Effects of field application of phosphate fertilizers on the availability and uptake of lead, zinc and cadmium by cabbage (*Brassica chinensis* L.) in a mining tailing contaminated soil

WANG Biling[1], XIE Zhengmiao[1,2,*], CHEN Jianjun[1,2], JIANG Juntao[1], SU Qiufeng[2]

1. College of Environmental Science and Natural Resources, Zhejiang University, Hangzhou 310029, China. E-mail: w-biling@sohu.com
2. Department of Environmental Science and Engineering, Hangzhou Dianzi University, Hangzhou 310018, China

Abstract

A field trial was conducted to evaluate the reduction of bioavailability of heavy metals including lead (Pb), zinc (Zn) and cadmium (Cd) in a soil contaminated by mining tailings in Shaoxing, Zhejiang, China. Three commercial phosphate (P) fertilizers including phosphate rock (PR), calcium magnesium phosphate (CMP), and single superphosphate (SSP) were applied to the plot at three P application rates, 50, 300, and 500 g/m^2 with 9 treatments and control (CK). Plants, water soluble and exchangeable (WE) extraction procedure and modified toxicity characteristic leaching procedure (TCLP) were employed. It was found that the addition of all three P fertilizers significantly decreased WE Pb (22.0%–81.4%), Cd (1.5%–30.7%) and Zn (11.7%–75.3%, exception of SSP treatments with no significant difference) and TCLP Pb concentration (27.1%–71.2%), compared with the control, leading to reduced uptake of Pb (16.0%–58.0%), Cd (16.5%–66.9%) and Zn (1.2%–73.2%) by cabbage (*Brassica chinensis* L.). It was suggested that P fertilizers induced immobilization of heavy metals such as Pb, Cd, and Zn. CMP provided the most effective method of reducing metal toxicity, mobility, and phytoavailability. At the P application rate of 300 g/m^2, CMP would be cost-effective to decrease WE Pb, Zn, and Cd concentration from 666 to 137 mg/kg, from 31.2 to 8.71 mg/kg, and from 1.69 to 1.36 mg/kg, respectively. SSP was more effective in reducing Pb bioavailability than PR but had variable effects on Zn bioavailability. Cd uptake by cabbage was negatively correlated with soil pH rather than with WE or TCLP, indicating that Cd uptake by cabbage was a complex process. It should be careful to evaluate the impact of phosphate application on Cd availability in soil.

Key words: bioavailability; *Brassica chinensis* L. (cabbage); cadmium; lead; phosphorus fertilizer; zinc

Introduction

Mining and smelting activities have contaminated soil and water resources with heavy metals throughout the world (Basta and McGowan, 2004). Accumulation of heavy metals in soils and subsequently in food chain are potential threats to human health (Hettiarachchi *et al*., 2000; Wu *et al*., 2000). Therefore, increasing awareness of the hazard makes it necessary to remediate metal contaminated soils.

Soil remediation technologies based on the excavation, transport, and landfill of metal contaminated soils and wastes are highly effective at a low risk, but the cost is high. *In situ* remediation techniques are of particular interest because they are relatively cost-effective compared to conventional techniques (Yang *et al*., 2001). The use of phosphates as stabilizing agent is for Pb particularly attractive (Theodoratos *et al*., 2002).

Lead phosphates, and in particular pyromorphite, are stable forms of Pb in soils under a wide range of environmental conditions (Nriagu, 1974; Lindsay, 1979). Based on its low solubility, a number of studies involving the use of phosphates including P fertilizers for the stabilization of Pb were carried out (Ma *et al*., 1995; Cotter-Howells and Capron, 1996; Hettiarachchi *et al*., 2000; Ryan *et al*., 2001; Tang *et al*., 2004). Formation of pyromorphite and significant reduction of Pb bioavailability in the soils upon addition of phosphate rock (PR) or single superphosphate (SSP) were observed in lead-contaminated soils (Ma *et al*., 1997; Hettiarachchi *et al*., 2001). Reduced plant uptake of Pb was also observed upon PR or SSP addition to lead-contaminated soils (Hettiarachchi and Pierzynski, 2002; Cao *et al*., 2002; Chen and Zhu, 2004). Besides, zinc and cadmium frequently occur simultaneously with lead in the lead and zinc mine, and hence they are detected in the soil contaminated by the mining tailing. Therefore, the effect of phosphate application on Cd and Zn availability is also important. It had been reported that rock phosphate mixed with soils was generally ineffective for reducing Cd and Zn elution with < 27% (Basta and McGowen, 2004). Similarly, the addition of PR or triple-superphosphate (TSP) reduce Cd and Zn uptake in shoot with 39.1%–42.4% and 31.2%–47.3%, respectively (Chen *et al*., 2007).

However, the addition of P source as monobasic calcium phosphate was found to immobilize Pb and Cd, whereas Zn was slightly mobilized (Theodoratos *et al.*, 2002). Although the knowledge of immobilization mechanism of metals, especially Pb, using P amendments is needed, implementation of this technology in the field for remediation of soils and vegetables contaminated by lead and zinc mining tailings is limited, especially in China. Therefore, it is necessary to undertake a field demonstration of this technology, at a site heavily contaminated by heavy metals such as Pb, Zn and Cd from lead and zinc mining tailings. The main objective of this field experiment was to evaluate the effectiveness of three kinds of P fertilizers PR, calcium magnesium phosphate (CMP), and SSP on *in situ* immobilization of Pb, Zn and Cd, and to observe concentration changes of water soluble and exchangeable heavy metals in the contaminated soil in relation to uptake and accumulation of heavy metals by a vegetable plant and to evaluate the feasibility of *in situ* immobilization of Pb in the contaminated soil using P fertilizers in China.

