properties were summarized here as: pH 7.66, CEC 20.1 cmol/kg, organic carbon 1.26%, total Cd 0.06 mg/kg, extractable Cd (DTPA-Cd) 0.02 mg/kg. Two oilseed rape genotypes used in this study were Zhongyou Za-1hao (Z1), Xikou Huazi (XK) and Indian mustard (IN). Plastic pots (13 cm × 12 cm) were used in the trial for the characteristics of Cd uptake in oilseed rapes. Porcelain pots containing 3 kg soil were used in the trial for the Cd distribution pattern in oilseed rapes.

1.2 Trial for the characteristics of Cd uptake in oilseed rapes

The experiment was conducted in greenhouse as soil culture. Treatments consisted of two oilseed rape genotypes and one Indian mustard with 5 different Cd levels (20, 40, 60, 80 and 100 mg/kg soil) as $CdSO_4$ and control. The soil was treated with N 0.30 g/kg soil in the form of $(NH_4)_2SO_4$, P 0.20 g/kg and K 0.30 g/kg soil in the form of KH_2PO_4 and K_2SO_4, respectively. The soil was sieved to 3 mm and air dried, manually mixed with $CdSO_4$ solution to ensure

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homogeneity and equilibrated again for 7 d. Subsamples of 500 g were introduced into plastic pots (13 cm × 12 cm). Treatments were arranged on a completely randomized design with three replicates per treatment. After germination, the seedlings were thinned to 4 plants per pot and allowed to grow for 42 d, with pots being watered daily using tap water. At the end of the growth period, plants were harvested by cutting the shoots at the soil surface and the roots were removed from the pots. The samples were washed thoroughly with tap water and rinsed with double deionized water before oven dried at 70℃ for 48 h, and dry weights were recorded. Oven-dried plant material was ground using a stainless steel mill. Subsamples of plant samples were digested with a mixture of conc. HNO_3-$HClO_4$, and total shoot Cd contents were determined using inductively coupled plasma atomic emission spectroscopy (ICP-AES). And total root Cd contents were determined using the atomic absorption spectrometry (AAS). Relationship between the soil Cd concentration and shoot dry weight (shoot Cd content and shoot Cd uptake amounts) of the two oilseed rape genotypes and the Indian mustard were simulated by linear or quadratic curve and the correlation between both were tested. Relative shoot dry weight and percentage of Cd uptake amounts were calculated.

1.3 Trial for Cd distribution pattern in shoot of oilseed rapes

The soil, plant and pots experiment methods used were as above. Differently treatments consisted of two oilseed rape genotypes and Indian mustard with 2 different Cd levels (20, 40 mg/kg soil) as $CdSO_4$. Each pot was filled with 3 kg of air-dried soil. At the end of the growth period, plants were harvested by cutting the shoots at the soil surface and the leaves and stem were separated. Leaf number was from the base of the plant, counting the first identifiable leaf as No. 1, No. 2, to core leaf and stem. Total shoot Cd contents were determined using atomic absorption spectrometry (AAS). All data were analyzed using a SAS statistical package by an IBM personal computer. One way ANOVA was carried out to compare the means of different treatments; where significant F values were obtained, differences between individual means were tested using the least significant difference test.

