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Bioaccumulation and translocation of cadmium in cole (*Brassica campestris* L.) and celery (*Apium graveolens*) grown in the polluted oasis soil, Northwest of China

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Abstract

A pot experiment was conducted to study the bioaccumulation and translocation of cadmium (Cd) in cole (*Brassica campestris* L.) and celery (*Apium graveolens*) grown in the Cd-polluted oasis soil, Northwest of China. The results showed that Cd in the unpolluted oasis soil was mainly bound to carbonate fraction (F2) and Fe-Mn oxide fraction (F3). However, marked change of Cd fractions was observed with increasing soil Cd concentrations, in which the concentration of Cd in F1 (exchangeable fraction), F2 and F3 increased significantly (p < 0.001 for F1, F2 and F3). The growth of cole and celery could be facilitated by low concentrations of Cd, but inhibited by high concentrations. The correlation analysis between the fraction distribution coefficient of Cd in the soil and Cd concentration accumulated in the two vegetables showed that Cd in F1 in the soil made the greatest contribution on the accumulation of Cd in the two vegetables. The high bio-concentration factor and the translocation factor of Cd in both cole and celery were observed, and Cd had higher accumulation in the edible parts of the two vegetables. Therefore, both cole and celery grown in Cd-polluted oasis soil have higher risk to human health. And the two vegetables are not suitable to be cultivated as vegetables consumed by human in the Cd-polluted oasis soil.

Key words: bioaccumulation; translocation; cadmium; cole and celery; oasis soil

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Introduction

There is increasing concern regarding food safety due to environmental pollution (Yang et al., 2009). Soil pollution with anthropogenic heavy metals coming from industry or agriculture such as smelting industries, residues from metalliferous mined, pesticide, fertilizers, municipal compost has received much attention in recent years (Lu et al., 2005; Liu et al., 2007b). Heavy metals in soils can not be biodegraded, but can be bio-accumulated and biotransformed by plants, and pose toxicity beyond certain limit. Excessive heavy metals may cause greater toxicity, furthermore, can pose a threat to human health through the food chains.

Many studies have found that the toxicity and mobility of heavy metals depended not only on their total amounts but also on their chemical fractionations (Ma and Rao, 1997; Li and Thornton, 2001; Greenway and Song, 2002;

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Chojnacka et al., 2005; Gupta and Sinha, 2006; Silveira et al., 2006; Kartal et al., 2006; Li et al., 2007; Tandy et al., 2009; Wang et al., 2009; Rodríguez et al., 2009). There are many classification methods to chemical fractions of heavy metals (Tessier et al., 1979; Ahnstrom and Parker, 1999; Qiao et al., 2003; Silveira et al., 2006). Tessier's five-step sequential extraction method has been widely used, in which heavy metals in soils were categorized in the five fractions: exchangeable fraction (F1), carbonate fraction (F2), Fe-Mn oxide fraction (F3), organic matter fraction (F4) and residual fraction (F5) (Tessier et al., 1979; Lucho-Constantino et al., 2005; Silveira et al., 2006; Rodríguez et al., 2009). F1 is bioavailable fractionation; F2, F3 and F4 are potential bioavailable fractionation; F5 is unbioavailable fractionation (Ma and Rao, 1997; He et al., 2005; Rodríguez et al., 2009).

The oasis soil mainly distributes in the oasis in the arid desert region including Xinjiang Autonomous Region and the Hexi Corridor, Northwest of China. In recent decades,

due to croplands have been irrigated by the wasterwater coming from the industry and agriculture activities in the arid area, the oasis soil has been seriously polluted by heavy metals, especially from Cd pollution (Nan and Zhao, 2000; Ding et al., 2008), which caused a serious threat to the oasis ecological environment and human health.

