The contribution of atmospheric deposition to cadmium and lead accumulation in rice grains

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Abstract

Purpose Over-accumulation of cadmium and lead in rice grain is a global concern as it has adverse health impacts. Atmospheric deposition is an important source of heavy metal accumulation in soil, but contribution to crops has not been quantified and the mechanisms of foliar Cd and Pb uptake via the stomata of rice leaves exposed to atmospheric fallout are unclear.

Methods To quantify the contribution of atmospheric deposition on Cd and Pb accumulation in rice grains, a rice pot experiment with three exposure treatments (T1, all day exposure without geotextile membranes; T2, all day exposure with geotextile membranes; T3, daytime exposure with geotextile membranes; and T4, night exposure with geotextile membranes) using severely (ZZ) and moderately (XT) polluted soils was conducted.

Results Cd content in shoots and roots was $T1 > T2$, $T3 > T4$ in XT soils, and $T1 > T2$, $T4 > T3$ in ZZ soils, while Pb content in both soils was $T1 > T2$, and $T4 > T3$. Cd and Pb contents in rice grains showed the same trend. Using the tracer of isotope tracer ratios ($^{114}/^{111}\text{Cd}$, $^{112}/^{111}\text{Cd}$, and $^{207}/^{206}\text{Pb}$, $^{208}/^{206}\text{Pb}$) the contribution of atmospheric deposition to rice grains was quantified as 63.55% and 18.01% for Cd, and 27.69% and 41.13% for Pb in XT and ZZ soils, respectively.

Conclusions Foliar uptake atmospheric deposition had substantial effect on Cd and Pb accumulation in rice grains and the control of heavy metal foliar uptake should be paid more attention in to maintain rice safety production.

1 Introduction

Cadmium and lead are highly toxic trace elements in the environment and can cause obvious damage to human health even at low concentrations (Bohdálková et al. 2020). Studies have proven that crops contaminated with Cd and Pb can be passed on through the food chain and pose a direct threat to humans due to biomagnification effect (Keller et al. 2015; Baycu et al. 2017). Therefore, food safety issues caused by over-accumulation of Cd and Pb in crops has drawn increasing attention recently. Rice (*Oryza sativa* L.) is the most important staple food for half of humanity. China (especially southwestern China) is one of the countries with maximum consumption and production of rice. Liu et al. (2016) collected and analyzed heavy metals in 101 pairs of soil and rice samples from the main rice-producing areas along the Yangtze River in China, and found that the average content of Cd in paddy soils was 0.73 mg kg^{-1} with an increasing rate of 0.01 mg/kg/year . Du et al. (2013) conducted a monitoring test in paddy fields in northern Hunan Province and found that the average content of Cd in soils was 0.64 (range from 0.13 to 6.02) mg kg^{-1} , with 0.46 (range from 0.01 to 2.77) mg kg^{-1} in rice grains, exceeding the limits specified in China's Food Safety and Quality Standards (GB/T 1354–2009). Therefore, it is critical to pay efforts to identify the source of origin of Cd and Pb and quantify their accumulation in rice grains.

Where are the Cd and Pb coming from? Metal elements can be assimilated through both the root and leaf (Schreck et al. 2014). Many studies have proved that the accumulation heavy metals in rice grains are

taken up by roots from the soil and then translocate to edible parts through xylem and phloem (Li et al. 2017). Moreover, foliar uptake can also be an important source responsible for heavy metal accumulation in grains, particularly when ultra-fine particles directly interact with plant leaves (Feng et al., 2011; Hu et al., 2011; Schreck et al., 2012). Airborne PM_{2.5} can be directly absorbed by leaves through the stomata and then translocated to other parts of plant, which is closely related to the characteristics of stomata. (Gao et al. 2021; Melotto et al. 2017). The stomata are sensitive to light changes, salt stress, and drought. (Xiong et al. 2018; Mekonnen et al. 2016). It was found that the opening and closing of stomata under light and dark conditions had a considerable influence on metal accumulation through foliar uptake (Gao et al. 2021). Zhou et al. (2020) conducted a field experiment and found that atmospheric metal deposition can lead to over-accumulation of Cu and Pb even in rice crop planted in clean soil. De Temmerman et al. (2012) found the levels of As and Cd in carrot and celery were significantly correlated with the amount of heavy metals in atmospheric deposition.

In recent decades, enormous anthropogenic emissions have resulted in a high level of heavy metals in the atmosphere. In 2012, the anthropogenic emissions of Cu, Pb, and Cd in China were approximately 9547 t, 14397 t, and 527 t, respectively (Tian et al. 2015). The large stock of heavy metals in atmosphere has already become one of the main contributors to heavy metal accumulation in agricultural soils through long-term rain and dust fallout (Luo et al. 2009; Bermudez et al. 2012; Hou et al. 2014; Feng et al. 2018; Zhang et al. 2018). Studies have shown that the activity of Cd and Pb in atmospheric particulate matter (PM) was active (Liu et al. 2019). For example, in a coastal city of northern Spain, the water-soluble exchangeable components of Cd and Cu in atmospheric deposition accounted for 52% and 43%, respectively (Ignacio et al., 2014). In Asia, 30–84% of Cd was in exchangeable form, while the components of Pb were mainly in carbonate and reduced form, indicating heavy metals have higher potential bioavailability in particulate matters than in soil (Lee et al., 2015). Therefore, except for root uptake, foliar uptake of Cd and Pb from atmospheric deposition can also be an important pathway of Cd and Pb accumulation in rice grain.

To explore the contribution of atmospheric deposition to Cd and Pb accumulated in rice grain, there have been many attempts by many scholars. Feng et al. (2019) found that Cd and Pb accumulation in rice grains planted in more polluted deposition zones were significantly higher than in zones with less polluted deposition, and the contribution of atmospheric deposition accounted for more than 35% for Pb, and 40% for Cd. Liu et al. (2019) conducted pot experiments with the same soils in three sites of graded atmospheric heavy metal deposition and found 52–62% of Pb accumulation in vegetable shoots was directly uptake through leaf stomata. Zhou et al. (2019) also conducted a similar experiment, and concluded that atmospheric deposition contributed 31–63% (in moderate deposition areas), and 66–84% (in high deposition areas) to Pb accumulation in rice grains. Cao et al. (2020) conducted a simulated wet precipitation experiment with hydroponic culture of rice at seedling stage and found that Cd concentration in rice shoots increase significantly when the annual Cd precipitation flux ≥ 50 g/ha (amount of the annual Cd flux in industrial areas was about 61.0 g ha^{-1}). These monitoring and

simulation experiments prove that foliar uptake can increase Cd and Pb accumulation in rice grains, but they fail to precisely quantify the contribution.

