Reduction of Cadmium Uptake of Rice Plants Using Soil Amendments in High Cadmium Contaminated Soil: A Pot Experiment

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ABSTRACT

The aims of this study were to investigate the effect of agricultural residues on reducing cadmium uptake in rice plants. The rice plants growing on no cadmium/free cadmium soils (N), Cd soils (Cds), and Cd soils each amended with 1% w/w of coir pith (CP), coir pith modified with sodium hydroxide (CPm) and corncob (CC) under high cadmium contaminated soil with an average 145 mg Cd kg^{-1} soil were investigated. The results showed that the cumulative transpiration of rice grown in various treatments under high cadmium contaminated soil followed the order: Cds > CPm ≥ CP ≥ CC. These transpirations directly influenced cadmium accumulation in shoots and husks of rice plants. The CC and CP seemed to work to reduce the cadmium uptake by rice plants indicated by accumulated cadmium in the husk that were 2.47 and 7.38 mg Cd kg^{-1} dry weight, respectively. Overall, transpiration tended to drive cadmium accumulation in plants for rice grown in high cadmium contaminated soil. The more that plants uptake cadmium, the lower cadmium that remains in the soil.

Keywords: *transpiration, cadmium uptake, rice plant, agricultural residues*

INTRODUCTION

The study concerned in the rice-based agricultural system on Mae Sot District, Thailand reported that the area contained natural Zn-Cd silicate ore which caused the total cadmium in soil to have wide ranges from 0.5 to 284 mg Cd kg^{-1} soil with most of them higher than European Economic Community (EEC) maximum permissible level for cadmium soil concentration of 3.0 mg Cd kg^{-1} soil. Most of the cadmium concentrations of grain yielded from rice farming ranged from 0.05 to 7.7 mg Cd kg^{-1} dry weight which is higher thanthe Thai standardof 0.15 mg Cd kg^{-1}dry weight. As a consequence, theestimation values of weekly in-take (WI) of cadmium from people living around the area ranged from 20-80 µg Cd kg^{-1} body weights [1].

Rice is a staple food that is rich in cadmium with the potential to cause negative health effects such as diarrhea, stomach pains, and severe vomiting, bone fractures, reproductive failure and possibly even infertility, damage to the central nervous system, damage to the immune system, psychological disorders, DNA damage or cancer development [2]. Cadmium not only causes chronic and acute effects to humans and animals as plant consumers but also disturbs plants themselves. A summary from many researchers and authors [3] stated that cadmium induced the inhibition of root Fe (III) reductase which led to Fe (II) deficiency and, finally, it seriously affected photosynthesis and/or caused chlorosis. Cadmium toxicity could also affect the plasmamembrane permeability which causes reduction in water content.

Considering the disturbing cadmium effects on humans, animals and plants, a solution is urgently needed to solve the problem of cadmium contamination on soil and irrigation water in rice farming, especially in high cadmium contaminated areas. The conversion of rice farming to other non-food plant farming can be an alternative solution, but in fact, farmers prefer to continue rice farming than change to one of the suggested non-food plants. The conventional approaches which aim to remove heavy metals from soil using several technologies including

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chemical precipitation, membrane filtration, reverse osmosis, ion exchange, and adsorption can be done but most of them are inefficient and expensive. On the other hand, one of the modern techniques that can be adapted to solve this problem is the aided phytostabilization technique [4, 5].

The aided phytostabilization technique emphasizes to immobilize or fix metals in soil which uses plant root absorption as the secondary role and soil amendment adsorption as the first role. This technique aims not to remove metal contaminants from a site, but rather to hold them in the soil; therefore, the risk to human health and the environment can be reduced by reducing metal mobility by rhizosphere-induced absorption, and precipitation processes [4, 5, 6]. Biomass raw materials used as amendments in this technique can be from agricultural residues which are abundant such as coir pith and corncob. Moreover, the enhanced adsorption capacity of the amend-ments was also studied focusing on biomass modification [7, 8, 9].

Investigation and development of agricultural residues to adsorb metal ions or other contaminants in aqueous solutions have been done by many researchers [10, 11, 12, 13, 14, 15] but a few studies have been conducted on the application of adsorbent on the plant-soil system. The aims of this study were to investigate whether a variety of agricultural residues as soil amendments reduced Cd uptake of rice plants in high cadmium contaminated soil.

