



Effects of cadmium on uptake and translocation of nutrient elements in different welsh onion (*Allium fistulosum* L.) cultivars



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ABSTRACT

The concentration of nutrient elements is an important quality characteristic of vegetables, and the variation in accumulation among cultivars can provide clues about the mechanism of low accumulation of heavy metals. Pot-culture experiments were arranged under four cadmium (Cd) treatments (CK, 1.0, 2.5 and 5.0 mg/kg) to explore influences of Cd on the accumulation of nutrient elements in 25 welsh onion cultivars. There were significant positive correlations ($p < 0.05$) between Cd and nutrient elements in the pseudostems and leaves. There were also significant positive correlations in nutrient elements ($p < 0.05$) among cultivars, which might be disturbed under high Cd treatments, especially for P, Fe and Mn. Our results suggested that there is a synergistic effect on the accumulation between Cd and nutrient elements, and within nutrient elements among cultivars. In addition the uptake and translocation process of Cd was closely related to Mn in welsh onion.

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1. Introduction

Cadmium (Cd), as an important heavy metal with high bioavailability in soils, easily enters into ecosystems and accumulates through food chain, thus posing a significant threat to human and animal health. It is one of the toxic metals under scrutiny by the US Environmental Protection Agency (USEPA). In China, Cd contamination has been considered as a serious problem for food safety (Bao et al., 2013). According to the China's Soil Pollution State Bulletin (the Ministry of Environmental Protection of PRC & Resources of PRC, 2014), the rate of Cd exceeding the standard was more than 7.0%. The primary concern with Cd is its transfer from vegetables to human bodies, because vegetables contribute more than 70% of Cd intake in human beings (Sarwar et al., 2010). At the same time, the techniques available for remediating the soil contaminated by Cd are costly and/or time consuming, which limits their application in most cases of lesser Cd contamination in the soil (Li, Zhou, Wei, & Ren, 2012). Recently, an alternative strategy to reduce the risk of soil Cd entering into food chain has been proposed, namely, the identification of Cd-excluding cultivars (Li et al., 2012). Cd-excluding cultivars are defined as

cultivars that accumulate Cd at a low enough level in the edible parts of a crop to allow for safe consumption, even when grown in soils with slight or moderate Cd contamination (Li et al., 2012; Liu, Zhou, Sun, & Liu, 2009; Zhou, Kong, & Zhu, 2004).

The concept of Cd-excluding cultivars is based on the theory that there is a significant difference in the uptake and distribution of toxic elements among crop species and cultivars within the same species, which has been clearly illustrated by several studies (Arivalagan et al., 2012; Chen et al., 2007; Fernandez-Ruiz, Olives, Camara, Sanchez-Mata, & Torija, 2011; Galdon, Gonzalez, Rodriguez, & Romero, 2008; Galdon et al., 2012; Singh, Sharma, & Singh, 2012). Differences in Cd accumulation levels among plant cultivars indicate variability in uptake and/or translocation of elements, including but not limited to Cd. Singh et al. (2012) found that both parentage and the breeding approach significantly affected the concentration of Fe, Zn, Cu, Mn, K and Ca in cabbage (*Brassica oleracea* var. *capitata* L.). The cultivars are considered to be one of the most important factors affecting the chemical composition of the potato (*Solanum tuberosum* L.) (Galdon et al., 2012). Significant differences in mineral composition among the eggplant (*Solanum melongena* L.) genotypes have also been observed by Arivalagan et al. (2012). Visible differences have been observed in the mineral contents among 202 rice (*Oryza sativa* L.) cultivars, and there was also a negative correlation between grain

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yield and mineral contents (Anandan, Rajiv, Eswaran, & Prakash, 2011). Similar results have also been reported in wheat (*Triticum aestivum* L.) (Zhang et al., 2010), Walnut (*Juglans regia* L.) (Cosmulescu, Baci, Achim, Botu, & Trandafir, 2009), onion (*Allium cepa* L.) (Galdon et al., 2008), tomato (*Lycopersicon esculentum* L.) (Suarez, Rodriguez, & Romero, 2007), grape (*Vitis vinifera* L.) (Yang, Duan, Du, Tian, & Pan, 2010), and banana (*Musa paradisiaca* L.) (Sulaiman et al., 2011).

The concentration of nutrient elements in plants and crops could also be influenced by other elements, especially by heavy metals. However, the response of uptake and translocation of nutrient elements to heavy metal pollution might be different, even in net direction, depending on the factor such as plant species and element types. Sikka and Nayyar (2012) found a decrease in the content of micronutrients (Mn, Fe, Cu, Zn) in Indian mustard [*Brassica juncea* (L.) Czern.] with the application of Cd, but a significant reduction in Fe occurred only in treatments with Cd concentrations above 50 mg/kg. Meanwhile, Liu, He, and Chen (2011) found a synergistic interaction in accumulation and translocation between Cd and Fe and a significant negative correlation between Cd and Cu or Zn concentrations in *Lonicera japonica* Thunb. Furthermore, the influence of heavy metals on nutrient elements vary by cultivar. Norton et al. (2010) found that Se, Cu, Mn and Ni concentrations in rice grain (*Oryza sativa* L.) showed significant genetic interaction with arsenic (As). Goncalves et al. (2009) also suggested that the influence of Cd on the nutrient contents in potato (*Solanum tuberosum*) was related to the level of Cd in the substrate, the potato cultivar, plant organ, essential element, growth medium and exposure time.

The nutrient elements not only are important quality characteristics of vegetables, but also provide clues on the mechanism of low Cd accumulation. With a cultivation history of approximately 2500 years, welsh onion is one of the most important vegetables in China and other countries of Northeast Asia, Europe, and Northern America. Its cultivation area and production accounted for approximately 3% of the total vegetables in China. Our previous study showed that significant differences occurred in Cd accumulation among welsh onion cultivars (Li et al., 2012), but the nutrient elements have yet to be examined in welsh onion cultivars with different Cd accumulation characteristics. Moreover, most of the studies on how the heavy metals influence on uptake and translocation of nutrients were carried out in hydroponic experiments with very few cultivars. Hence, the present study was carried out in the pot-culture with 25 welsh onion cultivars and focused on (1) the influence of Cd on the accumulation of nutrient elements in welsh onion; (2) the uptake/translocation relationship between nutrient elements and Cd in different welsh onion cultivars; and (3) the variation in the uptake and translocation of nutrient elements with cultivars as affected by Cd.

2. Materials and methods

2.1. Experimental design and soil characteristics

A pot-culture experiment was conducted under open field conditions at the Shenyang Station of Experimental Ecology, Chinese Academy of Sciences (41°31'N and 123°41'E). The meteorological conditions of the station are listed as follows: the average annual temperature is approximately 5–9 °C, the annual precipitation is 650–700 mm, and the frostless period lasts for 127–164 days.