Elements have fixed stable isotope ratios of homologous pollutants and can maintain this composition in the process of migration and reaction. Therefore, stable isotope ratios have been widely used as a “fingerprint” in source apportionment analyses in aerosols (Jaeckels et al. 2007; Yin et al. 2010), sediments (Tang et al. 2010), urban soils (Wong et al. 2004), and agricultural soils (Huang et al. 2015), but rarely in crops. Liu et al. (2019) used Pb isotope ratio analysis to quantify the sources of lead in soil and rice grains and showed that the sources of Pb in soil were background sources (20–84%), chemical fertilizers (0–42%), atmospheric deposition (16–42%), and irrigation water (0–28%), while that in rice grains, was atmospheric deposition (76–98%), and the soil (0–26%). Bi et al. (2009) analyzed Pb isotope ratios in soil and corn crops and concluded that the main source of Pb in corn grains and leaves was atmospheric, while Pb in roots and culms mainly came from the soil. However, the analysis of Cd isotope ratios was not determined.

Therefore, a full growth period, exposed pot experiment in rice was conducted along with long-term monitoring of atmospheric PM (PM₁₀ and TSP). The content of Cd and Pb in shoot, root, grain, soil, and atmospheric depositions, as well as ^{112/111}Cd, ^{114/111}Cd, ^{207/206}Pb, and ^{208/206}Pb were determined to explore the effects of stomata and exposure time on foliar uptake, and to quantify the contribution of atmospheric deposition on Cd and Pb accumulation in rice grains. The current study aimed to (1) explore the deposition fluxes of Cd and Pb in the study area; (2) to investigate the temporal change of Cd and Pb accumulation in rice with foliar uptake in a full growth period; (3) to assess and quantify the influence of atmospheric deposition on Cd and Pb accumulation in rice grains. The results will elucidate the foliar uptake mechanism of Cd and Pb in rice, and these metal accumulation in rice leaves through stomata, which can provide technical support to the control and management of heavy metal contamination in crops.

2. Materials And Methods

2.1 Site description and rice cultivation

The study was conducted at the experimental station of Hunan Agricultural University, Changsha City, southern China. This is a suburban area and not surrounded by typical sources of pollution, such as factories and mines. The leading wind direction prevails southeasterly in summer, and northeasterly in winter. The annual average temperature range was 16°C–18°C, with an annual average humidity of 70–80%. The average annual precipitation ranges from 1400 to 1700 mm.

2.2 Treatments design

To explore the contribution of atmospheric deposition on Cd and Pb accumulation in rice grain, a full plant growth outdoor exposure (all day exposure/day exposure/night exposure) pot experiments with two type of contaminated soils was set up. Moderately and seriously contaminated soils were collected from

paddy fields in Xiangtan city (XT) (the content of organic matter: 32.7 g kg^{-1} , content of Cd: 0.33 mg kg^{-1} , content of Pb: 61.9 mg kg^{-1} , pH: 5.42) and Zhuzhou city (ZZ) (the content of organic matter: 30.6 g kg^{-1} , content of Cd: 0.86 mg kg^{-1} , content of Pb: 65.2 mg kg^{-1} , pH: 5.42). To avoid direct input of Cd and Pb from atmospheric deposition to soil and then directly uptake by root, geotextile membrane was used to isolate atmospheric matters. Therefore, there were four treatments in the current study: T1 (all day exposure without geotextile membrane), T2 (all day exposure with geotextile membrane), T3 (daytime exposure with geotextile membrane) and T4 (night exposure with geotextile membrane). All treatments were conducted in triplicates 12 times.

The tested variety is Fengliangyou 4 of middle-season rice. The germinated seeds were firstly cultivated in uncontaminated soils for one month, then transplanted to the pots in mid-May 2019, and finally harvested in early-November 2019. Plants were exposed for the entire growth period and separately sampled in tillering (16th July 2019), heading (3rd August 2019), and ripening stage (7th November 2019).

2.3 Sample collection and analysis

2.3.1 Particulate matter (PM) samples

The PM samples were collected by an automatic precipitation and dust fall sampler each day from May 2019 to November 2019. The airflow rate was 28 L min^{-1} with 50% cutoff sizes of $> 10.0 \mu\text{m}$ (PM_{10}), and total suspended particles (TSP). The PM samples were digested using a $\text{HNO}_3\text{-HCl}$ (3:1) mixture at $105 \pm 5 \text{ }^\circ\text{C}$ for 2 h, cooled to room temperature ($25 \text{ }^\circ\text{C}$ – $27 \text{ }^\circ\text{C}$), and diluted with deionized water to 30 mL. The aqueous samples were first acidified by sub-boiling, quartz-distilled 6N HCl, and then digested using 2 mL HNO_3 (1 + 1) and 1 mL HCl (1 + 1) at $85 \text{ }^\circ\text{C}$ until the sample was evaporated to 20 mL. Concentrations of Cd and Pb were determined with an inductively coupled plasma mass spectrometer (ICP-MS, Model NEXION 350 X). Sample replicates and reagent blanks were included in each batch analyzed to ensure the quality of analysis.

2.3.2 Rice and soil samples

A whole plant in one pot was taken in each growing period, and the samples were rinsed thoroughly with deionized water to remove any attached soil/substrate particles before separating into shoots and roots. At the same time, surface soil (0–15 cm) samples in each pot were collected. The soil samples were dried at room temperature and homogenized using a mortar and pestle. One portion of the sample was sent through 2 mm sieves to measure the pH, while the other portion was passed through 0.145 mm sieves. Subsamples of grains, shoots, and roots were firstly dried in the oven at $105 \text{ }^\circ\text{C}$ for 30 min, then kept in the oven at $80 \text{ }^\circ\text{C}$ until they reached a constant weight, and then crushed into powders.

Soil samples were digested with a mixture of $\text{HNO}_3\text{-H}_2\text{O}_2\text{-HF}$ (6:3:3), and plant materials were digested with $\text{HNO}_3\text{-H}_2\text{O}_2$ (2:1). The digested solutions were diluted to 25 ml with 1% HNO_3 solution. All samples were stored in dark at $4 \text{ }^\circ\text{C}$ and were determined within one month. The concentrations of Cd and Pb in

solutions were determined using an inductively coupled plasma mass spectrometer (ICP-MS, Model NEXION 350 X).

