1	The application of biochar and oyster shell reduced cadmium uptake by crops
2	and modified soil biochemical properties in contaminated soil
3	Bin Wu ^{a*} , Jia Li ^a , Mingping Sheng ^b , He Peng ^b , Dinghua Peng ^b , Heng Xu ^{b*1}
4	^a State Key Laboratory of Geohazard Prevention and Geoenvironment Protection, College of
5	Ecology and Environment, Chengdu University of Technology, Chengdu, 610059, PR China
6	^b Key Laboratory of Bio-Resource and Eco-Environment of Ministry of Education, College of
7	Life Sciences, Sichuan University, Chengdu, 610065, PR China
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¹ Corresponding author: E-mail address: <u>wub@cdut.edu.cn</u> (Bin Wu); xuheng64@sina.com (Heng Xu)

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Abstract

28	Soil pollution with cadmium (Cd) has been threatening the human health. In this
29	study, we investigated the possibility of applying biochar and oyster shell to reduce
30	Cd uptake by crops and modify soil biochemical properties. A filed study based on the
31	rice-oilseed rape rotation was done and the treatments were comprised of without
32	amendments (PA0), 15000 kg/ha biochar (PA1), 15000 kg/ha oyster shell (PA2), and
33	7500 kg/ha biochar and 7500 kg/ha oyster shell (PA3). Results revealed that both
34	oyster shell and biochar reduced the HOAc-extractable Cd in soil. Compared to PA0,
35	the HOAc-extractable Cd in the PA1, PA2 and PA3 treatments was reduced by 4.76 -
36	20.79%, 17.86 - 38.61% and 5.95 - 10.89%, respectively. The cooperative application
37	of biochar and oyster shell reduced the Cd accumulation in brown rice and oilseed by
38	29.67% and 19.74%, respectively, compared to control, and thus decreased the Hazard
39	Quotient (HQ) by the consumption of brown rice and oilseed. The addition of biochar
40	slightly increased soil organic matter. In addition, the available P in the PA2 and PA3
41	treatments was significantly (p $< 0.05)$ increased by 200.96 - 295.92% and 187.46 -
42	280.04% compared to control. Moreover, the cooperative application of biochar and
43	oyster shell enhanced the activities of urease, catalase, and β -galactosidase by 139.44
44	- 147.56%, 10.71 - 34.31% and 82.08 - 244.38%, respectively. These results
45	demonstrated that the utilization of biochar and oyster shell might be an effective
46	pathway to reduce Cd uptake by crops and improve soil biochemical properties.
47	Keywords: Biochar; Oyster shell; Rice-oilseed rape rotation; In-situ remediation;

48 Enzyme activities; Cadmium

49 **1. Introduction**

Cadmium (Cd) contamination of agricultural soils is a worldwide environmental 50 51 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, 52 53 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 54 (7%) among all heavy metals (Mou et al., 2020). In southwest China, the intensive 55 industrialization is the resource of the farmlands being contaminated with Cd (Chen et 56 57 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 58 cost-effective and eco-friendly remediation technologies is crucial for food safety and 59 60 soil quality.

In recent years, the in-situ immobilization technology through the application of 61 soil amendments has raised wide attentions in the remediation of Cd contaminated 62 63 farmlands, which could reduce the Cd uptake by plants without delaying agricultural production (Palansooriya et al., 2020). Biochar derived from bio-wastes has been 64 widely recommended as a soil amendment (Zong et al., 2021). Amounts of nutrients 65 (such as C, N, P, K, and Mg) in biochar can improve soil fertility and promote plant 66 growth (Lu et al., 2015). Moreover, biochar has a large surface area and plenty of 67 functional groups (such as -COOH, C-O and C=O), which are reactive to immobilize 68 heavy metals, including Cd, lead (Pb), and nickel (Ni) (Wang et al., 2021). However, 69 the high price of biochar limits its large application. In addition, the application of 70

biochar can not effectively change soil pH in acidic fields, thus it can not effectively 71 reduce the bioavailability of Cd in soil (Liu et al., 2018). Oyster shell is a low-cost 72 73 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 74 adjustment and Cd immobilization in soil (Lee et al., 2008). In this sense, we think 75 that the joint use of biochar and oyster shell might be a low-cost and effective 76 pathway to decrease Cd uptake by crops and improve soil biochemical quality in 77 acidic fields. 78

