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RESEARCH ARTICLE

Effects of foliar application of Zn combined with organic matters on Cd accumulation and its chemical forms in rice

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Abstract

Rice consumption is a key Cd exposure pathway, which poses a health risk to humans. Reducing cadmium (Cd) concentrations in rice remains challenging. In this study, a pot experiment was conducted to examine the effects of foliar spray of Zn combined with organic matters (including Zn–lysine (Zn-Lys), Zn–fulvic acid (Zn-FA), Zn–amino acid (Zn-AA), and Zn combined with glutathione (Zn + GSH)) on Cd accumulation in rice grains. Compared with the control group, all treatment groups exhibited reduced Cd concentration in rice grains, while improving plant growth, and reducing Cd transport from other tissues to the grains. Zn–FA was found to be the most effective fertilizer, which considerably reduced Cd concentrations in grains from 0.77 ± 0.068 to 0.14 ± 0.021 mg/kg and yielded reductions of up to 81%, which is within the Chinese food maximum tolerable limit of 0.2 mg/kg. Furthermore, the analysis of the chemical forms of Cd of rice tissues indicated that the treatment groups had increased proportions of integrated with pectates and protein in the stems. Except for the group treated with Zn–Lys spray, the percentages of undissolved Cd phosphate in the leaves were increased in all treatment groups, which reduced Cd toxicity to rice plants. The foliar application of Zn combined with organic matters may be a promising strategy to decrease Cd concentration in rice grains cultivated in severely Cd-contaminated agricultural soil, particularly in the karst area in southwest China with limited available cultivable agricultural land.

Keywords Foliar spray · Rice grain · Agricultural soil · Cadmium pollution · Zinc · Remediation

Introduction

Cadmium (Cd) is a nonessential and toxic element that can cause renal failure, proximal tubular re-absorptive dysfunction, osteopenia, and cancer (Schwartz and Reis 2000; Wu et al. 2010). Different Cd forms exhibit differences in their ability to migrate to plants, which in turn affects Cd transport in vivo and causes Cd toxicity. Exchangeable Cd in soil showed positive correlations with Cd in plants because it is the most active form (Feng et al. 2020; Xu and Wang 2013). Cd migrates into agricultural soils through mining, metal smelting, atmospheric deposition, irrigation water, fertilizer, and metal-containing sewage sludge (Lu et al. 2023). On entering an agricultural ecosystem, Cd can accumulate in crops, decreasing plant growth, reducing the total dry weight, and posing a threat to animals' and humans' health (Du et al. 2020; Kaya et al. 2020; Ilyas et al. 2022). A study indicated that heavy metals are one of the abiotic stresses contributing to high-yield losses of crops (Mariyam et al. 2024).

In most Asian countries, rice (*Oryza sativa* L.) is widely cultivated. Annually, over 740 million tons of rice is produced. Higher expressions of Nramp5 proteins and *OsNramp5* in rice compared with other cereal grains contribute to higher Cd accumulation in rice (Sui et al. 2018). Consumption of Cd-contaminated rice is a major exposure pathway that resulted itai-itai illness outbreak in Japan (Jarup and Akesson 2009). Recently, a study in China revealed that Cd contents were as high as 65% in rice samples from the rural

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areas of southern provinces, such as Hunan, Guangxi, and Guizhou, which exceeded the Chinese food maximum tolerable limit of 0.2 mg/kg (Zhu et al. 2016). Thus, decreasing Cd concentrations of rice grains is critical for ensuring food security.

Water management, soil amendment, and foliar spray are generally used to reduce rice Cd concentration (Hussain et al. 2021). Among these methods, foliar spray has received considerable research attention because of its efficiency, ease of operation, rapid plant responses, and cost-effectiveness (Li 2019; Yadav et al. 2022). Recently, Kumar et al. (2023) applied a foliar spray of titanium dioxide nanoparticles to reduce hexavalent chromium accumulation, and foliar application of α-lipoic acid was reported to alleviate Cd toxicity (Yadav et al. 2022). Given the rapid development of unmanned aerial vehicle spraying technologies, which are effective sprayers, foliar spraying is increasingly being employed, especially in China (Yang et al. 2021).