Brown earth was used in the experiment. It is distributed widely in Northeastern Asia, which is also one of the most important production areas of welsh onion. The soil used in the experiment was collected from the agricultural area of the station. The surface layer of the soil (0–20 cm depth) was collected, air-dried,

grounded and sieved through a 4-mm mesh. According to China's environmental quality standard for soils (GB 15618-1995), the limiting value of soil Cd concentration is 0.30 mg/kg for farmland. And in most of the countries, this value is not more than 0.50 mg/kg. The triple limiting value (0.90 mg/kg) is considered to be the distinguishing value of the slight and moderate contamination. And more than 85% of the Cd contamination was classified as the slight level (the Ministry of Environmental Protection of PRC & Resources of PRC, 2014). As Cd posed a serious threat to human health, a strict screening experiment was carried out. Hence, in the present study, the minimum concentration of Cd addition was set as 1.0 mg/kg. Cd-excluding cultivars screened out under this experiment would ensure the food safety when grown in most of the Cd contamination soils. As one of the main purpose of the present study was to explore the relationship between Cd and nutrient elements accumulation in welsh onion, the two treatments with higher Cd addition were also implemented. Four treatments, including the control (without addition of Cd, marked with CK) and three treatments with 1.0, 2.5 and 5.0 mg/kg Cd addition (marked with T1, T2 and T3, respectively), were implemented in the present study. All of the treatments were carried out in triplicate. Solutions with different concentrations of Cd but the same concentration of NO_3^- (NaNO_3 was added to make sure that the same amount of NO_3^- was introduced to soils with different Cd concentrations) were prepared. A total of 2.5 kg soil was weighed into each plastic pot ($\varphi \times H = 20 \text{ cm} \times 15 \text{ cm}$) and aged for 2 months.

Seeds of 25 welsh onion cultivars were collected from the local market in Shenyang, Liaoning Province, China, planted in the pots directly in May, and cultivated under open field condition without fertilizer application. These pots were arranged in a randomized complete block design with 3 replicates. Groundwater (no Cd detected) was used in the irrigation every day (keeping the content of soil moisture at approximately 70% of the field water-holding capacity). When planted in fields, welsh onion seedlings are replanted to a vegetable field after breeding. Hence, the seedlings in the pot were removed and seven plants with uniform growth were selected and replanted back into the same pots in 8 weeks after the seeds were sown. The plants were harvested in mid-October.

2.2. Sample preparation and analysis

The soil samples were ground to a powder and sieved through a 0.85-mm mesh for analysis of pH and cation exchange capacity (CEC) and through a 0.15-mm mesh for the analysis of soil organic carbon, organic nitrogen, total nitrogen (TN), potassium (K), calcium (Ca), magnesium (Mg), iron (Fe), zinc (Zn), copper (Cu), manganese (Mn) and phosphors (P). The contents of organic C and organic N were determined by elemental analysis on a Vario EL III elemental analyzer (Germany). The soil samples were digested with a mixture of $\text{HNO}_3\text{--HClO}_4\text{--HF}$ (15:2:2, v/v) for the determination of the concentrations of P, K, Ca, Mg, Fe, Zn, Cu, and Mn using inductively coupled plasma optical emission spectrometry (ICP-OES, Optima 3000, PerkinElmer Ins. USA). Other characteristics were analyzed according to routine analytical methods for agricultural chemistry in soils. Some characteristics of the soil used in the experiment were as follows: pH value, 5.70; CEC, 17.99 cmol/kg; organic C, 1.14%; organic N, 0.11%; total N, 0.89%; total P, 3.50 g/kg; total K, 10.96 g/kg. The concentrations of Ca, Mg, Fe, Zn, Cu and Mn were 9.86, 8.32, 31.89 g/kg 65.68, 18.11 and 779.70 mg/kg, respectively.

After being harvested, the plants were rinsed with tap water to remove soil, washed with deionized water for 3 min., wiped clean with filter paper and then separated into roots, pseudostems and leaves to investigate the distribution of elements. The plant samples were dried at 105 °C for 5 min, and then at 70 °C until constant

weights were obtained. Fresh and dry weights of the samples were measured. The dried samples were powdered, and 0.50 g of the sample was digested with 15 ml of mixed acid containing concentrated HNO₃ and HClO₄ (87:13, v/v). The concentration of Cd in digested solution was determined using an atomic absorption spectrophotometer (FAAS, Varian AA 140/240, Varian Inc., Australia), and the concentrations of the nutrient elements including P, K, Ca, Mg, Fe, Zn, Cu and Mn were detected using ICP-OES (Optima 3000, PerkinElmer Ins. USA). The bush leaf material (GBW07603, Qinghai Province, China) was used as a certified reference material for the quality control of the analysis process. The chemicals used in the analysis process were all guarantee reagents.

2.3. Data processing and statistical analysis

Data processing was carried out using Excel 2007 (Microsoft, USA) and SPSS18.0 (SPSS Corporation, USA). The Kolmogorov–Smirnov test was applied to verify whether the distribution of the variables was normal. The significance analysis was performed using one-way ANOVA (LSD test and Student–Newman–Keuls test), and the correlation analysis was conducted using the Pearson method.

3. Results

3.1. Cd accumulation in edible parts of welsh onions

The pseudostem of welsh onion is the main edible part of the crop. The change in Cd concentration in edible parts among various cultivars is presented in Fig. 1 based on its fresh weight. The cultivars are listed in the increasing order of Cd accumulation in the T1 treatment. Significant differences ($p < 0.05$, LSD test) in the Cd accumulation in the pseudostem were observed. In the T1 treatment, the accumulation of Cd in the pseudostem ranged from 0.08 to 0.20 mg/kg with an average value of 0.14 mg/kg. Considering the maximum permission concentration (MPC) of Cd (0.10 mg/kg in fresh weight) for vegetables, only 8% (2/25) of the cultivars tested met the MPC under our strict screening experiment. For treatments with higher concentrations of Cd, all of the cultivars tested exceeded the MPC. The concentrations of Cd in the pseudostem varied from 0.18 to 0.41 mg/kg and from 0.26 to 0.61 mg/kg, with mean values of 0.29 in treatment T2 and

0.44 mg/kg in T3. Moreover, cultivars followed different orders of Cd accumulation in different treatments.

3.2. Nutrient element concentrations in welsh onions as affected by Cd

The concentrations of nutrient elements in the tissues of welsh onion were influenced by Cd and varied by the type of element and by the plant tissue. As shown in Fig. 2, the concentrations of the 8 elements in roots decreased in the T1 treatment, although significant differences ($p < 0.05$) were only found for P, Ca, Mg, Fe, Cu and Mn in some tissues. When the addition of Cd exceeded 2.5 mg/kg, the concentrations of K and Zn increased significantly in all tissues of welsh onion. The increases in K concentration in the roots, pseudostems and leaves were 21–24%, 111–116%, and 66–74%, respectively. The responding value was 28–34%, 27–29% and 40–58% for Zn, respectively. The concentrations of Ca, Mg and Fe were similarly influenced by Cd, and the effect was a bit different for those of K and Zn. Except for Fe in the pseudostems, the concentrations of these elements decreased under low Cd treatment (T1), but increased under T2 and T3. Under the T1 treatment, the decrease in the concentration of Ca in pseudostems and leaves, Mg in roots, and Fe in roots and leaves was significant ($p < 0.05$) in a range of 10–19%, while no significant difference ($p > 0.05$) was found for others. Under the T3 treatment, a significant increase was observed at the $p = 0.05$ level for Ca and Fe in roots and leaves, and for Mg in leaves compared with CK. When compared with T1, all concentrations of these 3 elements (Ca, Mg and Fe) significantly increased ($p < 0.05$) in all tissues (except for Fe in the pseudostems).

