

1 **The application of biochar and oyster shell reduced cadmium uptake by crops**
2 **and modified soil fertility and enzyme activities in contaminated soil**

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Abstract

Soil pollution with cadmium (Cd) has been threatening the human health. In this study, we investigated the possibility of applying biochar and oyster shell to reduce Cd uptake by crops and modify soil quality. A field study based on the rice-oilseed rape rotation was done and the treatments were comprised of without amendments (PA0), 15000 kg/ha biochar (PA1), 15000 kg/ha oyster shell (PA2), and 7500 kg/ha biochar and 7500 kg/ha oyster shell (PA3). Results revealed that both oyster shell and biochar reduced the HOAc-extractable Cd in soil. Compared to PA0, the HOAc-extractable Cd in the PA1, PA2 and PA3 treatments was reduced by 4.76 - 20.79%, 17.86 - 38.61% and 5.95 - 10.89%, respectively. The cooperative application of biochar and oyster shell reduced the Cd accumulation in brown rice and oilseed by 29.67% and 19.74%, respectively, compared to control, and thus decreased the Hazard Quotient (HQ) by the consumption of brown rice and oilseed. The addition of biochar slightly increased soil organic matter. In addition, the available P in the PA2 and PA3 treatments was significantly ($p < 0.05$) increased by 200.96 - 295.92% and 187.46 - 280.04% compared to control. Moreover, the cooperative application of biochar and oyster shell enhanced the activities of urease, catalase, and β -galactosidase by 139.44 - 147.56%, 10.71 - 34.31% and 82.08 - 244.38%, respectively. These results demonstrated that the utilization of biochar and oyster shell might be an effective pathway to reduce Cd uptake by crops and improve soil fertility and enzyme activities.

Keywords: Biochar; Oyster shell; Rice-oilseed rape rotation; In-situ remediation;

1. Introduction

Cadmium (Cd) contamination of agricultural soils is a worldwide environmental problem, which has been seriously threatening to human health (Yang et al., 2021). The excessive intake of Cd by food chain can cause serious damages to bones, thyroid, and kidneys (Ma et al., 2021b). According to the latest national survey on the status of soil environmental quality in China, Cd has been ranked as the highest contaminants (7%) among all heavy metals (Mou et al., 2020). In southwest China, the intensive industrialization is the resource of the farmlands being contaminated with Cd (Chen et al., 2018). Soil acidification also aggravated the bioavailability and solubility of Cd, thus enhancing Cd uptake by crops (Feng et al., 2020). Therefore, the development of cost-effective and eco-friendly remediation technologies is crucial for food safety and soil quality.

In recent years, the in-situ immobilization technology through the application of soil amendments has raised wide attentions in the remediation of Cd contaminated farmlands, which could reduce the Cd uptake by plants without delaying agricultural production (Palansooriya et al., 2020). Biochar derived from bio-wastes has been widely recommended as a soil amendment (Zong et al., 2021). Amounts of nutrients (such as C, N, P, K, and Mg) in biochar can improve soil fertility and promote plant growth (Lu et al., 2015). Moreover, biochar has a large surface area and plenty of functional groups (such as -COOH, C-O and C=O), which are reactive to immobilize heavy metals, including Cd, lead (Pb), and nickel (Ni) (Wang et al., 2021). However,

the high price of biochar limits its large application. In addition, the application of biochar can not effectively change soil pH in acidic fields, thus it can not effectively reduce the bioavailability of Cd in soil (Liu et al., 2018). Oyster shell is a low-cost and largely available bio-waste product from oyster farming (Li et al., 2020). Oyster shell is a promising slow-release alkaloid, which has the outstanding effects on pH adjustment and Cd immobilization in soil (Lee et al., 2008). In this sense, we think that the joint use of biochar and oyster shell might be a low-cost and effective pathway to decrease Cd uptake by crops and improve soil quality in acidic fields.

Rice and oilseed rape are the main food and economic crops in southwest China, and the rice-oilseed rape rotation is the dominant production model (Liu et al., 2014). Previous studies mainly focused on the effects of amendments on reducing the Cd uptake by rice (Tang et al., 2020; Yin et al., 2022), while the remediation efficiency of amendments under the rice-oilseed rape rotation is little known. Based on the above opinions, a field experiment under the rice-oilseed rape rotation was designed: (1) to investigate the effects of biochar and oyster shell on Cd immobilization in soil; (2) to evaluate the effects of biochar and oyster shell on decreasing human health risk of consuming contaminated crops; (3) to reveal the effects of biochar and oyster shell on soil fertility and enzyme activities.

2. Material and methods

2.1. Experimental site and soil properties

A field trial was conducted during 2019-2020 in a rice-oilseed rape rotation cultivated site where the soil was moderately contaminated with Cd. The field site

was located in a dominant agricultural cultivation region round industrial parks in Chengdu plain, Sichuan province, China (104°18'N, 31°81'E). This region belongs to a subtropical monsoon humid climate with an average temperature of 16.1 °C and annual rainfall of about 1000 mm. The main properties of the topsoil (0 - 20 cm) collected from the site in 2019 and 2020 were shown in Table S1.

2.2. Characteristics of experimental materials

Biochar was purchased from Zhenjiang Zedi agricultural and biological Co., Ltd., which was produced from rice straw in a reactor with N₂ and 500 °C for about 4 h. Oyster shell was purchased from Fujian Mata Co., Ltd (< 0.3 mm mesh). The main properties of biochar and oyster shell were presented in Table S1. The main composition of oyster shell was shown in Table S2. The surface structures of biochar and oyster shell were analyzed by Scanning Electron Microscope (SEM, JSM-7500F). The functional groups of biochar and oyster shell were measured by Fourier Transform Infrared Spectra (Nicolet 6700). The seeds of rice “Yixiang 2115” and seeds of oilseed rape “Yiyou 15” were obtained from Rice Research Institute, Sichuan Academy of Agricultural Science.

2.3. Experimental setup

The field experiment was conducted during 2019 - 2020. The treatments were PA0 (Control), PA1 (15000 kg/ha biochar), PA2 (15000 kg/ha oyster shell) and PA4 (7500 kg/ha biochar and 7500 kg/ha oyster shell). The concentrations of biochar and oyster shell used in this study were referred to the previous report (Ameloot et al., 2014). Each experimental plot was 56 m² (7 x 8 m) and arranged in a randomized

design with three replicates. Before rice planting, the amendments were sufficiently mixed with topsoil. After the harvest of rice, the oilseed rape was planted following the conventional tillage pattern without extra amendments.