79 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). 80 Previous studies mainly focused on the effects of amendments on reducing the Cd 81 82 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 83 opinions, a field experiment under the rice-oilseed rape rotation was designed: (1) to 84 85 investigate the effects of biochar and oyster shell on Cd immobilization; (2) to evaluate the effects of biochar and oyster shell on decreasing human health risk of 86 consuming contaminated crops; (3) to reveal the effects of biochar and oyster shell on 87 soil biochemical properties including pH, cation exchange capacity (CEC), total 88 organic carbon (TOC), organic matter (OM), available phosphate, available potassium, 89 available nitrogen, and the activities of soil enzyme, so as to estimate the correlation 90 91 of main parameters in the moderately polluted farmland.

92 2. Material and methods

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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.

101 **2.2.**

2.2. Characteristics of experimental materials

102 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. 103 104 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 105 composition of oyster shell was shown in Table S2. The surface structures of biochar 106 and oyster shell were analyzed by Scanning Electron Microscope (SEM, JSM-7500F). 107 The functional groups of biochar and oyster shell were measured by Fourier 108 Transform Infrared Spectra (Nicolet 6700). The seeds of rice "Yixiang 2115" and 109 seeds of oilseed rape "Yiyou 15" were obtained from Rice Research Institute, Sichuan 110 Academy of Agricultural Science. 111

112 **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

115 (7500 kg/ha biochar and 7500 kg/ha oyster shell). The concentrations of biochar and 116 oyster shell used in this study were referred to the previous report (Ameloot et al., 117 2014). Each experimental plot was 56 m² (7 x 8 m) and arranged in a randomized 118 design with three replicates. Before rice planting, the amendments were sufficiently 119 mixed with topsoil. After the harvest of rice, the oilseed rape was planted following 120 the conventional tillage pattern without extra amendments.

121 **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).

127 **2.5. Soil analysis**

Soil pH was determined by a pH meter (METTLER-S220) with a soil/water ratio 128 of 5 g/25 mL. The bioavailable Cd of soil was measured by the TCLP method (Xu et 129 al., 2020). Briefly, 2 g of soil sample was mixed with 40 mL of 0.11 M acetic acid 130 (HOAc) and shaken for 16 h at 25 °C, 150 rpm. The mixture was centrifuged for 5 131 min at 8000 rpm and then the supernatant was collected to determine Cd content by 132 AAS. Available P, available K, available N were measured according to the method 133 described by Wu et al. (2018). Soil TOC and OM were determined by the method 134 described by Walz et al. (2017). 135

136 In addition, activities of soil enzyme were analyzed to reflect the biological

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137	quality in this study. Dehydrogenase activity was evaluated by the production of
138	triphenylfornazan (TPF) at OD_{492nm} and expressed as μg TPF/g soil/24 h (Benefield et
139	al., 1977). Acid phosphate activity was assayed by the <i>p</i> -nitrophenol (pNP) release at
140	OD_{400nm} and expressed as μg pNP/g soil/24 h (Van Aarle and Plassard, 2010). Urease
141	activity was determined by the NH4-complex at OD_{578nm} and expressed as μg
142	NH ₄ -N/g soil/24 h (Yan et al., 2013). Catalase activity was measured by back titration
143	of $\rm H_2O_2$ added to soil with 0.1 M KMnO_4 and expressed as mL 0.1 M KMnO_4/g soil/h
144	(Zhang et al., 2011). Invertase activity was assayed by the amount of glucose
145	production at OD_{508nm} and expressed as µg glucose/g soil/24 h (Wu et al., 2019b).
146	β -galactosidase activity was measured by the released 4-methylumbelliferone (MUF)
147	and expressed as µg MUF µmol/g soil/h (Martínez-Iñigo et al., 2009).

