

RESEARCH ARTICLE

Investigating the mechanisms of rice husk biochar on the yield and quality of continuous cropping broad beans

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Continuous cultivation refers to planting crops continuously for multiple seasons or years on the same land, which has functions of improving soil structure, increasing soil fertility, and preventing soil erosion. However, the current continuous cultivation of broad beans faces problems such as immature cultivation techniques, unstable market demand, and insufficient germplasm resources. This study investigated the differential effects under different doses of rice husk biochar on the yield, quality, soil physicochemical properties, and enzyme activity of continuous cultivation of broad beans. Compared with the treatment without biochar, the addition of biochar significantly improved the yield and quality of broad beans. Throughout the entire growth period, soil pH, organic matter, alkaline nitrogen, and available potassium content gradually increased with the increase of biochar application and reached their peaks at the end of the broad bean growth. The results showed that there were significant differences in pH and organic matter between different treatments, while no significant differences were observed in available nitrogen and potassium content between the 2% and 4% treatments. The available phosphorus content in soil showed a trend of first increasing and then decreasing, indicating that the application of biochar effectively improved soil acidification conditions and promoted soil nutrient accumulation. During the harvesting period of broad beans, soil enzyme activities including peroxidase, sucrase, urease, and polyphenol oxidase reached their peaks under the 2% biochar treatment with the increases of 102.90%, 109.30%, 48.77%, and 39.21% compared to the control group, respectively. The correlation analysis of broad bean yield, quality, soil physicochemical properties, and enzyme activity showed that yield was significantly positively correlated with soil alkaline nitrogen, available phosphorus, available potassium content, urease activity, and polyphenol oxidase activity. In addition, the contents of vitamin C, soluble protein, soluble sugar, and fat were significantly positively correlated with pH, organic matter, alkaline nitrogen, available phosphorus, available potassium, peroxidase activity, sucrase activity, urease activity, and polyphenol oxidase activity. The addition of rice husk biochar effectively improved the soil conditions for continuous cultivation of broad beans, increasing yield and quality.

Keywords: rice husk biochar; broad beans; yield; quality; physicochemical properties; enzyme activities.

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Introduction

Broad beans are a pivotal component of the vegetable industry in Zhejiang province, China with an annual cultivation area exceeding 1,000,000 mu, approximately 164,736 acres [1-3]. Traditionally, broad beans are known as

rotation-sensitive crops [4-6]. In recent years, the hindrance of continuous cropping on broad beans has become increasingly prevalent and severe, resulting in stunted growth, frailty, exacerbated diseases, reduced pod formation, and ultimately lowered yields. Studies indicated that the accumulation of autotoxins in the soil

after successive cultivation of broad beans was a leading cause of these cropping impediments [7, 8]. In agricultural practice, effective methods to alleviate continuous cropping obstacles primarily include soil fumigation and crop rotation [9]. Soil fumigation demonstrates broad-spectrum efficacy by eradicating harmful microorganisms from the soil, which also adversely affects beneficial microorganisms. For example, commonly used soil fumigants like methyl bromide contribute to ozone layer depletion, leading to the gradual phasing out of soil fumigation techniques. Rotation of broad beans with wheat [10], potatoes [11], and rice [12] has been validated to effectively mitigate continuous cropping obstacles. However, the cultivation of wheat, rice, and potatoes by farmers yields relatively lower benefits and is time-consuming, making crop rotation challenging to be promoted in practical agricultural production. Therefore, it is important to seek safe and effective methods to overcome the hindrances of continuous cropping on broad beans.

Biochar has been proven to improve soil fertility, promote crop yield, reduce greenhouse gas emissions, regulate soil microbial activity, and absorb heavy metals and organic pollutants [13]. Meanwhile, biochar can effectively improve the soil for continuous crop cultivation, promote crop root development, and increase soil nutrients and microbial diversity for continuous cropping [14]. In addition, biochar can also improve the fungal community structure of soil for continuous cropping, increase soil enzyme activity, reduce phenolic acid content, and promote seedling growth [15]. Currently, the application of biochar in agriculture is receiving widespread attention [16-18]. Rawat *et al.* proposed the use of mint plant biochar (MPB) as an adsorbent to remove malachite green dye from aqueous solutions. The results showed that MPB had the best removal effect at pH 6.0 with a maximum adsorption capacity of 322.58 mg/g. The adsorption process followed a pseudo-second order kinetic model. The desorption results showed that about 50% of the dye could be recovered through 1N HCl [19]. However, it

was still an important issue to optimize the dosage of biochar to maximize its effect on crop yield and quality, while improving soil conditions.

This study aimed to address the yield decline and quality damage faced by continuous cultivation of broad beans by exploring the effects of adding different doses of rice husk biochar on broad bean yield, quality, soil physicochemical properties, and enzyme activity through pot experiments, soil sample analysis, and enzyme activity determination. The research findings would be important to understand the role of biochar in sustainable agricultural development and provide theoretical support for practical applications.

