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# Immobilization and phytotoxicity of Cd in contaminated soil amended with chicken manure compost

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#### ABSTRACT

The experiment was conducted to evaluate the effect of compost application on the immobilization and biotoxicity of Cd in winter wheat (*Triticum aestivum* L.) potted soils. Soils treated with various levels of Cd (0–50 mg Cd kg<sup>-1</sup> soil) were amended with 0, 30, 60 and 120 g compost kg<sup>-1</sup> soil. The fractions of Cd in soil were evaluated by a sequential extraction procedure. Compost application resulted in more than 70% lower soluble/exchangeable Cd (KNO<sub>3</sub>) but increased the concentration of organic-bound (NaOH) and inorganic precipitates (EDTA) Cd in soils. Addition of compost was effective in reducing the phytotoxicity of Cd by decreasing more than 50% Cd uptake by wheat tissue and improving wheat growth. Alleviation of Cd phytotoxicity by compost was attributed primarily to the increase of soil pH, complexation of Cd by the organic matter and coprecipitation with P content. Compost was effective in the immobilization of Cd in soils and can be used to remediate Cd-contaminated soils.

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# 1. Introduction

In many countries, cadmium (Cd) has been identified as a major toxic heavy metal reaching the food chain, directly through crop uptake and indirectly through animal transfer [1]. Cadmium is known as more mobile and soluble than many other metals in soils. Like most other metals, Cd does not undergo microbial or chemical degradation and therefore persists in soils for a long time after its introduction. There is sufficient evidence in humans for the carcinogenicity of Cd and Cd compounds, and for genotoxic effect of ionic forms in a variety of types of eukaryotic cells, including human ones [2].

Contaminated soil often presents an unacceptable risk to human and ecological health and must be remediated. Chemical immobilization is an in situ remediation method where inexpensive materials such as fertilizer and waste products are added to contaminated soil to reduce the solubility and bioavailablity of heavy metals. Traditionally, biosoild is viewed as one of major sources of metal accumulation in soils, and a large volume of work has been carried out to examine the mobilization and bioavailability of biosoild-borne metals in soil [3–8]. Many experiments have been conducted in order to study the effectiveness of different materials for the immobilization of heavy metals in contaminated soils [9–14]. Organic amendments such as composts or peat, which contain a high proportion of humified organic matter (OM), can decrease the bioavailability of heavy metals in soil by adsorption and by forming stable complexes with humic substances [15], thus permitting the re-establishment of vegetation on contaminated sites [16]. Furthermore, stabilization using alkaline materials has resulted in the immobilization of metals in biosoild. This alkalinestabilized biosoild compost can be used as an effective sink for reducing the bioavailability of metals in contaminated soils and sediments [11]. Basta et al. [14] demonstrated that alkaline organic treatments (Lime-stabilized biosolid and N-Viro soil) reduced the phytoavailability of Cd and Zn in soils contaminated by Zn and Pb milling and smelting operations in Oklahoma. Addition of biosolid compost in Egmont and Manawatu soils treated with various levels of Cd  $(0-10 \text{ mg Cd kg}^{-1} \text{ soil})$  was effective in reducing the phytotoxicity of Cd as indicated by the decrease in the concentration of NH<sub>4</sub>OAc extractable-Cd and soil solution-Cd [11]. Clemente et al. [17] reported that both fresh cow manure and compost having a high maturity degree amendments favoured Zn and Pb fixation in soil from a former Pb-Zn mine area at La Union (Murcia, SE Spain), particularly the manure. In an acidic, Cu-Zn minespoil, a yard waste compost rich in humic and fulvic acid was found to effectively bind heavy metals and reduce toxic bioavailable concentrations of Cu and Zn [18]. High Fe biosolids composts were shown to decrease in vitro and in vivo available Pb as well as phytoavailable Zn and Cd

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Table 1
Chemical characteristic of soil and compost

	pН	Total N (g kg <sup>-1</sup> )	Total P $(g kg^{-1})$	Available N (g kg $^{-1}$ )	Available P (mg kg <sup>-1</sup> )	Available K (g $kg^{-1}$ )	$OM(gkg^{-1})$	Total Cd (mg $kg^{-1}$ )
Soil	5.50	0.37	0.52	0.017	8.47	0.04	2.96	0.17
Compost	8.11	3.63	5.90	0.45	79.42	1.07	272.7	0.75

[19,20]. These findings can be integrated to guide the application of organic materials for heavy metal-contaminated soils revegetation projects.

Although quite a lot of investigations have been carried out for compost as potential contamination sources of heavy metals, limited work had been done regarding the beneficial effect of organic amendments as a sink for the immobilization of Cd in soils [11,14]. The objectives of the present study were to determine and compare the phytoavailability of Cd in soils applied with different rates of Cd and chicken manure compost, and to evaluate the remediating effect of compost on Cd-contaminated soils by pot experiments under greenhouse conditions.