2.4. Plant analysis

The rice grain and oilseed samples were dried and ground to powder. Then, 0.2 g samples were digested with HNO₃:HClO₄:HF in a mixture of 5:4:3 (v/v) and the mixture was then diluted into 10 mL with 1% HNO₃ (Wu et al., 2019b). The Cd concentrations in the mixture were determined by atomic absorption spectroscopy (AAS; VARIAN, SpecterAA-220Fs).

2.5. Soil analysis

Soil pH was determined by a pH meter (METTLER-S220) with a soil/water ratio of 5 g/25 mL. The bioavailable Cd of soil was measured by the TCLP method (Xu et al., 2020). Briefly, 2 g of soil sample was mixed with 40 mL of 0.11 M acetic acid (HOAc) and shaken for 16 h at 25 °C, 150 rpm. The mixture was centrifuged for 5 min at 8000 rpm and then the supernatant was collected to determine Cd concentrations by AAS. Available P, available K, available N were measured according to the method described by Wu et al. (2018). Soil TOC and OM were determined by the method described by Walz et al. (2017).

In addition, activities of soil enzyme were analyzed to reflect the biological quality in this study. Dehydrogenase activity was evaluated by the production of triphenylformazan (TPF) at OD_{492nm} and expressed as µg TPF/g soil/24 h (Benefield et al., 1977). Acid phosphate activity was assayed by the *p*-nitrophenol (pNP) release at

OD_{400nm} and expressed as µg pNP/g soil/24 h (Van Aarle and Plassard, 2010). Urease activity was determined by the NH₄-complex at OD_{578nm} and expressed as µg NH₄-N/g soil/24 h (Yan et al., 2013). Catalase activity was measured by back titration of H₂O₂ added to soil with 0.1 M KMnO₄ and expressed as mL 0.1 M KMnO₄/g soil/h (Zhang et al., 2011). Invertase activity was assayed by the amount of glucose production at OD_{508nm} and expressed as µg glucose/g soil/24 h (Wu et al., 2019b). β-galactosidase activity was measured by the released 4-methylumbelliferone (MUF) and expressed as µg MUF µmol/g soil/h (Martínez-Iñigo et al., 2009).

2.6. Human health risk assessment of consuming crops

The human health risks of consuming crops were assessed by the Hazard Quotient (HQ) according to the method introduced by Environmental Protection Agency (EPA) in the US (Wei et al., 2020). When HQ is lower than 1, it demonstrates no risk for human health (Mehdizadeh et al., 2021). Hazard Quotient values were calculated using the following equation:

$$HQ = (EF \times ED \times C \times IR) / (BW \times AT \times RfD)$$

EF (Exposure Frequency): 365 days/year.

ED (Exposure Duration): 70 years for adult, 7 years for children.

C: Cd concentrations in the rice grain and oilseed (mg/kg).

Where *IR* (Ingestion Rate): For rice grain, 0.3892 kg/day for adult and 0.1984 kg/day for children, respectively. For rape oil, 0.025 kg/day for adult and 0.0125 kg/days for children, respectively.

BW (Body Weight): 62.71 kg for adult male, 55.1 kg for adult female and 25.6 kg

for children.

AT (Averaging Time): 25550 days for adult and 2555 days for children.

RfD (Reference of Dose): 0.001 mg/kg for Cd.

2.7. Statistical analysis

In this study, statistical significance was analyzed using SPSS 18.0 package, and means values were considered to be different when $P < 0.05$ using least significant difference (LSD). Figures were performed using Origin 8.0 (USA).

3. Results

3.1. Characteristics of soil and amendments

The main characteristics of soil, biochar and oyster shell were shown in Table S1. The soil was acidic soil with pH values of 5.27 - 5.51. The biochar and oyster shell used in the field study were alkaline materials and their pH values were 8.22 and 8.52, respectively. The OM of biochar (54.15%) was significantly higher than that of soil (3.93%) and oyster shell (1.26%). The carbon percentage of biochar was 92.50%.

The surface of oyster shell (Figure 1a) was a filamentous layer with some disordered deposition, which might be calcium compounds. The structure of biochar (Figure 1b) was lamellar and polyporous, which might be in favor of Cd absorption. In addition, FTIR was operated to detect functional groups of oyster shell and biochar (Figure 1c). The characteristic peaks of calcium carbonate in oyster shell were observed at 1427 cm^{-1} and 879 cm^{-1} (Lu et al., 2021). Biochar showed obvious peaks at 1089 cm^{-1} and 790 cm^{-1} , which were related to C-O, and C-H bending vibration, respectively (Wu et al., 2019a). In addition, an obvious feature at 3436 cm^{-1}

corresponding to -OH was loaded on oyster shell and biochar (Lian et al., 2021).

3.2. Analysis of soil Cd bioavailability

To evaluate the effect of different amendments on Cd bioavailability, the concentrations of HOAc-extractable Cd in soils were determined by TCLP method (Halim, 2003). Figure 2 showed the variations of HOAc-extractable Cd with different amendments in the rice-oilseed rape rotation. Both biochar and oyster shell resulted in the reduction of HOAc-extractable Cd in soils. In the rice planting, the HOAc-extractable Cd in the PA1, PA2 and PA3 treatments was significantly decreased by 20.79%, 40.59% and 10.89%, respectively, compared to control. In the oilseed rape planting, the HOAc-extractable Cd in the PA1, PA2 and PA3 treatments was also reduced by 5.76%, 17.85% and 5.95% respectively, compared to control. The Cd immobilization efficiency in the PA3 treatment was higher than that in the PA1 treatment, which demonstrated that the addition of oyster shell could strength the Cd immobilization capacity of biochar.

3.3. Analysis of Cd concentrations in brown rice and oilseed

As shown in Figure 3, the application of biochar and oyster shell reduced the Cd concentrations in brown rice and oilseed. In the PA0 treatment, the Cd concentration in brown rice was 0.91 mg/kg. Compared to control (PA0), the Cd concentration in brown rice was decreased by 20.88% and 30.77%, respectively, in the PA1 and PA2 treatments. The Cd concentration in oilseed was reduced in the PA1 and PA3, about 27.63% and 19.74% lower than that in PA0, respectively. Moreover, the cooperative application of biochar and oyster shell contributed to higher reduction of Cd in brown

rice (29.67%) than that in signal biochar (20.88%).

3.4. Health risk assessment of consuming crops

Hazard Quotient values of consuming crops in different treatments were analyzed. The HQ order of consuming rice and oilseed was children > adult female > adult male, which indicated that children had more health risk than adults for the intake of contaminated crops (Figure 4). Without the application of amendments, the HQ values of consuming brown rice for adult male, adult female and children were 5.66, 6.44 and 7.07, respectively. For children, HQ values for brown rice intake in PA1, PA2 and PA3 were decreased by 20.87%, 31.11% and 29.76%, respectively, compared to control. In addition, it was also observed that the application of amendments decreased the HQ values of consuming oilseed by 17.27 - 28.14% compared to control.