Materials and methods

Broad bean seed, cultivation soil, and biochar

This study used Zhecan 1, a variety of broad beans obtained from Tianyu Dade Seed Company and produced in Zhejiang province as the research subject. This variety plays a crucial role in the vegetable industry in Zhejiang province, China with a vast annual cultivation area. The study was conducted at the Horticultural Technology Training Base of Lishui Vocational and Technical College (Lishui, Zhejiang, China) (28°28'41.26"N, 119°54'13.27"E), which is in the main production area of broad beans and has regional representativeness. The region features a subtropical monsoon climate with an average annual sunshine duration of 1,712 to 1,825 hours, an average temperature of 17.6°C, extreme maximum temperatures reaching 41.5°C, and extreme minimum temperatures of -7.7°C. The climate is warm and humid with an average annual precipitation of 1,400 to 2,275 mm, ample rainfall, and an average frost-free period of 180 to 280 days. The experimental soil was collected from the continuous cropping broad bean field in Lihe Village, Lishui City at a depth of 0 - 20 cm, representing five years of continuous cropping. The soil was classified as red soil and was air-dried and sieved through a 2 mm sieve to remove non-soil materials such as

stones, twigs, and fallen leaves. The soil pH was 5.10 with an organic matter content of 28.93 g/kg, available nitrogen content of 102.35 mg/kg, available phosphorus content of 23.46 mg/kg, and available potassium content of 39.58 mg/kg. The biochar provided by Henan Lize Environmental Protection Technology Co., Ltd was rice husk biochar carbonized at 450°C with a pH of 9.24, an organic carbon content of 284.38 g/kg, and a total nitrogen content of 2.35 g/kg. Biochar was mainly produced from biomass energy raw materials through thermal cracking, whose main component was carbon molecules.

Experimental groups and seed planting

This research consisted of four experimental groups including control group with no added biochar (CK), 1% biochar addition group (F1), 2% biochar addition group (F2), and 4% biochar addition group (F3) with 10 replicates in each group arranged in a randomized complete block design. The biochar was mixed thoroughly with the air-dried soil in proportion before the experiment. Each pot was filled with 5.5 kg of soil. The mixture was allowed to settle for one month before broad bean seeds were sown. Only uniformly plump, disease-free seeds were selected for planting with 3 seeds sown per pot and thinning to one seedling per pot after emergence. Standard cultivation practices were followed for crop management. The C-Life standardized planting management system was used in the study to achieve precise management of crops and optimal growth environment regulation. The current growth environment of crops was analyzed and calculated through standardized growth models, which could ensure that crops obtained suitable growth conditions, the temperature was maintained within the optimal range for crop growth (25 - 28°C), and the soil pH ranged from 6.0 to 6.5. The seeds were sown between March and April to ensure that crops could grow under the most suitable temperature and light conditions.

Sample collection

Soil samples were collected at three different stages including initial flowering stage (March 5),

harvest stage (April 5), and late growth stage (April 25). Three pots were randomly selected from each group for soil sampling. The soil from the cultivated layer was air-dried, ground, and sieved naturally before analyzing the physicochemical properties and enzyme activity. Additionally, broad bean pods were harvested for evaluating broad bean yield and quality indicators at the harvest stage.

Determination of broad bean quality and mass indices

Broad bean yield was determined by counting the total pods per plant. The evaluation of the quality indicators of broad beans is as follows. The determination of vitamin C content was carried out using the 2,6-dichlorodiphenol (LMAI Bio, Shanghai, China) titration method. The soluble protein content was determined using the commercially available Coomassie Brilliant Blue G-250 reagent kit and UV-Vis spectrophotometers at 595 nm. The fennel colorimetric method was used to determine the soluble sugar of broad beans using anthrone reagent [20]. The fat content was determined using ether d10 or petroleum ether 30-60 (Shanghai Baishun, Shanghai, China) [21].

Determination of soil physicochemical properties

The physical and chemical properties of soil were evaluated using the method reported previously [22]. The soil pH value was measured using the high-precision pH acid-base meter. The soil organic matter content was determined using the potassium dichromate volumetric method through titration. The soil alkaline nitrogen content was determined using the alkaline diffusion method by hydrolyzing the soil sample under 1.0 M NaOH alkaline condition to release alkaline nitrogen. The soil available potassium content was measured using FP6410 flame photometry (Shanghai Jingke, Shanghai, China) combined with 1 M NH₄OAc. The effective phosphorus content in soil was measured using the molybdenum antimony colorimetric method

Table 1. Effects of different treatments with biochar contents on broad bean yield and quality.

Groups	Yield (g)	Vitamin C (mg/100 g)	Soluble sugar (mg/100 g)	Soluble protein (mg/g)	Fat (mg/100 g)
CK	355.76 ± 5.55 ^d	1.70 ± 0.01 ^d	23.91 ± 0.10 ^d	21.98 ± 0.45 ^c	0.274 ± 0.009 ^d
F1	427.01 ± 5.57 ^c	1.90 ± 0.06 ^c	25.30 ± 0.64 ^c	24.71 ± 0.30 ^b	0.311 ± 0.008 ^c
F2	477.48 ± 4.70 ^a	2.33 ± 0.06 ^a	29.43 ± 0.16 ^a	29.35 ± 1.17 ^a	0.426 ± 0.013 ^a
F3	451.97 ± 9.56 ^b	2.06 ± 0.05 ^b	28.75 ± 0.17 ^b	28.42 ± 1.08 ^a	0.364 ± 0.014 ^b

Note: Different lowercase letters in the same row indicated significant differences among different groups ($P < 0.05$).

through 722 visible spectrophotometer (Tianjin Daman, Tianjin, China) and 0.5 M NaHCO₃ and molybdenum antimony anti-chromogenic agent.

