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Suppressive effect of magnesium oxide materials on cadmium accumulation in winter wheat grain cultivated in a cadmium-contaminated paddy field under annual rice–wheat rotational cultivation

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ABSTRACT

The effectiveness of two kinds of magnesium oxide (MgO) materials, commercial MgO (2250 kg ha^{-1}) and a material derived from MgO and magnesium silicate minerals named 'MgO-SH-A' (2250 and 4500 kg ha^{-1}1), in suppression of uptake and accumulation of cadmium (Cd) into grain of winter wheat (*Triticum aestivum* L. cv. Ayahikari) was examined in a Cd-contaminated alluvial paddy field under annual rice–wheat rotational system. The MgO materials were mixed into the plough-layer soil only once prior to the preceding rice cultivation. Cadmium concentration in wheat grain produced from the non-amendment control exceeded the maximum limit of Cd in wheat grain adopted by FAO/WHO (0.2 mg kg^{-1}). All of the treatments with the MgO materials significantly lowered plant available Cd fraction in the plough-layer soil. However, only the treatment with the commercial MgO at 2250 kg ha^{-1} produced wheat grain whose Cd concentration was not only significantly lower than that from the control but also less than 0.2 mg kg^{-1}. It is suggested that the significant suppressive effect of the commercial MgO on Cd accumulation in wheat grain would be mainly attributed to its high soil neutralizing capacity as compared to that of MgO-SH-A.

1. Introduction

Since 1972, the worldwide standards of cadmium (Cd) contents in various foodstuff has been discussed in Codex Alimentarius Commission (CAC) of FAO/WHO, in which Japan joined in 1966 [1]. In July 2006, 0.4 mg kg^{-1} ADW (air-dry weight) as the maximum limit of Cd in polished rice was adopted by CAC, whereas the value for wheat grain had been already adopted to be 0.2 mg kg^{-1} ADW [2,3]. According to the nationwide survey on Cd contents in domestic brown rice and wheat grain samples harvested in 1997 and 1998 ($n = 37{,}250$ and 382 for brown rice and wheat grain, respectively), 0.3% of the brown rice and 3.1% of the wheat grain exceeded the maximum limits by CAC [4]. Thus, the establishment of the new worldwide guideline for Cd contents may strongly affect wheat production in Japan, as well as that of rice.

For paddy rice, continuous flooding in paddy fields at the latter growth stage of rice plants, which includes the heading stage, has been found to significantly reduce Cd content in brown rice [5,6], and this way of water management is recommended as one of the most effective countermeasures against accumulation of Cd in rice grain [7]. In addition, the effectiveness of applications of soil amendments, such as lime, phosphatic fertilizers and calcium silicates, on suppression of Cd accumulation into rice grain has been confirmed, although it is usually unstable as compared to the effect of the water management practice [5,8–10].

On the other hand, wheat is cultivated under upland (non-flooded) condition, and such the strategy for suppression of Cd uptake as the case of paddy rice cannot be applied for wheat. It has often been documented that Cd uptake by wheat and durum wheat tend to increase with increasing soil acidity or salinity [11–14]. It was also reported that Cd content in wheat grain could be predicted reasonably well from soil total Cd and pH [15]. However, the universal and effective countermeasures to reduce Cd uptake into wheat grain have not been established.

The acreage of paddy rice in Japan was 1,669,000 ha in 2007, and it has been in a decreasing trend since 1970 [16]. On the other hand, the acreage of grain wheat, 209,700 ha in 2007, had increased continuously in the period from 1998 to 2006. In addition, 54.4% of

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the grain wheat (94.2% in case that Hokkaido Prefecture is excluded) was cultivated in paddy fields in 2007 [17]. Thus, the development of the strategies for suppressing Cd accumulation in grain of winter wheat cultivated in paddy fields is required.