Through reducing oxidative stress, modulating photosynthesis, and promoting nutrient balance, zinc (Zn) antagonizes and reduces Cd uptake in plants (Qin et al. 2020; Yasmin et al. 2021). Foliar application of Zn is more effective method for reducing Cd levels (Lv et al. 2019) and increasing Zn concentrations (Zulfiqar et al. 2020; Anwar et al. 2021) in grains compared to soil application. This is because Zn applied through foliar treatment can be readily absorbed by the leaf epidermis and subsequently transported to the developing grains via the phloem (Zhen et al. 2021). Foliar spray of $ZnSO_4$ (0.4–0.5%) and ethylenediaminetetraacetic acid disodium zinc salt tetrahydrate (ZnEDTA) has been reported to decrease Cd accumulation in rice grains (Fahad et al. 2015; Lv et al. 2019; Wang et al. 2020a, b). However, the application of these fertilizers can cause foliar burn of crops, attributable to the presence of salts and acids (Golden et al. 2016).

Natural organic substances are non-toxic for living organisms (Dolev et al. 2020). Research indicates that the utilization of Zn complexed with amino acids is more efficacious in enhancing crop growth, Zn uptake, and yield compared with the application of inorganic Zn fertilizers ($ZnSO_4$; Boonchuay et al. 2013). Wang et al. (2019) investigated the impact of foliar application of fulvic acid (FA) on Cd-exposed lettuce (*Lactuca sativa* L.) and observed increases in the growth, biomass, chlorophyll content, and photosynthesis capacity and a decrease in Cd concentration. Glutathione (GSH), similar to Zn, can play a crucial role in Cd tolerance in plants. Nakamura et al. (2019) discovered that GSH application can affect Cd behavior by Zn in oilseed rape (*Brassica napus*). However, limited studies have investigated the effects of foliar spray of Zn combined with organic matters (amino acids, FA, and glutathione) on Cd uptake in rice plants.

The migration of Cd in plants is affected by its chemical form (Wu et al. 2005). Studies have revealed that adding arbuscular mycorrhizal fungi to a hydroponic nutrient solution can reduce the proportion of inorganic and water-soluble Cd in rice plants (Luo et al. 2017). However, the changes in Cd forms when using foliar spray to decrease Cd accumulation in rice plants are yet to be fully understood. Therefore, in the present study, we examine the impact of foliar spray on Cd form and explore the mechanisms by which it decreases Cd concentration in rice grains.

In the present study, four types of Zn and natural organic matter-associated fertilizers, namely Zn–fulvic acid (Zn–FA), Zn–lysine (Zn–Lys), Zn–amino acids (Zn–AA), and Zn combined with glutathione (Zn + GSH), were used for foliar spray. The Cd levels, biomass, and Cd translocation factors (TFs) of rice were measured after foliar treatment. Additionally, Cd chemical forms in the roots, stems, and leaves of rice plants were studied. This study aims to provide a novel method for decreasing Cd levels in rice grains and reveal the possible mechanism of action of foliar spray. To the best of the knowledge, this study is the first to explore the effect of Zn combined with various organic matters on Cd accumulation in rice grains.

Materials and methods

Experimental soil and reagents

Surface paddy soil (0–20-cm depth) was collected from a Cd-contaminated Tianzhu Ba mining region in Guizhou Province, China (Lu et al. 2019). The soil was subsequently air-dried, ground, and passed through a 2-mm sieve in a laboratory. The average Cd concentration of the soil was 35 ± 3 mg/kg, which is higher than the Cd risk screening criterion of 0.3 mg/kg ($pH \leq 5.5$) for soil pollution on agricultural land (Ministry of Ecology and Environment of People's Republic of China (MEE) 2018). Soil parameters including pH, total carbon, and total nitrogen were assessed (Table 1). Soil pH was determined by transferring 5 g soil into 12.5 mL

Table 1 Initial characteristics of experimental soil

Soil parameters	pH	Total carbon (%)	Total nitrogen (%)	Carbon/nitrogen	Total Cd (mg/kg)
Values	4.97 ± 0.09	3.5 ± 0.063	0.33 ± 0.004	10.5 ± 0.14	35 ± 2.8

of deionized water (The Ministry of Agriculture of the People's Republic of China 2007). Total carbon and total nitrogen were measured using vario MACRO cube element analyzer following the method in Song et al. (2017).

We prepared the solutions of Zn–FA, Zn–AA (Beijing Bowei Shennong Technology Co., Ltd., China), Zn–Lys, glutathione (GSH; Heibei Liweisu Co., Ltd., China), and zinc sulfate ($ZnSO_4$; Sinopharm Chemical Reagent Co., China). Each solution contained 0.2% Zn (*m/v*). Specifically, 10 g Zn–FA was mixed with 1 L deionized water to prepare the Zn–FA solution; Zn–AA solution was prepared by adding 13 g Zn–AA to 1 L deionized water; Zn–Lys solution was obtained by adding 15 g Zn–Lys to 1 L deionized water; and Zn + GSH solution was prepared by adding 9 g zinc sulfate and 4 g GSH to 1 L deionized water.