Cadmium (Cd) is a toxic trace element to plants and animals, and can cause damage even at very low concentrations and can be taken up by crops easily. In China, a mount of soils and vegetables were polluted by Cd which even is considered of a most important and typical pollution of heavy metals. According to the report, about 14,000 hm² of agricultural soils were polluted with Cd (Chen, 1996; Su et al., 2009). Moreover, Cd may pose risk to human and animal health at plant tissue concentrations that are not generally phytotoxic (Liu et al., 2007a). In order to avoid the Cd pollution of the food chain, it is essential to assess its bioavailability in soil (Cornu et al., 2009).

Cole (*Brassica campestris* L.) and celery (*Apium graveolens*) have been widely cultivated as vegetables which were consumed for their edible parts in Northwest of China. People who consume vegetables grown in Cd-polluted soils in the area are at risk of an elevated Cd exposure. The proposed work aimed to study the bioaccumulation and translocation of Cd in cole and celery grown in the Cd-polluted oasis soil based on pot experiment. The health risks of the vegetables were also evaluated.

1 Materials and methods

1.1 Experimental design

Experimental reagents were selected with superior grade (Tianjin Kermel Chemical Reagents Corporation, China) and certified reference samples, GSS-1 (GBW07401) and GSB-6 (GBW10015) were used. The two parameters bioconcentration factor (BCF) and the translocation factor (TF) were calculated. BCF was the ratio of the metal concentration in plant tissues to that in their rooted soils, and TF was the ration of metal concentration in other plant parts to that in roots.

Pot experiments were conducted under open air conditions at Linze County, Hexi Corridor, Northwest China. The irrigation-silting soils are derived from oasis/desert soils, and textures mainly sandy. Soil properties are pH (H_2O) 8.47, 1.54% organic mater (OM) content, 7.39% $(CaCO_3)$ content, and cation exchange capacity (CEC) of 8.11 cmol/kg. The background concentration level of Cd in agrarian land is 0.118 mg/kg dry weight (dw).

The soil used was an irrigation-silt sandy soil, excavated from an oasis farm, mixed several times into one large heap, then passed through a 10 mm sieve to remove large stones and grass debris. Nine treatments (one control and eight amendments) were replicated three times in a randomized block design. The control was not amended with inorganic Cd. The others, one by one, had been spiked with different solutions of cadmium nitrate to elevate soil metal concentrations as follows: 0.35, 0.70, 1.05, 1.40, 2.10, 3.50, 4.90, and 7.00 mg/kg dw for Cd, which was

designed based on our field pollution investigation in the Cd-polluted oasis soil. The heavy metal was added by spraying a solution of metal salt over dry soil spread out on a large tray. The soil was turned over and sprayed several times then watered and left to equilibrate outdoors for two weeks before planting vegetables. The plants were seeded and taken care of according to cultivation system.

The whole vegetables were harvested after 60 days of growth. The vegetable was cut to separate parts grown under and above the soil. Leaves were washed three times using de-ionized water. Roots were abundantly washed with tap water to eliminate soil particles, and rinsed three times with de-ionized water. The fresh weight (fw) of roots and shoots were recorded. All plant samples were dried in an oven at 70°C, passed through 2 mm nylon sieve for further experiment.

Soil samples were taken from the pots after harvesting the vegetables. After removing crop debris, soil samples were air-dried at room temperature, passed through 100 mesh ($\phi = 0.149$ mm) nylon sieve.

1.2 Determination of Cd in soils and vegetables

Total Cd concentrations in soils were determined according to national standards of China (GE/T17141–1997). Total Cd in the vegetables was extracted using the acid digestion mixture (HNO₃-HClO₄-HF) (EPA3010A). The clear solution was obtained from the digestion, filtered, reconstituted to the desired volume, and analyzed by an atomic absorption spectrometer (AAS, M6MK2, Thermo Electron Corporation, USA). The solutions were acidified with HNO₃ and stored at 4°C until analyse.

1.3 Tessier's sequential extractions

Tessier's sequential extraction procedure was used in this experiment. Cd was extracted by MgCl₂ solution in F1; by HAc-NaAc solution in F2; by NH₂OH·HCl solution in F3; by HNO₃-H₂O₂-NH₄Ac solution in F4; and by HNO₃-HClO₄-HF solution in F5. The Cd concentrations were also determined by AAS.