2 Results and discussion

2.1 The characteristics of Cd uptake and accumulation in oilseed rape genotypes

The relationship between shoot dry weight yields of two oilseed rape cultivars and Indian mustard and soil Cd concentration could be simulated by mathematic model. Positive linear correlation and quadratic curve were found between both. Test results suggested that correlation of both relationship attained significant or very significant level (Fig. 1). With increases in the soil Cd concentrations, the shoot dry weight yields of oilseed rape Xikou Huazi and Zhongyou Za-1hao and Indian mustard decreased. But each curve was diverse in change extent. With increases in the soil Cd concentrations, the shoot dry weight of Indian mustard and oilseed rape Xikou Huazi decreased in parabolic trend, however the shoot dry weight of oilseed rape Zhongyou Za-1hao decreased linearly. The shoot dry weight yields of oilseed rape Zhongyou Za-1hao and Indian mustard decreased markedly at soil Cd concentration of 20—40 mg/kg, but the shoot dry weight yields of oilseed rape Xikou Huazi decreased slowly. On Cd contaminated soil, while the shoot dry weight of two oilseed rapes and Indian mustard attained more than 95% of maximum, the growth of two oilseed rapes and Indian mustard were hardly affected by Cd. While the shoot dry weight yields of two oilseed rapes and Indian mustard declined 5%, Xikou Huazi endured soil Cd concentration range of 0—51.8 mg/kg, Zhongyou Za-1hao endured soil Cd concentration range of 0—23.6 mg/kg, Indian mustard endured soil Cd concentration range of 0—9.5 mg/kg (Fig. 1). This suggested that Xikou Huazi had higher tolerance Cd level when the growth of two oilseed rapes and Indian mustard were not affected markedly by Cd. When shoot dry weight declined to 50% of maximum due to Cd toxicity, those soil Cd concentration were used to weigh the Cd tolerance ability of oilseed rapes. When shoot dry weight declined to 50% of maximum, soil Cd concentration for Zhongyou Za-1hao and Xikou Huazi were 144.1 mg/kg and 122.8 mg/kg, respectively; while Indian mustard was 145.2 mg/kg (Fig. 1). This suggested Zhongyou Za-1hao and Indian mustard had higher Cd toxicity tolerance ability. LSD methods was used to test the differences of the shoot dry weight yields of oilseed rapes and Indian mustard. Suggestting the shoot dry weight yields of oilseed rapes Zhongyou Za-1hao and Xikou Huazi were very significantly higher than Indian mustard in different Cd concentration soil, $F = 27.57^{**}$.

Cd content in plant is one of the most important characteristics of uptake and accumulate Cd in oilseed rape genotypes. The relationship between shoot Cd concentration of two oilseed rape and Indian mustard and different soil Cd concentration could be simulated by mathematic model. Both relationship were found to agree with quadratic curve. Test results suggested that both correlative coefficient attained very significant level. Cd uptake traits of two oilseed rapes and Indian mustard were different (Fig. 2). At soil Cd concentration of 0—100 mg/kg, with increase in the soil Cd concentrations, the shoot Cd concentration of oilseed rapes Zhongyou Za-1hao, Xikou Huazi and Indian mustard ascended in parabola trend; however the shoot Cd concentration of oilseed rape Zhongyou Za-1hao and Indian mustard always increased with increase in the soil Cd concentration. The shoot Cd concentration of Xikou Huazi first ascended. Maximum reached at soil Cd concentration of 84.3 mg/kg soil, then decreased with increases in the soil Cd concentration. At soil Cd concentration of 100 mg/kg, the shoot Cd concentration of oilseed rapes Zhongyou Za-1hao

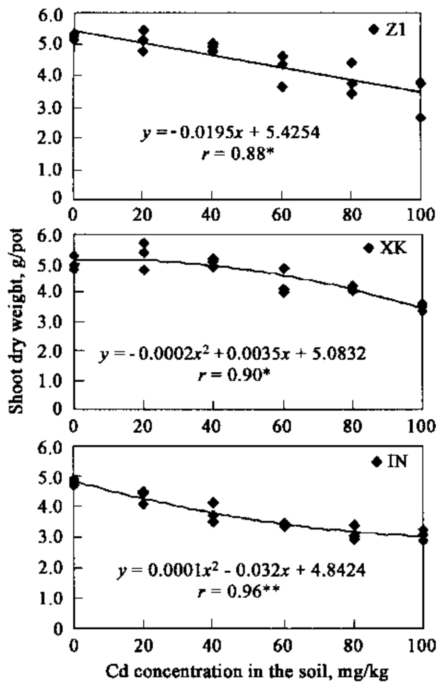

Fig.1 Shoot dry weight yield of two oilseed rapes and Indian mustard grown on different Cd concentration soil