Pot experiment

Rice was cultivated in plastic pots with approximately 10 kg of prepared soil in each pot. The pots were classified into five groups. Three replicates were set for each group (two plants in each pot). Drinking water (Cd concentration < detection limit) was used for irrigation.

For experiments, rice seedlings with similar growth were collected from a paddy field free from Cd contamination. Two seedlings were planted with urea (0.8 g) and compound fertilizer (0.5 g; Shandong Lvfeng Fertilizer Co., Ltd., China) into each pot. Next, 0.8 g urea and 1.6 g compound fertilizer were added at the tillering stage. Pots were flooded to a depth of 2 cm, and the water level was maintained throughout the rice-growing season.

Foliar treatments of Zn + GSH, Zn–FA, Zn–AA, and Zn–Lys, along with a control, were administered. Foliar applications of Zn combined with organic matters were achieved by spraying the leaves at five stages, namely seedling, tillering, heading, filling, and maturity. A sticking agent (0.1%, Tween-80) was used to maximize the period of stay of the solution on leaves. Tween-80 mixed with deionized water was administered to the control group.

Sample collection

At maturity, the height of rice plants was measured, and subsequently, rice was harvested. Fresh roots, stems, leaves, and panicles were collected from each pot; the collected samples were first washed with tap water and then with deionized water, followed by weighing. A portion of each sample was used to determine the moisture content, and a small amount of each fresh sample was immediately stored in an ultra-low temperature freezer (DW-86L828, China; – 80 °C) for the analysis of Cd chemical forms. The remaining parts of the plant were freeze-dried (EYELA model FDU-1100, Japan), powdered (IKA-A11; IKA, Germany), and stored in bags prior to analysis. The grains were separated into brown rice (JLGJ4.5, China) and ground into powder.

Sample analysis

Cd concentration analysis

Powdered samples (roots, stems, leaves, and brown rice) were digested with HNO_3/H_2O_2 for Cd analysis following the method reported by our previous article (Lu et al. 2019). Weighted samples were added to the Teflon crucible containing HNO_3, followed by heating in an oven at 150 °C for 36 h. Next, 2 mL H_2O_2 was added, and each sample was heated at 115 °C until the solution was completely evaporated. The Teflon crucible was then filled with deionized water (3 mL) and HNO_3 (2 mL) and stored at 150 °C for 5 h. Graphite furnace atomic absorption spectrometry was used to determine the Cd content (TAS 990, Puxi, Beijing, China).

Chemical forms of Cd

Six chemical forms of Cd were extracted and then quantified through graphite furnace atomic absorption spectrophotometry. The different Cd chemical forms extracted are as follows (Zhang et al. 2014):

Inorganic (F1): nitrate/nitrite, chloride, and aminophenol Cd were extracted with 80% ethanol.

Water-soluble (F2): water-soluble Cd in organic acid complexes and $Cd(H_2PO_4O_2)$ were extracted with deionized water.

Pectate and protein-bound (F3): 1 M NaCl was used to extract Cd combined with pectate and proteins.

Phosphate associated (F4): insoluble $CdHPO_4$, $Cd_3(PO_4)_2$, and other Cd phosphate complexes were extracted with 2% HAC (acetic acid).

Oxalate acid-bound (F5): 0.6 M HCl was used for the extraction of oxalate acid-bound Cd.

Residue (F6): residual Cd.

Quality assurance and quality control

Data validation was performed by using duplicates, method blanks, and standard materials. The average Cd content of 0.18 ± 0.03 mg/kg (certified value of 0.17 ± 0.02 mg/kg) was measured in the certified reference material (citrus leaf; GBW10020). The relative percentage difference was less than 5% between duplicate samples. The recoveries ranged from 92 to 106% for the diluted certificated standard stock solutions, with every 15 samples analyzed during the analysis.

Calculation of Cd TFs

Based on the Cd concentrations (C) in each tissue (roots, stems, leaves, and grains), the *TFs* of the corresponding rice plant tissues at maturity were calculated according to the following equations (Rehman et al. 2019):

$$TF_{root/leaf} = C_{leaf}/C_{root}$$

$$TF_{root/grain} = C_{grain}/C_{root}$$

$$TF_{stem/grain} = C_{grain}/C_{stem}$$

$$TF_{leaf/grain} = C_{grain}/C_{leaf}$$

where C is the concentration in each tissue.

Statistical analysis

Statistical analyses were performed using the one-way analysis of variance (ANOVA) in SPSS 27. Data are displayed as the mean and standard error based on the values in the experiments conducted in three replicates. Differences were considered significant at 5% level.