148 **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:

- 154 $HQ = (EF \times ED \times C \times IR)/(BW \times AT \times RfD)$
- 155 *EF* (Exposure Frequency): 365 days/year.
- 156 *ED* (Exposure Duration): 70 years for adult, 7 years for children.
- 157 C: Cd concentrations in the rice grain and oilseed (mg/kg).
- 158 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 159 kg/days for children, respectively. 160

161 BW (Body Weight): 62.71 kg for adult male, 55.1 kg for adult female and 25.6 kg for children. 162

AT (Averaging Time): 25550 days for adult and 2555 days for children. 163

RfD (Reference of Dose): 0.001 mg/kg for Cd. 164

2.7. Statistical analysis 165

In this study, statistical significance was analyzed using SPSS 18.0 package, and 166 means values were considered to be different when P < 0.05 using least significant 167 difference (LSD). Figures were performed using Origin 8.0 (USA). 168

3. Results 169

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170 3.1. Characteristics of soil and amendments

171 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 172 used in the field study were alkaline materials and their pH values were 8.22 and 8.52, 173 respectively. The OM of biochar (54.15%) was significantly higher than that of soil 174 (3.93%) and oyster shell (1.26%). The carbon percentage of biochar was 92.50%. 175 176 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. 178

In addition, FTIR was operated to detect functional groups of oyster shell and biochar 179

(Figure 1c). The characteristic peaks of calcium carbonate in oyster shell were 180

observed at 1427 cm⁻¹ and 879 cm⁻¹ (Lu et al., 2021). Biochar showed obvious peaks 181 at 1089 cm⁻¹ and 790 cm⁻¹, which were related to C-O, and C-H bending vibration, 182 respectively (Wu et al., 2019a). In addition, an obvious feature at 3436 cm⁻¹ 183 corresponding to -OH was loaded on oyster shell and biochar (Lian et al., 2021). 184

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3.2. Analysis of soil Cd bioavailability

To evaluate the effect of different amendments on Cd bioavailability, the 186 concentrations of HOAc-extractable Cd in soils were determined by TCLP method 187 (Halim, 2003). Figure 2 showed the variations of HOAc-extractable Cd with different 188 189 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 190 HOAc-extractable Cd in the PA1, PA2 and PA3 treatments was significantly 191 decreased by 20.79%, 40.59% and 10.89%, respectively, compared to control. In the 192 oilseed rape planting, the HOAc-extractable Cd in the PA1, PA2 and PA3 treatments 193 was also reduced by 5.76%, 17.85% and 5.95% respectively, compared to control. 194 The Cd immobilization efficiency in the PA3 treatment was higher than that in the 195 PA1 treatment, which demonstrated that the addition of oyster shell could strength the 196 197 Cd immobilization capacity of biochar.

3.3. Analysis of Cd contents in brown rice and oilseed 198

As shown in Figure 3, the application of biochar and oyster shell reduced the Cd 199 contents in brown rice and oilseed. In the PA0 treatment, the Cd content in brown rice 200 was 0.91 mg/kg. Compared to control (PA0), the Cd content in brown rice was 201 decreased by 20.88% and 30.77%, respectively, in the PA1 and PA2 treatments. The 202

Cd content 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%).

207 **3.4. Health risk assessment of consuming crops**

Hazard Quotient values of consuming crops in different treatments were 208 analyzed. The HQ order of consuming rice and oilseed was children > adult female > 209 210 adult male, which indicated that children had more health risk than adults for the 211 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 212 5.66, 6.44 and 7.07, respectively. For children, HQ values for brown rice intake in 213 PA1, PA2 and PA3 were decreased by 20.87%, 31.11% and 29.76%, respectively, 214 compared to control. In addition, it was also observed that the application of 215 amendments decreased the HQ values of consuming oilseed by 17.27 - 28.14% 216 217 compared to control.

218 **3.5. Analysis of soil biochemical properties**

219 **3.5.1. 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 theoilseed rape planting (Figure 5b).