3.3. Relationship between Cd and nutrient element concentrations among cultivars

A correlation analysis was carried out on nutrient elements and Cd concentrations in cultivars to examine the relationship between the uptake and translocation of Cd and between the nutrient elements among cultivars, and the results were listed in Table 1. Only the concentrations of Mn showed highly significant positive correlation ($p < 0.01$, Pearson test) with Cd concentrations in roots with correlation coefficients of 0.471, 0.561 and 0.393 under T1, T2 and T3 treatments, respectively, while most of the nutrient element concentrations displayed the significant positive correlation ($p < 0.05$, Pearson test) with Cd concentrations in the pseudostems and leaves. Furthermore, the correlation coefficients between the

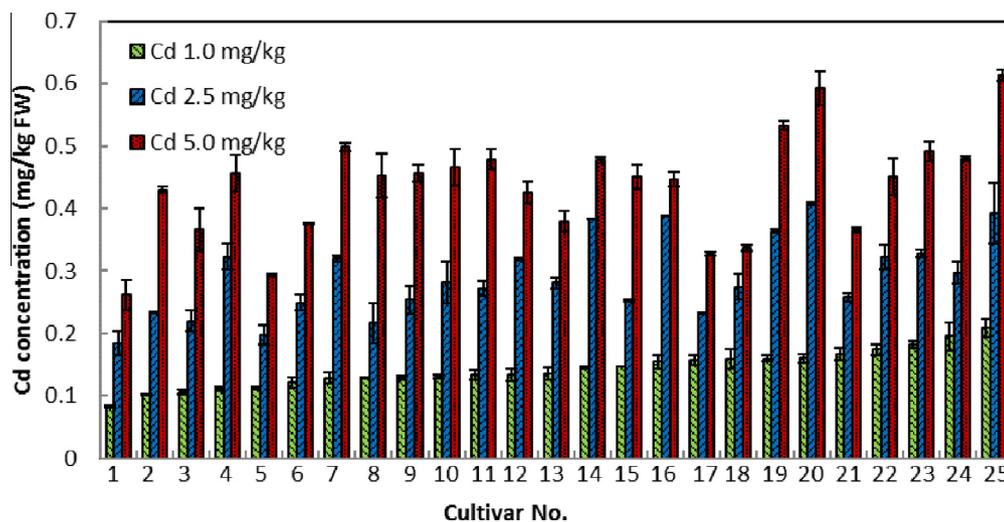


Fig. 1. Cd accumulation in edible parts of different cultivars (mean \pm SD).

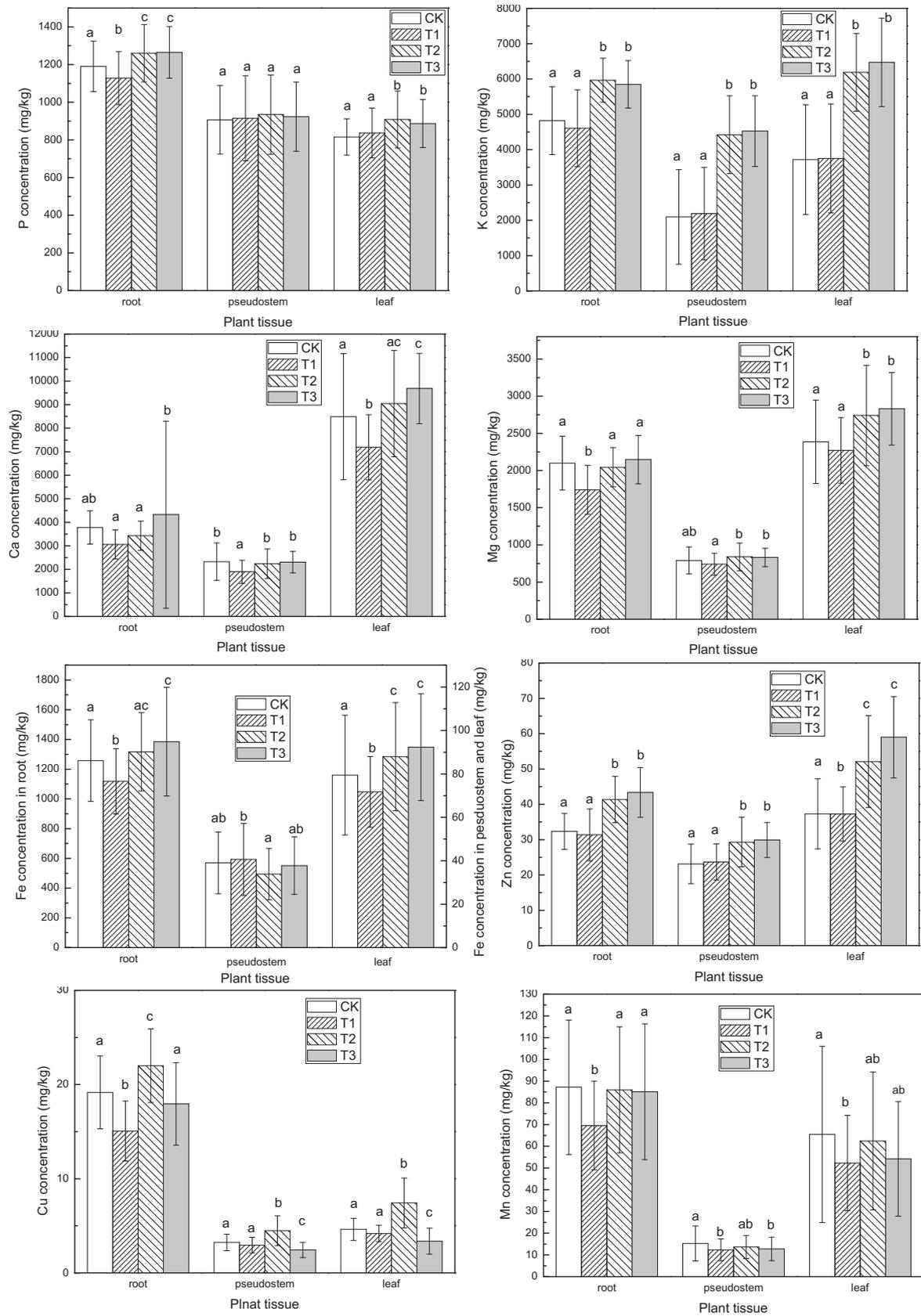


Fig. 2. The concentration of nutrient element in different welsh onion tissues under different Cd treatments (mean ± SD). * Results in the same tissue with the same letter were not significantly ($p < 0.05$, S-N-K test) different.

