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Journal of Hazardous Materials 154 (2008) 294–299
Journal of Hazardous Materials
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Suppressive effects of magnesium oxide materials on cadmium uptake and accumulation into rice grains
II: Suppression of cadmium uptake and accumulation into rice grains due to application of magnesium oxide materials

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Received 8 May 2007; received in revised form 8 October 2007; accepted 8 October 2007
Available online 13 October 2007

Abstract

The objective of this study is to assess the applicability of a commercial magnesium oxide (MgO) and a composite material containing MgO and natural minerals (‘MgO-SH-A’) as the soil amendments for suppression of cadmium (Cd) uptake and accumulation into rice grains. A cultivation experiment of rice plants (*Oryza sativa* L. cv. Kinuhikari) was conducted in an actual Cd-contaminated alluvial paddy field to evaluate the effectiveness of these materials. The ‘plant available’ fractions of Cd in the paddy soil significantly decreased by application of commercial MgO at 2250 kg ha^{-1} or MgO-SH-A at 4500 kg ha^{-1}. These decreases would be primarily attributed to the increase in soil pH due to applications of the MgO materials because these soil Cd fractions were significantly negatively correlated with the soil pH. Even under a suppressive condition for Cd uptake by rice plants, i.e., continuous flooding of the paddy field around the heading stage, applications of these materials further reduced Cd concentration in brown rice as compared to that from the control. It was concluded that the two MgO materials examined would be effective in preventing Cd contamination of rice grains grown in Cd-polluted paddy fields.

Keywords: Magnesium oxide; Cadmium; Rice; Paddy field; Suppression

1. Introduction

Cadmium (Cd) is one of the well-known toxic heavy metals for both plants and animals. It has been reported that Cd has a high carcinogenicity and an endocrine disrupting effect, as well as it can cause renal dysfunction and bone fracture, known as the “Itai-itai disease” as the severe case [1–5].

Rice is a staple crop for Japanese population. Consumption of food comprises most part of the entering pathway of Cd into

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doi:10.1016/j.jhazmat.2007.10.025

human body and rice was estimated to represent 36–50% of the total oral intake of Cd for Japanese population during 1998–2001 [6,7]. In 2006, 0.4 mg kg^{-1} as the maximum level of Cd in polished rice was adopted by Codex Alimentarius Commission (CAC) of FAO/WHO [8]. According to the nationwide survey on Cd contents in cereals produced from 'non-polluted' area of Japan in 1997 and 1998, 0.3% of the whole brown rice samples (n = 37,250) exceeded Cd concentration of 0.4 mg kg^{-1} [7]. On the other hand, another survey [9] informed that 22 brown rice samples out of 1909 ones, i.e., 1.2%, harvested in Japan in 2005 contained more than 0.4 mg kg^{-1} of Cd. Thus, the establishment of this new international guideline may strongly affect rice production throughout Japan.

The applicability of various materials, such as lime, phosphate fertilizers, hydroxyapatite, clay minerals, organic matter and industrial by-products has been examined to suppress uptake of Cd in contaminated soils by plants [10–12]. In particular, lime, calcium silicate and phosphate fertilizers have been widely utilized in Cd-polluted paddy fields of Japan. However, the percentage of Cd concentration in brown rice from the plot treated with these materials to that from the untreated plot varied from 39 to 205% in 1971 (n = 108) [13], indicating the instability of their effectiveness.

In the first part of this study [14], we reported the mineralogical and physicochemical properties and Cd sorption characteristics of a commercial magnesium oxide (MgO) and a composite material derived from MgO and natural minerals, named 'MgO-SH-A', and showed the possibility of their utilizations for remediation of Cd-contaminated soils due to their sufficient soil neutralizing capabilities and Cd sorption capacities. In this report, we evaluate the effectiveness of these materials as the soil amendment for reducing Cd accumulation into rice grains from the results of a rice cultivation experiment in an actual Cd-contaminated paddy field.