Application of liming materials to soil can be considered as one of the options to reduce uptake of Cd in soil by arable crops. However, this practice would also foment the imbalance between exchangeable calcium and magnesium in soil, which has been observed for the arable soils of Japan [18]. In the previous study [19], we examined the effectiveness of commercial magnesium oxide (MgO) and a material derived from MgO and magnesium silicate minerals, named 'MgO-SH-A', in suppression of Cd accumulation into grain of paddy rice grown in a Cd-contaminated alluvial paddy field, and showed that the applications of these two materials prior to transplanting of rice plants significantly lowered Cd concentration in brown rice when the paddy field had kept flooded at the latter growth stage of rice. Winter wheat was successively cultivated in the same experimental plots without additional applications of these materials. The suppressive effect of these materials applied only once at the beginning of annual rice–wheat rotational cultivation on Cd accumulation in the wheat grain will be discussed in this report on the basis of the maximum limit of Cd in wheat grain by CAC, i.e., 0.2 mg kg^{-1}. In addition, the comparison between winter wheat and paddy rice in the effectiveness of these materials on suppression of Cd accumulation into grain will be made.

2. Materials and methods

2.1. Site description and experimental design

The successive rice and winter wheat cultivation experiment had been performed in a paddy field of the Hommachi Farm, Field Science Center for Education and Research of Tokyo University of Agriculture and Technology in Fuchu City, Tokyo (35°39′N, 139°28′E) from June 2005 to May 2006. This farm is located on the Tama River alluvial lowland plain and the soil of this site was classified into Aquic Fluvents [20]. The texture of the plough-layer soil in the experimental paddy field was silt loam. The physico-chemical properties and nitrogen fertility of the paddy soil in this farm were reported by Sakagami et al. [21]. Spatial distributions of soil nutrients in this farm were also investigated by Tanaka et al. [22]. Detailed information on geography and the history of heavy metal pollution of this farm was described by Okazaki and Saito [23].

Four kinds of treatment were designed as follows: commercial MgO at 2250 kg ha^{-1} (c-MgO), MgO-SH-A at 2250 kg ha^{-1} (MgO-SH-A I) and 4500 kg ha^{-1} (MgO-SH-A II) and no amendment control (Control) (Table 1). Triplicate plots (2.5 m × 2.5 m each) were prepared for each treatment. The commercial MgO used was of industrial-use grade which was a product of Nihon Kaisui Kakou, Co. Ltd., Niigata, Japan and MgO-SH-A was developed and produced by Ams Engineering, Co. Ltd., Miyagi, Japan. The physico-chemical properties and Cd sorption characteristics of these materials were summarized by Okazaki et al. [24]. The application rates of these materials were determined to adjust the soil pH to 7.0 or lower in order to prevent the micronutrient metals in soil from precipitating under alkaline condition. These materials were mixed into the plough-layer soil (0–15 cm in depth) prior to the rice cultivation and not added in the subsequent winter wheat cultivation period.

Table 1
Design of the treatments for application of the MgO materials.

Treatment	Material applied	Application rate (kg ha^{-1})
Control	None	–
c-MgO	Commercial MgO	2250
MgO-SH-A I	MgO-SH-A	2250
MgO-SH-A II	MgO-SH-A	4500

2.2. Annual rotational cultivation of rice and wheat

In the rice cultivation, rice seedlings (*Oryza sativa* L. cv. Kinuhikari) were transplanted on June 7, 2005. Heading of the rice plants was on 12 August, and the rice plant samples for Cd analysis were finally harvested on October 3. The experimental paddy field had been kept flooded from transplanting to ripening stage of the rice plants, except for the period of mid-term drainage (27 July–1 August). Details of the rice cultivation are summarized in the previous paper [19].

In the winter wheat cultivation, the plough-layer soil of each plot in the experimental paddy field was ploughed on November 9. On November 10, 11 and 13, a compound fertilizer was applied into seven rows in each plot at the rate of $N:P_2O_5:K_2O = 25:25:25$ kg ha^{-1}, and successively seeds of wheat (*Triticum aestivum* L. cv. Ayahikari) were sown into the same rows. Four or five seeds were sown together into one point. Topdressing was not conducted. About 20 hills of the wheat as a whole plant with the soil around the roots were picked up from the center of each plot on May 29, 2006 and had been kept in a greenhouse to get the wheat plant at full-maturity because the next rice cultivation had to be started in the experimental paddy field.

