MARCEL DEKKER, INC. • 270 MADISON AVENUE • NEW YORK, NY 10016

COMMUNICATIONS IN SOIL SCIENCE AND PLANT ANALYSIS
Vol. 34, Nos. 9 & 10, pp. 1421–1434, 2003

Assessing Zinc Thresholds for Phytotoxicity and Potential Dietary Toxicity in Selected Vegetable Crops

X. X. Long,[1] X. E. Yang,[1,2,*] W. Z. Ni,[1] Z. Q. Ye,[1] Z. L. He,[2] D. V. Calvert,[2] and J. P. Stoffella[2]

[1]Department of Natural Resources, Zhejiang University, Huajiachi Campus, Hangzhou, China
[2]University of Florida, Institute of Food and Agricultural Sciences, Indian River Research and Education Center, Fort Pierce, Florida, USA

ABSTRACT

Heavy metal pollution in soils influences crop yield and quality, and metal accumulation in vegetables may pose a human health risk when consumed. Zinc (Zn), one of the heavy metals, is an essential element for plants, animals, and humans, but it is toxic at high levels. In this study, bioavailability of added Zn in a vegetable garden soil and critical Zn concentrations for phytotoxicity and potential dietary toxicity were determined for Chinese cabbage (*Brassica chinensis* L.), pakchoi (*Brassica chinensis* L.), and celery (*Apiumg graveolens* L.). Different Zn levels (0, 100, 200, 300, 400 $mg\,kg^{-1}$ soil, supplied as $ZnSO_4 \cdot 7H_2O$)

*Correspondence: X. E. Yang, Department of Natural Resources, Zhejiang University, Huajiachi Campus, Hangzhou, China; E-mail: xyang@mail.hz.zj.cn; xyang@mail.ifas.ufl.edu.

DOI: 10.1081/CSS-120020454 0010-3624 (Print); 1532-2416 (Online)
 www.dekker.com

were added to the soil samples, based on Zn adsorption-desorption characteristics of the soil, the availability of added Zn in the soil decreased with time, with minimal change after 10–12 weeks of incubation. The results from sand and soil culture experiments showed that shoot growth was significantly inhibited at Zn concentrations above 25 mg L^{-1} in nutrient solution or at DTPA-Zn above 170 mg kg^{-1} in the soil. The sensitivity to Zn toxicity differed among the three vegetable crops, changing in the order: celery > Chinese cabbage > pakchoi. Zinc concentration in shoots and edible parts varied with Zn supply levels and type of the vegetables. Negative correlations were noted between Zn concentrations in shoots and dry matter yields ($r = 0.90–0.98$, $P < 0.01$), the critical Zn concentrations in plant tissues at 10% reduction of biomass yield (PT_{10}) were 173.1, 167.5, 144.2 and 222.2 mg kg^{-1} (DW) for Chinese cabbage, pakchoi, celery (stem) and celery (leaf), respectively. Zinc concentrations in the edible parts were positively correlated with available and total Zn in the soil ($r = 0.91–0.99$, $P < 0.01$). Based on the threshold of human dietary toxicity for Zn (20 mg kg^{-1}), the critical concentrations of total and available Zn in the soil were 413 and 244 mg kg^{-1} for Chinese cabbage, 224 and 75 mg kg^{-1} for pakchoi, and 272 and 101 mg kg^{-1} for celery, respectively. These results indicate that some vegetable species like pakchoi might accumulate Zn in edible parts over human dietary toxic threshold before the dry matter yield reduction was observed.

Key Words: Genotypic difference; Human health; Toxic threshold; Vegetable crops; Zinc.

INTRODUCTION

As indispensable food for human life, vegetables play important roles in human health. With improvements in living standards, the demand for "non-contaminated" and high quality vegetables is increasing. However, heavy metal contamination of vegetable garden soils in suburban area has increased due to disposal of sewage sludge and municipal wastes, irrigation with industrial waste-water, and precipitation of pollutants from air to the soils.[1,2] Heavy metal pollution in vegetable garden soils can have adverse effects on crop growth, food quality and environmental health,[3,4] and its accumulation in vegetables may pose a human health risk when consumed.[5,6]