#### 2. Materials and methods

#### 2.1. Soil and compost

A soil (Ferralsol) from the top 20 cm was collected from the university farm of Huazhong Agricultural University, Hubei Province, China. It is a fine clay-loam soil with 8.53% sand, 31.09% clay and 60.4% silt. The soil samples were air-dried for 1 week and passed through 2 mm sieve before use in pot experiments. The compost which was made from poultry manure and chaff and matured for 6 months was obtained from The Biological Engineering Company of Wuhan, China. The compost was air-dried and ground to pass 0.149 mm sieve for subsequent analysis. The pH of soil and compost was measured using 1:2.5 (w/v) soil/water ratio. Organic matter was analyzed by potassium dichromate oxidation and titration with ferrous ammonium sulphate [21]. Soil and compost samples were digested by H<sub>2</sub>SO<sub>4</sub> [21]. The total N and P were measured by FIAstar 5000. Available N was determined by NaOH pervasion method. Available P was determined by NaHCO<sub>3</sub> method. Available K was analyzed by AAS (Varian AA240FS) after extraction with NH<sub>4</sub>OAc. Some chemical characteristics of soil and compost are listed in Table 1.

# 2.2. Pot experiment

The experiment was conducted in greenhouse. The soil was treated with four levels of Cd (0, 2, 10 and  $50 \text{ mg kg}^{-1}$  soil) using CdCl<sub>2</sub> solution. The Cd-treated samples were subsequently amended with compost at four rates (0, 30, 60 and 120 mg kg<sup>-1</sup> of compost). The soil samples treated with compost and Cd were thoroughly mixed, watered and placed for 2 days in order to get to the equilibration of Cd between compost and soil. The soil was then transferred to plastic pots (13 cm × 12 cm) and each pot contained 2 kg soil. Treatments were arranged in a completely randomized design with four replicates per treatment. The treated soils were maintained at 60% water holding capacity with distilled water.

 Table 2

 Sequential extraction of Cd in soil (according to Sposito et al. [22])

Twenty seeds of winter wheat (*Triticum aestivum* L.) were sown in each pot and after germination the seedlings were thinned to ten per pot. Soil moisture content was maintained at 60% of field capacity during the germination period and it was adjusted to field capacity after thinning. Wheat was harvested 4 months after sowing.

Wheat seeds and stems were dried at 65 °C for 48 h. The dry weights of seeds were recorded, and the seeds and stems were ground using a stainless steel grinder. Soil samples were collected from each pot after harvesting, air-dried and ground to pass 2 mm sieve for subsequent analysis.

# 2.3. Total Cd and fractionation of Cd in soils

The plant materials were digested with concentrated nitric acid and Cd was determined by graphite-furnace atomic absorption spectrometry (AAS). Total soil Cd was extracted using aqua regia and determined by AAS (Varian AA240FS).

Fractionations of soil Cd were obtained according to the sequential extraction scheme described by Sposito et al. [22]. A brief summary of the procedure is presented in Table 2.

# 2.4. Statistical analysis

The data sets were analyzed using the ANOVA procedure of SPSS and differences between means were determined using least significant difference (LSD) test.

## 3. Results

# 3.1. Soil chemical properties

Soil pH, organic matter (OM) and total P in the absence and presence of compost are shown in Table 3. For all Cd-treated soils, soil pH increased by 0.5-1.2 units after compost application. In general, compost-amended soils resulted in significantly (p < 0.05) higher OM and total P contents compared with the soils without compost. With the increase of compost application in Cd treated soils, the content of OM and total P increased by 0.2-1.9 and 0.2-3.2 times, respectively, comparing to compost 0 soils. Marked differences (p < 0.05) for OM and total P was also found among compost-amended soils. Apparently, compost addition increased soil pH values and the content of OM and total P.

#### 3.2. Cadmium fractions in soils

The amount of Cd in different soil fractions is presented in Fig. 1. In Cd 0 soils, Cd content in all fractions was below  $0.25 \text{ mg kg}^{-1}$ and no significant influence was observed for compost application

Steps	Fractionation	Expression	Reagent	Soil:solution ratio	Experiment condition
1 2 3 4	Soluble/exchangeable Organic-bound Inorganic precipitates Residual	KNO3 NaOH EDTA HNO3	0.5 mol 1 <sup>-1</sup> KNO <sub>3</sub> 0.5 mol 1 <sup>-1</sup> NaOH 0.05 mol 1 <sup>-1</sup> Na <sub>2</sub> EDTA 4 mol 1 <sup>-1</sup> HNO <sub>3</sub>	1:10 (w/v) 1:10 (w/v) 1:10 (w/v) 1:10 (w/v)	Shake 16 h at 20°C Shake 16 h at 20°C Shake 6 h at 20°C Shake 16 h at 89°C