Table 1Correlation analysis results between Cd and nutrient elements among welsh onion cultivars ($n = 75$, Pearson test).

Treatment	Tissue		Elements							
			P	K	Ca	Mg	Fe	Zn	Cu	Mn
T1	Root	Cor. Coe.	-0.045	-0.212	-0.014	-0.041	-0.049	-0.059	0.016	0.471 ^b
		Sig.	0.701	0.068	0.906	0.727	0.675	0.613	0.892	0.000
	Pseudostem	Cor. Coe.	0.192	0.082	0.510 ^b	0.496 ^b	0.222	0.352 ^b	0.092	0.589 ^b
		Sig.	0.098	0.485	0.000	0.000	0.056	0.002	0.430	0.000
	Leaf	Cor. Coe.	0.267 ^a	0.388 ^b	0.485 ^a	0.538 ^b	0.249 ^a	0.523 ^b	0.543 ^b	0.617 ^b
		Sig.	0.020	0.001	0.000	0.000	0.031	0.000	0.000	0.000
T2	Root	Cor. Coe.	0.164	0.187	-0.079	0.226	-0.128	0.011	0.135	0.561 ^b
		Sig.	0.159	0.108	0.500	0.051	0.275	0.926	0.248	0.000
	Pseudostem	Cor. Coe.	0.301 ^a	0.439 ^b	0.553 ^b	0.694 ^b	0.653 ^b	0.638 ^b	0.362 ^b	0.610 ^b
		Sig.	0.009	0.000	0.000	0.000	0.000	0.000	0.000	0.000
	Leaf	Cor. Coe.	0.062	0.351 ^b	0.392 ^b	0.481 ^b	0.541 ^b	0.535 ^b	0.364 ^b	0.742 ^b
		Sig.	0.598	0.002	0.001	0.000	0.000	0.000	0.001	0.000
T3	Root	Cor. Coe.	-0.210	-0.217	-0.172	0.243 ^a	0.030	0.273 ^a	0.045	0.393 ^b
		Sig.	0.071	0.061	0.140	0.036	0.796	0.018	0.704	0.000
	Pseudostem	Cor. Coe.	-0.172	0.316 ^b	0.393 ^b	0.283 ^a	0.150	0.288 ^b	0.084	0.628 ^b
		Sig.	0.139	0.006	0.000	0.014	0.199	0.012	0.474	0.000
	Leaf	Cor. Coe.	-0.072	0.201	0.753 ^b	0.776 ^b	0.508 ^b	0.763 ^b	0.144	0.656 ^b
		Sig.	0.538	0.083	0.000	0.000	0.000	0.000	0.219	0.000

Cor. Coe.: Correlation Coefficient.

^a Correlation is significant at the 0.05 level (2-tailed).^b Correlation is significant at the 0.01 level (2-tailed).

concentrations of nutrient elements and Cd in the leaves were higher than those in the pseudostems when significant correlations ($p < 0.05$, Pearson test) were observed, indicating that the correlation coefficients could be enlarged upon translocation of the nutrient elements.

3.4. Variation on concentration relationships between nutrient elements among cultivars under different Cd treatments

Correlation analysis was also carried out to examine the variation in concentrations among cultivars affected by Cd. Significant positive correlations ($p < 0.05$, Pearson test) were found among the concentrations of K, Ca, Mg, Zn and Cu in most of the welsh onion tissues under all treatments (except for those between K and Ca, Mg, Zn or Cu in roots under the T2 treatment, between Ca and K or Cu in roots, between K and Ca or Cu in pseudostems, and between Cu and K, Ca, Mg or Zn in leaves under the T3 treatment).

Although the concentrations of P, Fe or Mn and other 7 elements were also positively correlated ($p < 0.05$, Pearson test) in most samples, insignificant correlations ($p > 0.05$, Pearson test) or significant negative correlations ($p < 0.05$, Pearson test) results were also observed.

As shown in Table 2, significant positive correlations between P and Mn concentrations were found in leaves subjected to the CK treatment, in all tissues subjected to the T1 treatment and in pseudostems subjected to the T2 treatment ($p < 0.05$, Pearson test), but highly negative correlations ($p < 0.01$, Pearson test) were displayed in all tissues subjected to the T3 treatment with correlation coefficients of -0.387 , -0.399 and -0.288 for roots, pseudostems, and leaves, respectively. For the other six elements, highly significant positive correlations with P ($p < 0.01$, Pearson test) were widely observed in the treatments with soil Cd concentrations below 2.5 mg/kg. When the soil Cd concentration reached 5.0 mg/kg, much less positive correlations were observed, especially in leaves.

The concentration of Fe in the roots showed highly significant positive correlations ($p < 0.01$, Pearson test) with the concentrations of Mg, Zn, Cu, and Mn, and significant positive correlations ($p < 0.05$, Pearson test) with the concentrations of K and Ca in the CK treatment. After having added Cd, most of the highly significant or significant positive correlations disappeared. When the soil Cd

concentration reached 5.0 mg/kg, highly significant positive correlations ($p < 0.01$, Pearson test) were only found for Fe–Mg and Fe–Zn.

When Cd was added, the similar disappearance of significant positive correlations also disappeared for Mn and other elements. The concentrations of Mn showed highly significant positive correlations ($p < 0.01$, Pearson test) with the concentrations of Ca, Mg, Fe, Zn and Cu under the CK treatment, while there were no significant positive correlations ($p > 0.05$, Pearson test) in roots under the T3 treatment. Positive correlations on Mn and those 6 elements in pseudostems were observed at the highly significant level ($p < 0.01$, Pearson test) in treatments with soil Cd concentrations below 2.5 mg/kg. However, the highly significant correlations ($p < 0.01$, Pearson test) were only found between Mn and Ca in pseudostems and between Mn and Ca, Mg, Fe, and Cu in leaves under the 5.0 mg/kg Cd treatment. All these results indicated that the addition of Cd broke the balance of uptake and translocation of nutrient elements in welsh onion.

3.5. Uptake and translocation characters of nutrient elements

Some accumulation and transfer indexes of nutrient elements, such as the bioaccumulation factor of roots (BF_r), the translocation factor from roots to pseudostems (TF_{r-p}) and translocation factor from pseudostems to leaves (TF_{p-l}), were introduced to analyze the uptake and translocation of nutrient elements. $BF_{i,r}$, $TF_{i,r-p}$ and $TF_{i,p-l}$ were defined as the ratio of the concentration of Element I in roots ($C_{i,r}$) to that in soils ($C_{i,s}$) (Eq. (1)), that in pseudostems ($C_{i,p}$) to that in roots (Eq. (2)), and that in leaves ($C_{i,l}$) to that in pseudostems (Eq. (3)), respectively. All results were on the basis of dry weight.

$$BF_{i,r} = \frac{C_{i,r}}{C_{i,s}} \quad (1)$$

$$TF_{i,r-p} = \frac{C_{i,p}}{C_{i,r}} \quad (2)$$

and

$$TF_{i,p-l} = \frac{C_{i,l}}{C_{i,p}} \quad (3)$$

Table 2
Correlation analysis results between P, Fe Mn and other nutrient elements among welsh onion cultivars ($n = 75$, Pearson test).