Zinc is an essential micronutrient for plants, animals, and humans. It acts in metallo-enzymes or cofactors for a large number of enzymes such as anhydrases, dehydrogenases, oxidase, and peroxidases and plays an important role in regulating nitrogen metabolism and photosynthesis.[7,8] Although soil total Zn in non-polluted agricultural soils is generally below 500 mg kg^{-1}, total soil Zn concentrations in polluted soils could be higher than 3000 mg kg^{-1}. Excess Zn is toxic to plants and humans, causing disturbance of a wide range of biochemical and physiological processes, inducing iron (Fe) deficiency in plants, and resulting in leaf defoliation.[7,8] Plant species and genotypes differ greatly in their tolerance to high Zn concentrations. It is generally assumed that leaf Zn levels in excess of 300–600 mg kg^{-1} dry weight (DW) is considered to be toxic to plants.[7] However, the thresholds of leaf Zn levels, total soil and available Zn levels have not been established for phytotoxicity in different vegetable crops.

The knowledge of Zn toxicity in humans is scarce. The most important information reported is its interference with Cu metabolism.[9,10] The symptoms that an acute oral Zn dose may provoke include: tachycardia, vascular shock, dyspeptic nausea, vomits, diarrhea and pancreatic, and damage of hepatic parenchyma.[11] Although maximum Zn tolerance for human health has been established for edible parts of crops (20 mg kg^{-1}),[12] soil Zn threshold for producing safe vegetables is not available. The objectives of this study were to evaluate effects of excessive Zn on the growth and accumulation of Zn for Chinese cabbage, pakchoi, and celery, to assess the availability of added Zn in vegetable garden soil, and to establish critical Zn concentrations for potential human dietary toxicity.

MATERIALS AND METHODS

Sandy Culture Experiment

The seeds of three vegetable crops, Chinese cabbage (*B. chinensis* L.), pakchoi (*B. Chinensis* L.), and celery (*A. Graveolens* L. *var. dulce* DC) were purchased from a vegetable seed corporation in Hangzhou, China. Seeds were germinated on wetted filter paper for two days in the dark at 25°C. The germinated seeds were sown on quartz sand with nutrient solution for preparing seedlings.

Five uniform seedlings were transplanted into quartz sand with nutrient solution in a 3-liter plastic container. The composition of nutrient solution was modified from Yang et al.[13] (in mmol L^{-1}): KNO_3 6.00, $Ca(NO_3)_2 \cdot 4H_2O$ 3.50, KH_2PO_4 1.33, $MgSO_4 \cdot 7H_2O$ 2.00, NaCl 0.48, and (in μmol L^{-1}): H_3BO_3 10.00, $MnSO_4 \cdot H_2O$ 0.50, $ZnSO_4 \cdot 7H_2O$ 0.50, $CuSO_4 \cdot 5H_2O$ 0.20, $(NH_4)_6Mo_7O_{24}$ 0.01, Fe-EDTA 200. After growth of 15 days, plants were exposed to different Zn levels: CK(0.03), 25, 50, 100, 200 mg L^{-1}, supplied as $ZnSO_4 \cdot 7H_2O$. A randomized complete block experimental design was used with each treatment replicated three times. The pH was maintained at 5.8 by daily adjustment with 0.1 mol L^{-1} HCl or 0.1 mol L^{-1} NaOH, and nutrient solution was replaced every seven days. Plants were harvested after three weeks of treatment. At harvest, roots of intact plants were rinsed with deionized water, and were separated from shoots (leaf blades and petioles-edible portion for celery). The base of stalks and roots were rinsed thoroughly with deionized water, blotted dry, first dried at 105°C for 30 min, then dried at 70°C. Fresh weight (FW) and dry weight (DW) of shoots and roots were recorded. Dry plant samples were ground with a stainless steel mill and passed through a 60-mesh sieve for Zn analysis.

Incubation Experiment

Alluvial soil (Fluvio-marine yellow loamy soil) was collected from an old vegetable production area in Hangzhou suburb. The main agrochemical properties of the tested soil were as follows: pH 7.15, organic C 38.7 g kg^{-1}, cation exchange capacity (CEC) 13.93 cmol kg^{-1}, total N 2.60 g kg^{-1}, total P 0.90 g kg^{-1}, total and DTPA-extractable Zn 118.75 and 18.12 mg kg^{-1}, respectively. Different Zn levels (0, 100, 200, 300, 400 mg kg^{-1} soil, supplied as $ZnSO_4 \cdot 7H_2O$) were added to the soil samples, based on Zn adsorption-desorption characteristics of the soil (Long et al., unpublished data). The mixed soil samples were incubated in a large plastic container ($60 \times 40 \times 15$ cm) at approximately 70% of maximum field water capacity for 12 weeks. During the incubation, soil samples were collected at the intervals of 0, 1, 2, 4, 8 and 12 weeks, and available soil Zn (DTPA-extractable) concentrations were measured.