Determination of soil enzyme activity

The determination of urease activity was achieved through the indophenol blue colorimetric method using Shimadzu UV-1800 spectrophotometer (Shimadzu, Kyoto, Japan). The catalase activity was determined through potassium permanganate titration method using 10% H₂SO₄, 0.2 M phosphate buffer (pH 7.8), 0.1 M potassium permanganate standard solution, and 0.1 M H₂O₂. The activities of sucrase and cellulase were measured by applying the 3,5-dinitrosalicylic acid colorimetric method using Shimadzu UV-1800 spectrophotometer [23].

Data analysis

Microsoft EXCEL 2020 (Microsoft, Redmond, WA, USA) was used for data organization and visualization. Data Processing System (DPS) (http://www.dpsw.cn/dps_eng/) was employed for ANOVA analysis of multiple comparisons between different treatments using Duncan's test. Additionally, SPSS (Armonk, New York, USA) was utilized for Pearson correlation analysis to assess the relationships between broad bean yield, quality, soil physicochemical properties, and soil enzyme activity. The P value less than 0.05 was defined as significant difference, while P value less than 0.01 was very significant difference.

Results

Effects of biochar on broad bean yield and quality

The broad bean yield in the F2 treatment significantly exceeded that in the F1 and F3 treatments, while all three treatment groups significantly surpassed the CK group, showing the increase of 16.75%, 30.54%, and 23.57%, respectively, compared to CK (Table 1). The addition of rice husk biochar in continuously cropped broad bean soil demonstrated a significant enhancement in broad bean quality. The broad beans in the F1, F2, and F3 treatment groups exhibited significantly higher vitamin C, soluble sugar content, soluble protein, and fat than that in the CK group ($P < 0.05$). The most notable improvements were observed in the F2 treatment with 36.93%, 23.10%, 33.55%, and 55.58% increase of vitamin C, soluble sugar, soluble protein, and fat comparing to that in the CK group, respectively. There was no significant difference in soluble protein content between the F2 and F3 treatment groups, while other indicators showed significant disparities between them ($P < 0.05$). Therefore, the addition of rice husk biochar in continuously cropped broad bean soil appeared to alleviate the harm caused by continuous cropping obstacles, significantly enhancing both the yield and quality of broad beans.

Effects of biochar on soil physicochemical properties

The soil physicochemical properties including pH and organic matter, available nutrient contents including alkaline nitrogen, available phosphorus, and available potassium in the CK group exhibited a declining trend during the growth of broad beans. In contrast, soils treated

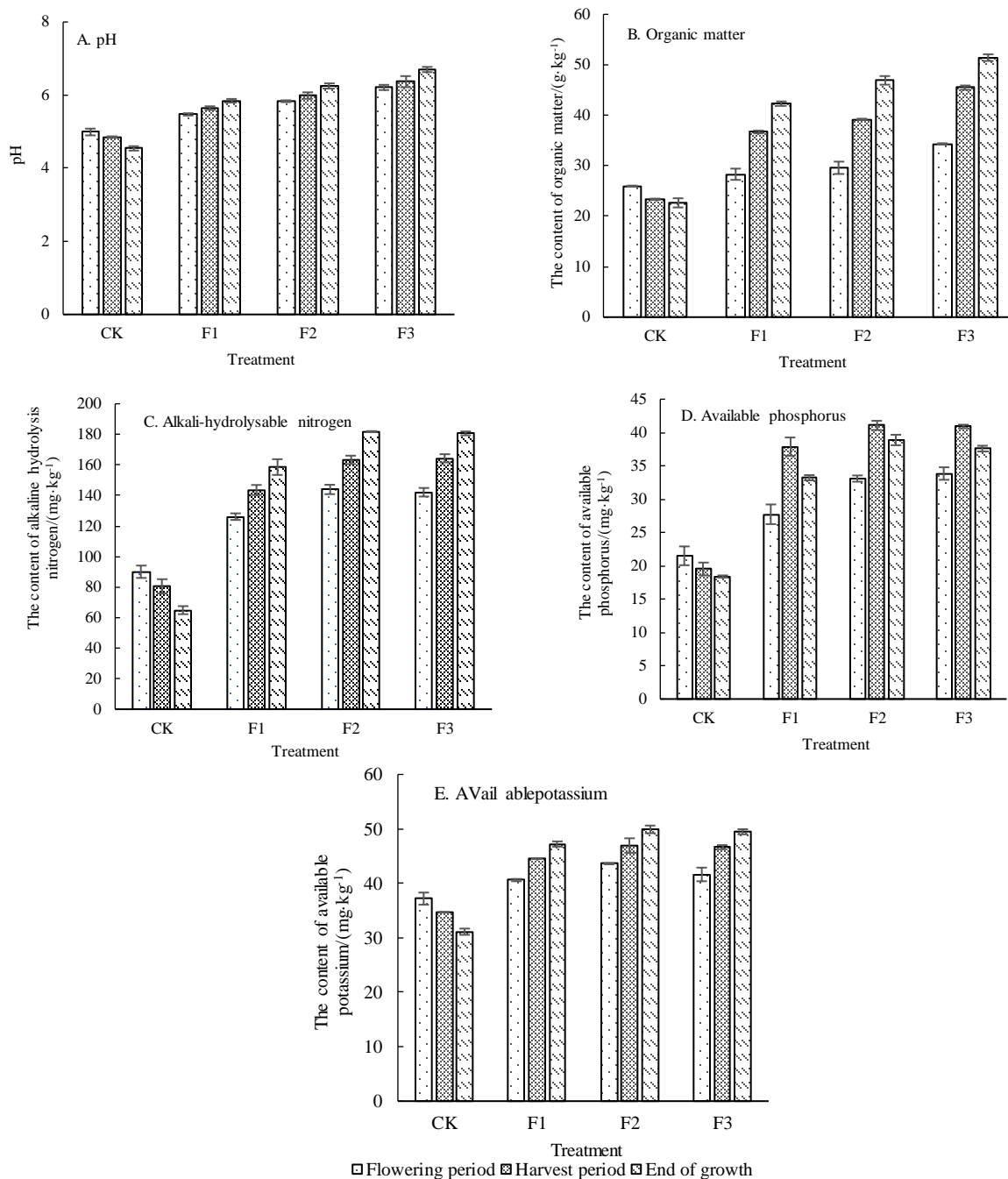


Figure 1. Soil physicochemical properties in different treatments.