Results and discussion

Effects of Zn combined with organic matters on plant biomass

The effects of Zn combined with organic matters foliar spray on the leaf weight, plant height, and grain yield are detailed in Fig. 1. Foliar spray did not considerably affect leaf weight and plant height ($p > 0.05$, Fig. 1A and 1B). For grain yield, compared with the control (72 g/pot), Zn–Lys (77 g/pot), Zn–AA (76 g/pot), and Zn + GSH (75 g/pot) showed slight effect after application, whereas Zn–FA (109 g/pot) increased grain yield, with the percentage of 11% (Fig. 1C).

FA-chelated Zn is more effective in increasing rice yield than other zinc fertilizers. Zinc application can improve the grain yield of crop mainly because Zn plays a crucial role in biochemical processes (Anwar et al. 2021). FA is a natural biostimulant that can promote the absorption of micronutrients by foliage in various plants by complexing micronutrients in the soluble form (Justi et al. 2019). Additionally, it can enhance the photosynthetic activity, leaf SPAD, stimulate cell division, and increase vegetative growth and dry matter accumulation, thereby improving plant yield (Anjum et al. 2011; Mei et al. 2022). FA serves as a carrier for many chemicals, transporting them from

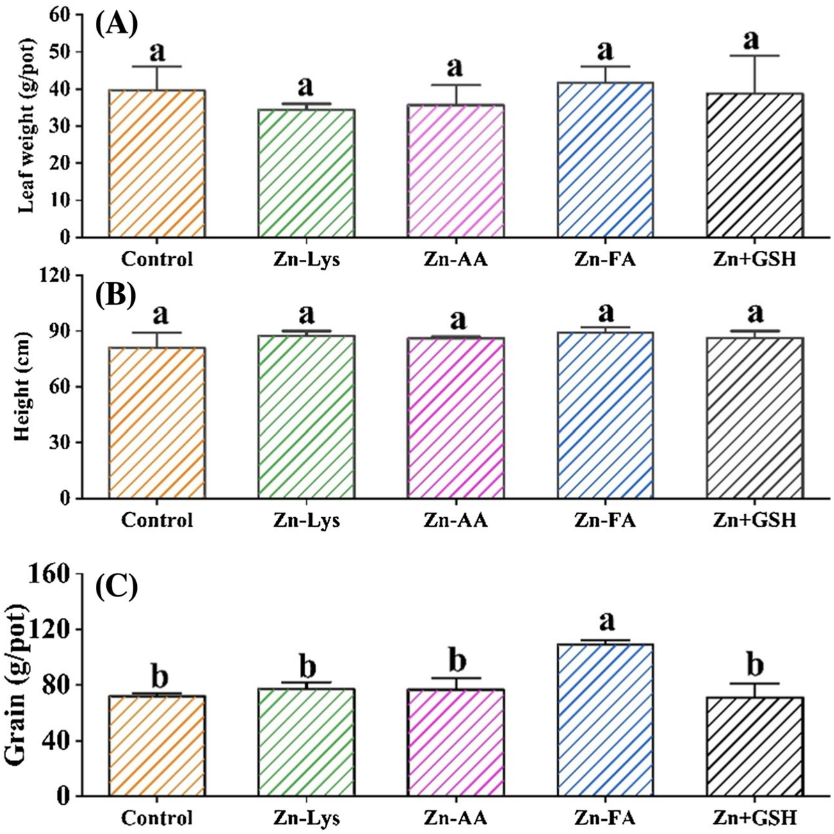

Fig. 1 Effects of the foliar application of Zn fertilizers on leaf weight (**A**), plant height (**B**), and grain yield (**C**). Bars with different letters indicate significant differences (at $P < 0.05$) based on one-way ANOVA analysis. Zn-Lys, Zn–lysine; Zn-AA, Zn–amino acid; Zn-FA, Zn–fulvic acid; Zn + GSH, Zn combined with glutathione

the surface of plant organs to plant cells. Due to its low molecular weight, it can pass through membrane pores and carry nutrients (Bocanegra et al. 2006; Braziene et al. 2021). Xu (1986) reported that FA spraying enhanced the leaf chlorophyll content and plant yield under drought conditions. Wang et al. (2020a, b) observed that the foliar application of Zn plus FA resulted in higher Zn bioavailability than foliar Zn application alone, which helped achieve agronomic biofortification. The mechanisms for these phenomena remain be comprehensively investigated.