Soil Culture Experiment

The incubated and initial air-dried soil samples were used to grow Chinese cabbage, pakchoi, and celery. Each pot had one kg of soil mixed with 1.0 g of urea, 0.48 g of $Ca(H_2PO_4)_2$ and 0.3 g of KCl as basal fertilization. The process of preparing seedlings was similar to the sand culture experiment. In this case, and five of the 20-day-old seedlings were transplanted. Plants were grown under glasshouse conditions with air temperature of 25 ± 3°C and humidity of approximately 75%. A randomized complete block experimental design was used with each treatment replicated four times. Soil moisture was maintained at 70% of the maximum field water holding capacity. Plants were harvested 35 days after the transplanting. Shoots were separated from the roots, and fresh and dry weights were recorded. Dry plant samples were ground with a stainless steel mill and passed through a 60-mesh sieve for Zn analysis.

Chemical Analysis

Determination of soil agrochemical properties mentioned above was performed using the methods described in *Physical and Chemical Analyses of Soils* by the Institute of Soil Science, Chinese Academy of Science.[14] Soil total Zn was measured by the method of Yuan.[15] Soil available Zn was extracted by 0.05 mol/L DTPA (soil: extractant of 1:20), and plant samples were dry ashed at 550°C, and dissolved with dilute HCl (HCl:water (v/v) of 1:30). Concentration of Zn in the soil and plant solution was determined using an Atomic Absorption Spectroscopy (AAS, Model 8081, Hitachi, Japan).

RESULTS AND DISCUSSION

Bioavailability of Added Zinc in the Vegetable Garden Soil

Metals such as Zn exist in soils in various fractions, chemical species or forms including: exchangeable, carbonate-bound, oxide-bound, organic matter-bound, and crystal lattice metals.[16] Availability of soil Zn to plants differs and may be governed by dynamic equilibrium among these fractions.[17] Total soil metals can be used to estimate the degree of soil exposure to heavy metal pollution, although this is not generally well correlated with metal mobility and bioavailability.[18] The biologically active fractions of Zn in soils mainly consist of its soluble, exchangeable and complexed forms. Many studies showed that DTPA-extractable Zn is correlated well with plant uptake Zn.[19,20] As shown in Fig. 1, DTPA extractable Zn decreased progressively with incubation time, and leveled off in 8–12 weeks of incubation. However, after 12-week incubation, 60–70% of added Zn was still extractable by the DTPA method (Fig. 1). The results showed that a major portion of the Zn added to the garden soil is phytoavailable, which is in agreement with other studies.[21,22]

Effects of Excess Zinc on Growth of Different Vegetable Crops

Excess Zn in growth media caused toxicity to all three vegetable crops. Toxicity symptoms included chlorosis of new leaves, and browning and stunting

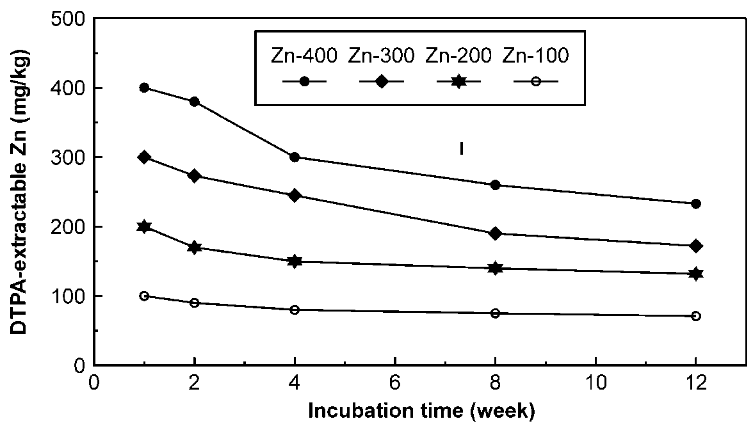

Figure 1. Changes of DTPA-extractable Zn with incubation time. All data are means of three replications. The bar depicts $LSD_{5\%}$. Zn-100, Zn-200, Zn-300, and Zn-400 refer to Zn additional levels of 100, 200, 300, and 400 mg Zn kg^{-1} soil, respectively.