1.4 Statistical analysis

All treatments were replicated three times. The means and standard deviations (SD) were calculated by Excel2003 for Windows. One-way analysis of variance was carried out with SPSS16.0. When a significant (p < 0.05) difference was observed between treatments, multiple comparisons were made by the LSD test.

2 Results and discussion

2.1 Chemical fractions of Cd in the oasis soil

The experimental results in Fig. 1 showed that planting various vegetables (cole and celery) had no effect on the Cd chemical fractions distribution in the oasis soil (Fig. 1). With increasing Cd concentrations in soils, the concentrations of Cd in exchangeable fraction (F1), carbonate fraction (F2) and Fe-Mn oxide fraction (F3) increased significantly (p < 0.001 for F1, F2 and F3) because of

the addition of Cd in the form of solution, which were high bioavailability in spite of two weeks age time. The parameter fraction distribution coefficient (FDC) usually was defined as the percentage of the each heavy metal fraction. The FDCs of Cd increased significantly in F1 and F2 (p < 0.001) with increasing soil Cd concentration probably because of greater mobility of Cd and higher CaCO₃ concentration in the oasis soil. FDC of Cd reduced significantly in F3 (p < 0.001) (Fig. 2).

Previous studies showed that in the unpolluted soil, F5 was mainly heavy metals fraction, which was not bio-available for the plants (Gao et al., 2001; Wang and Zhou, 2003; Guo and Zhou, 2005), but the percentage of heavy metals associated with available fraction increased with increasing total amount of heavy metals (Gao et al., 2001; Zhou et al., 2003; Wang and Zhou, 2003; Guo and Zhou, 2005). The similar results were observed in present study. The fraction of Cd in unpolluted soil was mainly in F3, however, the bioavailable fraction concentration increased significantly (p < 0.001) with increasing soil Cd concentrations.

2.2 Effects of Cd stress on vegetables growth

The change tendency of fresh weight of vegetables roots and shoots under Cd treatments are shown in Table 1.

The fresh biomass of vegetables in roots and shoots

increased at first, and reached peak value, then decreased with increasing soil Cd concentration. When the treatment level of Cd was 2.10 mg/kg, the fresh biomass of cole shoots reached the maximum value $(27.56 \pm 6.04 \text{ g/pot})$, increased by 88.6%, comparing with the control, indicating that this value was hazardous threshold of cole. Beyond it, the growth of cole would be inhibited. Similarly, when the treatment level of Cd was 1.05 mg/kg, the fresh biomass of celery shoots reached the maximum value $(6.73 \pm 2.74 \text{ g/pot})$, increased by 127.4%, comparing with the control, indicating that this value was hazardous threshold of celery. Beyond it, the growth of celery would be inhibited.

These results indicated that the toxic effect of Cd on vegetables had threshold values. Vegetables growth could be enhanced by low concentrations of Cd (\leq 2.10 mg/kg for cole, \leq 1.05 mg/kg for celery), but inhibited by high concentrations. This conclusion was in agreement with previous studies (Song et al., 1996; Li et al., 2002; Zong et al., 2007). The phenomena probably due to that high concentrations of Cd damaged vegetables roots, and inhibited uptake of nutrient elements in roots, then inhibited the growth of vegetables.

2.3 Accumulation of Cd in cole and celery

The amounts and distribution of Cd accumulated in vegetables under various treatments are shown in Table 2.

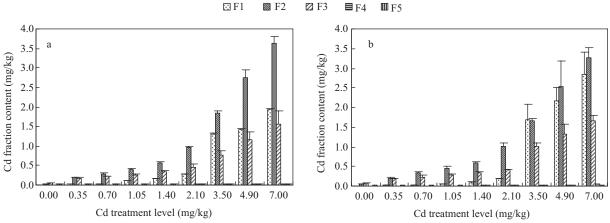


Fig. 1 Distribution of Cd fractions concentration in the oasis soil in which cole (a) and celery (b) was grown, respectively. F1: exchangeable fraction, F2: carbonate fraction, F3: Fe-Mn oxide fraction, F4: organic matter fraction, F5: residual fraction. Values were means ±SD of three replicates.