MATERIALS AND METHODS

Preparation of agricultural residues for soil amendment

There were three kinds of agricultural residues which have been used as soil amendments involving coir pith (CP), corn corb (CC), and coir pith modification (CPm). CP and CC have been available as remediation laboratory properties where the initial sources came from companies from Prachuapkhirikan and Bangkok provinces; however, CPm was prepared in this study. CPm was made from CP modified with 0.1 NaOH as follows: 300 g of CP was mixed with 3000 mL of 0.1 M NaOH for 2 hours. After treatment with 0.1 M NaOH, the excess NaOH from treated coir pith was washed with tap water until the pH was constant (7.75) [8].

Culturing rice plants

The rice variety that was used was known as 75 days rice. This variety is usually harvested af-

ter 65-75 days in the field. Scientific information of the rice is not provided because this variety seems to be under a development project. Rice seeds were soaked with tap water for 1 day and spread on normal soil. After 40 days of seeding, rice plants were transferred to pots.

Effect of agricultural residues on plant transpiration growth under high cadmium contaminated soil

The relationship between cumulative transpiration and plant cadmium uptake on soil amendment treatments were investigated. 40 day rice plants were used as the test plants. After preparation of the media which contained 700 g of solid materials mixed with 350 mL tap water, 2 rice plants were cultured in the media. After one day of incubation, 200 mL water was added into the experimental pots.

The combination of treatments for solid media were as follows: no cadmium/free cadmium soil (N) as the control, cadmium soil (Cds), Cds with CP 1% (w/w), Cds with CPm 1% (w/w) and Cds with CC 1% (w/w). The serial pot was made into three replicates for 50 days of observation. Pot media without plants were also provided as blanks which were made into three replicates. Transpiration was measured as water was lost from the plants every three to five days by weighing the blank and treated pots. This was done by following the modified gravimetric method [16], but the values were converted into milliliter units by multiplying by the water specific density.

Determination of cadmium distribution in plants

The plants were separated into two different parts, as roots and shoots (seeds were included). Both of them were dried at 60 °C for 48 hours. Then, they were burned in an electric muffle at 450 °C for 2 hours. Plant samples were digested with 65 % of HNO_3 and 95-97 % of H_2SO_4 and then furthermore analyzed by Inductive Coupled Plasma Spectroscopy (ICP). The concentration was reported in mg kg^{-1} of the dry weight.

Determination of remaining cadmium in soil

Soil samples were dried at 60 °C for 48 hours and then ground. Samples were digested with 65 % of HNO_3 and 95-97 % of H_2SO_4 and then furthermore analyzed by ICP. The concentration was reported in mg kg-1 of the dry weight.

Statistical analysis

Analysis of variance (ANOVA) with the Tukey test and correlation test were determined using the SPSS Program for Windows.

RESULTS AND DISCUSSION

Effect of agricultural residue additions in high cadmium contaminated soil on transpiration, dry weight and cadmium (cd) uptake in rice plants

To confirm the ability of CC, CPm and CP to reduce cadmium uptake in rice plants, the effect of these agricultural residues was investigated in rice grown under high cadmium concentrations. The initial cadmium concentration in contaminated soil was in the range of 96.26 to 215 mg Cd kg^{-1} soil with an average of 145.01 mg Cd kg^{-1} soil. The addition of agricultural residues affects the transpiration of rice grown under high cadmium concentrations; therefore, the treated rice transpired water in different levels which were in the following order: CPm ≥ CP ≥ CC (Figure 1). They were similar to rice transpiration grown under low cadmium concentratiosn which were in the following order: CPm > CP > CC (data not shown).

Previous research reported that the low cadmium concentration of soil that is equivalent to a range of 0.11 to 0.56 ppm can retard root growth without toxic effects in leaves resulting in increased transpiration [17]. Transpiration of rice grown in normal soil indicated that the plant metabolism reached the optimal level because there were no stress conditions. Therefore, the transpiration value was the highest (Figure 1 and 2). Contrarily, rice grown in Cds was assumed in metal stress conditions. High transpiration values probably indicated the defense mechanism of plants from high cadmium accumulation in its organs by such kinds of continuous dilutions; therefore, plants could minimize the toxic effects of cadmium. However, the detailed mechanism to explain the increase of transpiration affected by higher cadmium accumulation is still unclear.

The cumulative transpiration of rice grown in various treatments under high cadmium contaminated soil was in the following order: Cds > CPm ≥ CP ≥ CC (Figure 2). These transpiration values affected cadmium accumulations in the shoots which were in the following order: Cds > CPm ≥ CP ≥ CC and cadmium accumulations in the husk which were in the following order: Cds = CPm > CP > CC (Table 1 and Figure 3). By considering these results, CP and CC seemed to work in reducing the cadmium uptake by rice plants.