Effect of Zn combined with organic matters on Cd bioaccumulation

The concentrations of Cd in brown rice grains under Zn combined with organic matters treatments are displayed in Fig. 2. In the control, Cd concentration in grain was 0.77 ± 0.068 mg/kg. Foliar-sprayed Zn fertilizers reduced Cd accumulation in grains to varying degrees. Cd concentrations in the groups treated with Zn + GSH, Zn–FA, Zn–AA, and Zn–Lys were 0.34 ± 0.11, 0.14 ± 0.021, 0.24 ± 0.0064, and 0.23 ± 0.087 mg/kg, respectively, and the corresponding percentages of reduction in Cd concentrations compared with control were 56%, 81%, 68%, and 69%, respectively. Zinc shares chemical properties and many transporters and binding sites with Cd. For example, OsHMA2, OsLCT1, and OsZIP3 are expressed at node I and considerably affect Zn and Cd transport in rice. Thus, these compounds increase Zn transfer to rice grains and decrease Cd accumulation (Tian et al. 2019). Among the treatment groups, the Zn-FA group had the lowest Cd level in rice grains (0.14 ± 0.021 mg/kg). This concentration was found to be lower than the maximum permissible concentration of 0.2 mg/kg in rice in China (National Health Commission of the People’s Republic of China (NHC) 2017), indicating that Zn–FA has the most significant impact on reducing Cd in rice. FA application can decrease the expression of *IRT1*, *Nramp1*, *HMA2*, and *HMA3*, which can transport Cd ions (Chen et al. 2022).

Cadmium concentrations in the roots, stems, and leaves of the treatment groups and control group are displayed in Fig. 3. In all treatment groups and the control group, roots had considerably higher Cd concentrations than other parts of the plant. Cd enters root apoplasts, where it can be adsorbed onto the surfaces of pectin-containing root cell walls (Schroeder et al. 2001). Membrane transporters facilitate the entry of Cd into the root symplast through the plasma membrane, which is followed by its elimination and then retention in the roots (Khan et al. 2014). Therefore, rice root tissues tend to be enriched more with a high Cd level than other tissues. Generally, the root Cd concentrations were reduced after Zn combined with organic matters foliar application. Zn–Lys and Zn–AA application slightly reduced the root Cd concentration. However, Zn–FA and Zn + GSH foliar spray led to significant reductions ($P < 0.05$) in Cd accumulation in the roots ($P < 0.05$) compared with controls (40% and 41%, respectively, Fig. 3). Zn is an antagonistic element that limits Cd entry into plants because it is chemically similar to Cd and competes with it for root uptake and vacuolar storage in root cells, and both Zn and Cd have common transporters. Furthermore, FA can promote nutrient uptake by plants, leading to a competition between nutrients and

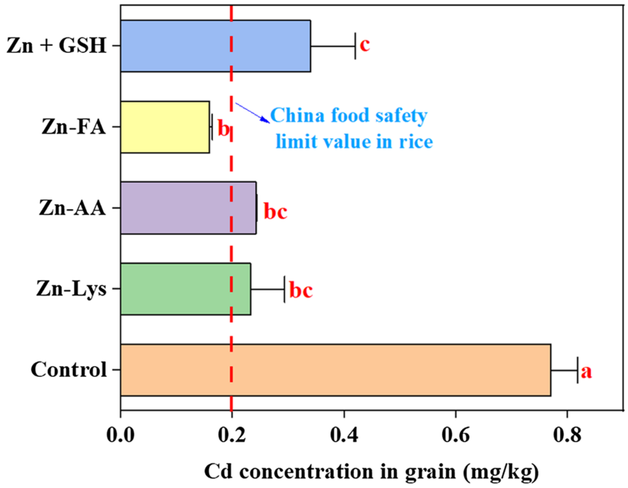

Fig. 2 Effects of the foliar application of different Zn fertilizers on Cd concentrations in rice grain. Bars with different letters indicate significant differences (at $P < 0.05$) based on one-way ANOVA analysis. Zn-Lys, Zn–lysine; Zn-AA, Zn–amino acid; Zn-FA, Zn–fulvic acid; Zn + GSH, Zn combined with glutathione

