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Research article

Effects of applying peanut shell and its biochar on the microbial activity and community structure of dryland red soil

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ABSTRACT

Due to its soil formation process, dryland red soil has certain characteristics that are unfavorable for crop growth, including acidity, fineness, plate structures, and erosivity. The use of large amounts of fertilizer can decrease fertility and biodiversity and increase acidification, thereby seriously restricting the sustainable utilization of dryland red soil resources. Therefore, there is an urgent need for techniques that improve the crop quality and yield in dryland red soil areas. Returning crop waste to fields as fertilizer is a promising approach to sustainable agriculture. In the present study, the effects of applying peanut shell and an associated biochar product to dryland red soil were investigated, with a focus on soil microbial activity and community structure. Field experiments were conducted in Jiangxi Province, southern China, in 2020, in field plots of sweet potato crops. Seven treatments were set up according to the principle of equal carbon return to farmland: Control: (conventional fertilization); S1, S2, S3 (peanut shell application of 3000, 4500 and 6000 kg hm^{-2}, respectively); and BC1, BC2, BC3 (peanut shell biochar application of 1000, 1500 and 2000 kg hm^{-2}, respectively). The application of peanut shell and its biochar improved soil basal respiration, with the greatest increase relative to controls of 161.06% found in treatment S3 at the root harvest stage. The most obvious increase in microbial biomass carbon content due to biochar application was 206.50% in treatment BC2 at the root harvest stage. The application of peanut shell and its biochar increased the phospholipid fatty acid (PLFA) contents of total soil microorganisms and different microbial groups. The maximum increases in the PLFA contents of total soil microorganisms, gram-positive bacteria, and gram-negative bacteria occurred at the early root formation stage in treatment BC2, which were 112.16%, 102.52%, and 115.64%, respectively. Both peanut shell and biochar increased the PLFA contents of soil actinomycetes, arbuscular mycorrhizal fungi (AMF), and other fungi to certain extents. The soil actinomycetes PLFAs increased by 120.08% at the early root formation stage in BC2, while the AMF PLFAs increased by 79.44% at the seedling stage in S2. This study provides theoretical and practical guidance for the comprehensive utilization of peanut shell and the implementation of circular agriculture in dryland red soil regions. It also provides a scientific basis for improving the fertility of dryland red soil.

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1. Introduction

Microorganisms are a vital component of the soil. They participate in most biochemical reactions in soil systems and play a key role in soil formation, organic matter metabolism, nutrient transformation, energy flow, and circulation [1–3]. Soil basal respiration (SBR) represents the immediate activity of soil microorganisms. Soil CO_2 release is closely related to the metabolism and community composition of soil microbes [4]. Soil microbial biomass carbon (MBC) is a critical component of the soil's active carbon pool [5]. It can reflect the abundance of soil microorganisms and helps promote soil microbial activity [6]. MBC also aids in soil nutrient transformation and organic matter decomposition. Soil microorganisms are sensitive to changes in the external environment, as well as to changes to soil substrate availability, temperature, humidity, physical and chemical properties, and above-ground vegetation [7]; [46]; [8].

Straw returning is an important type of soil modification. It reuses crop waste that would otherwise be incinerated, thereby providing active organic carbon to the soil and avoiding environmental damage. At the same time, it provides more available carbon for microbial growth and reproduction, which increases the number and activity of microorganisms [9]. Biochar is a solid substance produced by the pyrolysis (<700 °C) of crop straw and other biomasses under complete or partial hypoxic conditions. It is insoluble, stable, and highly aromatic. Due to its structural characteristics and physical and chemical properties, biochar can improve soil structure, enhance soil water retention and fertilizer and microbial activity, and acts as a good soil modifier [10]; [42]; [5,11,12]. Prayogo et al. (2014) found that biochar can significantly increase the number of bacteria and actinomycetes in soil. Xu et al. [13] found that soil bacterial diversity increased according to the dose of biochar added. Wang et al. [14] found that adding biochar to red soil decreased the phospholipid fatty acid (PLFA) content of gram-positive bacteria and fungi. Ameloot et al. [15] found that biochar can provide a suitable habitat for soil bacteria, thus stimulating changes in soil bacterial function and community diversity. Other studies have pointed out that biochar affects soil microbial activity and community structure because it changes the physical and chemical properties of the microbial colonization habitat [5,16].