3.5. Analysis of soil pH and CEC

It was observed that soil pH was weakly increased by biochar, but significantly increased by oyster shell (Figure 5a). After the oyster shell application, the soil pH increased from acidity (5.2 - 5.5) to neutral (6.9 - 7.3). Meanwhile, the cooperative application of biochar and oyster shell also increased soil pH to 7.10 - 7.24. The application of oyster shell slightly increased the CEC of soil in the rice planting, while both oyster shell and biochar had no significant effects on the CEC of soil in the oilseed rape planting (Figure 5b).

3.6. Analysis of soil fertility

To analyze the effects of amendments on soil fertility, the concentrations of TOC,

OM, available P, available K, and available N were determined during the rice-oilseed rape rotation (Table S3). Biochar application slightly increased TOC and OM in the rice-oilseed rape rotation. In the rice planting, soil TOC and OM in the PA3 treatment were increased by 10.09% and 9.92%, respectively, compared to control. In the oilseed rape planting, soil TOC and OM in the PA1 treatment were enhanced by 11.06% and 11.32%, respectively, compared to control. More obviously, available P was significantly increased by the addition of oyster shell. Compared to control, the available P significantly were increased by 200.96 - 295.92% and 184.73 - 187.46%, respectively, in the PA2 and PA3 treatments.

3.7. Analysis of soil enzyme activities

As shown in Figure 6, adding amendments variously changed the activities of soil enzyme. In the rice-oilseed rape rotation, the application of biochar (PA1) increased the dehydrogenase activity, about 20.12 - 25.49% higher than that of control (PA0). Urease activity was markedly enhanced by the oyster shell treatment. Compared to the control, urease activity was significantly increased by 205.56 - 268.88% and 139.44 - 147.56%, respectively, in the PA2 and PA3 treatments. However, biochar had no obvious effect on the activities of acid phosphate and invertase, but oyster shell significantly reduced the acid phosphate activity by 43.30% in the rice planting. In addition, the cooperative application of biochar and oyster shell enhanced the activities of catalase and β -galactosidase activity by 10.71 - 34.31% and 82.08 - 244.38%, respectively, compared to control.

3.8. Analysis of correlation coefficient

The Pearson correlation analysis was used to analyze the relationship among different parameters. As shown in Figure 7a, the Cd concentration in brown rice was positively correlated to Cd bioavailability ($r = 0.90$) but negatively correlated to soil pH ($r = -0.83$). Meanwhile, the activities of soil enzyme except acid phosphate were positively connected to available N, available P, available K, and TOC. The Figure 7b showed a weak correlation between Cd uptake by oilseed rape and Cd bioavailability. Soil pH was positively correlated to available P and β -galactosidase activity ($r > 0.95$), which further demonstrated that alkaline substances could increase available P and β -galactosidase activity by adjusting soil pH in acidic fields.

3.9. Cost approach for amendments

Considering the remediation of large areas of the contaminated agricultural soil, the cost of amendments is a key parameter in the practical application. The market price of biochar (> 1200 RMB/t) was much higher than that of oyster shell (500 RMB/t) (see Supplementary Materials). In this study, the dosage of amendments was 15000 kg/ha. The cost for biochar amendment was at least 1800 RMB/ha, while the joint use of biochar and oyster shell decreased the cost of amendments by 29.17%. Based on these results, the collaborative application of biochar and oyster shell might be an economical pathway to immobilize Cd and improve soil properties.

4. Discussion

Rice and oilseed rape are the most important crops over the globe. Simultaneously, the rice-oilseed rape rotation was the main cultivated model in China (Huang et al., 2020). However, the Cd contamination in agricultural lands, especially

in acidic soils, has severely threatened food safety production and human health (Shi et al., 2022). Cd accumulation in the crops poses a great human health risk due to the Cd uptake by crops may result in kidney damage and adverse effects on lung, cardiovascular, musculoskeletal systems (Wei et al., 2020).

In-situ immobilization was an effective pathway to decrease the Cd uptake by crops by the application of amendments (Kumpiene et al., 2008). In this study, two bio-wastes namely biochar and oyster shell were used to decrease the Cd uptake by crops and modify the soil quality. The application of biochar and oyster shell both reduced the HOAc-extractable Cd in soil (Figure 2). The HOAc-extractable Cd has widely used to evaluate the bioavailability of Cd in soils (Liu et al., 2021). Previous studies have revealed that biochar had a great potential on the Cd immobilization by surface absorption and co-precipitation (He et al., 2019; Liu et al., 2018). However, the reduction of the HOAc-extractable Cd in the oyster shell treatments was significantly higher than that in the biochar treatments, which might result from the enhancement of soil pH in the oyster shell treatments (Lee et al., 2008). Soil pH is one of the main factors influencing the bioavailability of Cd in soils (Huang et al., 2020). It has been widely verified that soil pH determines the solid-solution equilibria of heavy metals in soils (Zhao and Masaihiko, 2007). Oyster shell has been regarded as a low-release alkaloid in soils due to it primarily consisted of CaCO_3 (Ok et al., 2010). The dissolution of CaCO_3 from oyster shell can produce hydroxyl ion (OH^-) (Ok et al., 2010). The increase of soil pH can result to the increase of the negative soil surface charge, which easily causes an increased capacity of cationic metal adsorption (Ok et

al., 2010). The precipitants of metal oxy/hydroxides could be formed due to increased hydroxyl ions (Bolan et al., 2014). The Cd uptake by crops was positively related to the Cd bioavailability in soil (Huang et al., 2020). Similar to other reports (Jing et al., 2020; Mehdizadeh et al., 2021), the Cd concentrations in brown rice and oilseed were decreased after the application of biochar and oyster shell. Furthermore, the health risk related to the special polluted crops consumption with Cd has been estimated by HQ, and the decreased HQ values demonstrated that the human health risk of consuming crops was decreased by the application of amendments (Ma et al., 2021a).