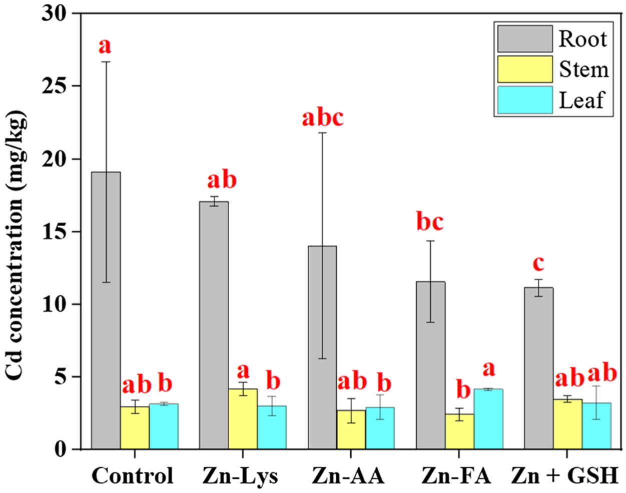

Fig. 3 Effects of the foliar application of Zn fertilizers on Cd concentrations in the roots, stems, and leaves. Bars with different letters indicate significant differences (at $P < 0.05$) based on one-way ANOVA analysis. Zn-Lys, Zn–lysine; Zn-AA, Zn–amino acid; Zn-FA, Zn–fulvic acid; Zn + GSH, Zn combined with glutathione

heavy metal ions, resulting in the reduction of heavy metal contents in roots (Wang et al. 2019). GSH can reduce Cd concentration in the symplast sap of root cells and promote Cd efflux from roots (Nakamura et al. 2019). No significant change in stem Cd concentration was observed in the treatment groups, except for the group treated with foliar sprays of Zn–FA. Studies have reported that Zn–Lys and Zn–AA application can decrease Cd accumulation in the stem (Ali et al. 2022). The variation could be attributed to the plant species. In the leaves, Cd concentrations increased by 32% after Zn–FA foliar spray, but no obvious effect was observed after the application of other fertilizers. The increase in leaf Cd concentration with Zn–FA application could be attributed to the change in the chemical forms of Cd in the leaves, which led to its reduced mobility and increased accumulation (see Effect of Zn combined with organic matters on Cd chemical forms in rice tissues of this study).

In addition to the influence of Zn combined with organic matters foliar spray on Cd uptake and accumulation in rice crop, Cd translocation between rice tissues may be altered. Hence, the TF values were calculated, as shown in Fig. 4. Compared with the control group, treatment groups significantly decreased Cd translocation from other tissues to rice grains (Fig. 4A–C) but increased the translocation of Cd from the roots to leaves (Fig. 4D). Higher Cd concentration was observed in leaves of the Zn–FA group than in those of the control group (Fig. 3), which is similar to that of Han et al. (2019) in which the foliar spray of Zn efficiently decreased Cd in grains by inhibiting Cd transport from plant organs and increased the transfer of Cd from roots to leaves. Zhen et al. (2021) also revealed that Zn foliar application enhanced Cd accumulation in rice leaves by increasing Cd chelation onto the cell wall of leaves.

Effect of Zn combined with organic matters on Cd chemical forms in rice tissues

The percentages of various chemical forms of Cd in the root, stem, and leaf of rice are displayed in Fig. 5. Pectate and protein-integrated Cd (F3) and undissolved Cd phosphate (F4) were abundant in the rice tissues of both treatment and control groups. The total percentage of these compounds was greater than 50%. Additionally, the percentage of F3 was up to 68% in the leaves after Zn–Lys application, and the proportion of F4 was up to 43% in the leaves treated with Zn–FA foliar spray. However, the proportions of F1 (inorganic form) and F2 (water-soluble form) were considerably lower than those of F3 and F4. For instance, the F1 and F2 percentages were only 1.0% and 1.2%, respectively, in the stem with Zn + GSH treatment. Overall, Cd in rice plants existed mostly in the undissolved Cd phosphate and pectate and protein-bound Cd forms. Additionally, the treatment groups exhibited significantly increased proportions of F3 in the stems from 23 to 37% (Zn–Lys), 23 to 52% (Zn–AA), 23 to 65% (Zn–FA), and 23 to 54% (Zn + GSH). Zn–FA foliar application significantly increased F4 proportion in the leaves (from 20 to 43%). However, compared with the control group, the total proportions of F1 and F2 were reduced in stems and leaves of the treatment group.

Cadmium toxicity to plants is related to its chemical forms. Cd combined with proteins/pectates exerts fewer negative effects on plant cells than its water-soluble and inorganic forms (Xu et al. 2011), thereby having a vital