		P			Fe			Mn		
		R	P	L	R	P	L	R	P	L
CK	P	–	–	–	0.008	0.358 ^b	0.337 ^b	0.146	0.106	0.251 ^a
	K	0.534 ^b	0.182	0.307 ^b	0.288 ^a	0.377 ^b	0.686 ^b	0.188	0.491 ^b	0.608 ^b
	Ca	0.432 ^b	0.349 ^b	0.350 ^b	0.284 ^a	0.417 ^b	0.813 ^b	0.389 ^b	0.519 ^b	0.761 ^b
	Mg	0.515 ^b	0.564 ^b	0.360 ^b	0.444 ^b	0.567 ^b	0.753 ^b	0.645 ^b	0.529 ^b	0.733 ^b
	Fe	0.008	0.358 ^b	0.337 ^b	–	–	–	0.464 ^b	0.524 ^b	0.643 ^b
	Zn	0.581 ^b	0.575 ^b	0.319 ^b	0.315 ^b	0.788 ^b	0.780 ^b	0.388 ^b	0.585 ^b	0.664 ^b
	Cu	0.343 ^b	0.691 ^b	0.420 ^b	0.310 ^b	0.681 ^b	0.816 ^b	0.380 ^b	0.492 ^b	0.694 ^b
	Mn	0.146	0.106	0.251 ^a	0.464 ^b	0.524 ^b	0.643 ^b	–	–	–
	T1	P	–	–	–	0.253 ^a	0.170	0.219	0.228 ^a	0.514 ^b
K		0.410 ^b	0.515 ^b	0.383 ^b	0.018	0.451 ^b	0.542 ^b	–0.296	0.591 ^b	0.533 ^b
Ca		0.601 ^b	0.416 ^b	0.386 ^b	0.104	0.119	0.648 ^b	0.223	0.666 ^b	0.701 ^b
Mg		0.475 ^b	0.758 ^b	0.469 ^b	0.253 ^b	0.183	0.523 ^b	0.175	0.792 ^b	0.755 ^b
Fe		0.253 ^a	0.170	0.219	–	–	–	–0.001	0.392 ^b	0.374 ^b
Zn		0.676 ^b	0.724 ^b	0.529 ^b	0.296 ^b	0.359 ^b	0.633 ^b	–0.013	0.707 ^b	0.747 ^b
Cu		0.516 ^b	0.757 ^b	0.662 ^b	0.240 ^a	0.455 ^b	0.542 ^b	–0.048	0.660 ^b	0.620 ^b
Mn		0.228 ^a	0.514 ^b	0.355 ^b	–0.001	0.392 ^b	0.374 ^b	–	–	–
T2		P	–	–	–	0.021	0.358 ^b	0.279 ^b	0.056	0.181 ^a
	K	0.291 ^b	0.539 ^b	0.407 ^b	–0.035	0.503 ^b	0.299 ^b	–0.182 ^a	0.334 ^b	0.175 ^a
	Ca	0.022	0.379 ^b	0.318 ^b	0.021	0.321 ^b	0.569 ^b	0.199 ^a	0.346 ^b	0.444 ^b
	Mg	0.206 ^b	0.499 ^b	0.334 ^b	0.092	0.507 ^b	0.629 ^b	0.227 ^b	0.542 ^b	0.487 ^b
	Fe	0.021	0.358 ^b	0.279 ^b	–	–	–	–0.042	0.413 ^b	0.352 ^b
	Zn	0.266 ^b	0.590 ^b	0.377 ^b	0.120	0.523 ^b	0.614 ^b	0.143	0.480 ^b	0.478 ^b
	Cu	0.421 ^b	0.509 ^b	0.359 ^b	0.173 ^a	0.490 ^b	0.565 ^b	0.150	0.494 ^b	0.436 ^b
	Mn	0.056	0.181 ^a	0.108	–0.042	0.413 ^b	0.352 ^b	–	–	–
	T3	P	–	–	–	0.218	0.572 ^b	0.227	–0.387 ^b	–0.399 ^b
K		0.666 ^b	0.668 ^b	0.675 ^b	0.165	0.484 ^a	0.377 ^b	–0.108	0.054	–0.084
Ca		0.136	0.141	0.073	0.055	0.189	0.653 ^b	0.216	0.312 ^b	0.544 ^b
Mg		0.451 ^b	0.523 ^b	0.054	0.333 ^b	0.558 ^b	0.622 ^b	0.035	0.157	0.539 ^b
Fe		0.218	0.572 ^b	0.227	–	–	–	0.164	0.030	0.468 ^b
Zn		0.411 ^b	0.629 ^b	0.270 ^a	0.303 ^b	0.620 ^b	0.694 ^b	–0.077	0.037	0.350 ^b
Cu		0.278 ^a	0.177	–0.047	–0.014	0.195	–0.218	–0.099	0.060	0.053
Mn		–0.387 ^b	–0.399 ^b	–0.288 ^b	0.164	0.030	0.468 ^b	–	–	–

R, P, and L represent root, pseudostem, and root of welsh onion, respectively.

^a Correlation is significant at the 0.05 level (2-tailed).

^b Correlation is significant at the 0.01 level (2-tailed).

The BF_r values were 0.32–0.36 for P, 0.41–0.54 for K, 0.31–0.44 for Ca, 0.21–0.26 for Mg, 0.35–0.43 for Fe, 0.47–0.65 for Zn, 0.82–1.05 for Cu, and 0.09–0.11 for Mn. Most of the BF_r values were less than 1.0 (except for Cu in a few cultivars). Because the BF_r values are the ratios of root to soil concentrations, and the same soil was used in our study, the BF_r values reflect the concentrations in the roots (shown in Fig. 2 and described in 3.2).

The variations in TF_{r-p} s and TF_{p-l} s under different treatments are described in Fig. 3. Similar to BF_r s, all of TF_{r-p} s were lower than 1.0. Low concentration of Cd (T1) enhanced the translocation from roots to pseudostems of P, Mg, Fe, Zn, and Cu significantly ($p < 0.05$). TF_{r-p} for K increased with the amount of Cd added, and a significant increase ($p < 0.05$) was observed for T2 and T3. For P, Mg, Fe and Zn, however, the enhancement decreased significantly when the soil Cd concentration reached 5.0 mg/kg, and no significant differences ($p > 0.05$) were observed between CK and T3. Furthermore, a significant decrease ($p < 0.05$) was observed between CK and T1 for Cu.