# Table 3

Soil pH, organic matter and total	P after compost amendments
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Cd treatments (mg kg <sup>-1</sup> )	Compost amendments (g kg <sup>-1</sup> )	рН	Organic matter (g kg <sup>-1</sup> )	Total P (mg kg <sup>-1</sup> )
0	0	5.95	3.2hijk <sup>a</sup>	0.44gh
	30	6.71	4.55fg	0.53g
	60	7.39	7.4c	1.34bc
	120	7.44	9.33a	1.49b
2	0	6.17	3.19hijk	0.44gh
	30	6.78	3.87ghij	0.80e
	60	7.28	6.37d	0.98d
	120	7.31	9.1a	1.69a
10	0	6.17	3.19hijk	0.34h
	30	6.92	4.1gh	0.64f
	60	7.32	6.14de	1.03d
	120	7.35	8.31b	1.49b
50	0	6.09	2.84ik	0.40h
	30	6.60	3.87gh	0.65f
	60	7.21	5.23ef	0.87e
	120	7.38	7.6bc	1.68b

<sup>a</sup> Means with	the same	letters (a	–k) withii	n a column	are not	significantly	different
( <i>p</i> < 0.05).							



**Fig. 1.** The amount of Cd fractions in soils treated with Cd and compost by sequential extraction scheme.



Fig. 2. The dry matter yield of winter wheat (*Triticum aestivum* L.) seeds at various amount of chicken manure compost application.

on the distribution of various fractions of Cd. For Cd 2 soils. substantial changes were observed in terms of Cd fractions exerted by compost. The amount of soluble/exchangeable Cd was 1.34 mg kg<sup>-1</sup> in the absence of compost. It lowered to  $0.16 \text{ mg kg}^{-1}$  in the presence of compost 30. No further decrease of soluble/exchangeable Cd was observed for compost 60 and 120 soils. Organic-bound Cd increased steadily from 0.28 to  $0.68 \text{ mg kg}^{-1}$  and inorganic precipitates Cd increased remarkably from 0.05 to 0.88 mg kg<sup>-1</sup> with increasing amount of compost addition. No significant difference was found for residual Cd with compost application. In Cd 10 and 50 soils, soluble/exchangeable Cd was 6.39 and 20.66 mg kg<sup>-1</sup> without compost, respectively. Compost 30, 60 and 120 decreased the amount of soluble/exchangeable Cd by 71.8-95.7%. Increases by 0.9-7.8 times were found for organicbound Cd in these high level Cd-treated soils after compost application. About 0.6-1.5 times increases were also detected for inorganic precipitates as a result of compost applications. No significant differences were observed for inorganic precipitates and residual Cd among compost 30, 60 and 120 in Cd 10 and 50 soils. Overall, with increasing rate of compost addition to soil, the amount of soluble/exchangeable Cd decreased considerably in Cd treated soils, whereas organic-bound, inorganic precipitates content were increased accordingly.

# 3.3. Plant growth

Fig. 2 shows that in Cd 0 soils, the wheat seed yields was  $2.98 \text{ g pot}^{-1}$  without compost, and it increased by 1.73, 2.44 and 3.34 times for compost 30, 60 and 120, respectively. Compost 30, 60 and 120 in Cd 2 soils improved wheat seed yield by 1.14, 1.8 and 2.34 times, respectively. There was almost no wheat seed yield in Cd 10 and 50 soils without compost. Compost application increased the wheat seed yields from 7.17 to 12.4 g/pot for Cd 10 soils and from 5.59 to 8.73 g/pot for Cd 50 soils. These results indicate that compost amendments increased significantly the wheat seed yield for all Cd treated soils. For high Cd-treated soils, the increase of wheat seed yield was more evident by compost amendments, showing more significant ameliorations of compost in heavily polluted Cd soils.

Plant height of wheat was also affected by compost in all Cd treated soils (Table 4). The wheat plant height was taller in the presence than in the absence of compost. No significant differences were found for plant height among compost 30, 60 and 120. These results show obviously the beneficial effects of compost on weight of thousand seeds and plant height in Cd treated soils.

#### Table 4

Plant height and Co	concentration in wheat (	(Triticum aestivum L.	) tissues

Soil Cd level (mg kg <sup>-1</sup> )	Compost treatments (g kg <sup>-1</sup> )	Plant height (cm)	Cd concen	tration (mg kg <sup>-1</sup> dw)
			Seed	Stem
0	0	44.75f <sup>a</sup>	0.17h	0.18h
	30	69ab	0.13h	0.34h
	60	63.75bcd	0.13h	0.4h
	120	63bcd	0.1h	0.8h
2	0	62cd	2.2e	16.8f
	30	74a	1.48f	7.34g
	60	72.75a	1.08fg	4.23gh
	120	67.5abc	0.85g	1.83h
10	0	35.5f	-	58.84c
	30	69ab	7.02b	34.9d
	60	67.25abc	4.57c	24.66e
	120	59.25d	3.5d	16.98f
50	0	32.25f	-	116.18a
	30	62.75bcd	9.58a	65.63b
	60	62.5bcd	6.6b	58.21c
	120	57.5d	4.62c	54.58c

<sup>a</sup>Means with the same letters (a–h) within a column are not significantly different (p < 0.05).