In recent years, research has primarily focused on the effects of different returning methods and the types and amounts of returned biochar or straw on soil microorganisms [17,18]; [42]; [5,19]. However, there have been few reports on the effects of applying both biochar and straw on the microbial activity and community structure of dryland red soil. Due to the different forms of organic carbon and the physical and chemical properties of straw and its resulting biochar, it is expected that returning both products rather than a single one may have different effects and mechanisms of soil fertility enhancement. The carbon in straw is readily utilized, while the carbon in biochar is difficult to decompose. Hence, we expect them to have different effects on soil microorganisms. In this study, a field experiment was conducted at the Jiangxi Institute of Red Soil where peanut shell and its corresponding biochar were returned to a cropping field containing dryland red soil. The effects on the soil microbial community and its activity were investigated by measuring the SBR, MBC, and contents of microbial PLFAs. This study aimed to provide a basis for improving the fertilization of dryland red soil with consideration of efficiency, safety, environmental protection, and sustainable development.

2. Materials and methods

2.1. Overview of the study area

The study plot was located at the Jiangxi Institute of Red Soil (116° 20′ 24″ E, 28° 15′ 30″ N). The terrain of this area has low hills with an elevation of 25–30 m and a slope of 5°. It has a mild subtropical humid climate with abundant rainfall and sufficient sunshine. The annual average temperature is 17.7–18.5 °C, the annual average rainfall is 1537 mm, and the annual evaporation is 1100–1200 mm.

2.2. Materials

The soil was derived from Quaternary red clay classified as Udic Ferralsols. The basic physical and chemical properties of the soil layer at depths of 0–15 cm were: pH = 4.56, soil organic carbon = 7.98 g kg^{-1}, total N = 0.97 g kg^{-1}, total P = 0.46 g kg^{-1}, and total K = 6.86 g kg^{-1}.

Peanut shells and biochar were purchased from Shangqiu Sanli New Energy Company (Henan Province, China). The shells were dried, crushed, and put through a 2 mm sieve for later use. The basic physical and chemical properties of the peanut shells were: organic carbon = 396.97 g kg^{-1}, total N = 10.76 g kg^{-1}, total P = 0.85 g kg^{-1}, and total K = 14.91 g kg^{-1}. Biochar was prepared as follows: Smashed peanut shells were oven-dried at 80 °C until they reached a constant weight. Biochar was produced by a slow pyrolysis procedure under N_2 protection. The pyrolysis procedure used a slow heating rate of 8 °C/min until reaching the final temperature of approximately 500 °C, which was maintained for 2 h. The basic chemical properties of the biochar were: pH = 10.35, organic carbon = 467.18 g kg^{-1}, total N = 5.90 g kg^{-1}, total P = 14.45 g kg^{-1}, and total K = 11.58 g kg^{-1}. Its physical characteristics were determined using Transmission electron microscope (TEM)Fig. S1, Fourier Transform Infrared Spectroscopy (FTIR, Fig. S2), X-Ray Diffraction (XRD, Fig. S3), and elemental distributions (Table S1).

2.3. Experimental design

In general, straw-returning strategies use peanut shell and biochar (carbonization rate 30–40%) in equivalent carbon amounts. The present study compared seven treatments of soil used to grow sweet potato: (1) conventional fertilization (control), (2) 3000 kg hm^{-2}

peanut shell (S1), (3) 4500 kg hm^{-2} peanut shell (S2), (4) 6000 kg hm^{-2} peanut shell (S3), (5) 1000 kg hm^{-2} peanut shell biochar (BC1), (6) 1500 kg hm^{-2} peanut shell biochar (BC2), and (7) 2000 kg hm^{-2} peanut shell biochar (BC3). Each treatment was applied to four 5 × 6 m plots, with all 28 plots being randomly distributed in space. Peanut shell or its biochar were spread on the surface of the soil and ploughed in evenly. The experiment was conducted from May 10 to October 30, 2020. In the seedling stage, early root formation stage, root enlargement stage, and root harvest stage, fresh soil from depths of 0–15 cm was randomly collected from each plot with a soil drill after the removal of surface litter. Particles such as residual roots were removed and screened through a 2 mm sieve. Part of each soil sample was stored in a refrigerator at 4 °C, and another part was dried naturally for later use.