2. Materials and methods

2.1. Experimental design

The rice cultivation experiment was carried out in the Hommachi Farm, Field Science Center for Education and Research of Tokyo University of Agriculture and Technology in 2005. This farm is located on the Tama River alluvial lowland plain in Fuchu City, Tokyo (35°39′N and 139°28′E), and the soil of this site is classified as Aquic Fluvents [15]. The soil of this farm had been contaminated with heavy metals, including Cd, from the polluted irrigation water by industrial effluents, which had been supplied into this site until 1970 [16]. The mean total Cd concentration in the plough-layer soil collected from 11 locations in this farm in 2003 was 1.5 mg kg^{-1} DW (Kikuchi, unpublished data).

Twelve square plots (6.25 m^2 each) were made in one of the paddy fields in this farm by surrounding each plot with a wave-shaped steel plate. Each plot had one opening as the inlet/outlet of irrigation water, which was closed when the MgO materials were mixed into the soil to prevent them from washing out. They were arranged in three rows, each of which consisted of four plots. The distance between the neighboring plots was 0.5 m. Four kinds of treatments (three replicates per treatment) were designed as follows: control (no amendment), commercial MgO at 2250 kg ha^{-1} and MgO-SH-A at 2250 and 4500 kg ha^{-1}. The application rates of 2250 and 4500 kg ha^{-1} for commercial MgO and MgO-SH-A, respectively, were determined to adjust the soil pH to 7.0, the effective pH to suppress Cd uptake by rice plants [17], based on the experimental data on the soil neutralizing capacities of these materials for the soil in this site [14]. These treatments were arranged randomly in each row.

2.2. Rice cultivation practices

On 6 June, the MgO materials were mixed into the plough-layer soil (0–15 cm) of each plot at the same time of paddling. The next day, rice seedlings (*Oryza sativa* L. cv Kinuhikari) were transplanted at totally 84 points (12 points × 7 rows) in each plot by hand. Three seedlings were transplanted together into each point. Heading of the rice plants was on 12 August. Basal fertilization was done at the rate of $N:P_2O_5:K_2O$ = 30:30:30 kg ha^{-1} using a compound fertilizer on the same day of applications of the MgO materials. Topdressing of fertilizer was conducted twice (3 August and 30 August). The first topdressing was conducted at the rate of $N:P_2O_5:K_2O$ = 10:10:10 kg ha^{-1} using the same compound fertilizer as that used in the basal fertilization, and a compound fertilizer of N and K was applied for the second topdressing at the rate of $N:K_2O$ = 10:10 kg ha^{-1}. The paddy field had been kept flooded from the day of transplanting to 27 July. Irrigation had been stopped from 27 July to 1 August for mid-term drainage, and afterwards the paddy field had been continuously irrigated beyond ripening stage of the rice plants.

The pH of the plough-layer soil in one of the triplicate plots for each treatment had been monitored from 0 to 42nd day after transplanting by directly inserting a glass electrode (PT-200, Custom, Tokyo, Japan), which was connected to a portable pH meter (TES-1380, Custom), into the soil (at 7.5 cm depth).

2.3. Sampling and analysis

The soil sample of the plough-layer in each plot was collected from ten locations per plot just after application of the materials and bulked into a composite sample. These samples were air-dried, sieved through a 2 mm nylon screen and stocked for the subsequent chemical analyses. The pH(H_2O) of the soil sample was measured for its suspension (soil:water = 1:2.5) by a pH meter (M-7, Horiba, Kyoto, Japan) equipped with a glass electrode (#6066-10C, Horiba). The concentrations of Cd in the soil samples extracted by three kinds of reagents (0.1 and 0.025 M HCl and 1 M NH_4Cl) were determined using an atomic absorption spectrophotometer (AAS) (Z-5010, Hitachi, Tokyo, Japan) with air–acetylene flame at 228.8 nm. The extraction of soil Cd by 0.1 M HCl is the official method adopted in Agricultural Land Soil Pollution Prevention Law in Japan, and 67% (as a mean) of the total Cd in Japanese soils can be extracted by this reagent [18]. The concentrations of Cd in soil extacted by 0.025 M HCl and 1 M NH_4Cl were reported to be signifi-