To ensure the reliability and quality of the data, standard reference soil (GBW07387, Cd: 0.34 mg kg^{-1} ; Pb: 28 mg kg^{-1}), and rice (GBW(E) 100360, Cd: 0.22 mg kg^{-1} ; Pb: 0.049 mg kg^{-1}) samples from the Center of National Reference Materials of China were measured together with soil and plant samples. The recovery of the spiked standard for each element ranged between 90 and 110%, and the limits of detection were $0.01 \text{ } \mu\text{g mL}^{-1}$ and $0.01 \text{ } \mu\text{g mL}^{-1}$ for Cd and Pb, respectively.

2.4 Scanning electron microscopy (SEM) analysis

Small strips of rice leaf (0.5 cm^2) were trimmed between margin and mid-rib. The small strips were first kept in 2.5% glutaraldehyde solution and left overnight for fixation (prefixation), and then kept in osmium tetroxide for post fixation for 1 h. After washing twice with buffer solution for 15 minutes, these samples were passed through a series of acetone solutions (30%, 50%, 70%, 95%, and 100%) for dehydration. Samples were then dried in a critical point drier (CPD) with CO_2 as a carrier gas. SEM was carried out using a 30 keV, JEOL JSM-6490 LV scanning microscope with standard automated features such as auto focus/stigmator, auto gun, and auto contrast with multiple live image displays. The samples were coated with carbon using a high vacuum system for wet specimens, the upper surfaces of the leaf segments were studied, and micrographs were taken at various magnifications.

2.5 Foliar photosynthetic parameter analysis

The foliar photosynthetic parameters (transpiration rate (T_r), and stomatal conductance (gs) were recorded simultaneously at tillering and heading stage using a portable infrared gas analyzer (LI-COR6400XT). The infrared gas analyzer system (IRGAs) was equipped with a clamp-on leaf chamber of 6 cm^2 . According to Majer and Hideg (2012), IRGAs measure the reduction in transmission of infrared wavebands caused by the presence of CO_2 between the radiation source and a detector. The reduction in transmission is a function of the concentration of CO_2 . Thus, the measurements of gas exchange are based on the differences in CO_2 and H_2O in an air stream that is flowing into a leaf cuvette (reference cell) compared to the air stream flowing out of it (sample cell). The rate of CO_2 uptake ($\mu\text{mol (m}^2\cdot\text{sec)}^{-1}$) is used to assess the rate of photosynthetic carbon assimilation while the rate of water loss ($\text{mol H}_2\text{O (m}^2\cdot\text{sec)}^{-1}$) is used to assess the rate of transpiration and stomatal conductance. Gas exchange measurements were performed under irradiance at $425 \text{ (m}^2\cdot\text{sec)}^{-1}$ and at a temperature of $23 \pm 2^\circ\text{C}$. During measurement, humidity was fixed at 65%, and CO_2 concentration was maintained at a constant level of $380 \text{ } \mu\text{mol mol}^{-1}$ using a LI-6400-01 CO_2 injector with a high-pressure liquid CO_2 cartridge source.

2.6 Cd and Pb isotope ratio analysis

To identify the contributions of atmospheric deposition and the soil to the Cd and Pb accumulation in the edible of rice, the stable Cd and Pb isotope ratios $^{112}/^{111}\text{Cd}$, $^{114}/^{111}\text{Cd}$, $^{207}/^{206}\text{Pb}$, and $^{208}/^{206}\text{Pb}$ in the rice samples, atmospheric deposition and soil were determined using Inductively coupled plasma mass

spectrometer (ICAP-QC) (Thermo Fisher Scientific, Waltham, MA, USA). The operating conditions of the ICAP-QC were: RF power 1550W, atomizing chamber rate 40 r min⁻¹, atomizing chamber temperature 2.5 °C, atomized gas 1.0749 L min⁻¹, integration time 0.02 s. In consideration of elemental abundance and isobaric elements, ^{112/111}Cd, ^{114/111}Cd, ^{207/206}Pb, and ^{208/206}Pb were chosen for isotope ratio analysis. T1 isotope standard solution (SRM NIST 997) was used to correct the instrumental mass bias. Standard solutions of Cd (GBW 04622), and Pb (SRM NIST 981) were measured after five unknown samples to verify the results.

In the current study, we hypothesized that Cd and Pb in rice were mainly from atmospheric deposition and soil. On the basis of the measured abundance of ¹¹¹Cd, ¹¹²Cd, ¹¹³Cd, ²⁰⁶Pb, ²⁰⁷Pb, and ²⁰⁸Pb in the samples, the Cd and Pb contributions of the atmospheric PM and soil can be identified. Therefore, the contributions of soil and atmospheric deposition could be calculated as:

$$\delta_{\text{rice}} = f_A \delta_A + f_s \delta_s, (1)$$

$$1 = f_A + f_s, (2)$$

Each element had two isotope systems (^{112/111}Cd, ^{114/111}Cd, ^{207/206}Pb, and ^{208/206}Pb), and two potential sources. Where δ_A , δ_s are isotopic signatures of the two major sources (atmosphere and soil), and f_A , f_s are their relative contributions, respectively.

2.7 Statistical analysis

Data analysis was performed using SPSS 16.0, R Studio and Origin Pro 8.0. The datasets of photosynthetic parameters were subjected to a factorial analysis of variance (ANOVA) using a least significant difference (LSD) test at a significance level of $p < 0.05$ and $p < 0.01$. Using forest package, the random forest algorithm was used to calculate the interpretation intensity of the various factors with Cd content in rice grains.

3. Results

3.1 Concentration and distribution characteristics of Cd and Pb in atmospheric particulates

Figure 1 showed the changing trend of Cd and Pb concentrations in atmospheric pm (TSP and PM₁₀) during the whole growing period of rice in the monitored area. The Cd and Pb content in atmospheric PM varies greatly over time, and there was no obvious pattern. The concentrations of TSP-Cd and PM₁₀-Cd were 0.0016 (0.0001–0.0245) µg m⁻³ and 0.0015 (0.0001–0.0056) µg m⁻³, respectively. The concentration of TSP-Pb and PM₁₀-Pb was 0.1339 (0.0047–0.4298) µg m⁻³ and 0.1657 (0.0163–0.4814) µg m⁻³, respectively. The concentration of TSP-Cd and PM₁₀-Cd did not exceed the second-class standard of annual average concentration of Cd in ambient air stipulated in the Ambient Air Quality

Standard (GB3095-2012, $0.005 \mu\text{g m}^{-3}$). TSP-Pb and PM_{10} -Pb were also lower than the first-class standard of the Ambient Air Quality Standard (GB3095-2012, $0.5 \mu\text{g m}^{-3}$). The concentrations of TSP-Pb and PM_{10} -Pb in the atmosphere was higher from July to September, and then lower in October. Moreover, the concentration of Pb in PM_{10} was significantly higher than that in TSP, which indicated that Pb was mainly enriched in fine particles ($p < 0.05$).