Soil nutrients play an important role on soil quality and plant growth. Phosphorus fractions are mainly dependent on soil pH, soil mineralogy and the application of phosphate fertilizer (Lee et al., 2008). The Fe-P and Al-P are the predominant forms in acidic soils, while calcium bound-P is the predominant form in alkaline soils (Dean, 1949). In acidic soils, the loosely bound phosphates are converted into Fe-P and Al-P fractions gradually owing to the re-precipitation process. Previous studies found that the concentration of available P reached the maximum at neutral pH soils (Lee et al., 2008). Our results showed that the addition of oyster shell markedly increased the concentration of available P in soils, which might be resulted from the enhancement of soil pH (Table S3). Correlation analysis (Figure 7) further demonstrated that available P was highly correlated to the changes of soil pH ($r > 0.99$). The concentrations of available K and available N also slightly increased with the application of biochar and oyster shell, indicating an improvement of soil fertility.

Activities of soil enzyme have been widely used to reflect soil biological quality

(Lin et al., 2021). In this study, the activities of dehydrogenase, urease, catalase and β -galactosidase were increased in the treatments of biochar and oyster shell (Figure 6). Dehydrogenase usually reflects the microbial degradation capacity for organic matter (Campos et al., 2019). Urease is often used as a biochemical indicator to reflect soil fertility, which played a crucial role on soil nitrogen mineralization (Lebrun et al., 2012). The addition of oyster shell increased the soil pH, which usually results in the enhancement of dehydrogenase and urease activities (Wen et al., 2021). Abd El-Azeem et al. (2013) reported that dehydrogenase activity was positively correlated to soil pH. Oyster shell can raise the urease activity, thus catalyzing the hydrolysis of urea to CO_2 and NH_3 with an optimum pH around 7.4 (Lee et al., 2008). Soil β -galactosidase plays an important role in the microbial glycometabolism, and the significant increase of β -galactosidase by the application of biochar indicates a shift in the use of soil organic carbon from plant-derived sugars towards more recalcitrant C compounds (Giagnoni et al., 2019). In addition, the porous structure and rich nutrients of biochar can contribute to the growth of soil microorganisms, and thus might increase the activities of soil enzyme (Liao et al., 2016). Moreover, the enhancement of enzyme activities in biochar and oyster shell treatments might also be related to the decrease of Cd biotoxicity in soil (Zhang et al., 2021). In conclusion, the enhancement of the activities of soil enzyme indicated that the cooperative application of biochar and oyster shell could improve the soil biological properties.

5. Conclusions

The current study revealed the impacts of applying oyster shell and biochar on

Cd bioavailability, Cd uptake by crops, human health risk of consuming crops, soil fertility and enzyme activities during the rice-oilseed rape rotation. The application of oyster shell significantly ($p < 0.05$) increased soil pH and thus decreased the bioavailability of Cd in soil. The cooperative application of biochar and oyster shell significantly reduced the Cd concentrations in crops and human health risk of consuming brown rice and oilseed. In addition, the application of biochar increased OM and TOC, while the addition of oyster shell was suitable to improve available P. Moreover, the activities of soil enzyme were markedly enhanced by the cooperative application of oyster shell and biochar. These results suggested that the joint application of biochar and oyster shell is a low-cost pathway to effectively reduce the Cd uptake by crops and improve soil quality.

Acknowledgements

This study was financially supported by Key Technologies Research and Development Program (CN) (2018YFC1802605), the Science and Technology Project of Sichuan Province (2022YFN0066), Chengdu Science and Technology Project (2021-YF05-00195-SN), State Key Laboratory of Geohazard Prevention and Geoenvironment Protection Independent Research Project (SKLGP2021Z030), and State Environmental Protection Key Laboratory of Synergetic Control and Joint Remediation for Soil & Water Pollution (GHBK-2020-012). The authors also wish to thank Professor Guanglei Cheng and Hui Wang from Sichuan University for the technical assistance.

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Figure captions:

Figure 1 SEM images of oyster shell (a) and biochar (b) and FTIR spectra (c) of oyster shell and biochar.

Figure 2 The effects of amendments on Cd bioavailability in soil. Dots represent the value of each sample. Bars followed with different lowercase letters (a - c) and capital letters (A, B) indicated significant ($p \leq 0.05$) difference among different treatments in rice planting and oilseed rape planting according to the LSD test. Values represent means \pm standard deviation.

Figure 3 The effects of amendments on the Cd concentrations in brown rice (a) and oilseed (b). Dots represent the value of each sample. Bars with different lowercase letters indicated significant ($p < 0.05$) difference among different treatments according to the LSD test. Values represent means \pm standard deviation.

Figure 4 The effects of different amendments on the HQ of grown rice and oilseed. Mean with different lowercase letter indicated significant ($p < 0.05$) difference from each other according to the LSD test. Values represent means \pm standard deviation.

Figure 5 The effects of different amendments on soil pH (a) and CEC (b). Dots represent the value of each sample. Bars followed with different lowercase letters (a - c) and capital letters (A, B) indicated significant ($p \leq 0.05$) difference among different treatments in rice planting and oilseed rape planting according to the LSD test. Values represent means \pm standard deviation.

Figure 6 The effects of different amendments on the activities of soil enzyme. Dots represent the value of each sample. Bars followed with different lowercase letters (a - c) and capital letters (A-C) indicated significant ($p \leq 0.05$) difference among different treatments in rice planting and oilseed rape planting according to the LSD test. Values represent means \pm standard deviation.

Figure 7 The correlation of investigated parameters in rice planting (a) and rice-oilseed planting (b)

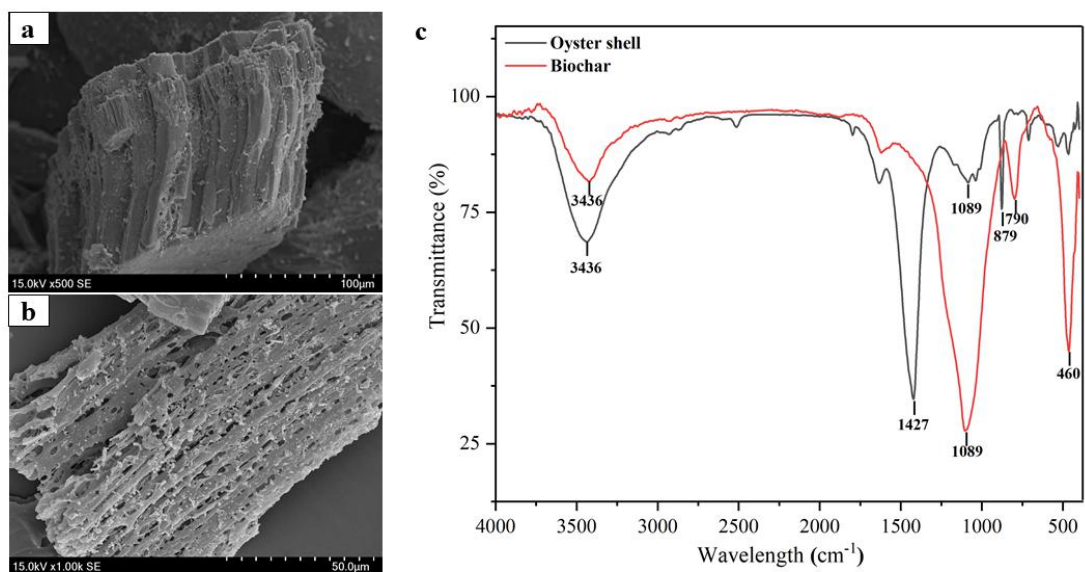


Figure 1 SEM images of oyster shell (a) and biochar (b) and FTIR spectra (c) of oyster shell and biochar.