Table 1
The mean pH(H_2O) and extractable Cd concentrations of the plough-layer soil samples from each treatment plots ($n = 3$ for each treatment)

Treatment	pH(H_2O)	Cd concentration in soil (mg kg^{-1} DW[a])		
		0.1 M HCl	0.025 M HCl	1 M NH_4Cl
Control	6.1 a	1.33 a	0.32 b	0.68 b
MgO 2250 kg ha^{-1}	6.9 b	1.24 a	0.05 a	0.25 a
MgO-SH-A 2250 kg ha^{-1}	6.4 ab	1.30 a	0.20 ab	0.50 ab
MgO-SH-A 4500 kg ha^{-1}	6.5 ab	1.23 a	0.12 a	0.40 a

Different letters (a and b) in each column indicate significant differences between treatments ($p < 0.01$). MgO: Commercial MgO.
[a] DW: Dry weight.

cantly correlated with the Cd contents in grains of wheat and durum wheat, respectively [19,20]. For 0.1 M HCl- or 0.025 M HCl-extractable Cd analysis, 5 g of soil sample was weighed into a 100 mL polypropylene vial and mixed with 25 mL of the HCl solution. The vial was shaken on a reciprocal shaker (SR-II, Taitec, Saitama, Japan) for 1 h at 25 °C. The soil suspension was filtered with a filter paper (No. 5C, Advantec, Tokyo, Japan) to obtain the extract for Cd determination by the AAS [18,19]. For 1 M NH_4Cl-extractable Cd analysis, 5 g of soil sample was taken in a 100 mL polypropylene vial and mixed with 30 mL of 1 M NH_4Cl solution. The soil extract was obtained by the same procedure described above, except for the shaking time of 16 h [20].

Twenty hills (5 hills × 4 rows) of rice plants for Cd analysis were harvested from the center of each plot on 3 October. The area in which the sampled rice plants had been planted was measured simultaneously to calculate the grain yield per unit area. The brown rice and chaff were separated using a rubbing machine, and the brown rice samples obtained from the 20 hills of rice plants were bulked, sieved through a 1.8 mm stainless screen and then weighed. The moisture contents of the brown rice samples were measured by a moisture meter (Riceter-L, Kett, Tokyo, Japan). The brown rice samples were successively washed with tap water and distilled water, dried at 70 °C for 24 h and ground with a food mill for subsequent Cd analysis. The ground samples were digested using concentrated HNO_3 and $HClO_4$, and the Cd concentrations in the digests were determined by a graphite furnace AAS (GFAAS) with a Zeeman-effect background correction system (Z-5010, Hitachi). The mixture of 2% (w/v) $NH_4H_2PO_4$ and 0.4% (w/v) $Mg(NO_3)_2$ dissolved in 0.3 M HNO_3 was used as the matrix modifier in the GFAAS analysis [21].

All reagents used were of special grade (Wako, Osaka, Japan), except for HCl and HNO_3 for the stock solution which were for analysis of poisonous metal (Wako). Distilled-deionized water was also used for preparation of reagents and sample solution.

2.4. Statistical analysis

The significant differences in the extractable Cd concentrations in soil and the Cd concentration in brown rice between treatments were detected by one-way ANOVA followed by Fisher's LSD. The test on correlation coefficients was performed according to the procedure by Snedecor and Cochran [22].