3.2 Cd content in rice shoots and roots

The content of Cd accumulated in shoots and roots significantly higher in seriously Cd polluted (ZZ: 0.86 mg kg^{-1} of Cd in soil) soils than that in moderately Cd polluted soil (XT: 0.33 mg kg^{-1} of Cd in soil) (Fig. 2). An increasing trend was observed in shoot and root concentrations during growth. In XT soil, there was no significant difference in shoot Cd content between T1 (0.43 mg kg^{-1} , 1.50 mg kg^{-1}) and T2 (0.42 mg kg^{-1} , 1.54 mg kg^{-1}) at tillering stage and heading stage, while T3 (1.57 mg kg^{-1}) > T4 (1.13 mg kg^{-1}) at heading stage ($p < 0.05$). The trend of shoot Cd content in ripening stage was T1 (2.93 mg kg^{-1}) > T2 (2.06 mg kg^{-1}) > T4 (1.78 mg kg^{-1}) $\approx$ T3 (1.75 mg kg^{-1}), and both T2 and T3 was significantly higher than T4 ($p < 0.05$). The Cd content in roots of the three stages were T1 > T2 but with no significant difference; T3 (3.95 mg kg^{-1} , 5.58 mg kg^{-1}) > T4 (2.90 mg kg^{-1} , 3.95 mg kg^{-1}) and there was a significant difference between the heading and ripening stages ($p < 0.05$). In ZZ soil, a similar trend was observed in the shoots and roots. The shoot Cd content in the three periods were T1 ($0.42\text{--}7.57 \text{ mg kg}^{-1}$) > T2 ($0.24\text{--}4.10 \text{ mg kg}^{-1}$), T4 ($0.34\text{--}2.81 \text{ mg kg}^{-1}$) > T3 ($0.19\text{--}1.78 \text{ mg kg}^{-1}$) ($p < 0.05$). At ripening stage, the Cd content in shoots and roots under T1 treatment was the highest. Therefore, the results indicated that exposure to atmospheric deposition was beneficial to the accumulation of Cd in rice. In addition, T3 was larger than T4 in shoots and roots in XT soils, which was different from the result that T4 > T3 in ZZ soils. This indicates that rice is highly affected by transpiration rate in ZZ soils, and roots absorb more Cd in soils with high Cd content.

3.3 Pb content in rice shoots and roots

There was no significant difference in Pb content in soils between XT (61.9 mg kg^{-1}), and ZZ (65.2 mg kg^{-1}). The content of Pb in shoots and roots of rice in the two kinds of soils both increased with the growing of rice (Fig. 3). In XT soil, the content of Pb in shoots showed no significant difference between all treatments at tillering stage, and T4 (4.18 mg kg^{-1}) was significantly higher than T3 (3.14 mg kg^{-1}) at heading stage ($p < 0.05$). At the ripening stage, the content in shoots decreased in the order of T1 (11.61 mg kg^{-1}) > T3 (7.88 mg kg^{-1}) > T2 (5.86 mg kg^{-1}) > T4 (4.87 mg kg^{-1}). T1 had the highest Pb content in shoots, and T3 was significantly higher than T2 and T4 ($p < 0.05$). For roots, T3 (96.77 mg kg^{-1} , 95.62 mg kg^{-1}) > T4 (76.60 mg kg^{-1} , 76.29 mg kg^{-1}) ($p < 0.05$) at tillering stage and ripening stage. At heading stage, the results were consistent with shoots. In ZZ soils, Pb content in the shoots and roots at tillering stage was T1 > T2 ($P < 0.05$), and T4 > T3, the trend of Pb content in shoots and roots at heading stage were T1 (3.20 mg kg^{-1} , 45.45 mg kg^{-1}) > T4 (2.62 mg kg^{-1} , 43.84 mg kg^{-1}) > T3 (1.90 mg kg^{-1} , 37.71 mg kg^{-1}) > T2 (1.72 mg kg^{-1} , 31.13 mg kg^{-1}). While that at ripening stage was T1 (11.08 mg kg^{-1}) > T2 (9.39

mg kg⁻¹) > T4 (4.62 mg kg⁻¹) > T3 (3.35 mg kg⁻¹) in shoots, and T1 and T2 were significantly higher than T3 and T4, but there was no significant difference between T1 and T2, T3 and T4. While for roots, T1 (166.86 mg kg⁻¹) was the highest, followed by T2 (113.88 mg kg⁻¹) and T4 (117.27 mg kg⁻¹), which were all significantly higher than T3 (85.80 mg kg⁻¹).

3.4 Cadmium and lead contents in rice grains

The contents of Cd and Pb in rice grains were significantly different among the four exposure conditions (Fig. 4a, b). Cd content in grain range from 0.20 mg kg⁻¹ to 1.48 mg kg⁻¹, and from 0.96 mg kg⁻¹ to 6.52 mg kg⁻¹ in XT and ZZ soil, respectively. For Pb, these contents range from 0.44 mg kg⁻¹ to 1.48 mg kg⁻¹ for XT soil, and from 0.37 mg kg⁻¹ to 2.18 mg kg⁻¹ for ZZ soil. Compared to the National Food Safety Standards (0.2 mg kg⁻¹ for Cd, 0.2 mg kg⁻¹ for Pb), the contents exceeded the standard values and indicated a particularly serious contamination. In general, Cd and Pb contents in rice grains under outdoor all-day exposure treatment (T1) was the highest. Additionally, for grain Cd content in XT soil, showed the trend of T1 (1.48 mg kg⁻¹) > T3 (0.64 mg kg⁻¹) ≈ T2 (0.60 mg kg⁻¹) > T4 (0.20 mg kg⁻¹). While in ZZ soil were T1 (6.52 mg kg⁻¹) > T2 (1.73 mg kg⁻¹) > T4 (1.33 mg kg⁻¹) > T3 (0.96 mg kg⁻¹), and there was no significant difference among the three treatments (T2, T3, T4). For Pb, grain content in XT soil were T1 (1.48 mg kg⁻¹) > T2 (0.58 mg kg⁻¹) > T4 (0.52 mg kg⁻¹) > T3 (0.44 mg kg⁻¹), and there was no significant difference among T2, T3, and T4. In ZZ soil, grain content in T1 (2.18 mg kg⁻¹) was higher than that of T2 (1.57 mg kg⁻¹), followed by T3 (0.52 mg kg⁻¹), and a little in T4 (0.43 mg kg⁻¹). Both all day exposure treatment (T1, T2) were significantly higher than daytime exposure (T3) and night exposure treatment (T4).