1 Materials and methods

1.1 Site description

The tested site is located in Shaoxing, Zhejiang, China (N30°00′14″, E120°46′39″). The climate style of this site is middle subtropical monsoon climate with the annual average parameters as follows: temperature 16.2°C, rainfall 1,335.9 mm, evaporation 1,260.7 mm, and relative humidity 75.1%. The studied soil (Tilth land, Typentiaqualf) was heavily contaminated with Pb. The polluted area is about 800 hm^2 around the abandoned mine. Based on the previous report, total Pb ranges from 5,271 to 16,369 mg/kg, with an average of 12,552 mg/kg (Xie *et al.*, 2003). The clay mineralogy of the soil was predominantly illite in nature with kaolin in minority. The physical and chemical characteristics are listed in Table 1.

1.2 Phosphate (P) fertilizers

The P fertilizers used in this study were PR, CMP, and SSP. Their main elemental concentrations are given in Table 2.

1.3 Vegetable

A Chinese green vegetable (local name: Shanghai Green) *Brassica chinensis* L. *campestris* was used in this study, as it is not only the most popular vegetable consumed by the local people almost throughout the whole year, but also quite tolerant to heavy metals in the above

Table 2 Concentrations of elements in the tested phosphorus fertilizers

Phosphate fertilizer	PR	CMP	SSP
Main components	$Ca_{10}(PO_4)_6Cl_2$	$Ca_3(PO_4)_2$	$Ca(H_2PO_4)_2$
Form	Farinose	Farinose	Farinose
Size	< 1 mm	< 1 mm	< 1 mm
State of deliquescent	Water-insoluble	Citric-soluble	Water-soluble
pH* (H_2O)	7.41	9.51	3.09
Water soluble P (g/kg)	1.00	14.2	43.0
Citric acid soluble P (g/kg)	24.4	52.0	73.6
Total P (g/kg)	330	129	152
Total Pb (mg/kg)	240	3.20	115
Total Zn (mg/kg)	228	168	234
Total Cd (mg/kg)	3.40	1.80	1.71
Total Cu (mg/kg)	51.3	62.8	61.0

* Measured in 1:2.5 fertilizer/water suspensions.

mentioned contaminated soil. Our preliminary investigation on three main species of local vegetables found that Chrysanthemum (*Dendranthema morifolium* L.) and Spinach (*Spinacia oleracea* L.) with greater enrichment coefficients (ratio of metal concentration in plant to metal concentration in soil) of heavy metals could not grow well, while *B. chinensis* with lower enrichment coefficients grew well on the soil (data not shown).

1.4 Experimental plot design

The field plots were established in the highly polluted zone of the site, after the surface soil (0–20 cm depth) was completely homogenized. Each plot was an area of 1.0 m^2, which were separated by 0.2 m from each other for avoidance of possible inter-plot contamination. P fertilizers including PR, CMP or SSP were applied to the plot at three P application rates (rate 1: 50 g/m^2, rate 2: 300 g/m^2, and rate 3: 500 g/m^2) with 9 treatments (Table 3). The amount of P added was calculated on the basis of the total P content of the P fertilizers. Each treatment had 6 replicates. The 60 plots including 9 treatments and 1 control (CK) were arranged in a completely randomized design (Fig.1).

The P fertilizers were ground to pass through a 1-mm sieve and then completely mixed with surface soil (0–20 cm depth) in experimental plots on April 27, 2004. Plots were watered to keep moist until vegetable sowing. After equilibrium of added P fertilizers with the soil in treated plots, 1,000 seeds of *B. chinensis* were sown in each plot on September 1st, 2004. Urea was applied to each plot 4 weeks after sowing.
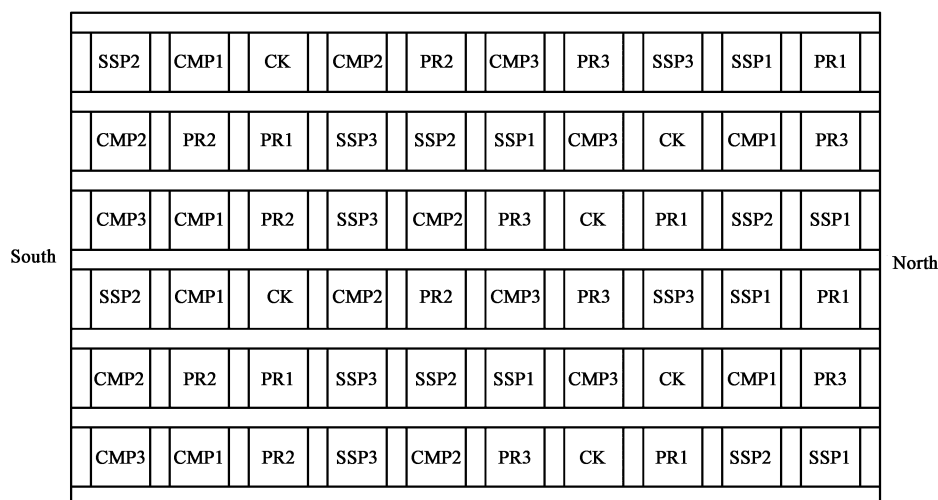
1.5 Soil and plant sampling