The cadmium movement from soil to roots and from roots to shoots of rice grown in high cadmium contaminated soil seemed to be influenced by the end product compounds of agricultural residue degradation by soil microbial activity. It was assumed that the end products of degradation contained high and low molecular the presence of humic acid (HA), as an example of a high molecular weight compound, decreased the
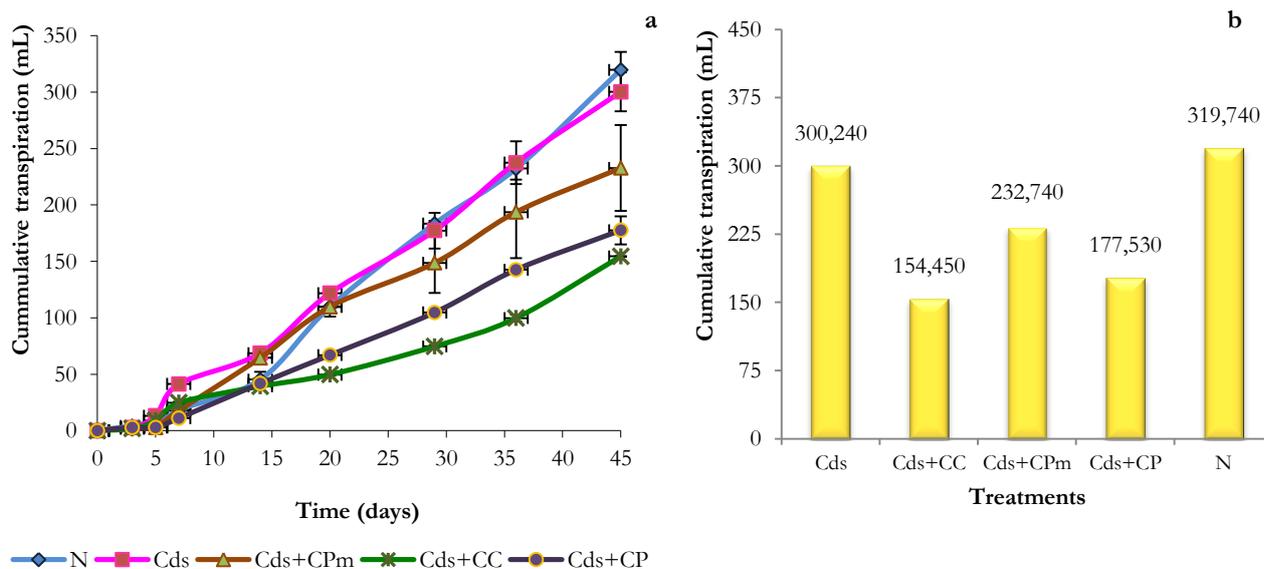

Figure 1. Cumulative transpiration of rice plants (P) grown in no cadmium/free cadmium soil (N) and high cadmium contaminated soil (Cds) in 45 days (a) and under various treatments: (CC: corncob, CPm: modified coir pith and CP: coir pith) (b). Note: Values followed by the same notation indicate that there is no significant difference (α = 0.05)

Table 1. Cumulative transpiration, dry weight and cadmium concentration of rice plants grown in high cadmium contaminated soils (Cds) under various treatments