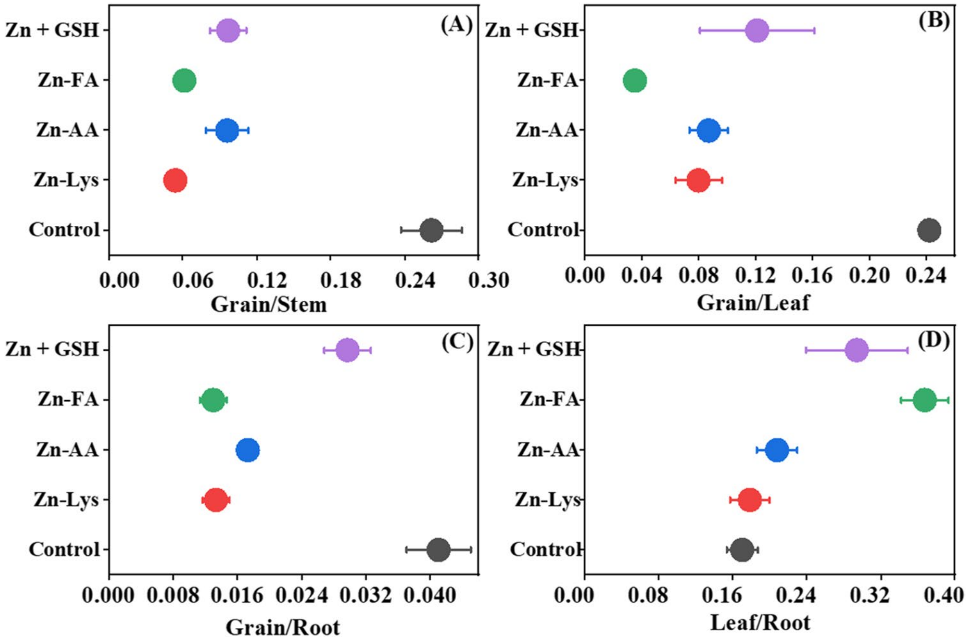

Fig. 4 Effects of the foliar application of Zn fertilizers on Cd translocation factors from the stems to grains (**A**), leaves to grains (**B**), roots to grains (**C**), and roots to leaves (**D**). Zn-Lys, Zn–lysine; Zn-AA, Zn–amino acid; Zn-FA, Zn–fulvic acid; Zn + GSH, Zn combined with glutathione

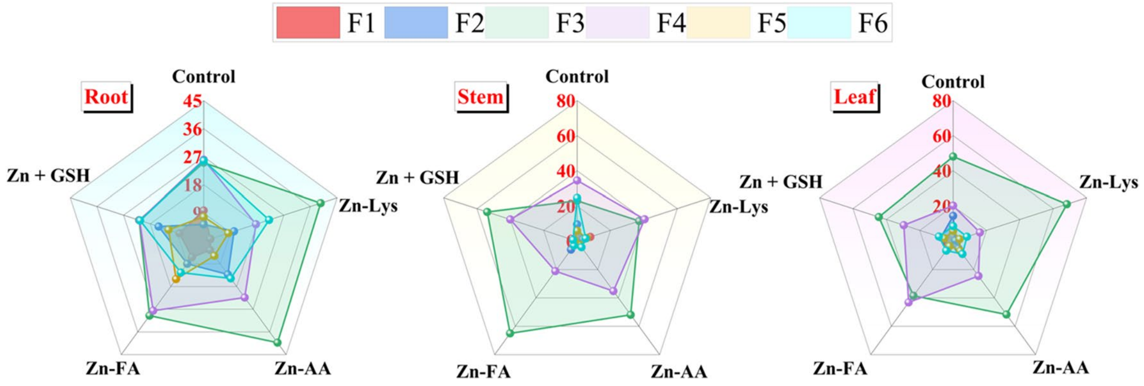

Fig. 5 Effects of the foliar application of Zn fertilizers on the chemical forms of Cd in rice tissues (including roots, stems, and leaves). Zn-Lys, Zn–lysine; Zn-AA, Zn–amino acid; Zn-FA, Zn–fulvic acid; Zn + GSH, Zn combined with glutathione; F1, inorganic Cd; F2, water-soluble Cd; F3, pectate and protein-integrated Cd; F4, Cd phosphate; F5, oxalate acid-bound Cd; F6, residue Cd

role in the physiological mechanism in Cd detoxification. Pectic fragments could induce defense response in plants (Meng et al. 2017). Ethanol- and deionized water-extracted Cd, combined with nitrates, dihydric phosphate, organic acids, and chlorides, could easily penetrate into symplast and localize to organelles (Zhang et al. 2014). These compounds exhibit a strong migration ability and are highly toxic to plants (Fu et al. 2011). Zheng et al. (2022) revealed that mannose application considerably reduced water- and ethanol-extracted Cd content in roots, which alleviated the toxic effects of Cd and improved wheat growth. Cd-sensitive barley cultivars exhibited higher contents of inorganic and water-soluble Cd and lower contents of pectate/protein-integrated Cd than Cd-resistant cultivars (Wu et al. 2005). KH_2PO_4 foliar application enhanced Cd tolerance in tall fescue by increasing the proportion of pectate- and protein-integrated Cd in leaves (Meng et al. 2017). Cd combined with phosphate (extracted by 2% HAC) is insoluble with less mobility and toxicity (Zhao et al. 2015). Jiang et al. (2007) reported that phosphorus input promoted the formation of Cd phosphate deposits in the cell wall and vacuoles, which eliminated Cd-related toxicity to leaves. Phosphorus is essential in the detoxification and immobilization of Cd in plants. Therefore, foliar application of Zn combined with organic matters changed the Cd chemical form, reduced the toxic effects of Cd, and decreased the transfer of Cd to the grains.