of coralloid roots, and serious inhibition on plant growth. Shoot fresh weight (FW) progressively decreased with increasing Zn concentrations (Fig. 2). Large differences in Zn tolerance were also noted among the three vegetable crops. Celery was more sensitive to higher Zn levels in reduced shoot growth than Chinese cabbage and pakchoi. Shoot FW decreased to approximately 63%, 73%, and 36% of the control for Chinese cabbage, pakchoi and celery, respectively, when plants were grown at the Zn level of 50 mg L^{-1} (Fig. 2).

Although no visible Zn toxicity symptoms were observed in the soil experiment, shoot growth was significantly inhibited at Zn levels above 200 mg kg^{-1} for celery and Chinese cabbage, and above 300 mg kg^{-1} for pakchoi (Table 1). Shoot DW decreased by 10% for Chinese cabbage and celery, but increased by 13% for pakchoi when grown at the soil DTPA-extractable Zn level of 72 mg kg^{-1}. However, at soil DTPA-Zn levels up to 172 mg kg^{-1}, similar yield reduction was observed with celery and pakchoi (Table 1). Root DW was reduced more than shoot DW for pakchoi grown at high Zn levels. Similar sensitivity of both root and shoot growth to Zn toxicity was noted for celery. The results showed that pakchoi required higher soil Zn concentrations for optimal growth and was more tolerant to Zn at soil available Zn concentrations less than 132 mg kg^{-1} than the other two vegetable species.

Significant and negative correlations were noted between shoot biomass and soil available or total Zn ($r = 0.95$, $P < 0.05$ to 0.99, $P < 0.01$). The regression equations between shoot biomass and soil available Zn were

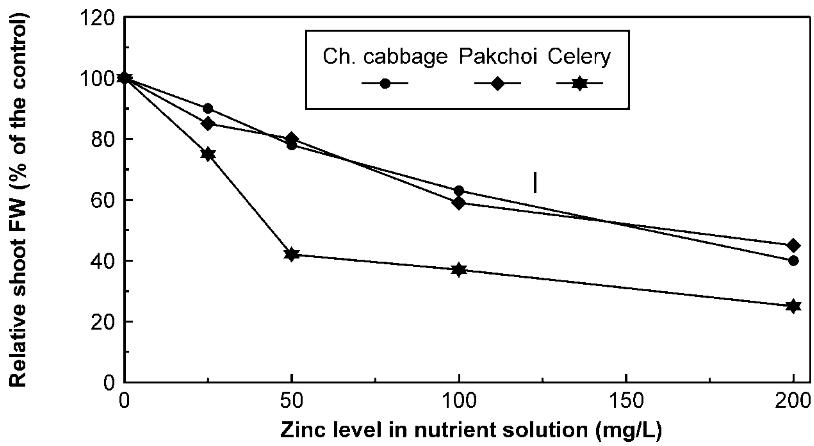

Figure 2. Growth response of different vegetable species to Zn levels in nutrient solution. All data are means of three replications, the bar depicts $LSD_{5\%}$.

$Y = -0.1062X + 79.605$ ($r^2 = 0.996$, $P < 0.01$), $Y = -0.2779X + 112.6$ ($r^2 = 0.888$, $P < 0.05$), and $Y = -0.1309X + 73.279$ ($r^2 = 0.998$, $p < 0.01$) for Chinese cabbage, pakchoi, and celery, respectively. The critical soil available Zn concentrations at 10% reduction of shoot DM yield were 85.59, 103.28, and 64.29 $mg\,kg^{-1}$ for Chinese cabbage, pakchoi, and celery, respectively. The results from both sand and soil culture experiments indicate that celery was the least tolerant to Zn toxicity, and pakchoi was the most tolerant vegetable species among the three species tested.