| Treatments	Cumulative transpiration at 45th day (mL)	Dry weight of plant (g)	Rice cadmium concentration at 50th day (mgCd/kgDW)		
			Root	Shoot	Husk
Cds	300.24±17.31 c	0.991±0.233 a	594.324±40.597 b	73.878±21.10 d	13.55±1.66 c
Cds+CC	154.45±12.52 a	0.626±0.131 a	139.651±37.315 a	11.071±3.70 ab	2.47±0.82 a
Cds+CPm	232.74±38.08 b	0.747±0.112 a	919.328±226.922 bc	40.47±3.96 c	15.63±3.10 c
Cds+CP	177.53±0.18 ab	0.780±0.145 a	1188.996±246.429 c	37.16±10.11 bc	7.38±0.77 b
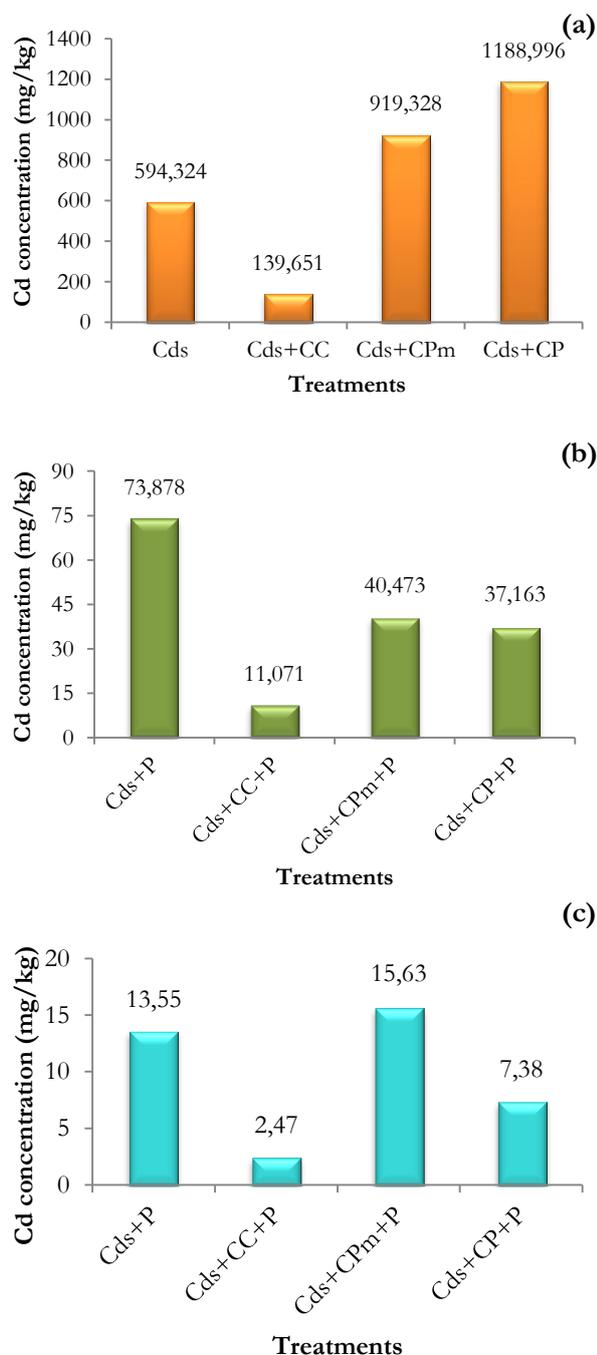
Figure 3. Cadmium concentration of the rice plants grown in high cadmium contaminated soil (Cds) under various treatments: (CC: corncob, CPm: modified coir pith and CP: coir pith). Values followed by the same notation above the bar indicate that there is no significant difference (α = 0.05). Root (a), shoot (b), and husk (c).
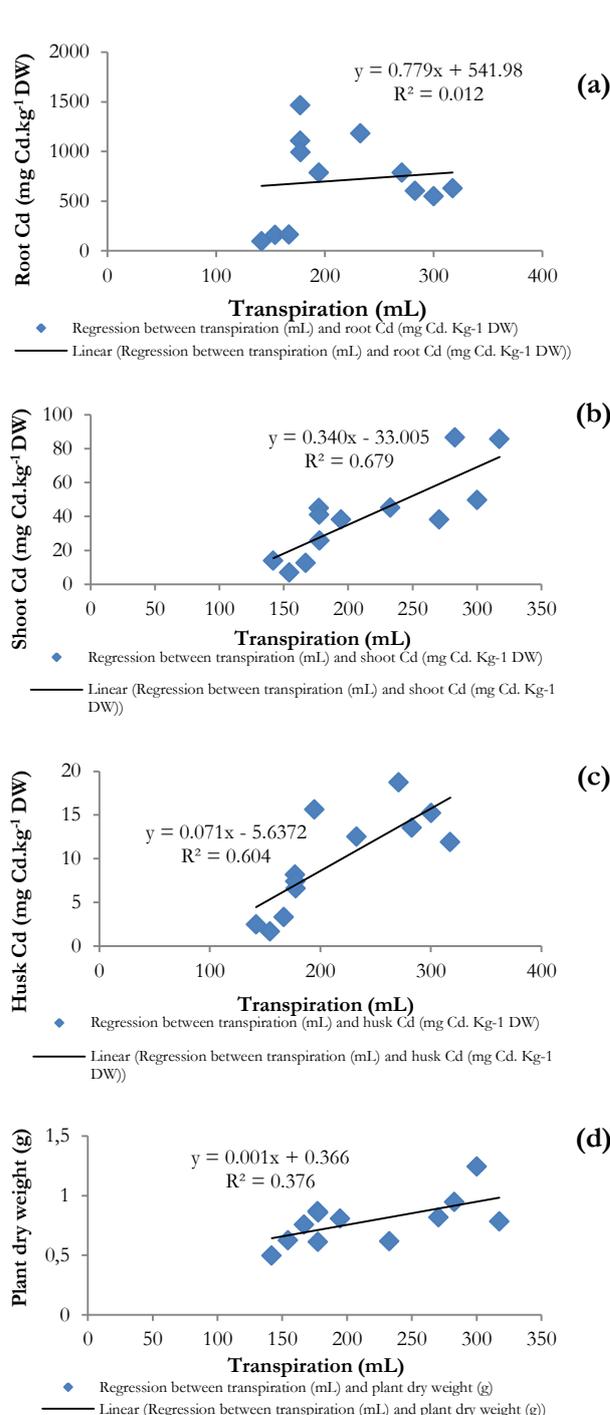