227 **3.5.2.** Analysis of soil nutrients

To analyze the effects of amendments on soil bioavailable nutrients, the contents 228 of TOC, OM, available P, available K, and available N were determined during the 229 rice-oilseed rape rotation (Table S3). Biochar application slightly increased TOC and 230 OM in the rice-oilseed rape rotation. In the rice planting, soil TOC and OM in the PA3 231 treatment were increased by 10.09% and 9.92%, respectively, compared to control. In 232 233 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 234 was significantly increased by the addition of oyster shell. Compared to control, the 235 236 available P significantly were increased by 200.96 - 295.92% and 184.73 - 187.46%, respectively, in the PA2 and PA3 treatments. 237

238 **3.5.3.** Analysis of soil enzyme activities

239 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) 240 increased the dehydrogenase activity, about 20.12 - 25.49% higher than that of control 241 (PA0). Urease activity was markedly enhanced by the oyster shell treatment. 242 Compared to the control, urease activity was significantly increased by 205.56 -243 268.88% and 139.44 - 147.56%, respectively, in the PA2 and PA3 treatments. 244 However, biochar had no obvious effect on the activities of acid phosphate and 245 invertase, but oyster shell significantly reduced the acid phosphate activity by 43.30% 246

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.

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3.6. Analysis of correlation coefficient

251 The Pearson correlation analysis was used to analyze the relationship among different parameters. As shown in Figure 7a, the Cd content in brown rice was 252 positively correlated to Cd bioavailability (r = 0.90) but negatively correlated to soil 253 pH (r = -0.83). Meanwhile, the activities of soil enzyme except acid phosphate were 254 positively connected to available N, available P, available K, and TOC. The Figure 7b 255 showed a weak correlation between Cd uptake by oilseed rape and Cd bioavailability. 256 Soil pH was positively correlated to available P and β -galactosidase activity (r > 0.95), 257 258 which further demonstrated that alkaline substances could increase available P and β -galactosidase activity by adjusting soil pH in acidic fields. 259

260 **3.7. Cost approach for amendments**

Considering the remediation of large areas of the contaminated agricultural soil, 261 the cost of amendments is a key parameter in the practical application. The market 262 price of biochar (> 1200 RMB/t) was much higher than that of oyster shell (500 263 RMB/t) (see Supplementary Materials). In this study, the dosage of amendments was 264 15000 kg/ha. The cost for biochar amendment was at least 1800 RMB/ha, while the 265 joint use of biochar and oyster shell decreased the cost of amendments by 29.17%. 266 Based on these results, the collaborative application of biochar and oyster shell might 267 be an economical pathway to immobilize Cd and improve soil properties. 268

269 **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).

277 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 278 bio-wastes namely biochar and oyster shell were used to decrease the Cd uptake by 279 280 crops and modify the soil biochemical properties. The application of biochar and oyster shell both reduced the HOAc-extractable Cd in soil (Figure 2). The 281 HOAc-extractable Cd has widely used to evaluate the bioavailability of Cd in soils 282 (Liu et al., 2021). Previous studies have revealed that biochar had a great potential on 283 the Cd immobilization by surface absorption and co-precipitation (He et al., 2019; Liu 284 et al., 2018). However, the reduction of the HOAc-extractable Cd in the oyster shell 285 treatments was significantly higher than that in the biochar treatments, which might 286 result from the enhancement of soil pH in the oyster shell treatments (Lee et al., 2008). 287 Soil pH is one of the main factors influencing the bioavailability of Cd in soils 288 (Huang et al., 2020). It has been widely verified that soil pH determines the 289 solid-solution equilibria of heavy metals in soils (Zhao and Masaihiko, 2007). Oyster 290

shell has been regarding as a low-release alkaloid in soils due to it primarily consisted 291 of CaCO₃ (Ok et al., 2010). The dissolution of CaCO₃ from oyster shell can produce 292 293 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 294 cationic metal adsorption (Ok et al., 2010). The precipitants of metal oxy/hydroxides 295 could be formed due to increased hydroxyl ions (Bolan et al., 2014). The Cd uptake 296 by crops was positively related to the Cd bioavailability in soil (Huang et al., 2020). 297 Similar to other reports (Jing et al., 2020; Mehdizadeh et al., 2021), the Cd content in 298 299 brown rice and oilseed was decreased after the application of biochar and oyster shell. Furthermore, the health risk related to the special polluted crops consumption with Cd 300 has been estimated by HQ, and the decreased HQ values demonstrated that the human 301 302 health risk of consuming crops was decreased by the application of amendments (Ma et al., 2021a). 303