Zinc Concentration and Accumulation Coefficient

Zinc uptake and accumulation by shoots and roots varied with Zn levels in growth media and vegetable types (Table 2). Both shoot and root Zn concentrations increased sharply with increasing Zn concentrations for three vegetable species. However, shoots contained over 3-fold less Zn than roots when grown under nutrient solution culture conditions. The three vegetable crops differed greatly in their ability to take up Zn from the growth media and to transport it to the shoots. At an external Zn level of 25 $mg\,L^{-1}$, shoot Zn concentration of Chinese cabbage was almost 2-fold lower than that of pakchoi or celery (Table 2). Zinc concentration in the edible part of celery was nearly 2-fold higher than that of the other two species when grown at higher Zn levels ($>50\,mg\,L^{-1}$).

Table 1. Effects of soil Zn levels on growth and yield of different vegetable crops grown under soil culture conditions.[a]

Species	Added Zn (mg/kg)	DTPA-Zn (mg/kg)	Plant height (cm)	Shoot FW (g/pot)	Root FW (g/pot)	Shoot DW (g/pot)	Root DW (g/pot)
Chinese cabbage	Ck	18.1	15.4 a	78.35 a A	2.83 a	5.16 a A	0.42 a
	100	71.7	14.8 ab	70.72 b B	2.78 a	4.55 b B	0.41 a
	200	132.2	14.3 ab	66.22 bc BC	2.77 a	4.32 b B	0.40 a
	300	172.3	13.9 a	61.29 c CD	2.76 a	3.82 c C	0.41 a
	400	233.2	14.7 ab	54.84 d D	2.74 a	3.69 c C	0.40 a
Pakchoi	Ck	18.1	18.1 aA	93.22 a A	3.66 a A	5.53 ab A	0.65 a A
	100	71.7	19.1 aA	104.96 a A	3.06 b B	6.22 a A	0.57 a AB
	200	132.2	18.5 aA	88.57 a AB	2.49 c C	5.51 ab A	0.46 b BC
	300	172.3	15.3 bB	61.77 b BC	2.11 d C	4.44 bc AB	0.38 b CD
	400	233.2	13.1 cB	40.11 c C	1.47 e D	3.35 c B	0.26 c D

	Added Zn (mg/kg)	DTPA-Zn (mg/kg)	Plant height (cm)	Stem FW (g/pot)	Leaf FW (g/pot)	Root FW (g/pot)	Stem DW (g/pot)	Leaf DW (g/pot)	Root DW (g/pot)
Celery	Ck	18.1	24.7 a	39.46 aA	32.61 aA	23.86 aA	3.68 aA	5.01 A	4.16 a A
	100	71.7	25.8 a	36.41 aA	28.85 bB	17.59 bB	3.04 bB	4.22 B	3.08 b B
	200	132.2	25.0 a	29.82 bB	22.56 cC	16.31 bBC	2.58 cC	3.33 C	2.88 bBC
	300	172.3	25.4 a	28.72bcB	20.37 dD	14.83 cCD	2.47 cdC	3.01 D	2.60 cCD
	400	233.2	24.9 a	26.25 cB	19.26 eD	13.34 dD	2.24 dC	2.73 E	2.32 d D

[a] All data are means of 4 replications, values followed by the same letter are not significantly different by Duncan's Multiple Range Test (low case - $P < 0.05$; upper case - $P < 0.01$) within the same column for each vegetable specie.

Table 2. Zinc concentration in the shoots and roots of different vegetable crops grown in nutrient solution.[a]

Zn supply level ($mg\,L^{-1}$)	Zinc concentration in shoots and roots ($mg\,kg^{-1}$ DW)						
	Chinese cabbage		Pakchoi		Celery		
	Root	Shoot	Root	Shoot	Root	Stem	Leaf
0.03	135.5 e	65.88 e	127.5 e	61.67 d	115.0 e	72.50 e	125.8 e
25	1167 d	494.4 d	1587 d	925.0 c	2308 d	870.0 d	1295 d
50	2313 c	1024 c	2623 c	976.7 c	4350 c	2820 c	2070 c
100	3595 b	1835 b	4642 b	1883 b	7567 b	4075 b	2342 b
200	12807 a	2975 a	7483 a	2375 a	12823 a	4514 a	2978 a

[a] All data are means of three replications, values followed by the same letter are not significantly different by Duncan's Multiple Range Test ($P < 0.05$).