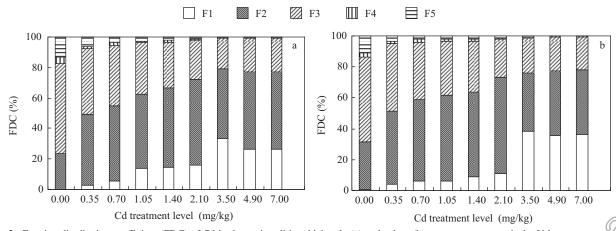


Fig. 2 Fraction distribution coefficient (FDC) of Cd in the oasis soil in which cole (a) and celery (b) was grown, respectively. Values were means of three replicates.

Table 1 Effects of Cd on growth of vegetables

Treatment level (mg/kg)	Fresh weight (g/pot)			
	Cole root	Cole shoot	Celery root	Celery shoot
0.00	0.46 ± 0.26 a	14.61 ± 1.90 a	$0.37 \pm 0.20 \text{ a}$	2.96 ± 1.81 a
0.35	0.59 ± 0.13 ab	$14.38 \pm 1.95 a$	$0.35 \pm 0.05 a$	1.50 ± 0.17 a
0.70	$0.49 \pm 0.07 a$	$13.77 \pm 3.16 a$	$0.88 \pm 0.15 \text{ ab}$	$5.82 \pm 0.74 \text{ b}$
1.05	0.55 ± 0.03 ab	$15.68 \pm 2.08 a$	$0.96 \pm 0.18 \mathrm{b}$	$6.73 \pm 2.74 \mathrm{b}$
1.40	$0.58 \pm 0.01 a$	$17.09 \pm 1.50 a$	$1.00 \pm 0.20 \text{ b}$	4.94 ± 0.42 ab
2.10	$1.08 \pm 0.53 \text{ b}$	$27.56 \pm 6.04 \text{ b}$	$0.86 \pm 0.56 \text{ ab}$	$3.85 \pm 1.97 \text{ ab}$
3.50	$1.07 \pm 0.48 \text{ b}$	21.11 ± 3.22 ab	$0.84 \pm 0.53 \text{ ab}$	$2.96 \pm 1.52 a$
4.90	0.73 ± 0.19 ab	$17.68 \pm 5.75 a$	$1.05 \pm 0.21 \text{ b}$	4.37 ± 0.62 ab
7.00	$0.87 \pm 0.40 \text{ ab}$	21.03 ± 10.17 ab	0.62 ± 0.42 ab	$3.10 \pm 2.69 a$

Values are presented as mean \pm SD of three replicates. One-way ANOVA (1 factor: different Cd treatments) was performed for each parameter on Cd concentration. Data in the same column followed by different letters are significantly different from each other (p < 0.05) according to the LSD test.