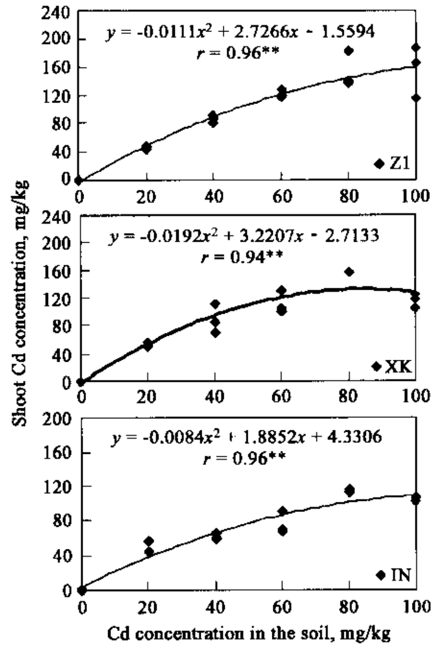

Fig.2 Shoot Cd concentration of two oilseed rapes and Indian mustard grown on different Cd concentration soil

reached 160.1 mg/kg, however the shoot Cd concentration of oilseed rape Xikou Huazi and Indian mustard reached 127.4 mg/kg and 108.9 mg/kg, respectively (Fig. 2). Here accumulation coefficient of Zhongyou Za-1hao was 1.6; Xikou Huazi was 1.3; Indian mustard was 1.1. LSD methods was used to test difference among the shoot Cd concentration of oilseed rapes and Indian mustard. Suggestting the shoot Cd concentration of oilseed rapes Zhongyou Za-1hao and Xikou Huazi were significantly higher than Indian mustard in different soil Cd concentrations, $F = 7.69^*$. Under the experiment condition shoot Cd concentration of two oilseed rape cultivars and Indian mustard followed the order: Zhongyou Za-1hao > Xikou Huazi > Indian mustard(according to maximum Cd concentration in plant).

Shoot Cd uptake amounts reflects uptake Cd ability of oilseed rape. It was one of the most general indices used to evaluate the phytoremediation potential of Cd accumulator oilseed rape. The relationship between shoot Cd uptake of two oilseed rape cultivars and Indian mustard and soil Cd concentration could be simulated by mathematic model. Positive quadratic curve was found between both. Test results suggested that both correlative coefficient attained significant significant level. From Fig.3 we can see that with increase in the soil Cd concentration, the shoot Cd uptake of two Cd accumulator oilseed rapes and Indian mustard ascended in parabola trend. At soil Cd concentration of 87.8 mg/kg, maximum shoot Cd uptake of Indian mustard reached 314.7 mg/kg; at soil Cd concentration of 84.0 mg/kg, the maximum shoot Cd uptake of Zhongyou Za-1hao reached 576.7 mg/kg; at soil Cd concentration of 69.1 mg/kg, the maximum shoot Cd uptake of Xikou Huazi reached 543.3 mg/kg(Fig.3). Here, the shoot Cd removal percentage of Indian mustard was 0.7%, Zhongyou Za-1hao was 1.4%, Xikou Huazi was 1.6%. This suggested that the cycle was shorter by two oilseed rapes to remediate the same Cd contaminated extent soil than by Indian mustard. Comparatively Xikou Huazi of oilseed rape demonstrated a higher phytoremediation potential for Cd comtaminated soil. LSD methods was used to test the differences of the shoot Cd uptake amounts of oilseed rapes and Indian mustard. The results suggested that the shoot Cd uptake amounts of oilseed rapes Zhongyou Za-1hao and Xikou Huazi were very significantly higher than Indian mustard in different soil Cd concentrations, $F = 16.54^{**}$.