Under soil culture conditions, the zinc accumulation coefficient (AF) in shoots increased for pakchoi, but decreased for celery and Chinese cabbage when soil available Zn was raised from 18 to 172 mg L^{-1} (Table 3). However, root Zn AF increased, to varied extents, with increasing soil Zn for all the vegetables. Celery showed the highest AF in edible parts at low soil Zn (CK), whereas pakchoi had the highest AF of Zn at higher soil available Zn levels. The AF for zinc in edible parts of the three vegetable crops decreased in the order: pakchoi > celery (stem) > Chinese cabbage. Significant positive correlations were noted between shoot Zn and soil available Zn level. The regression equations between shoot concentrations and soil available Zn were $Y = 0.1171X - 0.5674$ ($r = 0.953$, $P < 0.05$), $Y = -0.4067X - 10.469$ ($r = 0.948$, $P < 0.05$), $Y = 0.1055X + 9.3461$ ($r = 0.981$, $P < 0.01$) and $Y = 0.214X + 15.845$ ($r = 0.967$, $P < 0.01$) for Chinese cabbage, pakchoi, celery leaf, and celery stem, respectively.

Criteria for Zinc Phytotoxicity

Phytotoxicity of heavy metals is related to the amount of toxicants taken up and accumulated in plant tissues.[23,24] Significant and negative correlations were noted between shoot dry matter yield and Zn concentration in plant tissues for all three vegetable crops (Table 4). The critical tissue Zn

Table 3. Zinc accumulation coefficients (AF) of different vegetable crops grown at various soil Zn levels.[a]

		Zn accumulation coefficient (Zn AF)						
		Chinese cabbage		Pakchoi		Celery		
Zn rate added ($mg\,kg^{-1}$)	DTPA-Zn ($mg\,kg^{-1}$)	Roots	Shoots	Root	Shoots	Roots	Stem	Leaf
CK	18.1	0.158b	0.039b	0.169e	0.056e	0.290d	0.113c	0.195a
100	71.7	0.133c	0.030c	0.284d	0.078d	0.507c	0.068a	0.138b
200	132.2	0.135c	0.036b	0.292c	0.098c	0.419b	0.069a	0.114c
300	172.3	0.143d	0.042d	0.415b	0.119b	0.577a	0.066b	0.135b
400	233.2	0.235a	0.058a	0.709a	0.189a	0.572a	0.068a	0.130d

[a] AF = Zn in plant tissues ($mg\,kg^{-1}$)/total Zn in soil ($mg\,kg^{-1}$). All data are means of 4 replications, values followed by the same letter are not significantly different by Duncan's Multiple Range Test ($P < 0.05$).

Table 4. Regression between shoot dry matter yields and shoot Zn concentration for different vegetable crops grown in nutrient solution (n = 5).

Species	Variables	Regression equations	R^2	Critical Zn[a] (mg/kg)
Chinese cabbage	Shoot DM vs. Shoot Zn	$Y = -56.09\,\ln x + 583.05$	0.921*	173.1
Pakchoi	Shoot DM vs. Shoot Zn	$Y = -27.40\,\ln x + 301.81$	0.904*	167.5
Celery	Stem DM vs. Stem Zn	$Y = -12.56\,\ln x + 120.73$	0.989**	137.8
	Shoot DM vs. Stem Zn	$Y = -22.08\,\ln x + 214.62$	0.985**	144.2
	Leaf DM vs. Leaf Zn	$Y = -12.28\,\ln x + 112.79$	0.943*	230.6
	Shoot DM vs. Leaf Zn	$Y = -28.69\,\ln x + 259.95$	0.956*	222.8

* and ** indicate significant levels at 5% and 1%, respectively.
[a] Critical Zn = zinc concentration at 10% shoot dry matter reduction.

concentrations at 10% reduction of shoot dry matter yield were 173.1, 167.5, 137.8, and 144.2 $mg\,kg^{-1}$ (DW) for Chinese cabbage, pakchoi, celery stem, and celery shoots (leaf + stem), respectively. These results were in agreement with other studies showing that the relationship between Zn concentration in leaves and the percent growth retardation for corn, lettuce and bush bean was the highest linear correlation.[25–27] From both soil and solution culture experiments, we found that yield reduction by 10% was statistically significant (Table 1). Zinc concentrations that result in 10% yield reduction is considered as appropriate Zn thresholds for phytotoxicity. The critical Zn concentrations for phytotoxicity of the three vegetables crops were relatively lower than that reported from other crops.[7]