3. Results and discussion

3.1. Effect of the magnesium oxide materials on the pH of the cultivated soil

The mean pH(H_2O) values of the soil samples collected from each treatment plots ($n = 3$) are shown in Table 1, and the time course of the pH of the plough-layer soil in each treatment at the early stage of rice growth is described in Fig. 1. The soil pH(H_2O) value of the treatment with commercial MgO at 2250 kg ha^{-1} was significantly higher ($p < 0.01$) than that of the control, although the values of all the treatments with the MgO materials, ranging from 6.4 to 6.9, were not statistically different from each other. The *in situ* monitoring data of the pH of the plough-layer soil exhibited somewhat different tendencies from the pH(H_2O) values of the soil samples. The soil pH of all the treatments with the MgO materials had been kept to 7.0 or above from the beginning of rice cultivation, as compared to that of the control which had been lower than 7.0 throughout the monitoring period. The soil pH of the treatment with MgO-SH-A at 2250 kg ha^{-1}, which was designed as the half dose of the application rate to adjust the soil pH to 7.0 [14], had been kept to almost 7.0. The soil pH values of the treatments with commercial MgO at 2250 kg ha^{-1} and MgO-SH-A at 4500 kg ha^{-1} were consistent with each other. However, the soil pH values of all the treatments tended to converge at 7.0. This was considered to be due to the decrease in redox potential (Eh) of the soil under continuous flooding of the paddy field or to the neutralization of alkalinity by generated CO_2 gas in the soil [23]. The Eh value of the plough-layer soil in the paddy field measured on 21 June (2 weeks after transplanting) was −141 mV as the value of direct reading, indicating reductive condition in the soil.

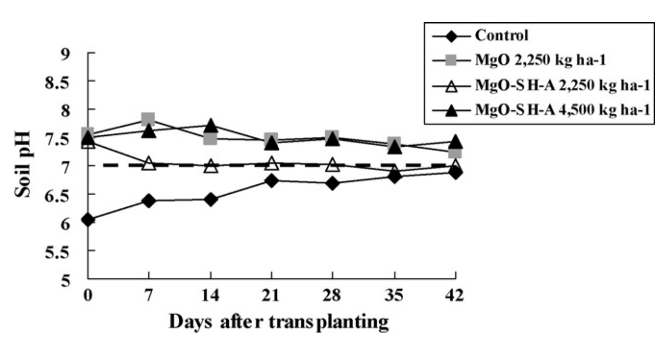

Fig. 1. The time course of the pH of the plough-layer soil at the early stage of rice growth under flooded condition. MgO: Commercial MgO.

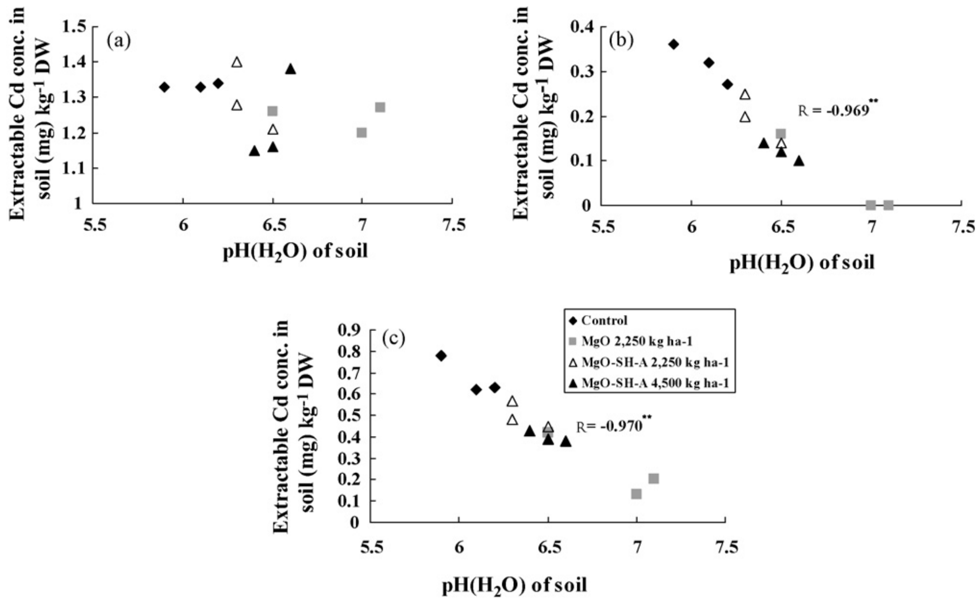

Fig. 2. The relationships between pH(H_2O) of the soil and Cd concentrations in the soil extracted by 0.1 M HCl (a), 0.025 M HCl (b) and 1 M NH_4Cl (c). Significant at $^{**}p < 0.01$. MgO: Commercial MgO.