P was the sole element with a $TF_{p-l} < 1.0$, indicating that all other elements had higher concentrations in the leaves than in the pseudostems. Compared with the influence on TF_{r-p} s of K, the addition of Cd had different influences on TF_{p-l} s, decreasing significantly ($p < 0.05$) under T2 and T3. Mg and Zn, Fe and Cu shared similar Cd influences on TF_{p-l} s. For Mg and Zn, the TF_{p-l} values increased with an increase soil Cd concentration, and significant differences ($p < 0.05$), with increases of 12% and 22%, respectively, were found for T3 and CK. The largest TF_{p-l} values of Fe and Cu

were observed in T2 treatment, with 27% and 15% increase from CK ($p < 0.05$).

Most TF_{r-p} s and TF_{p-l} s of nutrient elements showed significant positive correlations ($p < 0.05$, Pearson test, listed in Table 3). For the correlation analysis on TF_{r-p} s, 79% (22/28), 89% (25/28), 86% (24/28), and 61% (17/28) of the total results were significant positive correlations ($p < 0.05$, Pearson test) for CK, T1, T2, and T3 treatment, respectively. The data for TF_{p-l} s were slightly lower: 71% (20/28), 61% (17/28), 78% (22/28), and 61% (17/28), respectively. P and Mn were main elements of which insignificant correlations ($p > 0.05$, Pearson test) or significant negative correlations ($p < 0.05$, Pearson test) were found among the other seven elements. TF_{p-l} s on P showed weaker positive correlations to other elements than TF_{r-p} s, especially for low soil Cd treatment. Under CK treatment, TF_{p-l} s on P only showed a significant positive correlation to Cu ($p < 0.05$, Pearson test). Meanwhile, significant negative correlations were observed between P–Ca ($p < 0.01$, Pearson test) and P–Mn ($p < 0.05$, Pearson test). For the TF relationship between Mn and other elements, the amount of significant positive correlations ($p < 0.05$, Pearson test) decreased with an increase of the soil Cd concentration, while the amount of significant negative correlations ($p < 0.05$, Pearson test) increased. Significant positive correlations were only observed for TF_{r-p} s between Mn–P, Mn–Mg, Mn–Cu ($p < 0.01$), and for TF_{p-l} s between Mn–Mg ($p < 0.01$), Mn–Zn ($p < 0.05$) under treatment T2, while under treatment T3 they were only found between Mn and Ca, Mg, Fe or Zn for TF_{p-l} s. Moreover, significant negative correlations were found for

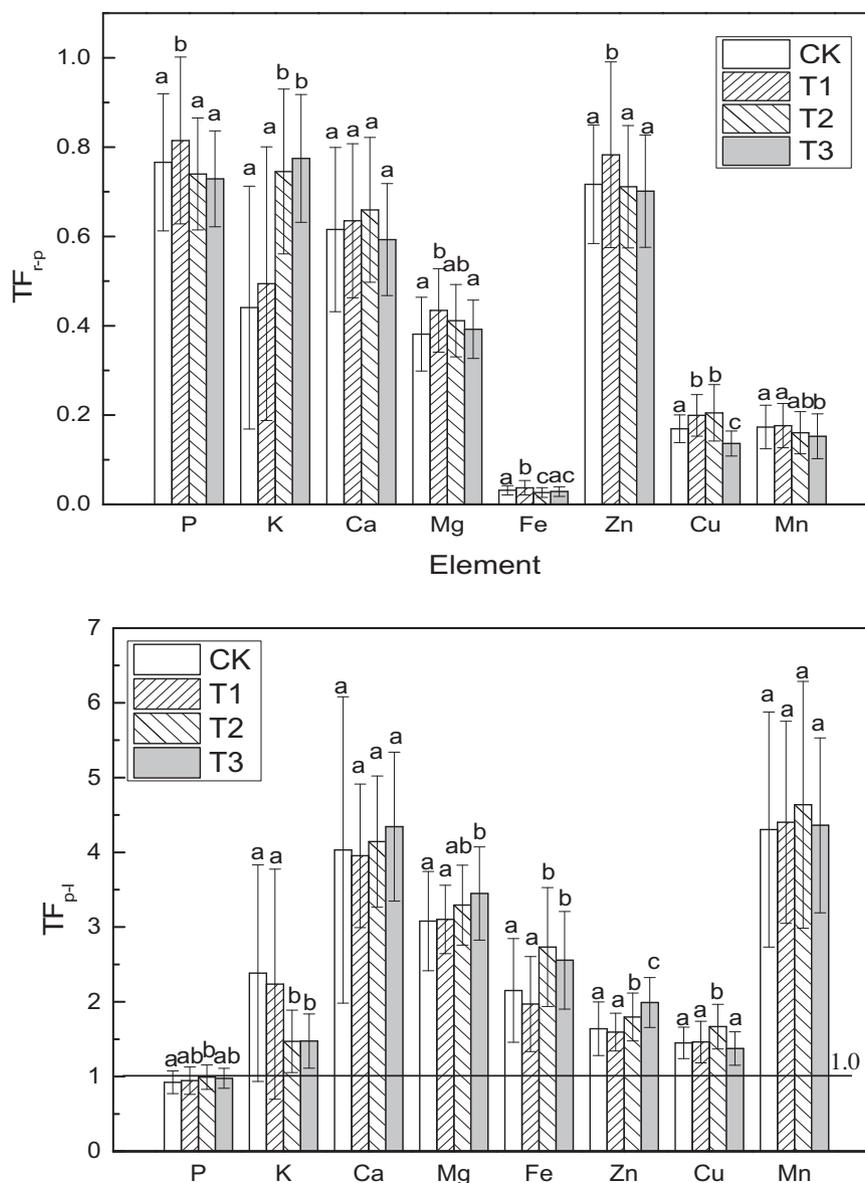


Fig. 3. Translocation factors under different Cd treatments (mean \pm SD). * Results for the same element with the same letter were not significantly ($p < 0.05$, S–N–K test) different.

TF_{r-p} s between Mn–P and Mn–K at the $p = 0.01$ level, between Mn–Mg and Mn–Zn at the $p = 0.05$ level under treatment T3.

4. Discussion

Screening-out for cultivars with low Cd accumulation might reduce the risk of Cd contamination in welsh onion, as indicated by their variability in uptake (Fig. 1). Although the soil Cd concentration was over three times higher than the limiting value of China's environmental quality standard for soils under the T1 treatment, two welsh onion cultivars met the MPC of Cd. As the Cd concentrations in more than 85% of Cd-contaminated soils were below 1.0 mg/kg (the Ministry of Environmental Protection of PRC & Resources of PRC, 2014), most Cd-contaminated soils could produce welsh onion for safe consumption via growing these two cultivars. Furthermore, Cd concentration in the edible part of the cultivar with the lowest Cd accumulation was only 57–62% of the average value, and 40–43% of the value of the cultivar with

the highest Cd accumulation. This indicated that the strategy of screening Cd-excluding cultivar might reduce nearly half of the human Cd intake via welsh onion. However, the ability of this strategy should not be overestimated. Even as a moderate accumulator (Li et al., 2012), more than 90% of the cultivars did not meet the MPC under low Cd treatment, and all cultivars exceed the MPC value in treatments with higher Cd concentrations, indicating that the safe food could not be obtained from soils with serious Cd contamination through the application of the screening-out of Cd-excluding cultivar strategy singly. To ensure food safety, some other techniques, such as soil flushing and the immobilization of heavy metals, should be employed coupled with screening-out of Cd-excluding cultivars growing in soils with serious Cd contamination.