Table 1 shows the root Cd concentration of two oilseed rapes and Indian mustard grown on different Cd concentration soil. With increases in the soil Cd concentrations, root Cd concentration of two oilseed rapes and Indian mustard significantly increased(Table 1). Heavy metals absorbed by most plants were accumulated in the roots. Heavy metals amounts absorbed by plants increased with increases of heavy metals concentrations in the soil. Generally speaking, root Cd concentration was 10—100 times that of shoot (Jiang, 2002). In our experiment, root Cd concentration of two oilseed rapes and Indian mustard were 2—3 times higher than that of shoot on different soil Cd concentrations. The results agreed with Jiang's(Jiang, 2002). Suggestting the most Cd uptake by two oilseed rapes and Indian mustard were accumulated in the shoot at each level of soil Cd concentration. Ulteriorly indicated two oilseed rapes and Indian mustard had the characteristics of hyperaccumulator.

2.2 The pattern of Cd distribution in oilseed rapes

On the Cd contaminated soil, relative shoot dry weight

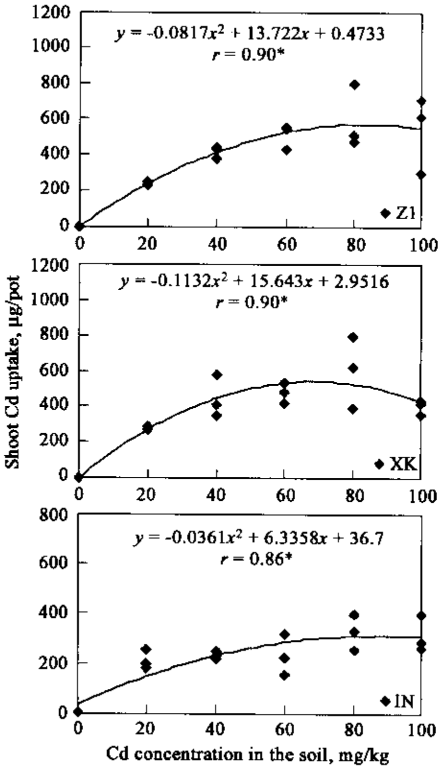

Fig.3 Shoot Cd uptake of two oilseed rapes and Indian mustard grown on different Cd concentration soil

Table 1 Root Cd concentration of two oilseed rapes and Indian mustard grown on different Cd concentration soil

Soil Cd concentration, mg/kg	Root Cd concentration, mg/kg		
	IN	Z1	XK
0	7.14 f*	12.74 e	7.16 f
20	81.85 e	93.02 d	88.44 e
40	169.80 d	173.99 c	156.07 d
60	251.21 c	236.11 b	237.72 c
80	305.54 b	353.43 a	314.60 b
100	349.38 a	346.01 a	357.81 a

Notes: LSD methods was used to test the differences of the shoot Cd concentration of two oilseed rapes and Indian mustard. Values in the same column having the same letter are not significantly different(p = 0.05, three replicates)

was one of the important indices showing plant tolerance of Cd toxicity ability. At soil Cd concentration of 100 mg/kg, shoot dry weight of Indian mustard descended 36%, while oilseed rapes Zhongyou Za-1hao and Xikou Huazi descended 35% and 19%, respectively(Table 2). In the artificially Cd contaminated soil, more than 56% of the Cd uptake by Indian mustard was accumulated in the shoot at each level of soil Cd concentration; Zhongyou Za-1hao was 75%; Xikou Huazi was 69%(Table 2). The results suggested two oilseed rapes had higher Cd translocation ability to shoot than Indian mustard.

Table 3 shows the distribution pattern of shoot Cd concentration of two oilseed rapes and Indian mustard on different soil Cd concentration. At soil Cd concentration of 20 and 40 mg/kg, Cd concentration of leaves decreased markedly from the base leaves of plant to top ones(Table 3). Salt *et al*. (Salt, 1995b) showed that Cd appeared to accumulate preferentially in the youngest leaves of both *B*. *juncea* and *T*. *caerulescens*. However, the result was contrary in our experiment. This could be because of different growth conditions. In the hydroponics available Cd was steady in the nutrition solution. Soil available Cd was variable with time in the pots trial. Soil available Cd was higher in the growth prophase. Here, plant Cd uptake was higher. Soil available Cd decreased with plant growth, plant Cd uptake amounts reduced. So translocation of Cd from older leaves to younger ones reduced. Cd concentration in younger leaves of two oilseed rapes and Indian mustard were same. However, Cd concentration of senescing leaves in two oilseed rapes were markedly higher than in Indian mustard (Table 3). This suggested that Cd accumulation ability of oilseed rape were higher in the growth prophase.