3.2. *Effects of the magnesium oxide materials on the extractable Cd concentrations in soil*

The mean Cd concentrations in the soil samples extracted by each of three kinds of extractants (0.1 and 0.025 M HCl and 1 M NH_4Cl) for each treatment, and the relationships between the extractable Cd concentrations and pH(H_2O) of the soil samples are given in Table 1 and Fig. 2, respectively. The extractable Cd concentrations in the soil, except for 0.1 M HCl-extractable Cd, significantly decreased ($p < 0.01$) due to application of commercial MgO at 2250 kg ha^{-1} or MgO-SH-A at 4500 kg ha^{-1} as compared to those of the no amendment control. Furthermore, 0.025 M HCl- and 1 M NH_4Cl-extractable Cd concentrations, referred to as the 'plant available' fractions of Cd in soil, were significantly negatively correlated with the soil pH ($p < 0.01$) irrespective of the treatment. From the results mentioned above, the decreases in the 'plant available' fractions of Cd in the soils of the MgO materials-amended plots were primarily attributed to the increase in soil pH due to applications of these materials. In general, the amount of Cd^{2+} adsorbed on the surface of soil colloids increases as the soil pH increases, due to the increase in the amount of negative charge on the surface [24,25]. In addition, the precipitation of $Cd(OH)_2$ on the surface of MgO particles as described in the first part of this study [14] can be considered as another mechanism of the reduction of plant availability of Cd in the materials-amended soils. The pH of the region near the surface of MgO particles is suggested to be higher than that of the bulk solution because the dissociation rate of $Mg(OH)_2$ formed on the surface of MgO is low, which would be appropriate for the formation of $Cd(OH)_2$ precipitates on the material's surface [14].

3.3. *Effect of the magnesium oxide materials on the yield and Cd concentration in brown rice*

The yield and Cd concentration of brown rice cultivated in each treatment are summarized in Table 2. The yield of brown rice in the treatment with MgO-SH-A at 4500 kg ha^{-1} was significantly higher than those in the other treatments ($p < 0.01$). Application of commercial MgO at 2250 kg ha^{-1} or MgO-SH-A at 4500 kg ha^{-1} also enhanced the growth of rice plants at the early growth stage (data not shown). The observed improvement

Table 2
The yield and Cd concentration of brown rice grown in each treatment

	Yield (t ha^{-1})[a]		Cd concentration (mg kg^{-1} DW[b])	
	Mean	S.E.[c]	Mean	S.D.[d]
Control	4.160 a	0.100	0.036 b	0.017
MgO 2250 kg ha^{-1}	4.850 a	0.125	0.020 a	0.007
MgO-SH-A 2250 kg ha^{-1}	4.865 a	0.290	0.016 a	0.004
MgO-SH-A 4500 kg ha^{-1}	5.390 b	0.190	0.018 a	0.010

Different letters (a and b) in each column indicate significant difference between treatments ($p < 0.01$). MgO: Commercial MgO.
[a] Grain weight at 14% of moisture content.
[b] DW: Dry weight.
[c] Standard error.
[d] Standard deviation.

of rice growth due to applications of the MgO materials would be attributed to the acceleration of organic nitrogen mineralization due to increase in soil pH [26], although Mg solubilized from the materials, which is an essential nutrient element to form porphyrin ring of chlorophyll, might be also partly responsible. The Cd concentrations in brown rice from the treatments were all less than 10% of the maximum level (0.4 mg kg^{-1}) for polished rice adopted by CAC [8]. Nevertheless, the Cd concentrations in brown rice from the treatments with the MgO materials were significantly lower than that from the control ($p < 0.01$), which confirmed the effectiveness of these materials in suppression of Cd accumulation into rice grain.