3.5 Accumulation of cadmium and lead in rice

Plant growth stages exerted significant effects on Pb and Cd accumulation in each rice plant, and the accumulation of Cd increased with higher soil Cd concentration (Fig. 5). In general, the maximum Cd and Pb accumulation in rice occurred at ripening stage. In both XT and ZZ soils, the accumulation of Cd of each plant in T1 (257.57 ug plant⁻¹ and 507.78 ug plant⁻¹) was higher than that in T2 (146.71 ug plant⁻¹ and 254.43 ug plant⁻¹), T3 (197.76 ug plant⁻¹ and 220.02 ug plant⁻¹) or T4 (160.49 ug plant⁻¹ and 302.58 ug plant⁻¹), while T1, T2 and T3 showed no significant difference. As for Pb, total Cd accumulation in XT soils was T1 (4049.50 ug plant⁻¹) > T3 (2349.73 ug plant⁻¹) > T2 (1946.09 ug plant⁻¹) > T4 (1913.97 ug plant⁻¹). For ZZ soils, it showed T1 (3523.29 ug plant⁻¹) > T3 (2726.49 ug plant⁻¹) > T4 (2527.45 ug plant⁻¹) > T2 (2040.21 ug plant⁻¹). Moreover, the accumulation of Cd in shoots was higher than in roots in both soils, while the opposite was true for Pb. More specifically, the amount of Pb accumulated in root was several times higher than in the shoots. However, the accumulation of Cd in roots at ripening stage remained at a relatively low level, although an increasing trend was observed for shoots at the same time. The phenomenon of Cd accumulation in root decreased at ripening stage, probably due to transportation to shoot, while Pb mainly accumulated in root probably due to poor

mobility. The main reason for these difference more likely resulted from atmospheric-soil-root transfer as well as the direct transfer of contaminants from the atmosphere to foliar uptake.

3.6 Cadmium and lead isotope ratio analysis

To find out the source of heavy metal accumulated in rice grains, $^{114}/^{111}\text{Cd}$ ratios were plotted against values of $^{112}/^{111}\text{Cd}$ ratio, and $^{208}/^{206}\text{Pb}$ against $^{208}/^{206}\text{Pb}$ for the possible pollutant sources (Fig. 6). The $^{112}\text{Cd}/^{111}\text{Cd}$ and $^{114}\text{Cd}/^{111}\text{Cd}$ ratios in grains (ZZ) were located at similar distance from atmospheric PM and soil, indicating that they were all contributors to grains Cd. For XT soils, the closer points were mainly atmospheric deposition samples, which resulting that atmospheric PM was the most likely source of grain Cd accumulation. In the case of Pb, the cluster of soil was close to the grains (XT), indicating that the accumulation of Pb in rice grains was greatly affected by soil content. For ZZ soils, both soil and atmospheric deposition substantially contributed to grain Pb accumulation. The contribution of atmospheric deposition and soil for Cd was 40.32–86.78% (63.55%), 13.22–59.68% (36.45%) in XT soils, and 4.2–33.56% (18.01%), 66.44–95.8% (81.99%) in ZZ soils, respectively. For Pb, atmospheric deposition and soil accounted for 18.72–36.65% (27.69%) and 63.35–81.28% (72.32%) in XT soils, and 29.25–56.45% (41.13%) and 43.55–70.75% (58.87%) in ZZ soils, respectively, which indicated that atmospheric deposition was the major source.

3.7 Stomatal characteristics of rice leaves

The stomatal aperture causes PM to easily enter the plants when stomata open in the presence of light (Uzu et al. 2010; Shahid et al. 2017). Primarily, stomata size plays a critical role in jamming fine particles (Mansfield and Majernik 1970). The stomata dimensions of rice leaves vary in the tillering, heading, and ripening stages. According to previous work, there is basically no difference in the stomatal distribution of the adaxial and abaxial epidermis of rice leaves; thus, in this study we only observed the front side of rice in each period. The width of stomata in the tillering, heading, and ripening stages were 1.33 μm , 2.16 μm , and 2.84 μm (Fig. 7). The length of stomata and guard cells was greater than 10 μm at tillering and heading stage, and the pore diameter and opening of stomata increased with plant growth, reaching 20 μm at ripening stage, indicating that it is more possible for atmospheric fine particles (PM_{10} and $\text{PM}_{2.5}$) to enter rice leaves directly when the stomata open at ripening stage.

Stomatal conductance (g_s) is a good indicator of the plant response to environmental changes (Gao et al. 2021). Fig. S1 shows that g_s of rice leaves at heading stage is decreased compared with that at tillering stage. At tillering stage, the g_s values of the leaves were 0.15–0.21 $\text{mmol mm}^{-2} \text{s}^{-1}$ in XT and 0.30–0.35 $\text{mmol mm}^{-2} \text{s}^{-1}$ in ZZ; but there was no difference between treatments. At heading stage, it was found that the g_s values of T3 (0.12 $\text{mmol mm}^{-2} \text{s}^{-1}$) were significantly higher than that of T4 (0.04 $\text{mmol mm}^{-2} \text{s}^{-1}$) in ZZ ($p < 0.05$).

4. Discussion

4.1 Characteristics of atmospheric fallout and its effects

Many studies have proved that Pb is a marker of traffic emissions since the process of fuels combustion as we as engines and catalysts loss would emit Pb to the environment (Guan et al. 2018; Li et al. 2013). However, Cd was more related to coal combustion and electronics industries (Guan et al. 2018; Huang et al. 2018). The experimental bases was located at the campus of Hunan Agricultural University, which was far away from any typical heavy metal emitters such as factories or mines. Compared with the limits stated in the Environmental Air Quality Standard (GB3095-2012), the average values of Cd and Pb in PMs (PM₁₀, TSP) in the study area were both lower than the secondary and primary class standards, indicating light pollution of atmospheric deposition in this area. Overall, the content of Pb in dry depositions was higher than that of Cd. Moreover, the contents of Cd and Pb in PM₁₀ were higher than those in TSP. Canepari et al. (2008) monitored the distribution of heavy metals in coarse particles (> 2.5 µm), and fine particles (< 2.5 µm) in Rome, Italy, and found that most heavy metals that came from natural sources (such as calcium, barium, iron, magnesium, and manganese) were located in coarse particles, while other elements, like Cd and Pb, which mainly came from anthropogenic emissions (such as vehicle emissions, non-exhaust sources, and railway emissions) were found adsorbed on fine particles. Among them, more than 50% of Cd and Pb were adsorbed on particles with a size < 1 µm. Louie et al. (2005) concluded the same conclusion with long time monitoring of PM_{2.5} and PM₁₀ in Hong Kong.