Previous studies have found conflicting results about the impact of Cd on uptake and/or translocation of other elements by previous studies, depending on factors such as species (even cultivar) of the test plant, the growth period, and the culture conditions. (Goncalves et al., 2009; Liu et al., 2003; Wang, Zou, Duan, Jiang,

Table 3
Correlation analysis results within translocation factors (TFs) of nutrient elements among welsh onion cultivars (n = 75; Pearson test).

	P		K		Ca		Mg		Fe		Zn		Cu		Mn			
	TF _{r-p}	TF _{p-l}	TF _{r-p}	TF _{p-l}	TF _{r-p}	TF _{p-l}	TF _{r-p}	TF _{p-l}	TF _{r-p}	TF _{p-l}	TF _{r-p}	TF _{p-l}	TF _{r-p}	TF _{p-l}	TF _{r-p}	TF _{p-l}		
CK	P	0.117	0.388 ^b	-0.21	0.415 ^b	0.469 ^b	0.616 ^b	0.391 ^b	0.187	-0.83	0.720 ^b	0.360 ^b	0.534 ^b	0.643 ^b	0.499 ^b	0.176	P	T1
	K	0.356 ^b	0.281 ^a	0.524 ^b	0.469 ^b	0.491 ^b	0.235 ^a	0.235 ^a	0.414 ^b	0.289 ^a	0.671 ^b	0.289 ^a	0.578 ^b	0.483 ^b	0.170	K		
	Ca	0.571 ^b	-0.061	0.666 ^b	0.718 ^b	0.706 ^b	0.755 ^b	0.668 ^b	0.087	-0.057	0.567 ^b	0.513 ^b	0.288 ^b	0.506 ^b	0.404 ^b	Ca		
	Mg	0.339 ^b	-0.055	0.582 ^b	0.292 ^a	0.371 ^b	0.553 ^b	0.631 ^b	0.124	-0.102	0.797 ^b	0.619 ^b	0.517 ^b	0.663 ^b	0.404 ^b	Mg		
	Fe	0.584 ^b	-0.037	0.489 ^b	0.300 ^b	0.232 ^a	0.566 ^b	0.451 ^b	0.587 ^b	0.538 ^b	0.308 ^b	0.191	0.558 ^b	0.488 ^b	-0.050	Fe		
	Zn	0.266 ^b	0.307 ^b	0.507 ^b	0.481 ^b	0.259 ^a	0.194	0.376 ^b	0.487 ^b	0.547 ^b	0.485 ^b	0.615 ^b	0.706 ^b	0.680 ^b	0.411 ^b	Zn		
	Cu	0.145	-0.242 ^a	0.440 ^b	0.294 ^a	0.214	0.528 ^b	0.556 ^b	0.272 ^b	0.287 ^a	0.211	0.342 ^b	0.261 ^a	0.695 ^b	0.726 ^b	Cu		
	Mn	0.728 ^b	0.583 ^b	0.475 ^b	0.130	0.398 ^b	0.176	0.235 ^a	0.285 ^a	-0.016	0.429 ^b	0.221	0.282 ^a	-0.379 ^b	-0.193	Mn	T3	
T2	P	0.580 ^b	0.380 ^b	0.645 ^b	0.036	0.297 ^b	0.153	0.400 ^b	-0.023	0.056	0.400 ^b	0.294 ^a	0.285 ^a	0.101	0.101	P		
	K	0.625 ^b	0.350 ^b	0.475 ^b	0.636 ^b	0.484 ^b	0.773 ^b	0.773 ^b	0.358 ^b	0.485 ^b	0.505 ^b	0.773 ^b	0.279 ^a	-0.124	0.559 ^b	K		
	Ca	0.423 ^b	0.127	0.548 ^b	0.636 ^b	0.651 ^b	0.445 ^b	0.445 ^b	0.528 ^b	0.587 ^b	0.711 ^b	0.715 ^b	0.341 ^b	-0.239 ^a	0.433 ^b	Ca		
	Mg	0.603 ^b	0.567 ^b	0.709 ^b	0.602 ^b	0.651 ^b	0.500 ^b	0.445 ^b	0.528 ^b	0.587 ^b	0.542 ^b	0.529 ^b	0.223	-0.022	0.379 ^b	Mg		
	Fe	0.570 ^b	0.343 ^b	0.438 ^b	0.649 ^b	0.600 ^b	0.789 ^b	0.810 ^b	0.637 ^b	0.454 ^b	0.388 ^b	0.640 ^b	0.283 ^a	0.318 ^b	0.318 ^b	Fe		
	Zn	0.373 ^b	0.219	0.186	0.448 ^b	0.553 ^b	0.502 ^b	0.698 ^b	0.465 ^b	0.558 ^b	0.181	0.297 ^b	0.645 ^b	-0.284 ^b	-0.072	Zn		
	Cu	0.373 ^b	0.219	0.186	0.185	0.421 ^b	0.042	0.266 ^a	0.226	-0.123	0.388 ^b	0.640 ^b	0.645 ^b	0.089	-0.072	Cu		
	Mn	0.373 ^b	0.219	0.186	0.185	0.421 ^b	0.042	0.266 ^a	0.226	-0.123	0.388 ^b	0.640 ^b	0.645 ^b	0.089	-0.072	Mn		

^a Correlation is significant at the 0.05 level (2-tailed).

^b Correlation is significant at the 0.01 level (2-tailed).

& Liu, 2007). This inconsistency implies the effects and mechanisms are complex. Toxic metals are absorbed by roots via transport proteins of cells in the root cortexes, which can be available for both nutrient elements and heavy metals (Cobbett, 2003; De Maria et al., 2011). Hence, competition for the same transporters between toxic metals (such as Cd and Pb) and essential nutrients could occur due to the low selectivity of these transport systems (De Maria et al., 2011; Sarwar et al., 2010). Low concentration of Cd might hyperpolarize plasma membranes at the root surface, thus increasing the transmembrane potential, which is an energy source for cation uptake (Kennedy & Gonsalves, 1987). Furthermore, Cd might also alter the conformation of proteins, such as enzymes, transporters, or regulator proteins, due to its strong affinity as ligand to sulfhydryl and carboxylic groups (Vanassche & Clijsters, 1990).