Cadmium in rice grains mainly has two sources: (1) re-translocation from old leaves and (2) node-based distribution after the flowering stage (Uraguchi and Fujiwara 2013). In this study, the percentage of Cd phosphate (F4) in the leaves increased with treatment, except for Zn–Lys application, indicating increased combination of Cd with phosphates, which has lower mobility (Zhao et al. 2015), thereby decreasing Cd re-translocation from leaves to grains. Overall, the application of Zn combined with organic matters reduced Cd toxicity to rice plants by changing the chemical forms of Cd. Additionally, they promoted the conversion of Cd into insoluble Cd phosphates in leaves, decreasing the amount of Cd re-translocated from leaves to rice grains. The investigation of the changes in Cd chemical form after Zn–Lys, Zn–AA, Zn–FA, and Zn + GSH applications is in the preliminary stage. A further study using synchrotron radiation technique is warranted for determining the changes in specific Cd species.

Environment implications

Cadmium pollution of rice has attracted considerable attention globally. Reducing Cd accumulation in rice to ensure food safety and human health is challenging. Zn is an essential element for plants, and zinc fertilizer foliar spray can decrease Cd concentration in rice grains. However, previous studies have mainly focused on the effectiveness of zinc sulfate (inorganic Zn fertilizer) foliar spray in reducing Cd levels in rice grains (Fahad et al. 2015; Lv et al. 2019). Zinc sulfate application may result in yield loss, foliar burn, and nutritional imbalances (Drissi et al. 2015; Golden et al. 2016). Organic Zn fertilizer is more effective than inorganic Zn fertilizer in increasing yield and improving the quality of crops (Ghasemi et al. 2013). Natural organic matter is non-toxic and eco-friendly to living organisms, and is thus suitable to sustainable agriculture (Dolev et al. 2020). This study is the first comprehensive report on the impact of Zn combined with organic matters (including Zn–Lys, Zn–AA, Zn–FA, and Zn + GSH) on the reduction of Cd content in rice grains grown in soil with severe Cd contamination. The most effective fertilizer combination identified in this study was Zn–FA, which could efficiently decrease grain Cd concentration from 0.77 ± 0.068 to 0.14 ± 0.021 mg/kg.

Therefore, the foliar application of Zn-FA can serve as a most potential measure for the remediation of highly Cd-contaminated soil and increasing rice yield.

Conclusions

This study revealed that foliar spray of Zn combined with organic matters (Zn–lysine, Zn–fulvic acid, Zn–amino acid, and Zn combined with glutathione) can decrease Cd levels in brown rice grains to varying degrees. Zn–fulvic acid application is the most effective that reduces the grain Cd value to less than 0.2 mg/kg (the Chinese safety standards) with 81% reduction percentage. These fertilizers improved plant growth, reduced Cd transport from other tissues to the grains, and increased the proportion of pectate- and protein-integrated Cd in the stems in all treatment groups and undissolved Cd phosphate in the leaves, except for the group treated with Zn–lysine foliar application. This study revealed that Zn–lysine, Zn–amino acid, Zn–fulvic acid, and Zn combined with glutathione foliar applications can decrease Cd levels in rice grains, with Zn–fulvic acid foliar application being the most promising measure for reducing Cd concentration in rice cultivated in severely Cd-contaminated paddy soil. Hence, Zn–fulvic acid foliar application might be an effective method to reduce Cd accumulation in rice grains. Given that this study was conducted ex-situ, the next step is to determine the efficiency of foliar application of these fertilizers under field conditions and explore the underlying molecular mechanisms.

Author contribution QL: investigation, writing—original draft, writing—reviewing and editing, funding acquisition; ZX: investigation, methodology, writing—reviewing and editing; ZC: investigation, formal analysis; GQ: supervision, project administration, funding acquisition, writing—reviewing and editing.

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Data availability All data generated or analyzed during this study are available from the corresponding author on reasonable request.

Declarations

Ethics approval This is not applicable.

Consent for participate This is not applicable.

Consent for publication Not applicable.

Competing interests The authors declare no competing interests.

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