with rice husk biochar showed an upward trend in soil physicochemical properties with all measured indicators significantly surpassing the CK group ($P < 0.05$). The available phosphorus displayed an initial increase followed by a decrease trend yet remaining significantly higher than the CK group ($P < 0.05$). The soil pH in the

F1, F2, and F3 treatments increased by 1.29, 1.61, and 2.16, respectively, compared to that in the CK group by the end of the broad bean growth period. The significant differences were observed among the treatments ($P < 0.05$) (Figure 1A). The soil organic matter content reached its peak at the end of the broad bean growth period with all

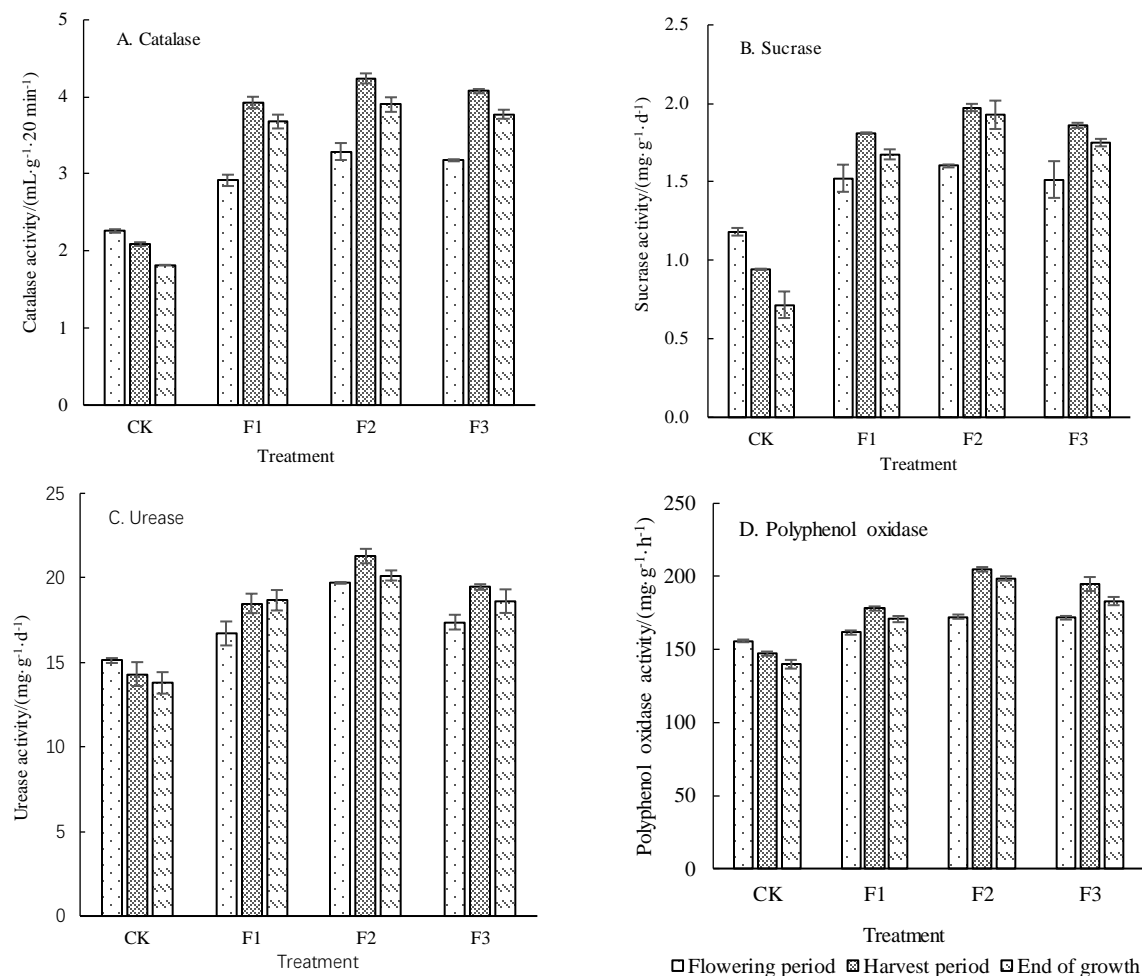


Figure 2. Soil microbial and enzyme activity in different treatments.

treatment groups increase of 86.82%, 107.18%, and 127.09%, respectively, compared to that in the control group ($P < 0.05$) (Figure 1B). The soil alkaline nitrogen, available phosphorus, and available potassium content in the F2 treatment were significantly higher than those in the CK and F1 treatment by the end of the broad bean growth period ($P < 0.05$), while no significant difference was observed between F2 and F3 treatments (Figures 1C to 1E). Additionally, the pH values and organic matter content in the CK reached the lowest points at the end of the broad bean growth period, which contrasted with the rice husk biochar treatment groups. The F2 treatment exhibited the highest levels of available nutrients nitrogen, phosphorus, and potassium, markedly surpassing the CK and F1

treatment. However, there was no significant difference between the F2 and F3 treatments. The addition of rice husk biochar effectively regulated the soil acidity in continuously cropped broad bean soil, ameliorating soil acidification conditions. Furthermore, it significantly enhanced the soil organic matter content and available nutrients with the F2 treatment showing the most favorable outcomes.