A previous study also indicated that heavy metals present in precipitation were more bioavailable (Sharma et al. 2008), which suggests that heavy metal deposition in the present study may cause a significant impact on the surrounding agro-ecological system even though the total amount was not excessive. In this study, a random forest analysis revealed that the duration of exposure of rice leaves to atmospheric deposition was one of the main factors affecting Cd and Pb content in rice grains in both XT and ZZ soils. On the contrary, the Cd and Pb contents in atmospheric deposition was not significant (Fig. 8). Therefore, the content of Cd and Pb in T2 was significantly higher than that in T3 or T4. More precisely, In XT, the duration of foliar exposure to atmospheric deposition was the main factor affecting Cd accumulation in grains. Different from ZZ, Cd content in soil is the key factor. However, for Pb, in both XT and ZZ soils, rice grains were mainly affected by Pb content in shoots and duration of leaves exposure, but not by soil content. Therefore, Pb in grains was mainly from atmospheric deposition absorbed by leaves.

Besides, metals from the atmosphere can settle into the topsoil, changing soil conditions and these metals can also be absorbed by rice roots. Therefore, in the current study, the content of Cd and Pb accumulated in T1 (all day exposure without geotextile membrane) was higher than that in T2 (all day exposure with geotextile membrane). Feng et al. (2019) used a retrospective model to predict the long-term accumulation of heavy metals in paddy soil for over 30 years and showed that long-term atmospheric deposition significantly increased heavy metal accumulation on the surface layer of paddy soils ($p < 0.05$). Liu et al. (2019) conducted ectopic soil experiments in three areas with different atmospheric deposition pollution gradients and reported that the content of Cd in soil increased significantly from $1.56 \pm 0.07 \text{ mg kg}^{-1}$ to $1.83 \pm 0.08 \text{ mg kg}^{-1}$ after exposure to high deposition inputs for

over six months, indicating that the accumulation of heavy metals was significantly higher in areas with serious deposition.

4.2 Mechanism of foliar uptake and translocation

In recent years, several studies have been carried out on the uptake of heavy metals in atmospheric precipitated particles through leaves, and the potential mechanisms have been discussed. Gajbhuye et al. (2016) suggested that heavy metals may enter leaves in two ways: (1) through the stomatal openings in the form of nanoparticles; and (2) through the hydrophilic pathway by diffusing through the aqueous pores of the stomata and cuticle. Therefore, to explore the accumulation mechanism of foliar uptake, healthy leaf samples were collected at tillering stage, heading stage, and ripening stage for SEM analysis to observe the morphological characteristics of the surface of rice leaves. Gao et al. (2021) found that $PM_{2.5}$ -Pb can be transferred from atmospheric fallout to leaves via the stomata, and larger stomata apertures were responsible for high Pb accumulation in the leaves. Gajbhuye et al. (2016) found large amounts of PM deposited on the leaf epidermis, covering the stomata and their openings on the front of leaves, and the open stomata and guard cells were completely covered by fine particles ($PM < 2 \mu m$). There were pores in stomata and the stratum corneum, through which PM-bound metals can penetrate to leaf tissues (Schreck et al. 2012). Tomasevic and Anicic (2010) analyzed the surface of plant leaves by SEM-energy dispersive X-ray (SEM-EDX) and found that Pb adsorbed by small PM ($< 2.5 \mu m$) could be captured on the leaf surface. Sawidis et al. (2012) studied the structure and density of stomata and glandular hairs on leaf surface (front and opposite) of different plants and found that foliar surface characteristics were crucial factors that determined capture efficiency. Therefore, the observation results based on SEM have provided strong evidence that PM particles and absorbed toxic heavy metals (Cd and Pb) can be effectively transferred through the stomata on leaves (Fig. 7).

Stomata is one of the important ways for leaves to absorb heavy metals and the only way to absorb a large amount of low solubility solid PM. Therefore, the openness of stomata greatly affects the efficiency of metal absorption (Kaiser et al. 2015). Moreover, the stomatal width-to-length ratio indicates the degree of stomatal opening (Bu et al. 2014). In general, stomata open in light and close in darkness. In addition, temperature has an effect on stomata, increasing the temperature can improve stomata opening, and stomata can reach the maximum opening at 30–50°C. In this study, except for heading stage, Cd and Pb content of rice plants exposed during the day (T3) was higher than that exposed at night (T4) in XT. This is due to more light and higher temperatures during the day than at night. Therefore, stomatal variation could be a key factor in regulating foliar Cd and Pb uptake and translocation in rice.

Temperature not only changes stomatal opening, previous studies have proved that increase in temperature effectively accelerates element migration and transport in rice from roots to shoots. In this study, T3 and T4 were exposed during the day and at night, respectively, and placed in a transparent enclosure for the rest of the time. Tested rice was grown in the study area where the average temperature was 34 °C and the highest temperature could reach 40 °C, and the temperature was as high as 50 °C inside the device. Fig. S1 shows that stomatal conductance (GS) and transpiration rate (Trmmol) of rice leaves at heading stage is decreased compared with that at tillering stage. It was found that the gs values

of T3 were significantly higher than that of T4 at heading stage in ZZ ($p < 0.05$) and there was no difference in XT. On the contrary, the transpiration rate of T4 was higher than that of T3 ($p < 0.05$). It indicated that as temperature rises, to prevent water loss, some stomata of rice leaves are closed, and stomatal conductance also decreases. Meanwhile, high temperature in T4 accelerates the transpiration rate and promotes the absorption and transport capacity of the root system through the xylem. Therefore, the Cd and Pb content of shoots and roots was $T4 > T3$, especially in ZZ soil with higher Cd content.