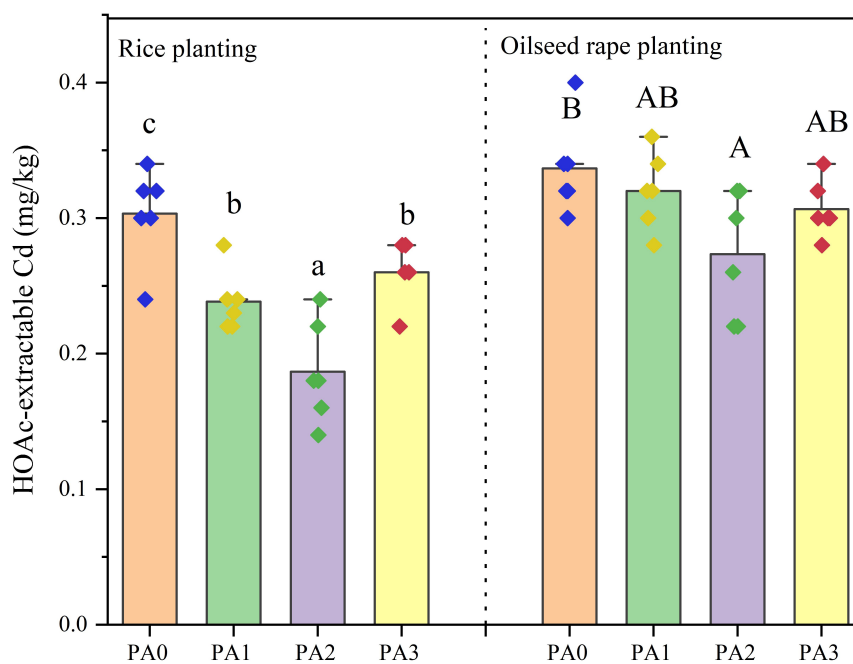


Figure 2 The effects of amendments on Cd bioavailability in soil. Dots represent the value of each sample. Bars followed with different lowercase letters (a - c) and capital letters (A, B) indicated significant ($p \leq 0.05$) difference among different treatments in rice planting and oilseed rape planting according to the LSD test. Values represent means \pm standard deviation.

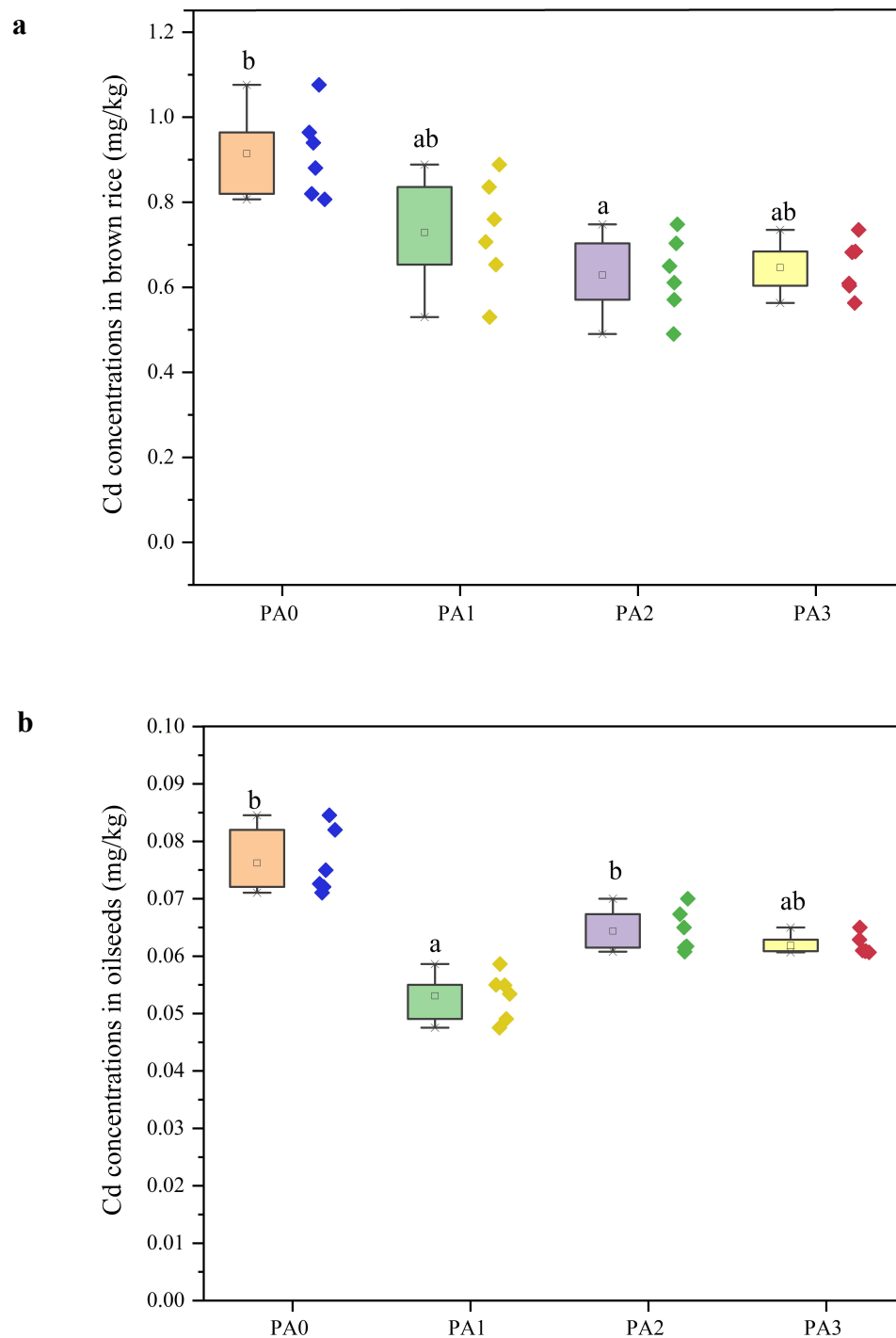


Figure 3 The effects of amendments on the Cd concentrations in brown rice (a) and oilseed (b). Dots represent the value of each sample. Bars with different lowercase letters indicated significant ($p < 0.05$) difference among different treatments according to the LSD test. Values represent means \pm standard deviation.