2.3. Sampling and analysis of soil and crops

The soil samples of the plough layer in each plot were collected randomly from 10 locations in the plot on June 6, 2005 (just after applications of the MgO materials prior to the rice cultivation) and February 28, 2006 (in the wheat cultivation period), and bulked into composite samples representative of the plot. (The soil around wheat root was also sampled when the wheat grain was harvested, but it was not analyzed in this study.) The composite samples were air-dried, lightly crushed with mortar and pestle and sieved through a 2-mm nylon screen. The pH(H_2O) of the soil sample was measured in a suspension (soil:water = 1:2.5) using a pH meter (M-7, Horiba, Kyoto, Japan) equipped with a glass electrode (#6066-10C, Horiba). The total Cd content in the plough-layer soil of the experimental paddy field, referred to as 'Cd_T', was analyzed for the soil sample collected from the same paddy field in 2004. The soil sample was digested with concentrated HNO_3 and $HClO_4$ under heat, and total Cd content in soil was determined for the soil digest using a graphite furnace atomic absorption spectrophotometer (GFAAS) (Z-5010, Hitachi, Tokyo, Japan) at 228.8 nm wavelength. Concentrations of Cd in the soil sample extracted with 0.1 mol L^{-1} HCl, named 'Cd_{AE}' (AE: Acid-Extractable), or 0.025 mol L^{-1} HCl, 'Cd_{PA}' (PA: Plant-Available), were also analyzed. The Cd_{PA} in soil was reported to be significantly correlated with Cd contents in grains of winter wheat [25]. The extraction procedures were described in the previous report [19]. Cadmium concentrations in the soil extracts were determined using an atomic absorption spectrophotometer (Z-5010, Hitachi) with air-acetylene flame.

The above-ground parts of rice plants were sampled from 20 hills of rice grown in the center of each plot. The brown rice samples were successively washed with tap water and distilled water, dried at 70 °C for 24 h and ground using a food mill. One gram of the ground sample was taken into a 200-mL conical beaker and digested with 10 mL of HNO_3 (60–61% (v/v)) and 5 mL of $HClO_4$ (60–62% (w/w)) under heat until remaining acid in the beaker was completely evaporated. The residue in the beaker was redissolved in 15 mL of 2 mol L^{-1} HNO_3 and decanted through filter paper (No. 5C, Advantec, Tokyo, Japan). The filtrate was transferred into a 100-mL volumetric flask and adjusted to the final volume with distilled-deionized water. Cadmium concentration in the sample solution was determined using a GFAAS (Z-5010, Hitachi). The mixture of 2% (w/v) $NH_4H_2PO_4$ and 0.4% (w/v) $Mg(NO_3)_2$ in 0.3 mol L^{-1} HNO_3 was used as the matrix modifier in the GFAAS analysis [26].

The wheat seeds at the maturing stage were separated from rachis branch using a steel thresher, and the grain and chaff were manually separated. The wheat grain samples obtained were washed in the same manner with the brown rice samples, dried at 60 °C for 36 h and ground with a mixer mill (MM301, Retsch, Haan, Germany) for subsequent Cd analysis. The powdered samples were digested and analyzed for Cd contents following the same ways as those of brown rice, except for the matrix modifier used in GFAAS analysis (the mixture of 0.5% (w/v) $NH_4H_2PO_4$ and 0.03% (w/v) $Mg(NO_3)_2$ in 0.3 mol L^{-1} HNO_3 [27]).