Figure 4. Regression between transpiration and plant cadmium and between transpiration and plant dry weight under high cadmium soil root Cd (a), shoot Cd (b), husk Cd (c), and plant dry weight (d)

uptake of Cd in the root on the base of total Cd concentration and free Cd ion activity. Two reasons may explain the decrease in the uptake of Cd. First, the reduced availability of cadmium may be due to complexation with HA, where insoluble precipitates are furthermore formed.

Second, the HA was adsorbed on the root surface and decreased the uptake sites for internalization of Cd, and thus decreased the uptake of Cd. Contrarily, [18] it was also argued that citric acid (CA), the small organic ligand as an example of a low molecular weight compound, increased the uptake and internalization of Cd. There are many reasons for this to be related. First, the dissociation of the Cd-citrate complex supplied free Cd ions on the root surface. Second, adsorbed citrate on the xylem cell wall had complexation with Cd; thus, the adsorbed citrate decreased Cd transferring to the cytosol. Third, the increased pH decreased the competition of H^+ with Cd^{2+} for the uptake sites on the surface root and enhanced the uptake of Cd.

The relationship between transpiration and accumulation of cadmium in the plant organs showed that transpiration mainly increased plant cadmium concentration in shoots and husks with the Pearson correlations 0.824 and 0.778, respectively (Figure 4 and Table 2).

Based on the dry weight measurement of rice grown under various treatments, it is assumed that the rice variety used in this experiment, namely 75 days rice, is a resistant variety of cadmium contamination. There was no significant difference among the dry weights of rice grown in all treatments.
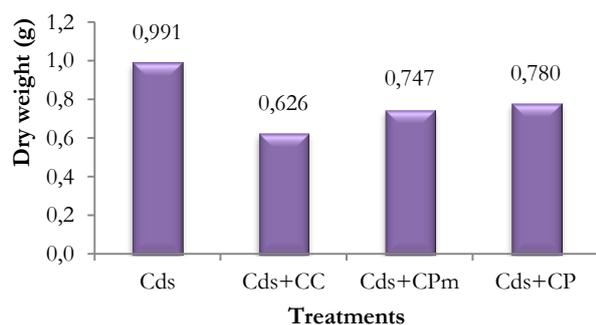

Figure 5. Total dry weight of 50th day rice plants grown in high cadmium contaminated soil (Cds) under various treatments: (CC: corncob, CPm: modified coir pith and CP: coir pith). Values followed by the same notation above the bar indicate that there is no significant difference ($\alpha = 0.05$)

Plant resistance might come from many defense mechanisms in plants. Besides producing phytochelation that restricts cadmium just in the roots and inhibiting its movement to the shoots, plants also produce exudates as a defense mechanism to prevent cadmium from entering into the plant roots. Root secretion includes organic ligands and inorganic ligands (e.g., Cl^-, SO_4^{2-}, NH_4^{+}, CO_3^{2-}, PO_4^{3-}, etc.). These substances function not only as the energy source of microorganisms, but also as ligands to be chelated with heavy metal ions and then influence the pH and Eh conditions as well as chemical characteristics in the rhizosphere [19].

Table 2. Correlation between transpiration with plant cadmium and plant dry weight of rice grown in high cadmium contaminated soil

| | Transpiration | |
	Pearson correlation	Sig.
Transpiration	1	
Husk Cd	0.778**	0.003
Shoot Cd	0.824**	0.001
Root Cd	0.111	0.732
Plant dry weight	0.613*	0.034

CONCLUSION

The results showed that the cumulative transpiration of rice grown in various treatments under high cadmium contaminated soil was in the following order: Cds > CPm ≥ CP ≥ CC. These transpiration values directly influenced cadmium accumulation in shoots and husks of rice plants. The CC and CP seemed to work to reduce the cadmium uptake by rice plants indicated by accumulated cadmium in the husk that were 2.47 and 7.38 mg Cd kg^{-1} dry weight, respectively. Overall, transpiration tended to drive cadmium accumulation in plants for rice grown in high cadmium contaminated soil. It was shown that the higher the plant uptake of cadmium, the lower remaining cadmium that was found in the soil.

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