Soil nutrients play an important role on soil biochemical quality and plant growth. 304 Phosphorus fractions are mainly dependent on soil pH, soil mineralogy and the 305 application of phosphate fertilizer (Lee et al., 2008). The Fe-P and Al-P are the 306 predominant forms in acidic soils, while calcium bound-P is the predominant form in 307 alkaline soils (Dean, 1949). In acidic soils, the loosely bound phosphates are 308 converted into Fe-P and Al-P fractions gradually owing to the re-precipitation process. 309 Previous studies found that the content of available P reached the maximum at neutral 310 pH soils (Lee et al., 2008). Our results showed that the addition of oyster shell 311 markedly increased the content of available P in soils, which might be resulted from 312

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 contents of available K and available N also slightly increased with the application of biochar and oyster shell, indicating an improvement of soil fertility.

317 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 318 β-galactosidase were increased in the treatments of biochar and oyster shell (Figure 6). 319 Dehydrogenase usually reflects the microbial degradation capacity for organic matter 320 321 (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., 322 2012). The addition of oyster shell increased the soil pH, which usually results in the 323 324 enhancement of dehydrogenase and urease activities (Wen et al., 2021). Abd El-Azeem et al. (2013) reported that dehydrogenase activity was positively correlated 325 to soil pH. Oyster shell can raise the urease activity, thus catalyzing the hydrolysis of 326 urea to CO₂ and NH₃ with an optimum pH around 7.4 (Lee et al., 2008). Soil 327 β-galactosidase plays an important role in the microbial glycometabolism, and the 328 significant increase of β -galactosidase by the application of biochar indicates a shift in 329 the use of soil organic carbon from plant-derived sugars towards more recalcitrant C 330 compounds (Giagnoni et al., 2019). In addition, the porous structure and rich nutrients 331 of biochar can contribute to the growth of soil microorganisms, and thus might 332 increase the activities of soil enzyme (Liao et al., 2016). Moreover, the enhancement 333 of enzyme activities in biochar and oyster shell treatments might also be related to the 334

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.

338 **5.** Conclusions

339 The current study revealed the impacts of the application of oyster shell and biochar on Cd bioavailability, Cd uptake by crops, and human health risk of 340 consuming crops as well as soil biochemical properties during the rice-oilseed rape 341 rotation. The application of oyster shell significantly (p < 0.05) increased soil pH and 342 343 thus decreased the bioavailability of Cd in soil. The cooperative application of biochar and oyster shell significantly reduced the Cd contents and human health risk of 344 consuming brown rice and oilseed. In addition, the application of biochar increased 345 346 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 347 application of oyster shell and biochar. These results suggested that the joint 348 application of biochar and oyster shell is a low-cost pathway to effectively reduce the 349 Cd uptake by crops and improve soil biochemical properties. 350

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559 Figure captions:

560 **Figure 1** SEM images of oyster shell (a) and biochar (b) and FTIR spectra (c) of 561 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 ($p \le 0.05$) 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 Cd contents 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 ($p \le 0.05$) difference among different treatments in rice planting and oilseed rape planting according to the LSD test. Values
- 578 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 ($p \le 0.05$) difference among different treatments in rice planting and oilseed rape planting according to the LSD test. Values represent means \pm standard deviation.
- 584 **Figure 7** The correlation of investigated parameters in rice planting (a) and 585 rice-oilseed planting (b)

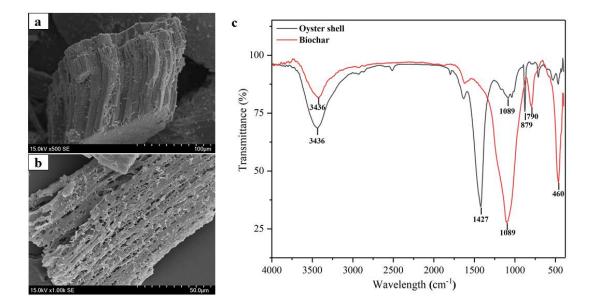


Figure 1 SEM images of oyster shell (a) and biochar (b) and FTIR spectra (c) of oyster shell and biochar.