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

3. Results

3.1. Effect of magnesium oxide materials on pH and availability of cadmium for plant uptake in plough-layer soil

The mean pH(H_2O) of the soil collected in the wheat cultivation period ranged from 6.1 to 6.5 among treatments and decreased in the following order: c-MgO ≥ MgO-SH-A II ≥ MgO-SH-A I ≥ Control. The pH values of c-MgO, MgO-SH-A I and MgO-SH-A II lowered by 0.4, 0.2 and 0.2, respectively, as compared to those for the soils collected just after application of these materials, whereas the pH of Control remained unchanged. The percentage of Cd_{PA} and Cd_{AE} to Cd_T for each treatment is illustrated in Fig. 1. Cd_T was determined to be 1.39 mg kg^{-1} DW (DW: dry weight), and the Cd fractions in soil of Cd_{PA}, Cd_{AE} minus Cd_{PA} and Cd_T minus Cd_{AE} are referred to as 'Weak-acid soluble', 'Strong-acid soluble' and 'Residual', respectively, in Fig. 1. Cd_{PA} decreased in the following order: Control > MgO-SH-A I ≒ MgO-SH-A II > c-MgO. The percentage of Cd_{PA} to Cd_T ranged from 10 to 26%. A significant negative correlation was found between Cd_{PA} and pH of the soil ($r = -0.901$, $p < 0.01$) (Fig. 2), which was also observed for the soil collected just after application of the MgO materials. There were no significant differences in Cd_{AE} between treatments, which was consistent with the case of the soil collected just after application of the MgO materials.

3.2. Effect of magnesium oxide materials on cadmium concentration in winter wheat grain

The mean Cd concentration in wheat grain of each treatment is shown in Fig. 3. Cadmium content in wheat grain grown in Control exceeded the maximum limit by CAC, i.e., 0.2 mg kg^{-1}, which implied that some countermeasures to suppress Cd uptake by winter wheat were necessary to be introduced in the wheat cultivation in this paddy field. Cadmium concentration in wheat grain decreased in the following order: Control ≥ MgO-SH-A I ≒ MgO-SH-A II ≥ c-MgO. Among the treatments with the MgO materials, only c-MgO produced wheat grain whose mean Cd concentration was significantly lower than that from Control ($p < 0.01$) and less than 0.2 mg kg^{-1}. There also existed a significant positive correlation between Cd concentration in wheat grain and Cd_{PA} ($r = 0.829$, $p < 0.01$) (Fig. 4), which was consistent with the observation by Ibaraki et al. [25].

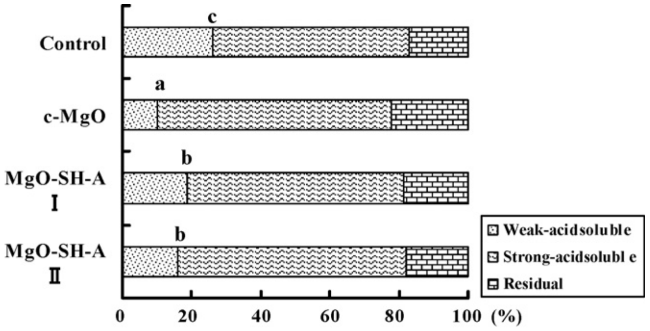

Fig. 1. Fractionation of Cd in the plough-layer soil of each treatment. "Weak-acid soluble", "Strong-acid soluble" and "Residual" represent the Cd fraction extracted with 0.025 mol L^{-1} HCl (Cd_{PA}), the Cd fraction extracted with 0.1 mol L^{-1} HCl (Cd_{AE}) minus Cd_{PA}, and the total Cd (Cd_T) minus Cd_{AE}, respectively. Different letters indicate significant differences between treatments in Cd_{PA} (Fisher's LSD, $p < 0.01$).