Effects of biochar on soil enzyme activity

The activities of peroxidase, sucrase, urease, and polyphenol oxidase in all treatment groups exhibited significant differences compared to that in the control group during the growth stages of broad beans ($P < 0.05$). The enzymatic activities in the CK group gradually decreased

Table 2. Correlation analysis of broad bean yield and quality and soil physicochemical properties.

Item	Yield	Vitamin C	Soluble protein	Soluble sugar	Fat
pH	0.537	0.746**	0.874**	0.876**	0.743**
Organic matter	0.508	0.713**	0.856**	0.832**	0.705*
Alkaline hydrolysis	0.600*	0.833**	0.899**	0.869**	0.814**
Available phosphorus	0.646*	0.875**	0.937**	0.916**	0.866**
Available potassium	0.593*	0.832**	0.891**	0.838**	0.807**
Catalase	0.562	0.814**	0.853**	0.805**	0.781**
Sucrase	0.570	0.825**	0.853**	0.803**	0.791**
Urease	0.667*	0.935**	0.923**	0.883**	0.912**
Polyphenol oxidase	0.677*	0.931**	0.947**	0.942**	0.918**

Note: *significant difference ($P < 0.05$). ** significant difference ($P < 0.01$).

throughout the broad bean growth stages, while the enzymatic activities in all treatment groups showed an initial increase then a decrease trend, reaching the maximum values during the harvesting period. The soil peroxidase activity reached its peak during the harvesting period with significant differences among the groups ($P < 0.05$) (Figure 2A). The F1, F2, and F3 treatments showed increases of 87.93%, 102.90%, and 94.99%, respectively, compared to that in the CK group with the F2 treatment exhibited the highest peroxidase activity by the end of the growth period, which was significantly higher than that in the F1 treatment and CK group with no significant difference to the F3 treatment. Similarly, the variation in soil sucrase activity mirrored that of peroxidase activity with the significant differences being observed among the groups during the harvesting period and the F1, F2, and F3 treatments showing increases of 91.95%, 109.30%, and 97.20%, respectively, compared to the CK ($P < 0.05$) (Figure 2B). The soil urease activity peaked during the harvesting period with the F2 treatment significantly surpassing the other groups ($P < 0.05$). The F1 and F3 treatments showed no significant differences. All treatment groups exhibited increases of 29.20%, 48.77%, and 35.99% compared to the CK group (Figure 2C). Moreover, the addition of rice husk biochar in all treatment groups showed no significant differences among them in the later growth period, but the sucrase activities of these treatments were significantly higher than that in the CK. During the harvesting period, the soil

polyphenol oxidase activities in the F1, F2, and F3 treatments peaked as 20.97%, 39.21%, and 32.37% higher than that in the CK. Significant differences among the treatments were also observed during both the harvesting and later growth periods ($P < 0.05$) (Figure 2D). The addition of rice husk biochar significantly enhanced the activities of peroxidase, sucrase, urease, and polyphenol oxidase in continuously cropped broad bean soil. Enzymatic activity reached its peak during the harvesting period, notably with the F2 treatment demonstrating the most prominent stimulation of enzyme activity.

The influence of rice husk biochar on broad bean yield and quality factors

The correlation analysis results between broad bean yield, quality, and soil physicochemical properties during the broad bean harvesting period were shown in Table 2. The yield demonstrated a significant positive correlation with soil alkaline nitrogen, available phosphorus, available potassium content, urease activity, and polyphenol oxidase activity with the correlation coefficients of 0.600, 0.646, 0.593, 0.667, and 0.677, respectively. There were no significant correlations with soil pH, organic matter content, peroxidase activity, and sucrase activity. Vitamin C, soluble protein, and soluble sugar exhibited significant positive correlations with pH, organic matter, alkaline nitrogen, available phosphorus, available potassium, peroxidase activity, sucrase activity, urease activity, and polyphenol oxidase activity with the correlation coefficients ranging

from 0.713 to 0.935, 0.853 to 0.947, and 0.803 to 0.942, respectively. Furthermore, the correlation coefficient between fat content and organic matter content was 0.705, showing a significant positive relationship. The correlation coefficients with pH, alkaline nitrogen, available phosphorus, available potassium, peroxidase activity, sucrase activity, urease activity, and polyphenol oxidase activity were 0.743, 0.814, 0.866, 0.807, 0.781, 0.791, 0.912, and 0.918, respectively, indicating highly significant positive correlations.