3. Methods

SBR was measured using the alkali absorption method. Briefly, 10 g of soil/waste from each sample was incubated in a 1 L glass jar containing 0.1 M NaOH solution in a vial to absorb the CO_2 produced during incubation. After 24 h of incubation at a temperature of 28 °C, 2 mL of 1 M $BaCl_2$ was added to the NaOH, and the excess hydroxide was titrated with 0.05 M HCl in the presence of phenolphthalein indicator [20]. Soil MBC was extracted by chloroform fumigation (K_2SO_4) and determined by total organic carbon (TOC) analyzer as follows. A 5.0 g soil sample was weighed in two different containers. The first set of containers received 40 mL of 0.05 M K_2SO_4 solution and was shaken for 1 h. The samples were centrifuged at 4000 rpm for 5 min before being filtered through a Whatman #42 filter. The other set of samples was fumigated in a desiccator with chloroform for 24 h and then extracted following the same procedure as the non-fumigated samples. Fumigated and non-fumigated K_2SO_4 extracts were acidified with 0.2 mL 1 M phosphoric acid and analyzed for total organic C and N using a TOC analyzer [21]. The soil microbial PLFA content was determined using improved Bligh and Dyer methods: 5.0 g of dry soil was put in a Teflon centrifuge tube and the lipids extracted with chloroform-methanol-phosphoric acid (1:2:0.8). The PLFAs were separated and purified by a silica gel column. After methylation and cleaning, the samples were dissolved in 200 μL n-hexane. The PLFA content of the microorganisms was determined using a gas chromatograph (7890A). A 19:0 ratio of the PLFA contents of different microorganisms to the concentration of an internal standard was used to calculate the microbial biomass and relative abundance [8].

3.1. Data analysis

The data were analyzed and plotted using Microsoft Excel 2013. One-way ANOVA tests were performed using SPSS 19.0. Differences between treatments were tested by LSD multiple comparisons, with the significance level set at $p < 0.05$.

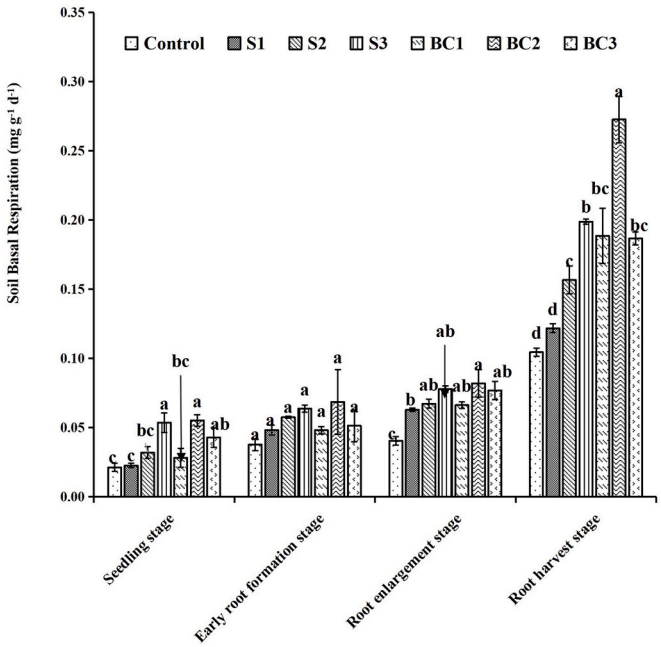

Fig. 1. Effects of peanut shell and its biochar application on basal respiration of dryland red soil control: conventional management; S1, S2, S3: peanut shell dosages of 3000, 4500, and 6000 kg hm^{-2}, respectively; BC1, BC2, BC3: peanut shell biochar dosages of 1000, 1500, and 2000 kg hm^{-2}, respectively. Different letters indicate a significant difference at $p < 0.05$. The same below.

4. Results

4.1. Effects of peanut shell and its biochar on soil microbial activity in dryland red soil

4.1.1. Effects on SBR

SBR increased with time in all treatments and peaked at the root harvest stage. SBR was higher in all treatments than in controls. It increased with peanut shell dosage, while it increased and then decreased with biochar dosage (Fig. 1). In the seedling stage, the SBR of treatments S3, BC2, and BC3 was significantly higher than that of the control by 152.20%, 159.52%, and 102.05%, respectively ($p < 0.05$). In the root enlargement stage, the SBR of treatments S1, S2, S3, BC1, BC2, and BC3 was significantly higher than that of the control by 55.48%, 66.34%, 92.73%, 63.87%, 102.71%, and 90.22%, respectively. Finally, in the root harvest stage, the soil basal respiration of treatments S2, S3, BC1, BC2, and BC3 was significantly higher than that of the control by 49.95%, 90.34%, 80.58%, 161.06%, and 78.72%, respectively.