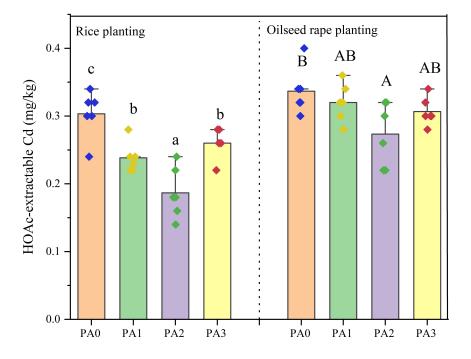


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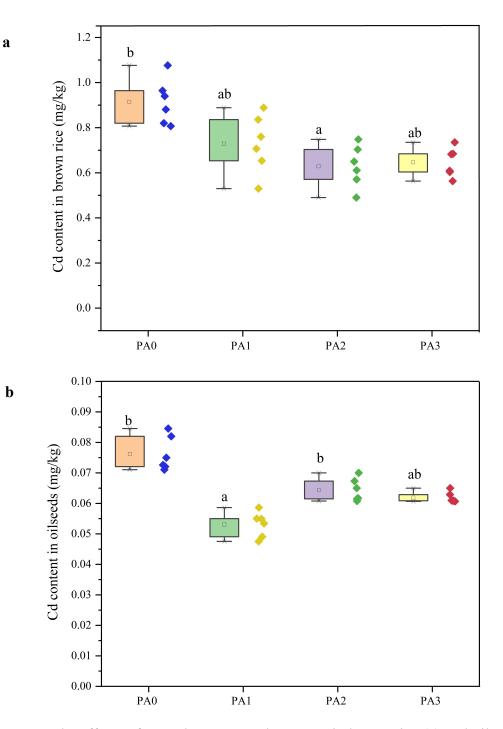


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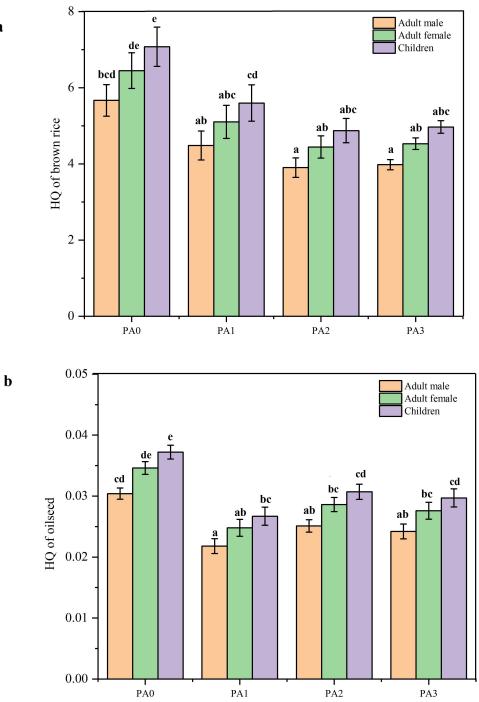
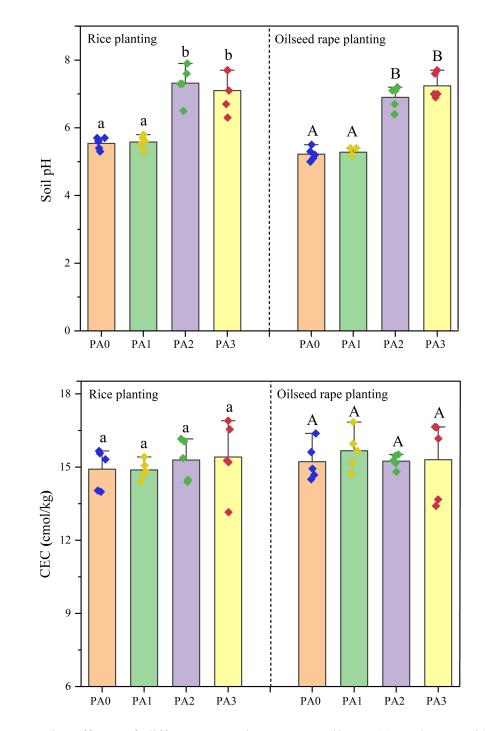