Discussion

The influence of rice husk biochar on yield and quality of continuous cropping broad beans

The yield and quality of broad beans are influenced by various factors. Continuous cropping can lead to soil compaction, soil acidification, decreased soil nutrients, and structural damage, hindering the normal growth and development of broad beans, resulting in decreased yield and quality. The results of this study indicated that the addition of rice husk biochar significantly enhanced the yield and quality of broad beans in continuous cropping soil with the most notable improvement observed in a 2% rice husk biochar addition. Rice husk biochar has an expansive surface area, porous structure, and carbon-rich composition, which can enhance soil structure, regulate soil pH, improve soil environmental conditions, and promote crop root growth and development to facilitate crop yield increase and quality improvement. Studies showed that adding 3% biochar increased tomato yield by 30%, while field application of 40 t/hm² biochar resulted in a 51.6% yield increase and 60 t/hm² biochar led to a 49.6% yield increase, suggesting that excessively high biochar application rates might reduce crop yield [24, 25]. Conversely, research indicated that moderate biochar application enhanced crop quality [26]. Consistent with these findings, this study demonstrated a 30.54% increase in continuous cropping broad bean yield upon adding 2% rice husk biochar, concurrently boosting broad bean vitamin C, soluble sugar,

soluble protein, and fat contents by 36.93%, 23.10%, 33.55%, and 55.58%, respectively. The enhanced nutrient absorption and utilization improved growth of roots and above-ground parts and, therefore, increased photosynthetic rates and transport of nutrients within the plant, which were likely contributing to the observed quality improvements. Guo *et al.* reported that biochar improved corn yield in relation to soil organic carbon, available phosphorus, potassium, and enzyme activity [27]. Another study confirmed that the relationship between biochar application and enhanced tomato quality was linked to improved soil water retention and increased levels of available nitrogen, phosphorus, and potassium [28]. In this study, the soil alkaline nitrogen, available phosphorus, available potassium content, and the activities of peroxidase, sucrase, urease, and polyphenol oxidase in soils treated with rice husk biochar were significantly higher than that in the control group. Correlation analysis revealed a significant positive relationship between soil available nutrient content, urease, and polyphenol oxidase activities and yield. Moreover, soil organic matter, alkaline nitrogen, available phosphorus, available potassium, peroxidase, sucrase, urease, and polyphenol oxidase activities were highly positively correlated with broad bean vitamin C, soluble protein, and soluble sugar content, providing further support for these findings.

The influence of rice husk biochar on the soil physicochemical properties of continuous cropping broad beans

The introduction of biochar into impaired soil environments effectively enhanced soil conditions. The results of this study illustrated that various treatments involving the addition of rice husk biochar significantly ameliorated the soil acidification resulting from continuous cropping. These treatments elevated soil pH levels and markedly increased organic matter content, as well as available nitrogen (N), phosphorus (P), and potassium (K) levels. Specifically, the 2% rice husk biochar treatment demonstrated a significant improvement in available phosphorus content, while the 4%

treatment excelled in enhancing available nitrogen and potassium levels. Rice husk biochar, with its elevated pH characteristics, can release more salts, oxides, and hydroxides into the soil upon application, consequently elevating soil pH. In the presence of soil microorganisms, biochar transforms into humus, enriching soil organic matter content. Additionally, biochar itself contains elements such as nitrogen, phosphorus, and potassium, which are gradually released into the soil upon application, thereby boosting available nutrient levels in the soil. Wang *et al.* demonstrated that biochar significantly enhanced acidic soil pH and increased available nutrient content, aligning well with the findings of this research [29]. Other research also indicated that biochar enhanced the retention ability of soil nitrogen and potassium elements, thereby improving crop nutrient utilization efficiency [30]. Furthermore, Zhang *et al.* suggested that high biochar application enhanced the fixation of soil nitrogen, leading to excessively high C/N ratios in the soil, which resulted in decreased microbial vitality and soil enzyme activity, subsequently lowering the content of available nitrogen, phosphorus, and potassium in the soil [31]. This study confirmed that the nutrient levels in the soil under the F3 treatment did not significantly differ from those in the F2 treatment.

The influence of rice husk biochar on the enzyme activity of continuous cropping broad bean soil

Soil enzyme activity reflects the health status and quality of soil, promotes soil nutrient cycling, and directly impacts the growth and development of crops. In this study, rice husk biochar enhanced enzyme activity in continuous cropping soil with its effects linked to the dosage of biochar applied. Additionally, enzyme activity varied in strength at different growth stages of broad beans. An analysis spanning the entire growth period revealed a trend of initially increasing and subsequently decreasing of enzyme activities. The activity of catalase, closely related to soil biological oxidation capacity, played a role in mitigating the toxic effects of hydrogen peroxide

on crops [32]. The results indicated a significant impact of rice husk biochar on catalase activity in the soil, influenced by the applied amount. Across various growth stages, the F2 treatment exhibited the strongest catalase activity, while the control group showed a gradual decline in activity, affecting both the yield and quality of broad beans. Sucrase in the soil catalyzes sucrose hydrolysis. Its activity is intimately linked to soil carbon cycling with enhanced sucrase activity effectively increasing soil nutrient content [33]. Gu *et al.* showed that the application of biochar increased soil sucrase activity, which was consistent with the results of this study [34]. Urease in the soil catalyzes organic nitrogen hydrolysis. Its enhanced activity promotes the conversion of organic nitrogen into an available form. Cheng *et al.* studied the effect of biochar on the urease activity of continuous cropping facility soil and demonstrated that biochar enhanced soil urease activity, mirroring the findings of this study [35]. Polyphenol oxidase in the soil degrades phenolic substances. The increased polyphenol oxidase activity effectively catalyzes the accumulation of phenolic substances in the soil [36]. The results of this study indicated that the application of rice husk biochar to continuous cropping broad bean soil significantly enhanced soil polyphenol oxidase activity, thereby effectively alleviating the impacts of continuous cropping obstacles and improving broad bean yield and quality. Correlation analysis revealed a significant positive relationship between soil polyphenol oxidase activity and broad bean yield and broad bean quality indicators. Biochar, with its porous structure and large surface area, absorbed a significant amount of enzyme-substrate complexes in the soil, increased soil enzyme binding sites, and thereby enhanced soil enzyme activity [37]. Additionally, biochar effectively promoted soil moisture retention, regulated soil bulk density, increased soil porosity, and consequently elevated soil enzyme activity [38]. Research showed that the addition of biochar to soil enhanced soil carbon content, regulated the water-fertilizer balance system, provided suitable conditions and sufficient substrates for