Soil samples (0–20 cm depth) were collected on October 27, 2004 which is 6 months after P application. Collected

Table 1 Properties and heavy metal contents of the tested soil

Item	Soil texture	Land use	pH	Organic matter (g/kg)	Total Pb (mg/kg)	Total Zn (mg/kg)	Total Cd (mg/kg)	Total Cu (mg/kg)
Tested soil	Loamy soil	Tilth land	5.51	21.8	16,362	871	5.81	103
Environmental quality standard for soils in China[a]			<6.50		250	200	0.300	50.0
Soil background levels of heavy metals, Zhejiang Province[b]					24.5	84.8	0.202	19.8

[a] SEPA, 1995.

Fig. 1   Arrangements for the experimental plots in the field. Each plot was an area of 1 m^2, which were separated by 0.2 m in distance.

Table 3   Experimental plot design

Treatment	Application rate of P (g/m^2)	Application quantity of P fertilizers (g/m^2)	Application method
CK	–	–	–
PR1	50	150	Rototill treated*
PR2	300	900	Rototill treated
PR3	500	1,500	Rototill treated
CMP1	50	380	Rototill treated
CMP2	300	2,300	Rototill treated
CMP3	500	3,800	Rototill treated
SSP1	50	330	Rototill treated
SSP2	300	2,300	Rototill treated
SSP3	500	3,300	Rototill treated

PR, CMP, and SSP mean three commercial phosphate fertilizers; 1, 2, and 3 stand for three treatments of 50, 300 and 500 gP/m^2 soil. * The top 20 cm soil of the plots was rototilled prior to phosphate fertilizers application, and then phosphate fertilizers were uniformly applied to each plot and rototilled into soil.

soils were air-dried, passed through a 1-mm sieve, and stored in plastic bottles for further analyses. The edible portion of the vegetable was harvested on October 27th, 2004 which is 2 months after sowing and rinsed with deionized water, and then dried in an oven at 65°C for 3 d. The dried vegetable was ground to pass through a 1-mm sieve for digestion.

1.6 Soil chemical analysis

1.6.1 Inorganic P fraction

The fraction of inorganic P in soils was determined with a modified procedure of Jiang and Gu (1989). Soil samples (1.000 g) were sequentially extracted with 0.25 mol/L $NaHCO_3$ for Ca_2-P (dicalcium phosphate bound P) determination, 0.5 mol/L NH_4OAc for Ca_8-P (octocalcium phosphate bound P), 0.5 mol/L NH_4F for Al-P (inorganic P associated with aluminum hydrous oxides), 0.1 mol/L NaOH and 0.1 mol/L Na_2CO_3 for Fe-P (inorganic P associated with iron hydrous oxides), 0.5 mol/L NaOH before digesting with H_2SO_4-$HClO_4$-HNO_3 (1:2:7, *V/V/V*) mixture for O-P (occluded P), and 0.5 mol/L H_2SO_4 for Ca_{10}-P (phosphorite bound P). The amount of inorganic P

was calculated based on the sum of Ca_2-P, Ca_8-P, Al-P, Fe-P, O-P and Ca_{10}-P. P in the supernatant was measured with the molybdenum blue method.

1.6.2 Single chemical extraction

Metal bioavailability and mobility were two major concerns in metal immobilization technique (Melamed *et al.*, 2003). It was well known that water soluble and exchangeable (WE) heavy metals were the most mobile and active species in the soil, and were closely correlated with phytoavailability (Xian, 1989). Therefore, WE leaching test was employed to evaluate the effects on reduction of heavy metals mobility and activity after P fertilizers application. WE heavy metals in the treated soils were obtained from following leaching test: soil samples were extracted at 25°C for 1 h with 1 mol/L $MgCl_2$ (pH 7.0) with 1:8 of soil:solution ratio at 250 r/min. After extraction, separation was done by centrifuging (J2-21, Beckman, USA) at 5,000 r/min for 30 min.

1.6.3 Modified toxicity characteristic leaching procedure (TCLP)

Extracting solution No. 2 of the TCLP was used (USEPA, 1991) in the leaching test. A 5.7-ml aliquot of glacial acetic acid was diluted to 1.0 L with deionized water. The pH of the extracting solution was 2.88 ± 0.01. A 20-ml extracting solution was added to a 50-ml polycarbonate centrifuge tube containing 1.000 g of test soil. The tube was sealed with a cap and placed on an end-over-end shaker at 30 ± 2 r/min for 18 h at 25°C. After extraction, separation was done with centrifuging (J2-21, Beckman, USA) at 5,000 r/min for 30 min.