4.2. Effects on soil MBC

The content of soil MBC in all treatments decreased and then increased, peaking at the seedling stage. Application of peanut shell and its biochar increased soil MBC to a certain extent. The MBC increased with peanut shell dosage and decreased with biochar dosage, being higher in all treatments than in the control (Fig. 2). At the seedling stage, the soil MBC of treatments S1, S2, S3, BC1, BC2, and BC3 was significantly higher than that of the control by 60.69%, 62.36%, 95.95%, 32.22%, 184.02%, and 43.45%, respectively. At the early root formation stage, the soil MBC of treatments S1, S2, S3, BC1, BC2, and BC3 was significantly higher than that of the control by 21.02%, 59.39%, 73.27%, 53.17%, 129.22%, and 69.71%, respectively. At the root enlargement stage, the soil MBC of treatments S2, S3, BC1, BC2, and BC3 was significantly higher than that of the control by 27.77%, 92.05%, 58.55%, 162.94%, and 67.14%, respectively. Finally, at the root harvest stage, the soil MBC of treatments S2, S3, BC1, BC2, and BC3 was significantly higher than that of the control by 82.86%, 183.56%, 95.72%, 206.50%, and 100.15%, respectively.

4.3. Effects of peanut shell and its biochar on the microbial community structure of dryland red soil

At the seedling stage, the PLFA content of the total soil microorganisms in treatments S2 and S3 was significantly higher than that of the control by 55.01% and 17.30%, respectively, with that of treatment S2 being significantly greater than that in all other treatments (Fig. 3A). At the early root formation stage, the PLFA content in treatments S2, S3, BC1, BC2, and BC3 was significantly higher than that of the control by 106.64%, 64.37%, 68.38%, 112.16%, and 72.70%, respectively. At the root enlargement stage, the PLFA content in treatments S1, S2, S3, and BC2 was significantly higher than that of the control by 27.41%, 41.58%, 30.26%, and 22.66%, respectively. At the root harvest stage, the PLFA content in treatments S1, S2, S3, BC2, and BC3 was significantly higher than that of the control by 24.11%, 58.25%, 49.02%, 33.68%, and 17.39%, respectively.

At the seedling stage, the PLFA content of gram-positive bacteria in treatment S2 was significantly higher than in other treatments and was 47.31% greater compared with the control treatment (Fig. 3B). At the early root formation stage, the PLFA content in treatments S2, S3, BC1, BC2, and BC3 was significantly higher than that of the control by 95.33%, 59.16%, 66.12%, 102.52%, and 67.62%, respectively. At the root harvest stage, the PLFA content in treatment S2 was significantly higher than that of the control by 21.18%.

At the seedling stage, the PLFA content of gram-negative bacteria in treatments S2 and S3 was significantly higher than that of the control by 81.22% and 31.02%, respectively (Fig. 3C). At the early root formation stage, the PLFA content in treatments S2, S3, BC1, BC2, and BC3 was significantly higher than that of the control by 108.72%, 67.38%, 63.60%, 115.64%, and 73.01%, respectively. At the root harvest stage, the PLFA content in treatment BC1 was significantly lower than that of the control by 29.46%.

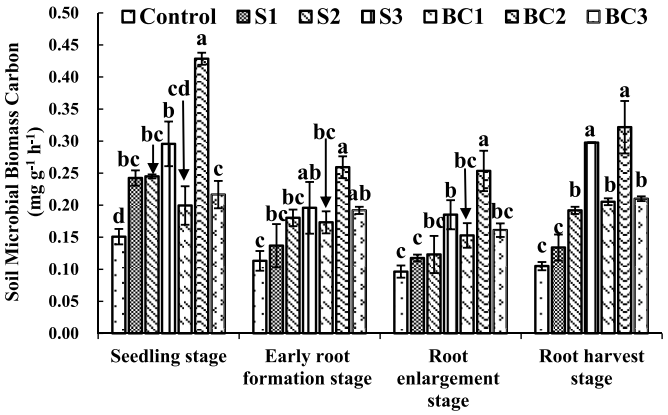

Fig. 2. Effects of peanut shell and biochar application on the microbial biomass carbon content of dryland red soil.