In the present study, a decrease in the concentrations of the eight elements in roots was observed under T1 treatment, which might be caused by the competition for absorption between nutrient elements and Cd (Fig. 2). For low Cd concentrations in soil (1.0 mg/kg), the capacity of transport systems was not seriously affected; hence, the competition for the transmembrane carriers with Cd reduced the concentrations of the eight elements in the roots. Thus, lower concentrations of nutrient elements were observed under the T1 treatment compared with CK. With the increase in the Cd concentration in the soil, other interactions between Cd and the roots played more important roles. As a synthesized result of these interactions, the concentrations of these elements in roots mostly increased, especially for K and Zn. The increase in the concentrations of K and Zn was also found in other plants (De Maria et al., 2011; Street, Kulkarni, Stirk, Southway, & Van Staden, 2010). Liu et al. (2011) suggested that the increase in the concentrations of essential elements might be attributed to the fact that these elements were involved in the detoxification processes of Cd. Meanwhile, the changes in the concentrations of these elements in roots might be the main factor in the changes in their concentrations in pseudostems and leaves, displaying similar tendencies of concentrations in welsh onion tissues as affected by different levels of Cd contamination. This also indicated that the selectivity was lower for elementary translocation for uptake.

The significant positive correlations between the concentrations of Cd and Mn in roots among cultivars (Table 1), which was also observed in barley (*Hordeum vulgare* L.) by Chen et al. (2007), suggests that the variation in uptake of Mn accounted for the cultivar-dependence of Cd uptake for welsh onion. This might be because that some transporter gene families, including members of the Nramp and ZIP families, function in both Mn and Cd transport processes (Goncalves et al., 2009; Wang et al., 2007). Meanwhile, Ramos, Esteban, Lucena, and Garate (2002) suggested that higher Mn concentrations might lead to a more powerful defense against Cd toxicity, thus promoting Cd uptake in cultivars with higher Mn concentrations. For the other six elements with positive charges (K, Ca, Mg, Fe, Zn, and Cu), the significant positive correlations with Cd concentrations in the shoots demonstrated that the charges might play important roles during translocation processes of nutrient elements in different welsh onion cultivars.

Cd exposure can damage the structure of chloroplast and reduce the production of chlorophyll in plants. P is involved in the detoxification of Cd (Sarwar et al., 2010; Zhou, 1995) and its uptake might help to reduce both effects by enhancing vacuolar and cell wall sequestration of Cd (Jiang, Yang, & Zhang, 2007; Zhou, 1995). In other words, higher P concentrations might improve Cd detoxification capacity. This might be the reason for the significant or highly significant positive correlation between P and Cd among cultivars in shoots for soil with less than 2.5 mg/kg of Cd. However, the detoxification did not maintain the P–Cd correlation among cultivars under the 5 mg/kg Cd treatment.

Many authors have found significant positive concentration correlations of nutrient elements among cultivars (Arivalagan et al., 2012; Fernandez-Ruiz et al., 2011; Galdon et al., 2008; Liu et al., 2003, 2011; Suarez et al., 2007; Zhang et al., 2010), similar to the results for welsh onion in the present study (Table 2). This implies that there was a synergistic effect in elementary accumulation in different cultivars, which benefited the screening-out of microelements-rich cultivars (Arivalagan et al., 2012), while limiting the application of heavy metal-excluding cultivars in soils with low or moderate levels of heavy metals, as selection for one trait leads to the selection of genetically correlated traits (Wricke & Weber, 1986). Moreover, the correlation would be affected by Cd, especially for P, Fe and Mn, due to the imbalance of uptake and/or translocation of nutrient elements caused by Cd (Liu et al., 2011; Sikka & Nayyar, 2012; Street et al., 2010; Wang et al., 2007). The change in the P–Mn correlation from positive to highly significantly negative when the Cd concentration reached 5.0 mg/kg suggests that the balance of P–Mn in different cultivars was seriously disturbed by Cd. The uptake balance of Fe and Mn to other elements was also affected by Cd, as shown in the decrease in the significant positive correlations with the increase in the concentration of Cd in soil.

BFs and TFs are widely used in the analysis of accumulation of elements, especially for heavy metals (Liu et al., 2009, 2011). Welsh onion had low root uptake capacity and root-pseudostem translocation capacity for the elements in the present study, as suggested by the $BF_{r,s} < 1.0$ and $TF_{r-p,s} < 1.0$. This result is shown in Table 3 and is convenient for heavy metal-excluding cultivars. Low concentrations of Cd stimulated the translocation of element from roots to pseudostems, of which the mechanism might be detoxification of these elements (Liu et al., 2011; Ramos et al., 2002). However, under higher Cd concentrations, the competition of transporters with Cd during the translocation of these elements reduced the TF_{r-p} values (except for K) (De Maria et al., 2011; Sarwar et al., 2010). In contrast to the $TF_{r-p,s}$, the TF_{p-s} of all the elements (K, Ca, Mg, Fe, Zn Cu, and Mn) with positive charges were all above 1.0, indicating that these elements were enriched in the leaves. Leaves are the main tissue of transpiration, where water departs from the leaves, but the dissolved solid mass is retained. Thus, higher elementary concentrations were observed in leaves. At the same time, the leaves, where photosynthesis and respiration occur, participate in more biological activities. More nutrient elements are needed for the enzymes involved in these activities, which explains observation of higher concentrations of element in leaves.

The correlation analysis on TFs provides detailed information about the relationship and translocation of different elements. The significant positive correlations in more than 60% of results were in accordance with higher correlation coefficients in leaves than those in pseudostems. There were fewer positive correlations results between P and other elements with positively charges, indicating that charge might play an important role in the translocation of nutrient elements.

5. Conclusions

The strategy of screening-out of Cd-excluding cultivars might solve most of the Cd contamination issues in agricultural fields. However, food safety could not be assured via the singly application of screening out Cd-excluding cultivars growing in soils with heavily Cd contamination, and some other techniques should be employed simultaneously.

The BF and TF_{r-p} values of the nutrient elements were less than 1.0 in welsh onion, while the TF_{p-l} values were greater than 1.0 (except for P). The concentration of the nutrient elements in welsh

onions can be influenced by Cd, though the effect varies by type of element, plant tissue and cultivar.

There were positive correlations between Cd and nutrient element concentrations in most samples of pseudostems and leaves. The correlation coefficients in leaves were higher than those in pseudostems for the significant correlations. However, Mn was the only element that was positively correlated with Cd in roots. Many of the concentrations of nutrient elements in this study were positively correlated. The increase in the Cd concentration in the soil disturbed the balance of nutrient elements, especially for P and Mn.

A synergistic effect was observed in the translocation of the nutrient elements. Overall, 79% (22/28), 89% (25/28), 86% (24/28), and 61% (17/28) of the $TF_{r-p,s}$, and 71% (20/28), 61% (17/28), 78% (22/28), and 61% (17/28) of the TF_{p-s} presented significant positive correlations under the CK, T1, T2 and T3 treatments, respectively.

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