soil enzyme-catalyzed reactions, and ultimately increased soil enzyme activity [39]. Tu et al. demonstrated that 1% biochar significantly increased soil catalase activity and soil urease activity. 5% biochar reduced enzyme activity due to the potential harmful effects on soil microorganisms [40]. The results of this study confirmed that 1% and 2% rice husk biochar effectively increased soil urease and soil catalase activities. 4% rice husk biochar reduced the activities of both enzymes compared to 2% biochar, corroborating the previous report. In this study, the addition of rice husk biochar promoted soil enzyme activity. The 2% rice husk biochar treatment showed the most significant enhancement in soil enzyme activity, indicating that adding 2% rice husk biochar to continuous cropping broad bean soil would be an appropriate dosage.

Conclusion

The results of this study showed that the yield and quality of continuous cropping broad beans exhibited a trend of first increasing and then decreasing with the increasing dosage of rice husk biochar and reached the peak with the application of 2% rice husk biochar. The addition of rice husk biochar significantly increased the organic matter and readily available nutrient contents in continuous cropping broad bean soil, adjusted the soil acidity resulting from continuous cropping, and enhanced soil nutrient levels. The incorporation of rice husk biochar notably enhanced the activities of catalase, sucrase, urease, and polyphenol oxidase in the continuous cropping soil and improved the soil's microecological environment, thus promoting the growth of broad beans and effectively enhancing both yield and quality.

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References

- Jensen ES, Peoples M, Hauggaard-Nielsen H. 2010. Faba bean in cropping systems. *Field Crop Res.* 115(3):203-216.
- Liu YM, Yang ZS, Dong Y. 2017. Effect of intercropping on fusarium wilt of faba bean and antioxidant enzyme activity of roots under the p-hydroxybenzoic acid stress. *J Nucl Agric Sci.* 31(5):987-995.
- Liu X, Zhang SL, Liu GH, Qiu H, Wang D, Zhang J. 2015. Soil fumigation and bio-organic fertilizer application promotes potato growth and affects soil biochemical properties in a continuous cropping system. *Acta Prataculturae Sin.* 24(3):122-133.
- Mou M, Wu FY, Xu WM. 2018. The effects of soil fumigation and biochar application on the survival rate of Sanqi seedling. *J Yunnan Norm Univ (Nat Sci Ed).* 1(38):35-39.
- Zhong ST, Lv NN, Sun YF. 2015. Screening eco-fumigants for banana orchards with serious fusarium wilt disease and their influences on soil microflora. *Soils.* 47(6):1092-1100.
- Zhang T, Yan PM, Li Y. 2013. Effects of NaCl stress on the seed germination and physiological characteristics of tobacco. *Chin Agric Sci Bull.* 29(3):116-120.
- Wei YJ, Zhang JW, Zhao YP, Wang JQ. 2014. Effects of eugenol and intercropping on root morphology of wheat and faba-bean. *J Desert Res.* 34(2):413-418.
- Hu GB, Dong K, Dong Y, Zheng Y, Tang L, Li XR. 2016. Effects of cultivars and intercropping on the rhizosphere microenvironment for alleviating the impact of continuous cropping of faba bean. *Acta Ecol Sin.* 36(4):1-10.
- Dong Y, Dong K, Zheng Y, Li T, Yang Z. 2014. Faba bean fusarium wilt (*Fusarium oxysporum*) control and its mechanism in different wheat varieties and faba bean intercropping system. *Chin J Appl Ecol.* 25(7):1979-1987.
- Yang ZS, Tang L, Zheng Y, Dong K, Dong Y. 2014. Effects of different wheat cultivars intercropped with faba bean on faba bean Fusarium wilt, root exudates and rhizosphere microbial community functional diversity. *J Plant Nutr Fertil.* 20(3):570-579.
- Dong Y, Dong K, Tang L. 2013. Relationship between rhizosphere microbial community functional diversity and faba bean fusarium wilt occurrence in wheat and faba bean intercropping system. *Acta Ecol Sin.* 33(23):7445-7454.
- Chen L, Dong K, Yang ZS, Dong Y, Tang L, Zheng Y. 2017. Allelopathy autotoxicity effect of successive cropping obstacle and its alleviate mechanism by intercropping. *Chin Agric Sci Bull.* 33(8):91-98.
- Kang YC, Yang XY, Zhang JL. 2020. Effects of ridge-sowing with plastic film mulching and rotation of broad bean on root exudates and allelopathic effects in field of continuous cropping potato. *Acta Agric Boreali-Occident Sin.* 29(8):1148-1158.
- Sun MY, Liu JH, Zhao BP, Gao Y. 2018. Effects of full film mulching and ridging planting on the rainfed potato growth and soil characteristics. *J Soil Water Conserv.* 32(5):262-269,276.
- Li L. 2009. A new farming model of bean-sweet corn-selenium-rich rice intercropping. *Shanghai Vegetables.* 2009(3):57-58.