Table 2 Concentration of Cd in vegetables under Cd treatments

Treatment level (mg/kg)	Cd concentration (mg/kg fw)				
	Cole root	Cole shoot	Celery root	Celery shoot	
0.00	0.52 ± 0.22 a	0.08 ± 0.00 a	0.03 ± 0.02 a	0.01 ± 0.00 a	
0.35	0.61 ± 0.11 a	0.42 ± 0.09 a	1.13 ± 0.07 ab	0.40 ± 0.03 a	
0.70	$0.72 \pm 0.05 a$	0.81 ± 0.16 a	$2.34 \pm 0.73 \text{ b}$	0.65 ± 0.20 at	
1.05	$3.00 \pm 0.72 \text{ ab}$	1.42 ± 0.19 ab	$3.40 \pm 0.54 \mathrm{b}$	0.79 ± 0.04 at	
1.40	$4.30 \pm 0.60 \mathrm{b}$	1.48 ± 0.01 ab	$3.21 \pm 0.43 \text{ b}$	0.82 ± 0.16 at	
2.10	$4.68 \pm 2.45 \mathrm{b}$	2.21 ± 0.86 bc	$5.57 \pm 1.11 \mathrm{c}$	1.54 ± 0.23 b	
3.50	$4.75 \pm 1.00 \mathrm{b}$	3.25 ± 0.89 bc	$9.28 \pm 2.87 \mathrm{c}$	2.49 ± 0.55 bo	
4.90	$8.45 \pm 2.05 \mathrm{c}$	3.23 ± 0.71 bc	$12.43 \pm 1.11 d$	2.90 ± 0.72 bo	
7.00	11.72 ± 3.39 c	4.54 ± 0.64 c	$14.30 \pm 5.32 d$	$3.69 \pm 3.07 \mathrm{c}$	

Values are presented as mean \pm SD of three replicates. One-way ANOVA (1 factor: different Cd treatments) were performed for each parameter on Cd concentration. Data in the same column followed by different letters are significantly different from each other (p < 0.05) according to the LSD test.

Cd concentration accumulated in vegetables increased significantly (p < 0.001) with increasing soil Cd concentrations. Compared with the control, cole and celery shoots Cd concentrations increased by 55.8 folds and 368.0 folds, respectively at the Cd level of 7.00 mg/kg.

According to the China Environmental Quality Standard for Soils (GB 15618-1995), Cd limits for the soils used for vegetable production are 0.3 and 0.6 mg/kg for pH < 7.5 and pH > 7.5, respectively. At a Cd level of 0.6 mg/kg, Cd concentration in vegetables was 1.24 mg/kg for cole (calculated by statistical regression equation $y = 1.0737 \ln x + 1.79$, $R^2 = 0.949$, p < 0.001, where y is Cd concentration in shoots, x is the concentration of Cd in soils) and 0.75 mg/kg for celery (calculated by $y = 0.8575 \ln x + 1.1871$, $R^2 = 0.931$, p < 0.001). At this level of Cd addition, the national allowable limits of Cd in vegetables (GB2762-2005, maximum levels of contaminants in foods, 0.2 mg/kg for both cole and celery) were exceeded in both cole and celery. In general, the two vegetables accumulated higher Cd concentrations in their edible parts.

Many studies indicated that cole (*Brassica*) had high accumulation for Cd, and even could be considered as a Cd hyperaccumulator (Chen et al., 1990; Su and Wong, 2002; Liu et al., 2002; Wang et al., 2003, 2005; Ru et al., 2005; Xu et al., 2007; Xiang et al., 2009; Wu and Su, 2009; Zhang et al., 2009). Moreover, some researchers observed the BCF values of Cd for cole in acidic soils were higher than that in the basic soils. And their studies showed the BCF of Cd was ranged from 6.62–472.15 for roots, 6.05–284.83 for shoots, respectively in red soil (Xu et al., 2007); 2.12–4.09 for roots, 1.03–2.90 for shoots in Cinnamon

soil (Liu et al., 2002); 1.98–5.95 for roots, 1.29–1.26 for shoots, in Lou soil (Zhang et al., 2009). Also, Li et al. (2002) found Cd had higher removal rate from red soil to celery, ranged from 0.522% to 8.796%.

Similar results were observed in the present study. The BCF of Cd in vegetables roots and shoots under various treatments are shown in Table 3. The BCF value of Cd incole was ranged between 0.26–3.94 for roots, 0.81–1.85 for shoots; while 1.66–4.11 for roots, 0.72–1.01 for shoots in celery. In general, the BCF of Cd in cole root was lower than in celery root, and that in cole shoot was higher than that in celery shoot.

Sun et al. (2003) found that the cumulative coefficient of heavy metals was higher under low heavy metal doses, on the contrary, the cumulative coefficient of heavy metals was low under high dose, but the absolute amount of heavy metals accumulated increased with increasing heavy metals dose. Similar results were observed in the present study, the BCF in roots and shoots increased firstly, and reached maximum value, then decreased with increasing soil Cd concentrations.