#### 3.4. Cd uptake by plant

The concentration of Cd in winter wheat seeds and stems was listed in Table 4. In Cd 0 soils, no significant changes of Cd content were observed for wheat seeds and stems after compost application. In Cd 2 soils, Cd concentration decreased by 33-61.4% in wheat seeds and by 56.3-89.1% in wheat stems with increasing compost application. In Cd 10 and 50 soils, compost 120 resulted in a more than 50% lower Cd concentration in wheat seeds than that of compost 30. For wheat stems, Cd content decreased by 40.5-71.2% in Cd 10 soils for compost 30, 60 and 120. In Cd 50 soils, compost 30 decreased Cd content in wheat stems by nearly 50% and increasing amount of compost application (60 and 120) resulted in further decrease in Cd concentration. The concentration of Cd in wheat seeds and stems was the lowest for the amendment of compost 120. These data reveal that addition of compost into Cd-treated soils decreased significantly Cd concentration in wheat seeds and stems, the effect being more pronounced at higher soil Cd levels.

#### 4. Discussions

Our results show the promotive effect of compost application on the seed yield of wheat grown in Cd-polluted soils. The uptake of Cd by wheat seeds and stems decreased substantially after compost amendments in these soils. The decreasing effect of compost on Cd accumulation by plants was more remarkable at higher soil Cd levels. The alleviating effect of compost on the toxicity of Cd to wheat could be assigned to the redistribution of Cd species in soils affected by compost. Compost application resulted in decrease of soluble/exchangeable Cd by 71.8-95.7% and increased organicbound and inorganic precipitates Cd by 0.4-18.4 times. These observations suggest that soluble/exchangeable Cd was converted to organic-bound and inorganic precipitates forms. The conversion of bioavailable Cd species to inert ones is vital in governing the uptake of Cd by plants and the phytotoxicity of the element. Several mechanisms may explain the transformation of Cd species in soils. Firstly, the pH of Cd-treated soils without compost increased from 5.95 to 7.44 in compost 120. Increase of soil pH may facilitate the adsorption of Cd on various soil binding sites, thus decreasing the partition of Cd to soil solution [23-25]. Secondly, the high P content of the compost (Table 1) probably contributed to the immobilization of Cd in soil. With increasing amount of compost application, inorganic precipitates Cd increased by 0.6-1.5 times, suggesting that a large proportion of soil mobile Cd may react with inorganic phosphorous compounds to form insoluble precipitates and decrease the availability of Cd. In highly metal-contaminated soils, precipitation as metal phosphates can play a major role in metal immobilization. The release of phosphates, carbonates and other salts mineralized from organic matter may form insoluble metal compounds and limit metal solubility which ultimately increase the proportion of inorganic precipitates Cd [9,26,27]. In an experiment with compost and fresh cow manure, Walker et al. [10] suggested that changes in soil pH and the presence of phosphorus and inorganic salts could contribute more to the change in heavy metal fraction in soil than the nature and the humification degree of OM. Thirdly, the increase of soil organic matter (OM) following compost application is responsible for the increment of organic bound Cd in soils. Thus, some heavy metals may change from soluble and exchangeable forms to fractions associated with organic matter. In the present study, the concentration of soil OM in Cd-polluted soils increased by 0.2-1.9 times after compost application (Table 3), and the organic bound Cd increased by 0.4–7.8 times accordingly. Organic materials provide a large number of non-specific and specific sorption sites for metals from which they may be difficult to displace [15]. The redistribution of Cd was strongly influenced by soil pH, organic matter, total P content, which in turn was influenced by compost application.

# 5. Conclusions

Addition of compost was effective in reducing Cd uptake by wheat seeds and stems, and increasing wheat growth in Cd-treated soils. The alleviating effect of compost on the phytotoxicity of Cd was mainly assigned to the redistribution of labile soil Cd. Compost amendments resulted in more than 70% lower of soluble/exchangeable Cd in soils. The remediation effect of compost on Cd-contaminated soil was influenced by the amount of compost addition and soil Cd level. Therefore compost amendment of Cdpolluted soil not only reduced the uptake of Cd by wheat seeds and stems, but also improved the performance of crops. It can be safely and effectively used for the restoration of Cd-contaminated soils.

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