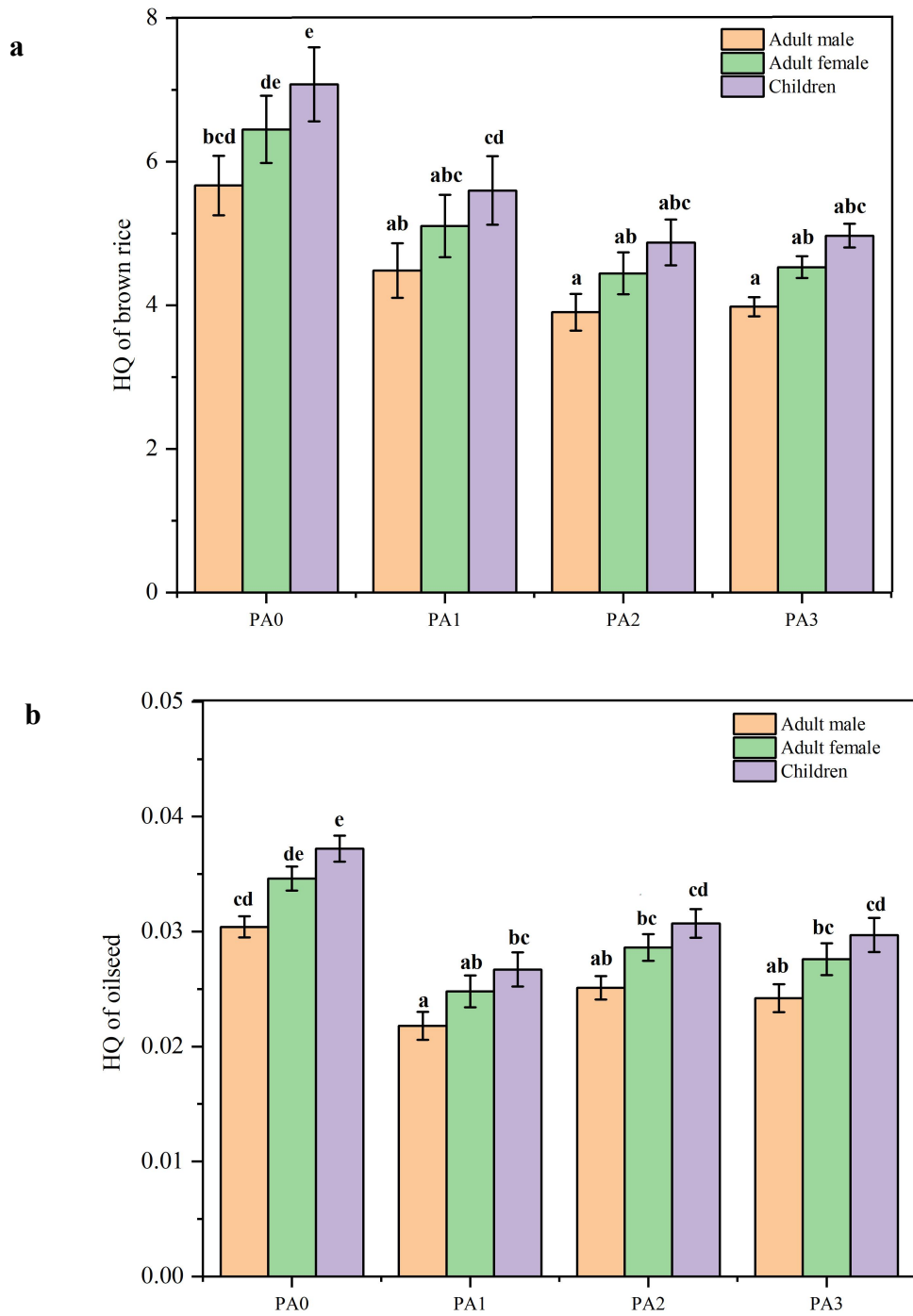


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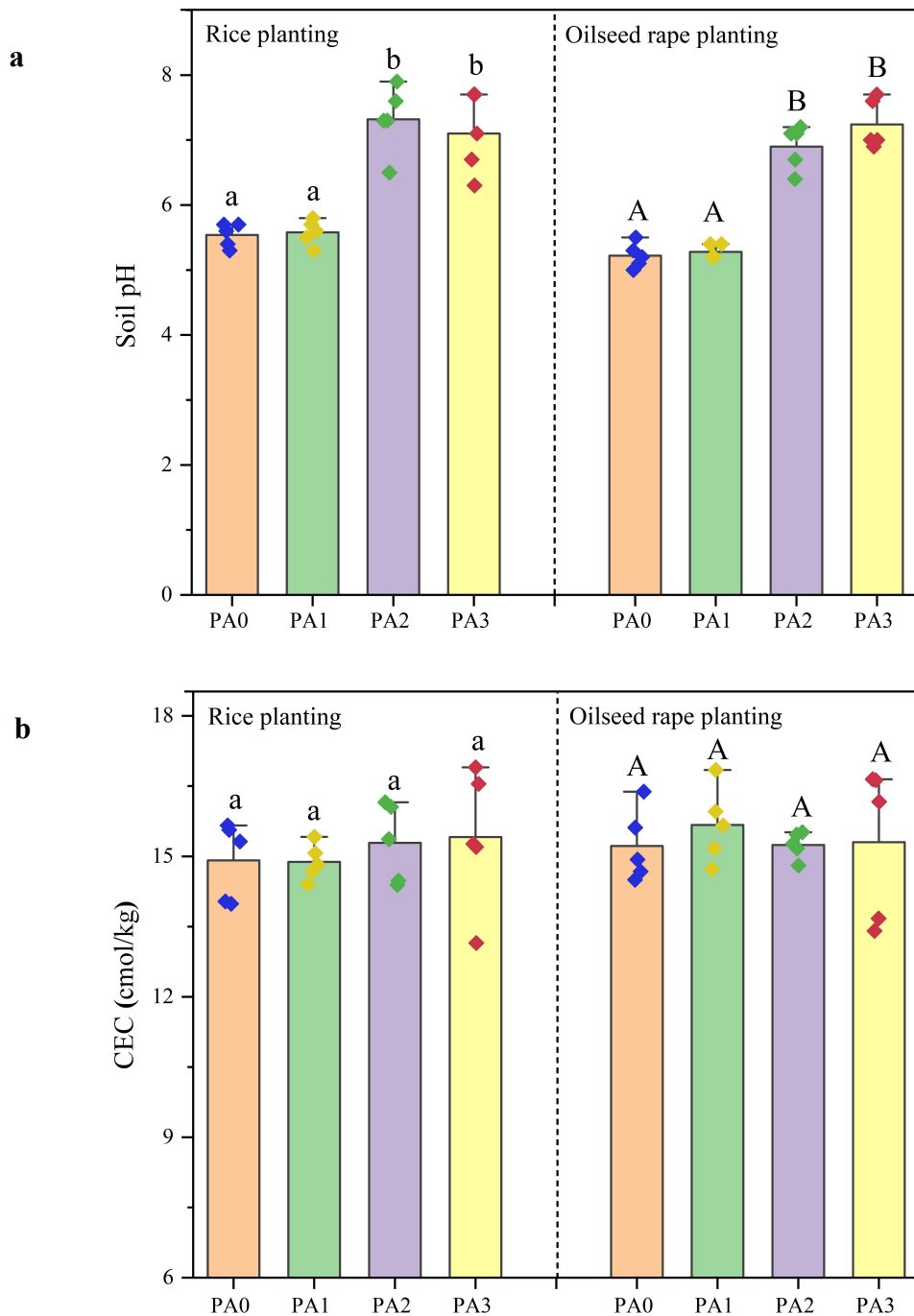