Soil Thresholds for Potential Zinc Dietary Toxicity

Soil thresholds for heavy metal toxicity are an important factor affecting soil environment capacity of heavy metal and determining heavy metal cumulative loading limit. For the soil-plant system, the heavy metal toxicity threshold is the highest permissible content in the soil (total or bioavailable concentration) that does not produce any phytotoxicity (i.e., inhibition of plant growth and decrease of yield), or the heavy metal in the edible parts of crops does not exceed the critical dietary Zn threshold for human health, which has been established to be 20 $mg\,kg^{-1}$.[12] According to the regression equations between shoot yield or Zn concentration and soil total or available Zn, soil Zn thresholds for yield reduction (decreased 10%) and potential dietary toxicity

Table 5. Soil Zn thresholds for yield reduction and potential dietary toxicity in edible parts of the vegetable crops.

	Total Zn ($mg\,kg^{-1}$)		Available Zn ($mg\,kg^{-1}$)	
Crop species	PDT $\leq 20\,mg\,kg^{-1}$	SDMYR by 10%	PDT $\leq 20\,mg\,kg^{-1}$	SDMYR by 10%
Chinese cabbage	413	244	176	85.6
Pakchoi	224	277	74.9	103
Celery (stem)	272	220	101	73.0
Celery (leaf)	122	187	19.4	55.8

PDT = potential dietary toxicity; SDMYR = shoot dry matter yield reduction.

in edible parts of the vegetables were calculated (Table 5). The total soil Zn thresholds for shoot dry matter yield reduction were higher for pakchoi and only slightly lower for celery (stem), than that for potential dietary toxicity. Soil available Zn thresholds for Zn potential dietary toxicity were 175.6, 74.9, and 101.0 $mg\,kg^{-1}$ for Chinese cabbage, pakchoi, and celery (stem), respectively. For pakchoi, a higher soil available Zn threshold for yield reduction (10%) (103 $mg\,kg^{-1}$) was again noted relating to that for potential dietary toxicity (74.9 $mg\,kg^{-1}$). The lower soil available Zn threshold for Zn potential dietary toxicity for pakchoi than for the other vegetable species is mainly associated with its greater ability to absorb Zn from the soil and to translocate and accumulate Zn in the shoots. These results indicate that some vegetable species, like pakchoi, may accumulate Zn in the edible part over Dietary Toxic Threshold before yield reduction occurs.

ACKNOWLEDGMENTS

This study was financially supported by a grant (#39925024) from the Outstanding Young Scientist Fund of the National Natural Science Foundation of China and a grant (#2002 CB410804) from the Science and Technology Ministry of China. Florida Agricultural Experiment Station Journal Series No: R-08901.

REFERENCES

1. MacNicol, R.D.; Beckett, P.H.T. Critical tissue concentrations of potentially toxic elements. Plant Soil **1985**, *85*, 107–109.