The TFs of Cd in cole were greater than those in celery (Table 3), suggesting that Cd had a greater mobility in cole than that in celery.

The above-mentioned results showed that the BCF of Cd in cole and celery had threshold values, beyond them, the roots of cole and celery would be damaged by Cd, although the uptake and accumulation of Cd in cole and celery had being continued, but their uptake rates gradually reduced, hence the BCF of Cd in cole and celery increased at first, then decreased. The above results also suggested

Table 3 Bio-concentration factor (BCF) and translocation factor (TF) of Cd in vegetables under Cd treatments

Treatment level (mg/kg)		Cole		Celery		
	BCF _{root}	BCF _{shoot}	TF	BCF _{root}	BCF _{shoot}	TF
0.00	0.26	0.81	3.11	1.66	0.72	0.43
0.35	1.27	1.04	0.82	2.62	0.94	0.36
0.70	1.31	1.44	1.10	3.60	1.01	0.28
1.05	3.94	1.85	0.47	4.11	0.96	0.23
1.40	3.46	1.48	0.43	3.01	0.76	0.25
2.10	2.33	1.42	0.61	3.52	0.89	0.25
3.50	1.56	0.88	0.56	2.08	0.55	0.26
4.90	1.52	0.65	0.43	2.10	0.48	0.23
7.00	1.64	0.59	0.36	1.69	0.42	0.25

Table 4 Correlation coefficient (*R*) between the F1 distribution coefficient (FDC) of Cd in the oasis soil and concentrations of Cd in vegetables root and shoot

Chemical fractions	R (cole)		R (celery)	
	Root	Shoot	Root	Shoot
F1	0.849**	0.909**	0.925**	0.959**
F2	0.481	0.569	-0.162	-0.195
F3	-0.817**	-0.900**	-0.868**	-0.880**
F4	-0.665*	-0.739*	-0.817**	-0.837**
F5	-0.719*	-0.808**	-0.666*	-0.679*

^{*} Significant at level p < 0.05, ** significant at level p < 0.01.

that Cd mainly accumulated in the edible parts of the two vegetables above the ground in polluted oasis soil. Therefore, cole and celery grown in the Cd-polluted oasis soil, consumed by human have high risk to human health.

2.4 Relationship between the accumulation in vegetables and chemical fractions of Cd in the oasis soil

Significantly positive correlations were found between the FDCs of Cd in F1 in the oasis soil and Cd concentrations in vegetables roots and shoots (Table 4). The corresponding correlation coefficients (*R*) were 0.849 for roots and 0.909 for shoots of cole; and 0.925 for roots and 0.959 for shoots of celery.

Zhu et al. (2002) reported exchangeable Cd were highest available and made the most contribution to the lettuce plant in purple soil. Cui et al. (2005) found that exchangeable Cd were extractable by reed plant. Similar results were observed in present study due to exchangeable Cd (F1) was available fraction to both cole and celery, which indicated that Cd in F1 made the greatest contributions on the accumulation of Cd in the roots and shoots of both cole and celery.

3 Conclusions

Cadmium (Cd) in the unpolluted oasis soil was mainly bound to the Fe-Mn oxide fractionation (F3) and carbonate fractionation (F2). However, the fractions distribution of Cd changed significantly with increasing Cd concentration in the soil, in which concentrations of Cd in exchangeable fractionation (F1), those bound to carbonate fractionation (F2) and Fe-Mn oxide fractionation (F3) increased significantly.

Low concentrations of Cd could facilitate the growth of cole and celery, but their growth was inhibited by high concentrations. The Cd in F1 made the greatest contributions on the accumulation of Cd in the two vegetables. The translocation factor of Cd in cole was greater than in celery, and Cd had higher accumulation in the edible parts of both cole and celery.

Therefore, cole and celery grown in the Cd-polluted oasis soil have highly potential risk to human health.

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