1.7 Analytical methods

All extractions of treated samples were conducted in triplicate in acid-washed (5% HNO_3) polycarbonate centrifuge tubes and filtered through a 0.2-μm membrane (Schleicher & Schuell, Germany). All chemicals used in this study were of analytical grade or better. Double deionized water was used. All edible vegetable samples were

digested with concentrated nitric acid in a microwave digester (MARS, CEM Corporation, USA) according to modified USEPA method 3051 (USEPA, 1994). The Pb, P, Zn and Cd in vegetable and soil solution were measured by means of an inductively coupled plasma (ICP)-Emission Spectroscopy (Perkin Elmer, USA) using high-purity standard solution (EPA, USA). Standard reference plant material (GBW07602) from the Ministry of Earth and Mine, China was used to verify the accuracy of metal determination. The recovery rates for Pb, P, Zn and Cd were within $(90\pm10)\%$. Solution pH value was determined by Orion Mode EA 940 pH meter (Orion Research, USA). Other soil properties were measured with conventional methods (Lu, 1999).

1.8 Data analysis

All results were expressed as an average of triplicates and treatment effects were determined by analysis of variance using SPSS software (version 10.0). Differences among treatment means were compared by least significant difference (LSD) and Duncan's multiple range tests. For mean separations, LSD values were used at $p < 0.05$.

2 Results and discussion

2.1 Effects of application of P fertilizers on soil pH

Compared with the control, soil pH was significantly changed with the addition of various P fertilizers (Fig.2). At the P application rate of 300 g/m^2, the addition of CMP increased soil pH significantly from 5.23 to 7.42; and the addition of PR increased soil pH by around 0.5 units, while the addition of SSP significantly reduced the soil pH by 0.6 units.

2.2 Effects of application of P fertilizers on WE Pb, Zn and Cd concentrations

Without P fertilizers treatment, WE Pb (666 mg/kg), Zn (31.2 mg/kg) and Cd (1.69 mg/kg) in the contaminated soil, were much higher than the average values in the natural soils, especially Pb. It is well known that the WE fraction is the most bioavailable and phytoavailable (Xian, 1989; Bolan *et al.*, 2003). Thus, heavy metals especially Pb in WE fraction in the studied soil could be easily mobilized and are potential threat to eco-environment.
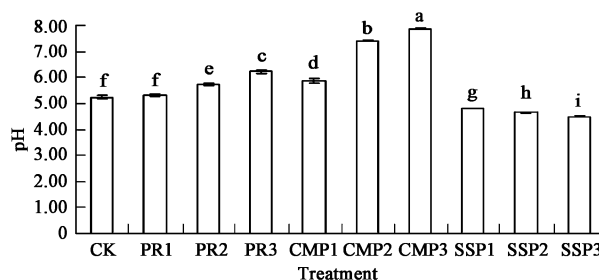

Fig. 2 Effects of the addition of three P fertilizers on soil pH. Means with the same letter are not significantly at $p < 0.05$.

As expected, Pb associating with WE fraction in soil was significantly reduced after application of PR, CMP, and SSP with range 22.8%–35.0%, 59.4%–81.4%, and 22.0%–53.6%, respectively (Table 4). It strongly demonstrated that the effectiveness of P fertilizers on reducing Pb availability was obvious in the field. Similar results have been reported that Pb availability and mobility were reduced by phosphate amendments application (Cao *et al.*, 2002; Brown *et al.*, 2004). It has also been reported that the formation of pyromorphite-like mineral was the probable mechanism for P-induced immobilization (Ma *et al.*, 1995, 1997; Hettiarachchi *et al.*, 2001; Chen *et al.*, 2006), and the dissolution of water-insoluble P compounds was a prerequisite for the immobilization of Pb (Laperche and Traina, 1998). We hypothesized that the reduction of WE Pb concentration by three P fertilizers was mainly through Pb phosphate precipitation, especially pyromorphite-like mineral. Among all three P fertilizers, CMP treatment was most effective in reducing WE Pb concentration. This is partly like that the increased soil pH from 5.7 to 6.5–7.8 after application CMP led to Pb phosphate precipitation from WE. On the other hand, PR is a water-insoluble fertilizer, which could not provide as much soluble phosphorus for decreasing WE Pb concentration as CMP or SSP did. In addition, the trend of the reduction of WE Pb concentration was similar to that of Ca$_2$-P ($r = -0.794^{**}$) and Ca$_8$-P ($r = -0.792^{**}$) (Table 5). It indicated that the effect of Pb immobilization in P fertilizers treated soils may be principally controlled by the solubility of P sources, and that the diffusion of phosphate from dissolving of the hydroxylapatite appeared to be the rate-limiting step in the overall lead pyromorphite formation process (Lower

Table 4 Effects of P fertilizers application on exchangeable Pb, Zn and Cd concentration in the soil

Treatment	Pb		Zn		Cd	
	Concentration (mg/kg)	Reduction (%)	Concentration (mg/kg)	Reduction (%)	Concentration (mg/kg)	Reduction (%)
CK	666 ± 13 a	0	31.2 ± 3.5 ab	0	1.69 ± 0.12 a	0
PR1	451 ± 22 c	32.3	27.5 ± 0.3 bc	11.7	1.66 ± 0.02 ab	1.5
PR2	514 ± 22 b	22.8	25.8 ± 0.9 c	17.1	1.17 ± 0.05 d	30.7
PR3	433 ± 7 c	35.0	26.0 ± 0.3 c	16.8	1.25 ± 0.14 cd	26.2
CMP1	270 ± 8 f	59.4	14.0 ± 0.6 d	55.2	1.51 ± 0.27 abc	10.5
CMP2	137 ± 1 g	79.4	8.71 ± 0.1e	72.1	1.36 ± 0.11 cd	19.5
CMP3	124 ± 3 g	81.4	7.71 ± 0.8 e	75.3	1.37 ± 0.09 cd	18.8
SSP1	520 ± 20 b	22.0	34.0 ± 2.2 a	−8.93	1.44 ± 0.04 abcd	15.0
SSP2	393 ± 1 d	41.0	32.0 ± 0.6 a	−2.7	1.38 ± 0.07 bcd	18.6
SSP3	309 ± 9 e	53.6	33.5 ± 2.2 a	−7.73	1.45 ± 0.08 abcd	14.0

* Values are means ± SE. Means with the same letter in the same column are not statistically different at $p < 0.05$.