2. Jinadasa, K.B.P.N.; Milham, P.J.; Hawkkins, C.A.; Cornish, P.S.; Williams, P.A.; Kaldor, C.J.; Conroy, J.P. Survey of cadmium levels in vegetables and soils of greater Sydney. Aust. J. Environ. Qual. **1997**, *26*, 924–933.
3. Fergussen, J.E. *Chemistry, Environmental Impact and Health Effects*; Pergamin Press: Oxford, England, 1990; 382–399.
4. McLaughlin, M.J.; Hamon, R.E.; McLaren, R.G.; Speir, T.W.; Rogers, S.L. Review: a bioavailability-based rationale for controlling metal and metalloid contamination of agricultural land in Australia and New Zealand. Aust. J. Soil Res. **2000**, *38*, 1037–1086.
5. Boon, D.Y.; Soltanpour, P.N. Lead, cadmium, and zinc contamination of Aspen garden soils and vegetation. J. Environ. Qual. **1992**, *21*, 82–86.
6. Pichtel, J.; Salt, C.A. Vegetative growth and trace metal accumulation on metalliferous wastes. J. Environ. Qual. **1998**, *27*, 618–624.
7. Marschnar, H. *Mineral Nutrition of Higher Plants*, 2nd Ed.; Academic Press: London, 1995.
8. Swietlik, D. Zinc nutrition in horticultural crops. In *Horticultural Reviews*; Janick, J., Ed.; John Wiley & Sons, Inc.: New York, 1999; Vol. 23, 109–179.
9. Barone, A.; Ebesh, O.; Harper, R.G.; Wapnir, R.A. Placental copper transport in rats: effects of elevated dietary zinc on fetal copper, iron and metallothionien. J. Nutr. **1998**, *128*, 1037–1041.
10. Gyorffy, E.J.; Chan, H. Copper deficiency and mycrocytic anemia resulting from prolonged ingestion of over-the-counter zinc. Am. J. Gastroenterol. **1992**, *87*, 1054–1055.
11. Salgueiro, M.J.; Zubillaga, M.; Lysionek, A.; Sarabia, M.I.; Caro, R.; Paoli, T.D.; Hager, A.; Weill, R.; Boccio, J. Zinc as an essential micronutrient: a review. Nutr. Res. **2000**, *20*, 737–755.
12. Protection Institute, Chinese Academy of Science, *Thresholds for Food Hygiene*; Standard Press: Beijing, China, 1995.
13. Yang, X.; Baligar, D.C.; Martens, D.C.; Clark, R.B. Influx, transport, and accumulation of cadmium in plant species grown at different Cd(II) activities. Environ. Sci. Health **1995**, *B30* (4), 569–583.
14. Institute of Soil Science, Chinese Academy of Science, *Physical and Chemical Analyses of Soils* (in Chinese); Shanghai Academic Press: Shanghai, China, 1980.
15. Yuan, D.W. Compared study on the pretreament methods for measuring soil total copper, zinc, lead, cadmium, nickel, and manganese. Agro-Environ. Protect **1988**, *7* (1), 34–36.

16. Shuman, L.M. Chemical forms of micronutrient in soils. In *Micronutrient in Agriculture*, 2nd Ed.; Soil Science Society of America: Madison, WI, 1991; SSSA Book Series, 113–144.
17. Kiekens, L. Zinc. In *Heavy Metals in Soils*; Alloway, B.J., Ed.; John Wiley & Sons: New York, 1990; 261–279.
18. Kuo, S.; Heilman, P.E.; Baker, S. Distribution and forms of copper, zinc, cadmium, iron, and manganese in soils near a copper smelter. Soil Sci. **1983**, *35*, 101–109.
19. Arnesen, A.K.M.; Singh, B.R. Plant uptake and DTPA-extractability of Cd, Cu, Ni and Zn in a Norwegian alum shale soil as affected by previous addition of dairy and pig manures and peat. Can. J. Soil Sci. **1998**, *78*, 531–539.
20. Miner, G.S.; Gutierrez, R.; King, L.D. Soil factors affecting plant concentration of cadmium, copper, and zinc on sludge-amended soils. J. Environ. Qual. **1997**, *26*, 989–994.
21. Cajuste, L.J.; Cruz-Diaz, J.; Garcia-Osorio, C. Extraction of heavy metals from contaminated soils. I. Sequential extraction in surface soils and their relationships to DTPA extractable metals and metal plant uptake. J. Environ. Sci. Health **2000**, *A35*, 1141–1152.
22. Darmawa, M.; Wada, S.I. Kinetics of speciation of copper, lead, and zinc loaded to soils that differ in cation exchanger composition at low moisture content. Commun. Soil Sci. Plant Anal. **1999**, *30*, 2363–2375.
23. Chang, A.C.; Granato, T.C.; Page, A.L. A methodology for establishing phytotoxicity criteria for chromium, copper, nickel, and zinc in agricultural land application of municipal sewage sludges. J. Environ. Qual. **1992**, *21*, 521–536.
24. Bould, C.; Hewitt, E.J.; Needham, P. *Diagnosis of Mineral Disorders in Plants. 1. Principles*; Chemical Publishing: New York, 1984.
25. Giordano, P.M.; Mortvedt, J.J.; Mays, A.D. Nitrogen effects on mobility and plants uptake of heavy metals in sewage sludge applied to soil columns. J. Environ. Qual. **1976**, *5*, 165–168.
26. Mortvedt, J.J.; Giordano, P.M. Response of corn to zinc and chromium in municipal wastes applied to soil. J. Environ. Qual. **1975**, *4*, 170–174.
27. Maclean, A.J.; Dekker, A.J. Availability of zinc, copper, and nickel of plants grown in sewage-treated soils. Can. J. Soil Sci. **1978**, *58*, 381–389.