Figure 5 The effects of different amendments on soil pH (a) and CEC (b). Dots represent the value of each sample. Bars followed with different lowercase letters (a - c) and capital letters (A, B) indicated significant ($p \leq 0.05$) difference among different treatments in rice planting and oilseed rape planting according to the LSD test. Values represent means \pm standard deviation.

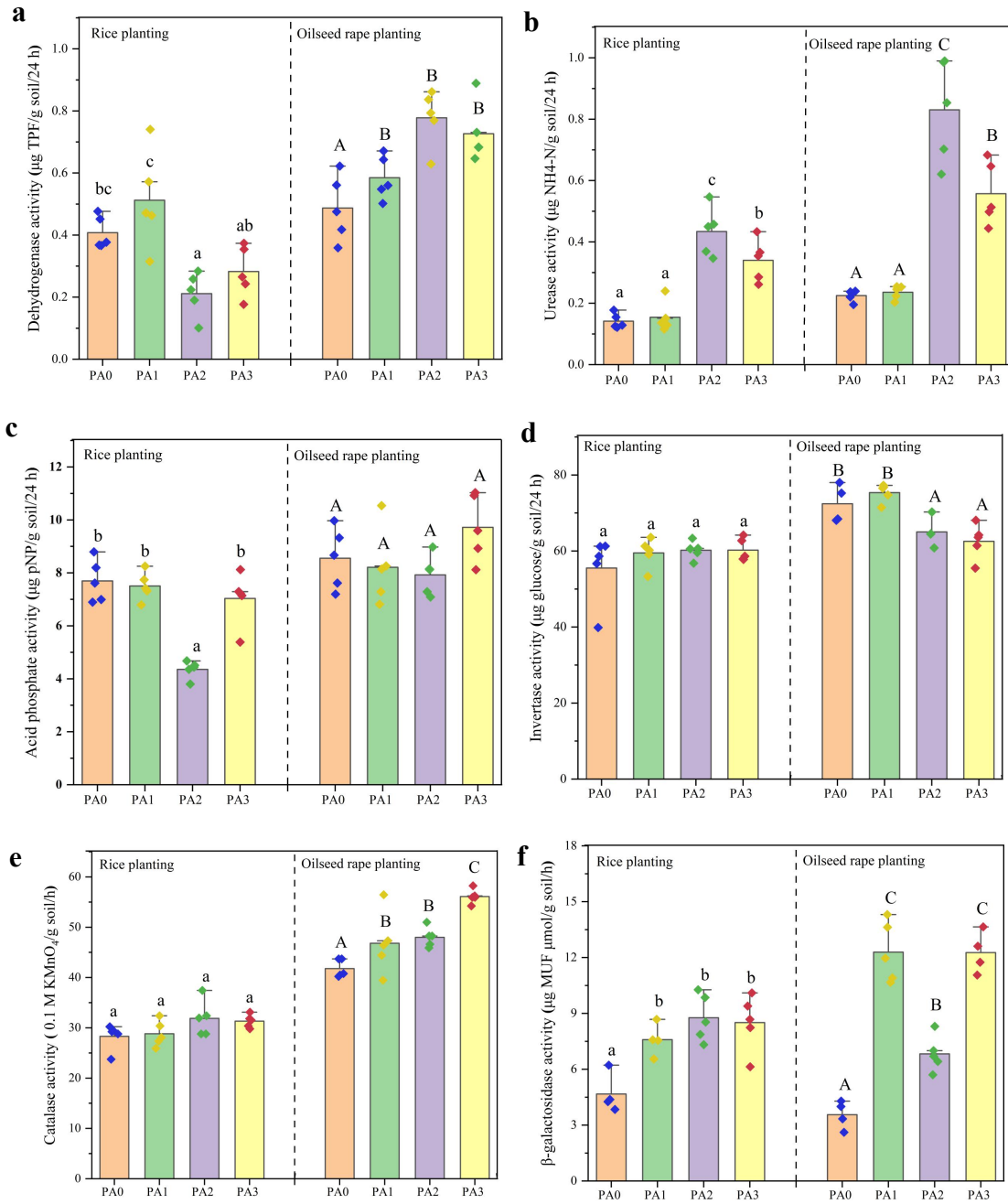


Figure 6 The effects of different amendments on the activities of soil enzyme. Dots represent the value of each sample. Bars followed with different lowercase letters (a - c) and capital letters (A-C) indicated significant ($p \leq 0.05$) difference among different treatments in rice planting and oilseed rape planting according to the LSD test. Values represent means \pm standard deviation.