Table 5 Correlation coefficient (r) among pH, soil inorganic P fractions concentrations, soil WE heavy metals concentrations and vegetable uptake content (mg/kg) in a contaminated soil after application of SSP, PR and CMP at 3 rates (n=10)

	pH	Ca_2-P[a]	Ca_8-P	Al-P	Fe-P	Ca_{10}-P	WE Pb	WE Zn	WE Cd	V-Pb	V-Zn	V-Cd
pH	1											
Ca_2-P	0.447	1										
Ca_8-P	0.775**	0.875**	1									
Al-P	−0.241	0.675*	0.275	1								
Fe-P	−0.420	0.154	0.074	0.949**	1							
Ca_{10}-P	0.197	−0.353	−0.209	−0.226	−0.196	1						
WE Pb	−0.665*	−0.794**	−0.792**	−0.374	−0.229	0.170	1					
WE Zn	−0.924**	−0.460	−0.730*	0.202	0.345	0.028	0.790**	1				
WE Cd	−0.322	−0.206	−0.228	−0.118	−0.104	−0.634*	0.273	0.201	1			
V-Pb	−0.425	−0.780**	−0.664*	−0.540	−0.429	0.076	0.900**	0.561	0.520	1		
V-Zn	−0.925**	−0.476	−0.720*	0.099	0.278	−0.187	0.789**	0.941**	0.444	0.664*	1	
V-Cd	−0.779**	−0.508	−0.630	−0.104	0.023	−0.233	0.874**	0.855**	0.529	0.807**	0.935**	1

[a] Ca_2-P, Ca_8-P, Al-P, Fe-P, and Ca_{10}-P were the soil inorganic phosphorus fractions, and Ca_2-P stands for dicalcium phosphate bound P, Ca_8-P stands for octo-Ca phosphate bound P, Al-P stands for inorganic P associated with Al hydrous oxides, Fe-P stands for inorganic P associated with iron hydrous oxides, O-P stands for occluded P, and Ca_{10}-P phosphorite bound P. WE Pb stands for water soluble and exchangeable Pb of the soil, and the same as to WE Zn and WE Cd; V-Pb stands for cabbage uptake Pb, and the same as to V-Zn and V-Cd; *p < 0.05; **p < 0.01.

et al., 1998). The WE Pb concentration was decreased along with increased application rate of P fertilizers with an exception in PR treatment. There was little difference in WE Pb concentration reduction of PR treatment between application rate of 50 and 500 g/m^2, while both greater than that of 300 g/m^2 (Table 4). It has been reported that lead retention by PR was primarily attributed to lead phosphate precipitation, surface adsorption or complexation (Cao *et al.*, 2004). Therefore, it was possible that the Pb desorption induced by the increasing of Ca^{2+} was greater than Pb phosphate precipitation induced by phosphorus increment because of the least P solubility, resulting in the increment of WE Pb when increasing the P application rate of PR from 50 to 300 g/m^2.

Compared to CK, WE Zn concentration was significantly reduced in CMP (by 55.2%–75.3%) and PR (by 11.7%–17.1%) treated soils, while slightly increased in SSP treated soils (Table 4). It was obviously related with the acidification of soil. With increasing the SSP application rate the soil pH decreased from initial value of 5.23 to 4.80–4.50. Therefore, SSP application must be carefully designed to reduce Zn mobilization and co-application of liming materials with SSP may be necessary to offset potential soil acidification. In addition, trend of WE Zn concentration reduction by increasing CMP application rate was similar to that of WE Pb. In contrast, there was not significant effect in PR and SSP treated soils when increasing application rate. It was reported that Zn retention by PR was mainly attributable to the surface adsorption or complexation (Cao *et al.*, 2004). If surface complexation on functional groups is a primary factor controlling Zn availability, it would be expected that soil pH value would be a dominant factor controlling the number of specific adsorption sites. Data showed (Table 5) that WE Zn concentration in the soil was strongly inversely correlated (r = −0.924**) with soil pH values, indicating that all three P fertilizers induced Zn reduction were primarily attributable to adsorption and surface complexation.
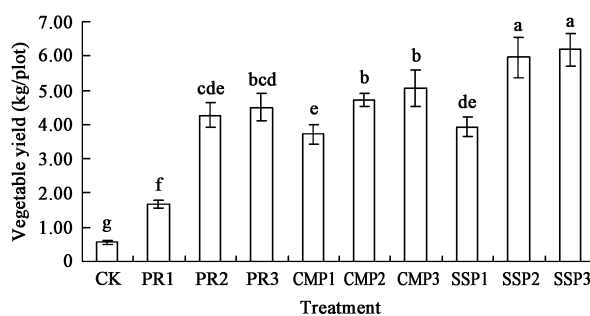
Not surprisingly, there were significant reductions of WE Cd concentration in the treated soils after P fertilizer application (Table 4), and the trend of reduction followed:

PR (up to 30.7%) > SSP (14.0%–18.6%), CMP (10.5%–19.5%). A reduction in Cd concentration by three P fertilizers would be in agreement with the reduction in exchangeable Cd by KH_2PO_4 (Zwonitzer *et al.*, 2003). It was well known that PR not only provided huge surface areas, but also increased the soil pH values and subsequently enhanced the Cd adsorption induced by PR application. The WE Cd concentration was inversely correlated with Ca_{10}-P (r = −0.634*) (Table 5), confirming that Cd retention by phosphates was via surface adsorption.