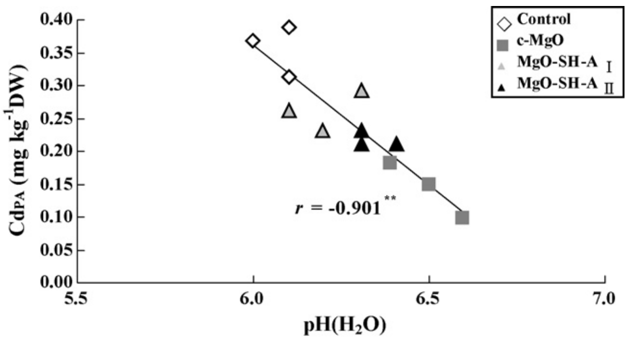

Fig. 2. Relationship between pH(H_2O) and 0.025 mol L^{-1} HCl-extractable Cd concentration (Cd_{PA}) in the plough-layer soil collected in the wheat cultivation period. **Significant at $p < 0.01$.

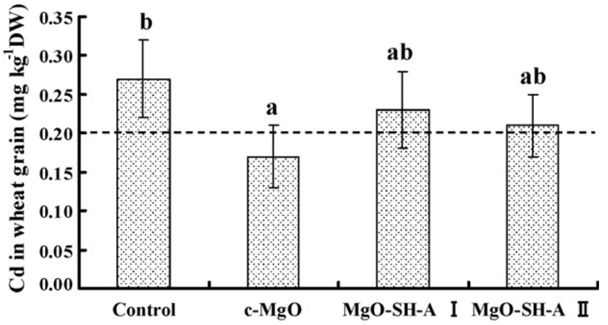

Fig. 3. Cadmium concentration in wheat grain from each treatment. Dotted line represents the maximum limit of Cd in wheat grain by CAC (0.2 mg kg^{-1}). An error bar represents standard deviation ($n = 3$). Different letters on error bars indicate significant differences between treatments (Fisher's LSD, $p < 0.01$).

3.3. Difference between cadmium concentrations in grains of winter wheat and paddy rice affected by magnesium oxide materials

The relationship between Cd concentrations in brown rice and wheat grain harvested from the same plot in the present study

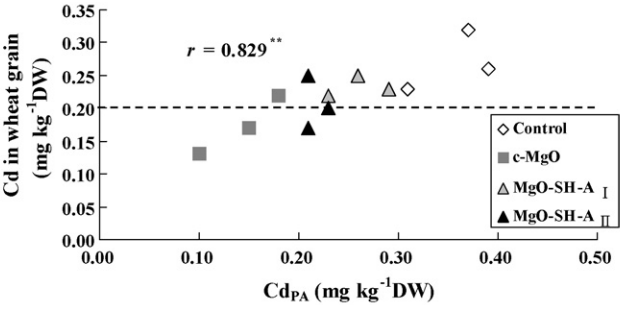

Fig. 4. Relationship between 0.025 mol L^{-1} HCl-extractable Cd concentration (Cd_{PA}) in the soil sample collected in the wheat cultivation period and Cd concentration in wheat grain. Dotted line represents the maximum limit of Cd in wheat grain by CAC. **Significant at $p < 0.01$.

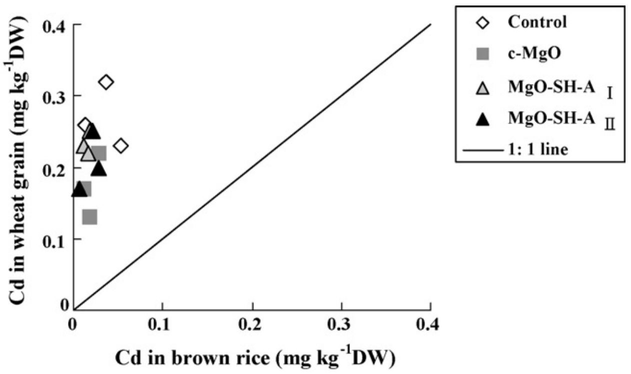

Fig. 5. The relationship between Cd concentrations in brown rice and wheat grain harvested in the same plot under annual rice–wheat rotational cultivation.

is illustrated in Fig. 5. The differences between the mean Cd concentrations in grain from treatments with the MgO materials and that from Control (absolute values) were larger for wheat grain than for brown rice by 2–6.5 times. In addition, the "suppression rate of Cd accumulation (CdASR)" was defined and calculated as follows:

$$\text{CdASR (\%)} = \frac{\text{Cd in grain from Control} - \text{Cd in grain from treatment with MgO material}}{\text{Cd in grain from Control}} \times 100 \qquad (1)$$

CdASE for wheat grain were 37, 15 and 22% for c-MgO, MgO-SH-A I and MgO-SH-A II, respectively, and these values were smaller than those for brown rice (44, 56 and 50%).