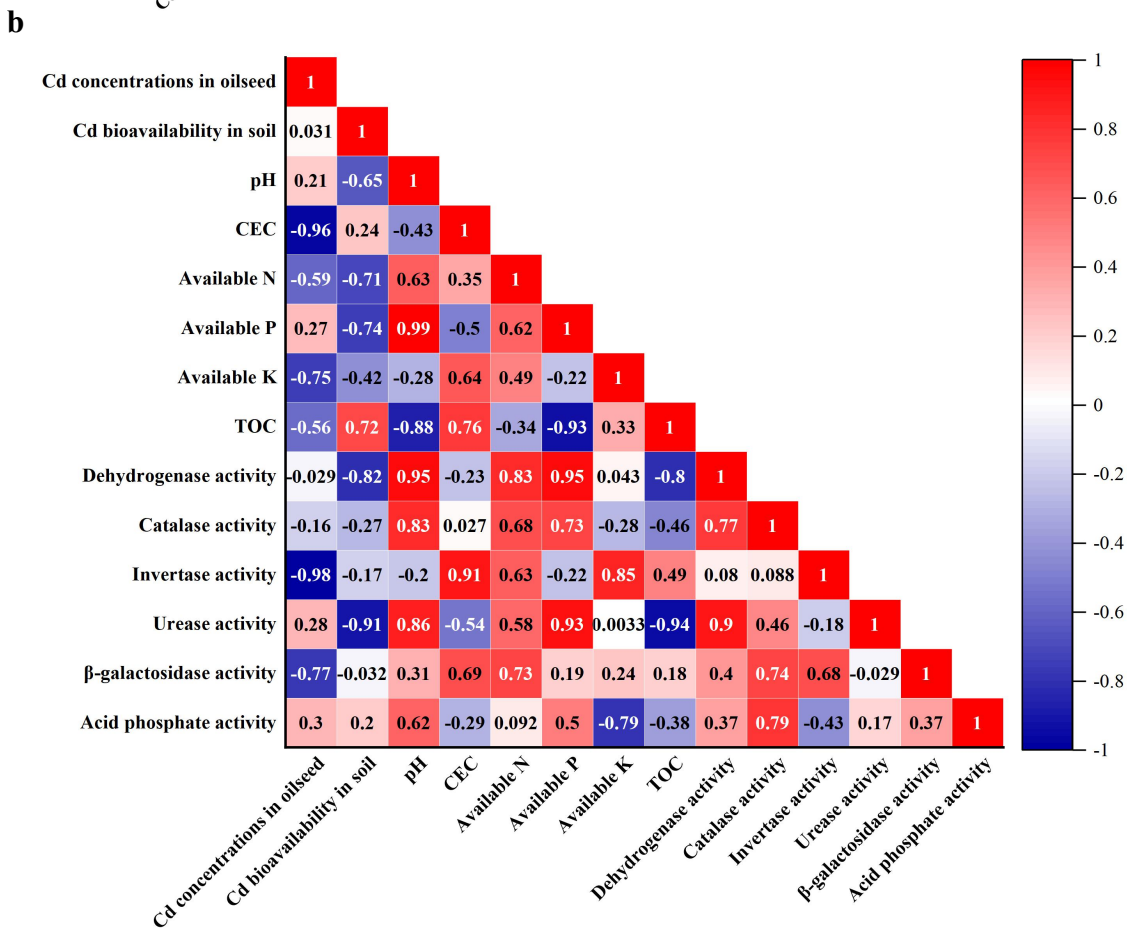
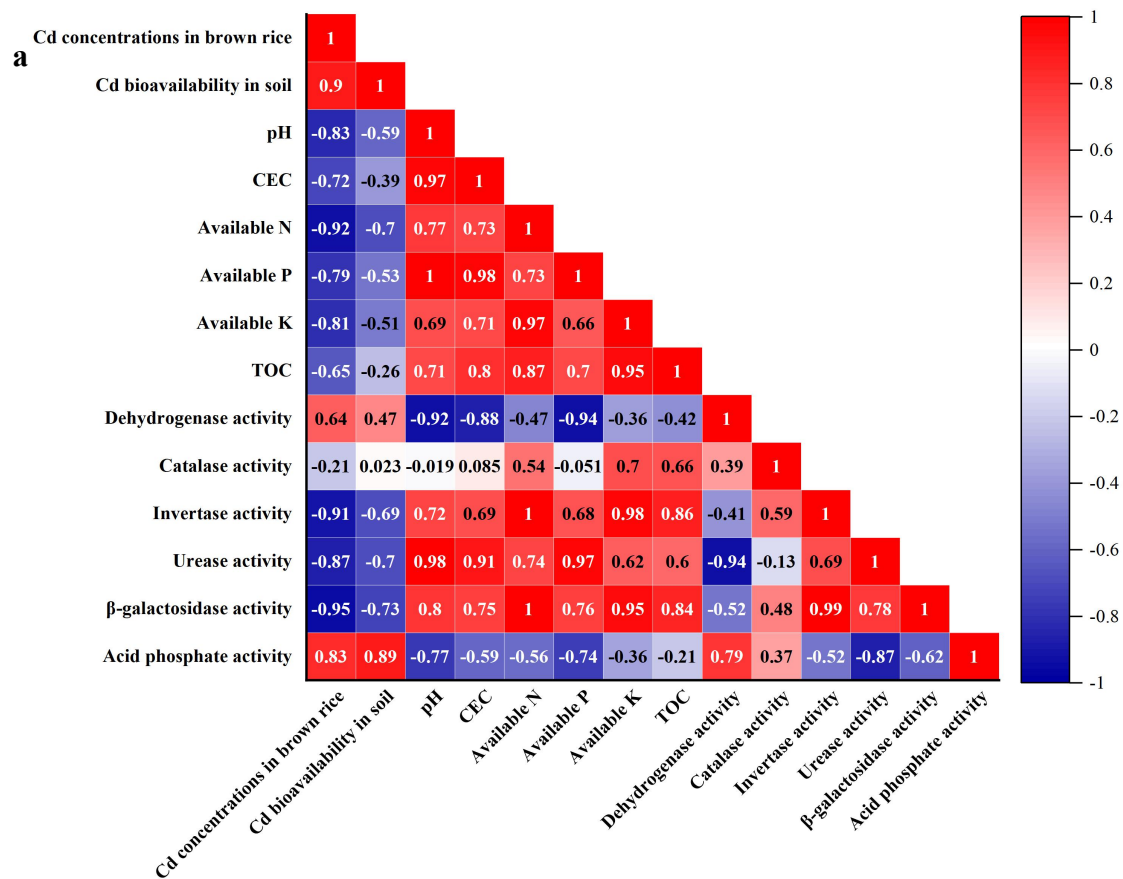


Figure 7 The correlation of investigated parameters in rice planting (a) and rice-oilseed rape planting (b).

Code/Data availability

Data are available upon request to the authors.

Author contribution

Bin Wu: Investigation, Writing Original Draft, Supervision

Jia Li: Writing - Review & Editing

Mingping Sheng: Investigation

He Peng: Investigation, Visualization

Dinghua Peng: Investigation, Data Curation

Heng Xu: Conceptualization, Resources, Funding acquisition

Declaration of interests

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.