2.3 Effects of P fertilizer application on Pb, Cd and Zn uptake by cabbage

Biomass can be used as a useful indicator for the overall health of plant growing on the Pb, Zn and Cd contaminated soil. Compared to CK, all treatments of SSP, PR and CMP resulted in significantly higher biomass (Fig.3). Although the vegetable is tolerant to heavy metals, its biomass was low. It is probably due to the high concentrations of metals in the soil (Table 1). There were obvious toxicity symptoms (burning at leaf edges) in the vegetable grown on the CK plot.
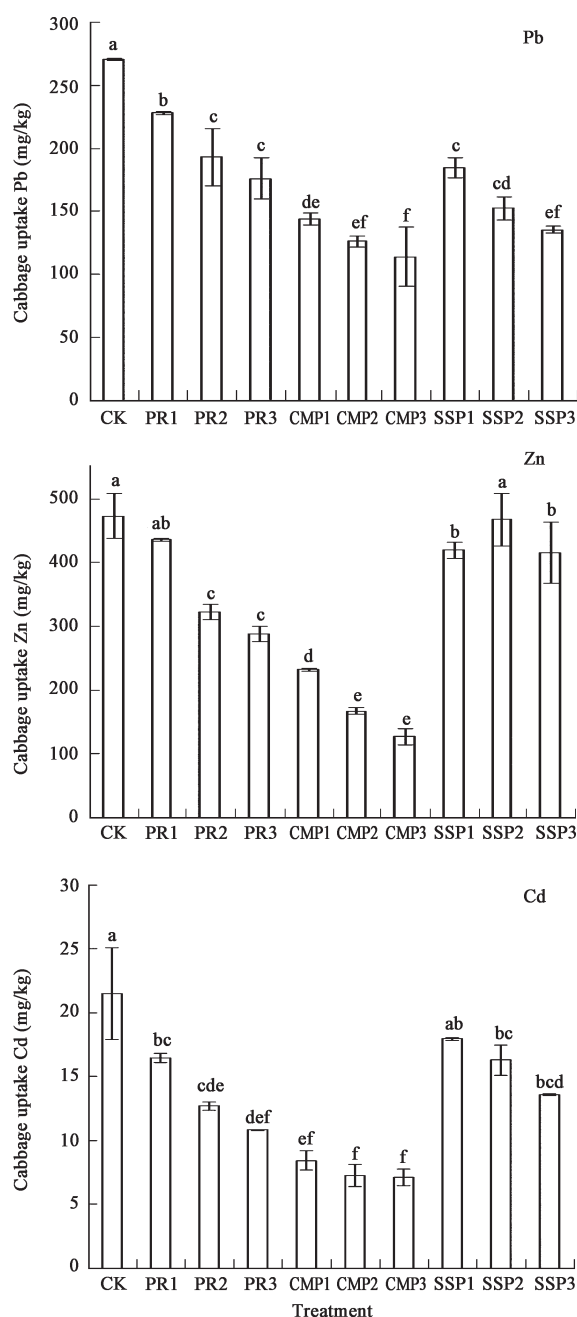
The concentrations of Pb (V-Pb), Zn (V-Zn) and Cd (V-Cd) in shoot tissue of cabbage grown in the treated soil plots were shown in the Fig.4. Pb, Zn, Cd accumulations varied between 114 and 271 mg/kg dry matter, 127 and 473 mg/kg dry matter, and 7 and 22 mg/kg dry matter,

Fig. 3 Effects of P fertilizers application on the biomass of cabbage (*Brassica chinensis* L.). Means with the same letter are not significantly at p < 0.05.

respectively, without obvious correlation with the application rates. Generally, the uptakes of these contaminants were significantly reduced with PR, CMP or SSP treatments.

Compared with CK, the addition of PR, CMP, and SSP significantly decreased the Pb concentration in the cabbage by 16.0%–35.0%, 47.0%–58.0%, and 32.0%–50.0%, respectively. Similar results were obtained in which Pb concentration in the tissue of St. Augustine grass (*Stenotaphrum secundatum*) was reduced after application of P to Pb polluted soils, which may be due to the formation of Pb-P precipitate on the root surface, within the root rhizosphere and in the bulk soil (Cao *et al.*, 2002).