4. Discussion

Applications of the MgO materials increased soil pH because MgO in the materials would hydrate to produce alkaline $Mg(OH)_2$. The X-ray fluorescence analysis revealed that Mg contents (as MgO) in the commercial MgO and MgO-SH-A were 90.17 and 61.41% (w/w), respectively [24]. Using these values, the amounts of MgO incorporated into the plough-layer soil due to applications of the commercial MgO at 2250 kg ha^{-1} (c-MgO) and MgO-SH-A at 2250 kg ha^{-1} (MgO-SH-A I) and 4500 kg ha^{-1} (MgO-SH-A II) are calculated to be 2030, 1380 and 2760 kg ha^{-1}, respectively. However, MgO-SH-A contains not only pure MgO but also Mg in minerals, one of which was identified to be antigorite ($Mg_3Si_2O_5(OH)_4$) through X-ray diffraction analysis [24]. Thus, it is suggested that the amount of reactive MgO, which can readily produce $Mg(OH)_2$, entered into the soil associated with application of the commercial MgO at 2250 kg ha^{-1} is still larger than that supplied from MgO-SH-A at 4500 kg ha^{-1}. In order to compare the abilities of the MgO materials to suppress Cd accumulation into grain on the basis of MgO applied, the "suppression efficiency of Cd accumulation per unit MgO applied ($\text{CdASE}_{\text{MgO}}$)" was defined and calculated as follows:

$$\text{CdASE}_{\text{MgO}}\ (\text{mg ha kg}^{-2}) = \frac{\text{Cd in grain from Control} - \text{Cd in grain from treatment with MgO material}}{\text{MgO incorporated into soil due to application of MgO material}} \qquad (2)$$

$\text{CdASE}_{\text{MgO}}$ for wheat grain were 4.9×10^{-5}, 2.9×10^{-5} and 2.2×10^{-5} mg ha kg^{-2} for c-MgO, MgO-SH-A I and MgO-SH-A II, respectively. This result revealed that the commercial MgO was the most efficient amendment for suppressing Cd accumulation into wheat grain among the MgO materials tested. It was also suggested that simply increasing the application rate of MgO-SH-A is not necessarily effective in enhancing its suppressive effect on Cd uptake into wheat grain.

In the experimental paddy field, it is considered from Figs. 2 and 4 that the decrease in Cd concentration in wheat grain would be related to the increase in the pH of the cultivated soil, which would decrease the fraction of Cd in the soil available for uptake by wheat, as shown in Fig. 1. This result implies that production of wheat grain containing less than 0.2 mg kg^{-1} of Cd would be possible by supplementation of the MgO materials into the soil in the wheat cultivation period to raise the soil pH to the adequate range. In addition, Ono and Wada [28] suggested from the result of their laboratory experiments that a part of copper or nickel ion in the mixture of MgO, active silica (SiO_2) and the corresponding metal nitrate solution was incorporated in the octahedral sites of the newly formed layer Mg-silicate minerals, and that the amount of the metal ions incorporated into the octahedral sites would increase as the crystallinity of the newly formed Mg-silicate increase by aging. Formation of such Mg-silicate minerals is highly probable in soil amended with MgO because SiO_2 and siliceous materials are quite common in soil [28]. It is thus speculated that a portion of Cd in the soil with the MgO materials could be also gradually incorporated into the newly formed Mg-silicate minerals, and the fixed Cd in the minerals would be less susceptible for the change in the chemical condition of the soil, e.g., acidification, than the Cd adsorbed on soil minerals or precipitated as carbonate or hydroxide.