Table 2 The pattern of Cd accumulation in two oilseed rapes and Indian mustard grown on different Cd concentration soil

Soil Cd concentration, mg/kg	Relative shoot dry weight, %			Shoot Cd uptake percent, %		
	IN	Z1	XK	IN	Z1	XK
0	100	100	100	34	27	39
20	91	98	106	67	77	75
40	79	94	101	61	75	73
60	71	80	87	56	82	75
80	65	74	84	62	77	74
100	64	65	81	58	81	69

Table 3 Distrtibution of Cd in two oilseed rapes and Indian mustard grown on different Cd concentration soil

Leaf No.[a]	Cd concentration, mg/kg					
	IN		XK		Z1	
	20 mg/kg Cd	40 mg/kg Cd	20 mg/kg Cd	40 mg/kg Cd	20 mg/kg Cd	40 mg/kg Cd
1	87.33 a*	120.72a	147.94a	192.16 a	154.12 a	214.77 a
2	72.37 ab	92.00 b	132.51 b	136.25 b	110.54 b	142.02 b
3	61.19 bc	79.59 bc	98.67 c	119.40 c	87.64 c	121.51 c
4	52.80 c	68.65 cd	78.80 d	94.18 d	68.37 d	94.72 d
5	47.43 cd	55.87 de	71.11 d	74.85 e	61.35 d	70.05 e
6	43.38 cde	50.02 def	52.14 e	61.53 ef	48.10 e	51.41 f
7	42.52 cde	56.57 de	47.81 e	50.19 fg	44.29 e	34.32 g
8	30.23 de	31.01 f	34.09 f	44.12 gh	25.44 f	20.76 g
9	31.48 de	35.35 f	28.41 f	32.48 h	—	—
Stem	26.83 e	45.94 ef	—	—	—	—

Notes: * values in the same column having the same letter are not significantly different(p = 0.05, three replicates); [a] leaf number was from the base of the plant, counting the first clearly identifiable leaf as No. 1

3 Conclusions

The relationship between shoot dry weight yields, shoot Cd content, shoot Cd uptake amounts of the two Cd accumulator oilseed rape cultivars and soil Cd concentration could be described by close linear or quadratic curve correlation.

While the growth of two oilseed rapes and Indian mustard were not markedly affected by Cd, Xikou Huazi endured soil Cd concentration: 51.8 mg/kg, Zhongyou Za-1hao endured soil Cd concentration: 23.6 mg/kg, Indian mustard endured soil Cd concentration: 9.5 mg/kg. While dry weight declined to 50% of maximal dry weight, soil Cd concentration of Zhongyou Za-1hao was 144.1 mg/kg, Xikou Huazi was 122.8 mg/kg, Indian mustard was 145.2 mg/kg, respectively.

At soil Cd concentration of 100 mg/kg, shoot Cd concentration of oilseed rape Zhongyou Za-1hao reached 160.1 mg/kg. However, shoot Cd concentration of oilseed rapes Xikou Huazi and Indian mustard reached 127.4 mg/kg and 108.9 mg/kg, respectively. The shoot Cd concentration of two oilseed rapes were significantly higher than that of Indian mustard.

At soil Cd concentration of 84.0 mg/kg, 69.1 mg/kg and 87.8 mg/kg, maximum shoot Cd uptake amounts of oilseed rapes Zhongyou Za-1hao, Xikou Huazi and Indian mustard reached 576.7 μg/pot, 543.3 μg/pot and 314.7 μg/pot, respectively.

Root Cd concentration of two oilseed rapes and Indian mustard were 2—3 times higher than shoot Cd concentration in different Cd concentration soil. Shoot Cd distribution pattern of two oilseed rapes and Indian mustard were similar in that Cd concentration of leaves decreased significantly from the older leaves to younger ones when the plants were grown in soil.

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