16. Li M, Hu Y, Huang XM. 2016. Effect of biological carbon on nutrient and bacterial communities of rhizosphere soil of facility cucumber. *J Agric Mach.* 47(11):172-178.
17. Gu MY, Liu HL, Li ZQ, Liu XW, Tang GM, Xu WL. 2014. Impact of biochar application on soil nutrients and microbial diversities in continuous cultivated cotton fields in Xinjiang. *Sci Agric Sin.* 47(20):4128-4138.
18. Wang XY, Liu JQ, Hu Y, Li F, Wang R, Wang X. 2018. Effect of biochar on microorganism, nutrient content and enzyme activity of cucumber rhizosphere Soil. *J Nucl Agric Sci.* 32(2):370-376.
19. Rawat AP, Singh DP. 2018. Decolourization of malachite green dye by mentha plant biochar (MPB): A combined action of adsorption and electrochemical reduction processes. *Water Sci Technol.* 77(5-6):1734-1743..
20. Wang XK. 2008. Principles and techniques of plant physiological and biochemical experiments. Beijing: Higher Education Press. Beijing, China.
21. Du ZX, Liu XN. 2017. Quality inspection of horticultural products. Beijing: China Agricultural Publishing House. Beijing, China. 2017:51-52.
22. Lu RK. 2000. Methods for agrochemical analysis of soil. Beijing: China Agricultural Science and Technology Press. Beijing, China.
23. Guan SY. 1986. Soil enzyme and research methods. Beijing: Agricultural Press. Beijing, China.
24. Akhtar SS, Li GT, Andersen MN, Liu F. 2014. Biochar enhances yield and quality of tomato under reduced irrigation. *Agric Water Manag.* 138:37-44.
25. Li CJ, Qu ZY, Gou MM, Su Y, Huo X. 2014. Effects of biochar amendment on soil water and nutrient utilization efficiencies and tomato growth. *J Agro-Environ Sci.* 33(11):2187-2193.
26. Guo LL, Yu HW, Kharbach M, Zhang W, Wang J, Niu W. 2021. Biochar improves soil-to- mato plant, tomato production, and economic benefits under reduced nitrogen application in northwestern China. *Plants.* 10(4):759.
27. Guo SY, Shang S, Zhang Y. 2022. Effects of biochar application after five years on soil biochemical properties and summer maize yield. *Soils Crop.* 11(3):290-297.
28. Agbna G, Ali AB, Basheer AK, Eltoum F, Hassan MM. 2017. Influence of biochar amendment on soil water characteristics and crop growth enhancement under salinity stress. *Int J Eng Works.* 4:2409-2770.
29. Wang KY, Guan HL, Lu J, Jun L. 2020. Effects of biochar on physicochemical properties of dry land acid red soil. *Soils.* 52(3):503-509.
30. Cui H, Wang LX, Ou Y, Yan B, Han L, Li Y, *et al.* 2019. Effect of the combined application of bio- char and chemical fertilizer on the migration and transformation of nitrogen and phosphorus in paddy soil. *J Agro-Environ Sci.* 38(2):412-421.
31. Zhang RH, Lan CJ, Liu W, Jin Q, Guo Y, Yu JH, *et al.* 2019. Effect of biochar on growth, yield and quality of open-field cherry tomato in counter season. *Mol Plant Breed.* 17(14):4831-4839.
32. Su RL, Zhao BQ, Zhu LS, Xu J, Zhang FD. 2003. Effects of long-term fertilization on soil enzyme activities and its role in adjusting-controlling soil fertility. *Plant Nutr Fertil Sci.* 9(4):406-410.
33. Liu SX, Ding FH, Chen WX, Xu GF, Cheng J. 2014. Effects of organic fertilizers on soil nutrients and enzyme activities of continuous cropping soil of asparagus bean. *Zhejiang Agric Sci.* 26(3):770-774.
34. Gu MY, Ge CH, Ma HG. 2016. Effects of biochar application amount on microbial flora and soil enzyme activities in sandy soil of Xinjiang. *Agric Res Arid Areas.* 34(4):225-230, 273.
35. Cheng XY, Lan Y, Ren XF. 2017. Effects of biochar on enzymatic activities and root characteristics of cucumber in continuous cropping soil of greenhouse. *J Shenyang Agric Univ.* 48(4):418-423.
36. Martine-Toledo MV, Delarubia T, Moreno J, Gonzalez-Lopez J. 1988. Root exudates of zeamays and production of auxins, gibberllins and cytokinins by azotobacter chroococccum. *Plant and Soil.* 110(1):149-152.
37. Du Q, Huang R, Li B. 2021. Effect of biochar returning on labile organic carbon and enzyme activity in tobacco-growing soil. *J Nucl Agric Sci.* 35(6):1440-1450.
38. Oleszczuk P, Josko I, Futa B, Pasiieczna-Patkowska S, Pałys E, Kraska P. 2014. Effect of pesticides on mi- croorganisms, enzymatic activity and plant in biochar-amended soil. *Geoderma.* 214/215:10-18.
39. Wang Y, Hu Y, Ma YH, Li JX. 2020. Effect of biochar addition on soil available cadmium and enzyme activities. *Chin J Soil Sci.* 51(4):979-985.
40. Tu C, Wei J, Guan F, Liu Y, Sun Y, Luo Y. 2020. Biochar and bacteria inoculated biochar enhanced Cd and Cu immobilization and enzymatic activity in polluted soil. *Environ Int.* 137:105576.