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a

b

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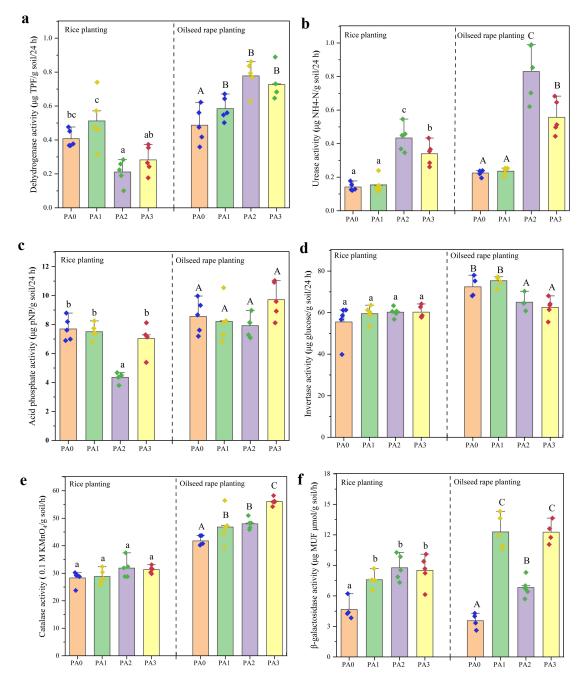


Figure 6 The effects of different amendments on the activities of soil enzyme. Dots represent the value of each sample. Bars followed with different lowercase letters (a - c) and capital letters (A-C) indicated significant ($p \le 0.05$) difference among different treatments in rice planting and oilseed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

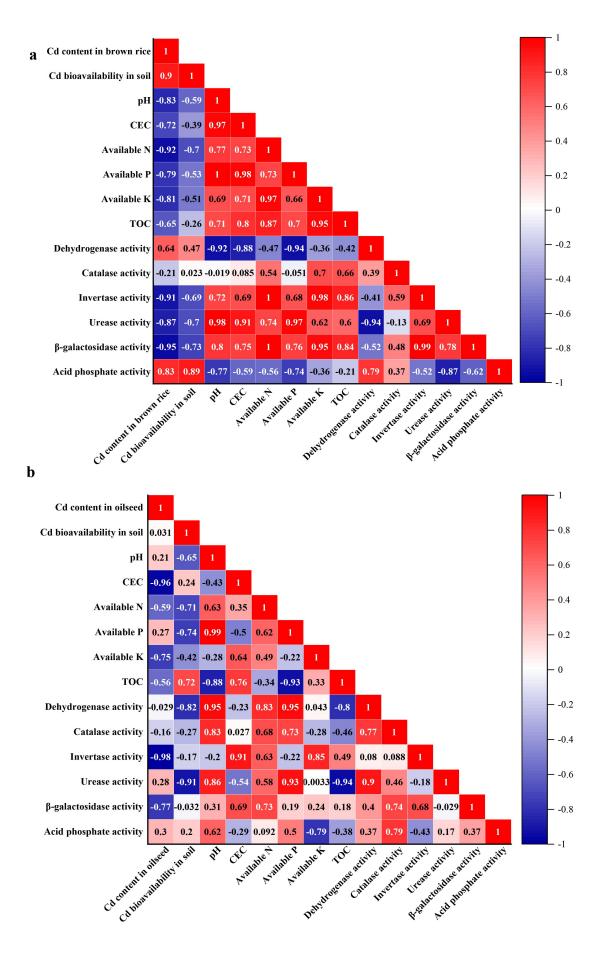


Figure 7 The correlation of investigated parameters in rice planting (a) and rice-oilseed rape planting (b).

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.