Pb-Ca-phosphates had been identified on the cell wall of the plant roots using SEM (scanning electron microscope), TEM (transmission electron microscope), and XRD (X-ray diffraction) in the P treated soils (Chen *et al.*, 2006). There was a strong correlation ($r = 0.900**$) between V-Pb and soil WE Pb (Table 5). This indicated that it would be possible for P fertilizers to reduce Pb uptake by cabbage via reduction of soil WE Pb concentration. Therefore, it was hypothesized that the formation of pyromorphite-like mineral (or Pb-Ca-phosphates) in the soils and on the root cell wall of cabbage may be responsible for decreasing Pb translocation from the roots to shoots, thereby reducing Pb concentration in shoot after P fertilizers application. Data showed that at P application rate of 300 g/m^2 soils, there was no significant difference between SSP and PR in the reduction of cabbage Pb uptake, while both were much smaller than CMP (Fig.4). It was possible that addition of CMP not only underwent sufficient dissolution to immobilize Pb via precipitation of pyromorphite-like mineral, but also increased soil pH and induced Pb phosphate precipitation.

The Cd concentrations in shoots generally decreased by 16.5%–66.9% after application of PR, CMP, or SSP indicating that Cd uptake was influenced by the addition of P fertilizers. The addition of P may reduce Cd phytoavailability through a combination of several mechanisms, such as sorption (including phosphate-induced Cd adsorption and surface complexation), precipitation, or co-precipitation (Laperche and Traina, 1998; Valsami-Jones *et al.*, 1998). However, there were no Cd phosphate minerals formed freshly by the end of experiment identified with XRD (Ma *et al.*, 1994), and it was speculated that a solid residue containing Cd was Ca-Cd phosphate or Ca-Pb-Cd phosphate (Ma *et al.*, 1994; Valsami-Jones *et al.*, 1998). Chen *et al.* (1997) suggested that reduction in aqueous Cd concentrations with apatite addition occurred primarily because of sorption mechanisms, such as surface complexation and ion exchange rather than precipitation of Cd phosphate. However, the lack of a strong relationship ($r = 0.529$) between V-Cd and soil WE Cd concentration and a strong inverse correlation ($r = -0.779**$) between V-Cd and soil pH values suggested that it was not sufficient to evaluate the effect of P fertilizers application on Cd immobilization only by soil WE extraction procedure.

The Zn concentration in the aboveground biomass of cabbage decreased significantly (31.8%–73.2%) with an exception of SSP (1.2%–12.2%) which may be owing to its lower pH. Hettiarachchi and Pierzynski (2002) reported that plant tissue concentrations of Pb, Zn and Cd were consistently reduced in the presence of soluble P, possibly through the formation of mixed-metal phosphates. Similarly, addition of CMP and SSP decreased Zn concentration in the aboveground biomass of cabbage may possibly through the formation of mixed-metal phosphates in soil (such as Zn-Pb-Cd phosphate). Hettiarachchi and Pierzynski (2002) also reported that the addition of PR reduced plant tissue concentrations of Zn was possibly through enhanced sorption mechanisms such as surface complexation and ion exchange. Meanwhile, Cao *et al.*

Fig. 4 Effects of P fertilizers application on uptake of Pb, Zn and Cd by cabbage (*Brassica chinensis* L.). Means with the same letter are not significantly at $p < 0.05$.

(2004) had a similar report that Zn immobilization by PR was mainly attributable to the surface adsorption or complexation in the soil system.

The results suggest that there are some relationships between the mechanisms controlling the phytoavailability of Pb, Zn and Cd (Table 5). As would be expected due to their similar characteristics, the relationship between V-Zn and V-Cd ($r = 0.935^{**}$) was stronger than V-Zn and V-Pb ($r = 0.664^{*}$). At the P application rate of 300 g/m^2, effects of three P fertilizers on reducing Cd and Zn concentrations in cabbage followed the same order: CMP > PR > SSP. It was possible that addition of CMP not only underwent sufficient dissolution to immobilize Pb via precipitation of mixed-metal phosphates (such as Zn-Pb-Cd phosphate) mineral, but also increased soil pH and then induced Cd or Zn phosphate precipitation such as $Zn_3(PO_4)_2 \cdot 4H_2O$, because CMP is an alkaline citric-acid-soluble fertilizer. Like CMP, addition of PR could also increase soil pH and then induce Cd or Zn phosphate precipitation, and immobilize Cd and Zn via surface adsorption or complexation at the same time.