It is well known that the reductive condition in paddy soils, which usually develops under flooded condition, markedly suppresses uptake of Cd in the soils by rice plants probably due to precipitation of the plant-available Cd in soil as insoluble cadmium sulfide (CdS). Cadmium sulfide, however, will resolubilize and become available for plant uptake under the oxidative condition, which usually occurs after drainage of surface water from paddy fields [27–30]. As mentioned in Section 2.2, the experimental paddy field had been flooded throughout the heading stage of rice plants, when uptake and translocation of Cd into the above-ground part of rice became the most active [31,32]. The continuous flooding of the paddy soil in this period was considered to be the major cause for the overall suppression of Cd accumulation into brown rice in the present study. The decrease in the 'plant available' fraction of Cd in soil due to applications of the MgO materials might be responsible for the further reduction of Cd accumulations into rice grains produced from the treatments with these materials. However, the Cd concentration in brown rice was not significantly correlated with the extractable Cd concentration in soil by 0.025 M HCl or 1 M NH_4Cl (data not shown). The effect of applications of the MgO materials in reduction of the 'plant-available' Cd in soil due to increase in soil pH might be weakened by continuous flooding of the paddy soils which would make the soil pH converge at neutral, as described in Section 3.1. The formation of hardly soluble $Cd(OH)_2$ precipitates on the surface of MgO particles as mentioned in the previous section could then be considered as one of the mechanisms involving in the suppression of Cd uptake by rice plants grown in the soils amended with the MgO materials.

The percentages of the mean Cd concentration in brown rice from the treatments with the MgO materials to that from the no amendment control were 56, 45 and 50% for commercial MgO at 2250 kg ha^{-1} and MgO-SH-A at 2250 and 4500 kg ha^{-1}, respectively. Hoshino [33] reported that these percentages for fused phosphate, calcium silicate and autoclaved lightweight concrete were 44–66, 38–83 and 45–71%, respectively, in the rice cultivation experiments conducted under continuous flooding around the heading stage. The repeated annual applications of alkaline silicate fertilizers did not significantly reduced the Cd content in brown rice when the paddy field was continuously flooded during the latter growth stage of rice, including heading stage [26]. The suppressive effect of the MgO materials on Cd accumulation into brown rice would be comparable or even superior to that of the existing soil conditioners.

4. Conclusions

A rice cultivation experiment was conducted to evaluate the applicability of the MgO materials (commercial MgO and MgO-SH-A) as the soil amendment for suppression of Cd uptake and accumulation into rice grain in an actual Cd-contaminated paddy field, and the following results were obtained:

1. The 'plant available' fraction of Cd in the soil which was extracted by 0.025 M HCl or 1 M NH_4Cl significantly decreased by application of commercial MgO at 2250 kg ha^{-1} or MgO-SH-A at 4500 kg ha^{-1}. These decreases were primarily attributed to the increases in soil pH due to applications of the materials.
2. When rice plants were cultivated under the suppressive condition for Cd uptake by rice plants, i.e., continuous flooding of the paddy field around the heading stage, applications of the MgO materials further reduced Cd concentration in brown rice.

In conclusion, the MgO materials can be utilized as the soil amendments for suppression of transfer of Cd into the edible part of rice plants from the contaminated soils, without any adverse effects on the yield of brown rice. However, the appropriate application rates of these materials should be carefully determined prior to their actual uses by examining their soil neutralizing capacities, in order to avoid precipitation of other effective nutrient elements for rice plants in the soil under unnecessarily high pH [14]. In addition, these materials should be utilized in combination with the adequate management practice of the surface water in the paddy field to secure the effective and stable reduction of Cd uptake by rice plants.

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