The suppression rates of Cd accumulation into grain due to applications of the MgO materials, defined as "CdASR" in Section 3.3, were lower for winter wheat than for paddy rice. However, the effect of the MgO materials on reduction of Cd concentration in wheat grain to less than the maximum limit by CAC should be regarded to be more important than that for rice grain because the production of brown rice whose Cd content is less than the maximum limit was possible without using the MgO materials by the proper water management practice in the paddy field (Fig. 5).

The use of Mg-containing materials as soil conditioners would be beneficial in improving the imbalance between exchangeable calcium and magnesium in soil, which has been recognized in the agricultural soils of Japan [18]. Tanaka et al. [22] reported that the average concentrations of exchangeable Ca, Mg and K in the paddy soils of the Hommachi Farm, which were collected in the spring of 2006, were 15.8, 2.9 and 0.6 cmol$_c$ kg^{-1}, respectively. These values can be converted to the molar equivalent ratio as Ca:Mg:K = 81.9:15.0:3.1, which represents the excess of Ca in the paddy soils of the study site because the recommended molar equivalent ratio of these exchangeable cations in the paddy soil of Japan is Ca:Mg:K = 65–75:20–25:2–10 [18]. Thus, the MgO materials can be utilized as an effective soil amendment for not only suppressing Cd uptake by crops but also optimizing the balance between exchangeable bases in the soil of this farm. However, a 2250 kg ha^{-1} of the commercial MgO applied into the experimental paddy field in this study would add 6.7 cmol$_c$ kg^{-1} of exchangeable Mg in the soil, assuming that the whole MgO in the material (2030 kg ha^{-1} as described above) was turned into exchangeable Mg. This would result in the change in the molar equivalent ratio of exchangeable bases in the soil as Ca:Mg:K = 60.8:36.9:2.3, representing the excess of Mg. Even though it is the extreme case (in other words, the whole MgO in the material should not be immediately converted to exchangeable Mg in the soil), the application rates of the MgO materials should be carefully determined from the both perspectives of suppressing Cd uptake by crops and maintaining the adequate balance of exchangeable bases in soil.

5. Conclusions

Winter wheat (*T. aestivum* L. cv. Ayahikari) was cultivated in a Cd-contaminated paddy field under annual rice–wheat rotational system to investigate the suppressive effects of the MgO materials (commercial MgO and MgO-SH-A), which had been applied prior to the preceding rice cultivation, on Cd accumulation in the wheat grain. The pH of the plough-layer soil in the wheat cultivation period decreased in the following order: commercial MgO at 2250 kg ha^{-1} (6.5) ≥ MgO-SH-A at 4500 kg ha^{-1} (6.3) ≥ MgO-SH-A at 2250 kg ha^{-1} (6.2) ≥ no amendment control (6.1), and this sequence had remained unchanged since the preceding rice cultivation. Although all the treatments with the MgO materials lowered Cd concentration in wheat grain as compared to the control, only the treatment with commercial MgO at 2250 kg ha^{-1} produced wheat grain whose Cd content was less than the maximum limit adopted by CAC, i.e., 0.2 mg kg^{-1}. In the experimental paddy field, pH of the plough-layer soil was negatively correlated with 0.025 mol L^{-1} HCl-extractable Cd concentration in the soil, which was positively correlated with Cd content in wheat grain. Consequently, the reduction in Cd concentration in wheat grain from the treatment with the MgO materials would be mainly attributed to the increase in soil pH due to incorporation of reactive MgO in the material and the subsequent reduction in availability of Cd in soil for uptake by wheat.

In conclusion, the MgO materials examined in this study can be utilized as the soil amendments for suppression of Cd accumulation into grains of paddy rice and winter wheat, although their application rates have to be carefully considered in order not to lead the imbalance of exchangeable Ca, Mg and K (excess in exchangeable Mg) in the paddy soil due to their applications.

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