2.4 Effects of application of P fertilizers on soil inorganic P fraction

Soil P enrichment and potential leaching can be an environmental concern for *in situ* metal immobilization using P fertilizers. More details of P distribution in the soil can be obtained from the soil inorganic P fraction procedure after P fertilizers application. Therefore, it was meaningful to evaluate soil P availability and potential risk (Fig.5). Without P fertilizers treatment, P in the contaminated soil was primarily associated with O-P fraction (47.6% of inorganic P) and Fe-P fraction (42.5% of inorganic P), following Ca_{10}-P fraction with 8.3% of inorganic P. Data showed that after the application of P fertilizers, there was some difference in P distribution in the treated soils among three different P fertilizers treatments. CMP was primarily transferred into Ca_8-P, Al-P and Ca_2-P, and P concentration was significantly increased in four fractions including Ca_8-P, Al-P, Ca_2-P and Fe-P in SSP treated soils. However, there was not influenced by PR application on P fraction except the increase of P concentration in Ca_{10}-P. SSP was a water-soluble and fast-release fertilizer, and followed by CMP, but PR is a water-insoluble (slow-release) fertilizer
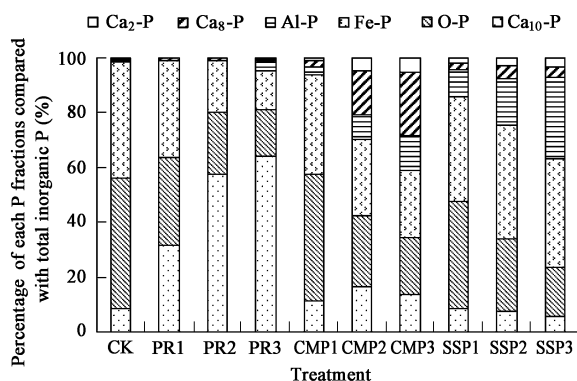
(Bolan *et al.*, 2003). This is also supported by the data of soluble P in Table 2. It was suggested that SSP supply the most P availability, followed by CMP and PR, so as to the potential risk. A bivariate correlation analysis was carried out among soil inorganic P fraction and WE metals, V-metals. The result indicated that Ca_2-P was most closely correlated with WE Pb, ($r = -0.794^{**}$), and V-Pb ($r = -0.780^{**}$), Ca_8-P was most closely correlated with WE Pb ($r = -0.792^{**}$), WE Zn ($r = -0.730^{*}$), V-Pb ($r = -0.664^{*}$), and V-Zn ($r = -0.720^{*}$), and Ca_{10}-P was most closely correlated with WE Cd ($r = -0.634^{*}$) (Table 5).

2.5 Effects of P fertilizer application on TCLP Pb, Zn, and Cd

The results of TCLP tests (Table 6) showed that all three P fertilizers reduced TCLP Pb by 27.1%–71.2%. The results were similar to the reports that the addition of P as $Ca(H_2PO_4)_2 \cdot H_2O$ reduced extractable Pb in treated soils in a laboratory study and all P amendment reduced extractable Pb in the top 10-cm profile in a field study (Hettiarachchi *et al.*, 2000, Chen *et al.*, 2003). Similar to WE Pb, the effect of extractable Pb reduced by P fertilizers was in the order: CMP > SSP > PR (Table 6). Addition of P fertilizers reduced extractable Pb significantly, although the level of Pb was still higher than the critical level of 5 mg/L. The fact that TCLP Pb concentration in untreated soil was 701 mg/kg which was much higher than those reported by Hettiarachchi *et al.* (2000) or Chen *et al.* (2003), e.g., 34.3 mg/kg. It may because the levels of P addition used in this study were much lower.

Nevertheless, addition of P fertilizers had a negative impact on TCLP Zn and Cd (Table 6), which increased from 14.8 to 32.4 mg/kg, and 0.559 to 2.707 mg/kg, respectively. A great increase of Ca^{2+} concentration (data was not shown) in the soil along with P fertilizers application and the low pH (2.88) of leaching solution used in this study may be responsible for it. However, the uptake of Zn and Cd by the vegetable was significantly reduced after P fertilizers application in despite of increased TCLP Zn and Cd concentrations, indicating that TCLP Zn and Cd could not be a decisive index but as a consulting parameter for the assessing the environmental risk of P fertilizers application on Zn and Cd activity.

Fig. 5 Effects of the addition of three P fertilizers on soil inorganic P fraction.

Table 6 Effects of P fertilizers application on TCLP Pb, Zn and Cd concentrations in the soil

	Pb (mg/kg)	Zn (mg/kg)	Cd (mg/kg)
CK	701 ± 58* a	14.8 ± 0.14 f	0.559 ± 0.023 c
PR1	500 ± 20 bc	23.4 ± 0.64 cd	1.553 ± 0.027 b
PR2	441 ± 5 cd	19.2 ± 0.21 e	2.088 ± 0.021 b
PR3	511 ± 44 b	32.2 ± 0.85 a	2.707 ± 0.638 b
CMP1	467 ± 19 bc	21.6 ± 0.28 d	1.551 ± 0.042 b
CMP2	321 ± 47 e	25.6 ± 0.42 b	1.673 ± 0.540 b
CMP3	202 ± 14 f	31.8 ± 0.57 a	1.882 ± 0.122 b
SSP1	401 ± 1 d	19.2 ± 0.14 e	0.969 ± 0.015 c
SSP2	342 ± 6 e	24.8 ± 0.57 bc	0.910 ± 0.035 c
SSP3	299 ± 20 e	32.4 ± 1.70 a	1.943 ± 0.118 b
TCLP limit	5	25	1

* Values are means ± SE. Means with the same letter in the same column are not statistically different at $p < 0.05$.

3 Conclusions

Results from this field trial indicate that: (1) P fertilizers induced immobilization of heavy metals Pb, Cd, and Zn. CMP provided the most effective method of reducing metal toxicity, mobility, and phytoavailability. SSP was more effective in reducing Pb bioavailability than PR but had variable effects on Zn bioavailability. (2) The level of P fertilizers at application rate of 300 g/m^2 was enough to reduce metals availability in the soil (with a reduction of up to 79% in WE Pb) and phytoability (up to 47% in V-Pb) at the first stage of remediation. (3) Cd uptake by cabbage was a complex process and it should be careful to evaluate the impact of phosphate application on cadmium availability in soil. (4) SSP application must be carefully designed to reduce Zn mobilization and co-application of liming materials with SSP may be necessary to offset potential soil acidification.

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