5. Conclusions

To investigate and quantify the influence of atmospheric deposition on Cd and Pb accumulation in rice grains, a rice pot experiment of three exposure treatments (all day exposure, daytime exposure, and night exposure) in both seriously and moderately polluted soils was conducted. Although the total Cd and Pb fluxes in the study area were lower than those of the National Environmental Quality Standard, atmospheric deposition was still an important source of Cd and Pb accumulation in rice. The present experiment proved that long-term exposure to atmospheric deposition could significantly increase the content of Cd and Pb in rice shoots and grains. The results of the isotope ratio analysis proved that the contribution of atmospheric deposition in rice grains for Cd was quantified as 63.55% and 18.01%, and was 27.69% and 41.13% for Pb in XT(moderately) and ZZ soils (severe), respectively. These results indicated that the influence of atmospheric deposition on crop Cd and Pb accumulation should be paid more attention, and that foliar control management can be an effective way to maintain food safety.

Declarations

Declaration of Internet interests

The authors declare that they have no conflict of interest. This manuscript has not been published or presented elsewhere in part or in entirety and is not under consideration by another journal. We have read and understood your journal's policies, and we believe that neither the manuscript nor the study violates any of these. There are no conflicts of interest to declare.

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References

1. Baycu G, Gevrek-Kürüm N, Moustaka J, Csatári I, Rognes SE, Moustakas M (2016) Cadmium-zinc accumulation and photosystem II responses of *Nocca caerulea* to Cd and Zn exposure. *Environ Sci Pollut Res* 24(3):2840–2850
2. Bi XY, Feng XB, Yang YG, Li XD, Shin GPY, Li FL, Qiu GL, Li GH, Liu TZ, Fu ZY (2009) Allocation and source attribution of lead and cadmium in maize (*Zea mays* L.) impacted by smelting emissions. *Environ Pollut* 157(3):834–839
3. Bohdálková L, Novák M, Krachler M, Míková J, Přechová E (2020) Cadmium contents of vertically and horizontally deposited winter precipitation in central Europe: spatial distribution and long-term trends. *Environ Pollut* 265:114949
4. Canepari S, Perrino C, Olivieri F et al (2008) Characterisation of the traffic sources of PM through size-segregated sampling, sequential leaching and ICP analysis. *Atmos Environ* 42(35):8161–8175
5. Cao XY, Tan CY, Wu LH et al (2020) Atmospheric deposition of cadmium in an urbanized region and the effect of simulated wet precipitation on the uptake performance of rice. *Sci Total Environ* 700:134513
6. De Temmerman L, Ruttens A, Waegeneers N (2012) Impact of atmospheric deposition of As, Cd and Pb on their concentration in carrot and celeriac. *Environ Pollut* 166:187–195
7. Du Y, Hu XF, Wu XH et al (2013) Effects of mining activities on Cd pollution to the paddy soils and rice grain in Hunan province, Central South China. *Environ Monit Assess* 185(12):9843–9856
8. Feng JF, Wang YX, Zhao J et al (2011) Source attributions of heavy metals in rice plant along highway in Eastern China. *J Environ Sci* 23(7):1158–1164
9. Feng WL, Guo ZH, Peng C et al (2018) Modelling mass balance of cadmium in paddy soils under long term control scenarios. *Environ Science: Processes & Impacts* 20:1158–1166
10. Feng WL, Guo ZH, Xiao XY et al (2019) Atmospheric deposition as a source of cadmium and lead to soil-rice system and associated risk assessment. *Ecotoxicol Environ Saf* 180:160–167
11. Gao PP, Xue PY, Dong JW et al (2021) Contribution of PM_{2.5}-Pb in atmospheric fallout to Pb accumulation in Chinese cabbage leaves via stomata. *J Hazard Mater* 407:124356
12. Guan QY, Wang FF, Xu CQ et al (2018) : Source apportionment of heavy metals in agricultural soil based on PMF: a case study in Hexi Corridor, northwest China, *Chemosphere*, 193: 189–197
13. Hou Q, Yang Z, Ji J et al (2014) Annual net input fluxes of heavy metals of the agro-ecosystem in the Yangtze River delta, China. *J Geochem Explor* 139(1):68–84
14. Hu X, Zhang Y, Luo J et al (2011) Accumulation and quantitative estimates of airborne lead for a wild plant (*Aster subulatus*). *Chemosphere* 82(10):1351–1357

15. Huang Y, Deng MH, Wu SF et al (2018) A modified receptor model for source apportionment of heavy metal pollution in soil. *J Hazard Mater* 354:161–169
16. Huang Y, Li TQ, Wu CX et al (2015) An integrated approach to assess heavy metal source apportionment in peri-urban agricultural soils. *J Hazard Mater* 299:540–549
17. Jaekels JM, Bae MS, Schauer JJ (2007) Positive matrix factorization (PMF) analysis of molecular marker measurements to quantify the sources of organic aerosols. *Environ Technol* 41(16):5763–5769
18. Keller C, Rizwan BM, Davidian C et al (2014) Effect of silicon on wheat seedlings (*Triticum turgidum* L.) grown in hydroponics and exposed to 0 to 30 μm Cu. *Planta* 241(4):847–860
19. Lee PK, Choi BY, Kang MJ (2015) Assessment of mobility and bio-availability of heavy metals in dry depositions of Asian dust and implications for environmental risk. *Chemosphere* 119:1411–1421
20. Li F, Huang J, Zeng G et al (2013) Spatial risk assessment and sources identification of heavy metals in surface sediments from Dongting Lake, Middle China. *J Geochem Explor* 132:75–83
21. Li H, Luo N, Li Y et al (2017) Cadmium in rice: Transport mechanisms, influencing factors, and minimizing measures. *Environ Pollut* 224:622–630
22. Liu G, Yu Y, Jing H et al (2014) An ecological risk assessment of heavy metal pollution of the agricultural ecosystem near a lead-acid battery factory. *Ecol Ind* 47:210–218
23. Liu HL, Zhou J, Li M et al (2019) Study of the bioavailability of heavy metals from atmospheric deposition on the soil-pakchoi (*Brassica chinensis* L.) system. *J Hazard Mater* 362:9–12
24. Liu J, Wang D, Song B et al (2019) Source apportionment of Pb in a rice-soil system using field monitoring and isotope composition analysis. *J Geochem Explor* 204:83–89
25. Liu ZP, Zhang QF, Han TQ et al (2016) Heavy metal pollution in a soil-rice system in the Yangtze river region of China. *Int J Environ Res Public Health* 13(1):1–16
26. Louie P, Watson JG, Chow JC et al (2005) Seasonal characteristics and regional transport of $\text{PM}_{2.5}$ in Hong Kong. *Atmos Environ* 39(9):1695–1710
27. Luo L, Ma YB, Zhang SZ et al (2009) An inventory of trace element inputs to agricultural soils in China. *J Environ Manage* 90:2524–2530
28. Majer P, Hideg éva (2012) Existing antioxidant levels are more important in acclimation to supplemental UV-B irradiation than inducible ones: Studies with high light pretreated tobacco leaves. *Emirates J Food Agric* 24(6):598–606
29. Mekonnen DW, Flügge UI, Ludewig F (2016) Gamma-aminobutyric acid depletion affects stomata closure and drought tolerance of *Arabidopsis thaliana*. *Plant Sci* 245:25–34
30. Melotto M, Zhang L, Oblessuc PR et al (2017) Stomatal Defense a Decade Later. *Plant Physiol* 174:561–571
31. Sapkota B, Cioppa MT (2012) Using magnetic and chemical measurements to detect atmospherically-derived metal pollution in artificial soils and metal uptake in plants. *Environ Pollut* 170:131–144

32. Sawidis T, Krystallidis P, Veros D et al (2012) A study of air pollution with heavy metals in Athens city and Attica basin using evergreen trees as biological indicators. *Biol Trace Elem Res* 148:396–408
33. Schreck E, Dappe V, Sarret G et al (2014) Foliar or root exposures to smelter particles: Consequences for lead compartmentalization and speciation in plant leaves. *Sci Total Environ* 476–477:667–676
34. Schreck E, Foucault Y, Sarret G et al (2012) Metal and metalloid foliar uptake by various plant species exposed to atmospheric industrial fallout: Mechanisms involved for lead. *Sci Total Environ* 427–428:253–262
35. Shao X, Cheng H, Li Q et al (2013) Anthropogenic atmospheric emissions of cadmium in china. *Atmos Environ* 79(11):155–160
36. Sharma RK, Agrawal M, Marshall FM (2008) Heavy metal (Cu, Zn, Cd and Pb) contamination of vegetables in urban india: a case study in varanasi. *Environ Pollut* 154(2):254–263
37. Tang WZ, Shan BQ, Zhang H et al (2010) Heavy metal sources and associated risk in response to agricultural intensification in the estuarine sediments of Chaohu Lake Valley, East China. *J Hazard Mater* 176(1–3):945–951
38. Temmerman LD, Ruttens A, Waegeneers N (2012) Impact of atmospheric deposition of As, Cd and Pb on their concentration in carrot and celeriac. *Environ Pollut* 166:187–195
39. Tian HZ, Zhu CY, Gao JJ et al (2015) Quantitative assessment of atmospheric emissions of toxic heavy metals from anthropogenic sources in china: historical trend, spatial distribution, uncertainties, and control policies. *Atmos Chem Phys* 15(8):12107–12166
40. Tomasevic M, Anicic M (2010) Trace element content in urban tree leaves and SEM-EDAX characterization of deposited particles. *Facta Universitatis-series Physics Chemistry and Technology* 8:1–13
41. Wong C, Li XD (2004) Pb contamination and isotopic composition of urban soils in Hong Kong. *Sci Total Environ* 319(1–3):185–195
42. Xiong DL, Douthe C, Flexas J (2018) Differential coordination of stomatal conductance, mesophyll conductance, and leaf hydraulic conductance in response to changing light across species. *Plant Cell & Environment* 41:436–450
43. Yin J, Harrison R, Chen Q et al (2010) Source apportionment of fine particles at urban background and rural sites in the UK atmosphere. *Atmos Environ* 44(6):841–851
44. Zhang YL, Zhang SX, Zhu FP et al (2018) Atmospheric heavy metal deposition in agro-ecosystems in China. *Environ Sci Pollut Res* 25:5822–5831
45. Zhou J, Du BY, Liu HL et al (2020) The bioavailability and contribution of the newly deposited heavy metals (copper and lead) from atmosphere to rice (*oryza sativa* L.). *J Hazard Mater* 384:121285

Figures

Figure 1

Characteristics of atmospheric fallout during entire growth period of rice. Variations of Cd concentration in PM₁₀ and TSP (a); Variations of Pb concentration in PM₁₀ and TSP (b).

Figure 2

Contents of Cd in rice shoots and roots for different exposure conditions (T1, T2, T3, and T4) in XT (a, c) and ZZ (b, d) soils at three different growing stages. (data are means $\pm$ SE, $n=4$). Lowercase letters indicate significant differences ($p < 0.05$) in Cd contents between the four exposure treatments in each growth stage, and asterisks (*) indicate a significant difference ($p < 0.05$) in the Cd contents of the treatments in the same growth stage.

Figure 3

The contents of Pb in rice shoots and roots for different exposure conditions (T1, T2, T3 and T4) in XT (a,c) and ZZ (b,d) soils at three different growing stages. (data are means $\pm$ SE, $n=4$). Lowercase letters indicate significant differences ($p < 0.05$) in Pb contents between the four exposure treatments in each growth stage, and asterisks (*) indicates a significant difference ($p < 0.05$) in the Pb contents of the treatments in the same growth stage.

Figure 4

Contents of Cd and Pb in rice grain (data are means $\pm$ SE, $n=4$). Lowercase letters indicate significant differences ($p < 0.05$) in Pb contents among the four exposure treatments in each growth stage.

Figure 5

Amount of Cd (a) and Pb (b) accumulated in each single rice plant (leaves and roots in tillering and heading stage; leaves, roots, and grains in ripening stage) throughout the entire growth (data are means, $n=4$).

Figure 6

Biplots of $^{112}\text{Cd}/^{111}\text{Cd}$ ratio vs. $^{114}\text{Cd}/^{111}\text{Cd}$ and $^{207}\text{Pb}/^{206}\text{Pb}$ ratio vs. $^{208}\text{Pb}/^{206}\text{Pb}$ for rice grains, roots, shoots, soil and atmospheric deposition (air) in XT and ZZ soils.

Figure 7

Scanning electron micrographs of leaves of rice in tillering stage, heading stage and ripening stage (a, b, c), the size of stomata were shown with arrows.

Figure 8

Random forest analysis to identify the main predictors of factors affecting Cd concentrations in grains. Influencing factors include atmospheric deposition duration, Cd concentrations in air, soil, roots and shoots. Red represents Cd and green represents Pb. And a, c are XT soils; b, d are ZZ soils. The importance of variables in the figure is measured by the value of "Percentage of increase of mean square error" (increase in MSE(%)) in the random forest.

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