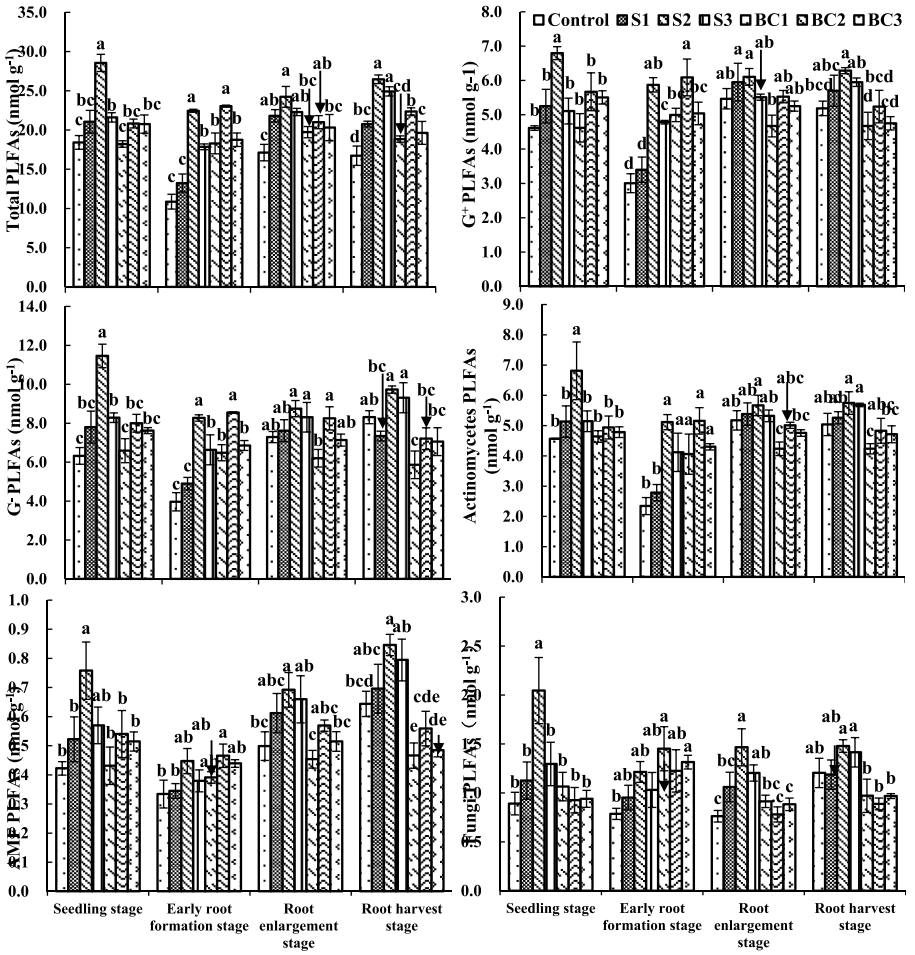

Fig. 3. Effects of peanut shell and biochar application on the PLFA contents of total soil microorganisms (A), gram-positive bacteria (B), gram-negative bacteria (C), actinomyces (D), arbuscular mycorrhizal fungi (E), and fungi (F) in dryland red soil.

At the seedling stage, the PLFA content of soil actinomyces in treatment S2 was significantly higher than that in other treatments and was 49.23% higher than that of the control (Fig. 3D). In the early root formation stage, the PLFA content in treatments S2, S3, BC1, BC2, and BC3 was significantly higher than that of the control by 118.29%, 75.75%, 73.19%, 120.08%, and 83.50%, respectively. In the root enlargement stage, the PLFA content in treatment BC1 was significantly lower than that of the control by 18.12%.

At the seedling stage, the content of soil arbuscular mycorrhizal fungal (AMF) PLFAs in treatment S2 was significantly higher than that of other treatments and was 79.44% greater than that of the control (Fig. 3E). At the early root formation stage, the content these PLFAs in treatment BC2 was significantly higher than that of the control by 39.40%. At the root enlargement stage, the AMF PLFA content in treatments S1, S2, S3, BC2 and BC3 was significantly higher than that of the control by 22.69%, 38.76%, 32.25%, 14.15%, and 3.18%, respectively. At the root harvest stage, the AMF PLFA content in treatments S2 and BC1 was significantly different from that of the control: S2 was 31.43% higher, while BC1 was 27.55% lower.

At the seedling stage, the PLFA content of soil fungi in treatment S2 was significantly higher than that of other treatments and was 129.61% greater than that of the control (Fig. 3F). At the early root formation stage, the PLFA content in treatments BC1 and BC3 was significantly higher than that of the control by 84.83% and 67.39%, respectively. At the root enlargement stage, the PLFA content in treatments S2 and S3 was significantly higher than that of the control by 92.44% and 57.99%, respectively.

5. Discussion

SBR data can reflect soil microbial activity and provide a sensitive and accurate estimate of the metabolic rates of microbes and their ability to transform and supply nutrients [22]. The results of this study show that the application of peanut shell improved SBR: it was higher in all treatments than in controls and increased with peanut shell dosage, which is consistent with the results of He et al. [23]. Soil active organic carbon increased effectively after the addition of peanut shell, and the mineralization of active carbon

components provided a source of sufficient carbon for microbial activities. As active carbon is the main source of soil microbial respiration, it thus enhanced soil basal respiration [24]. In addition, the surfaces of the peanut shells used in this experiment were broken down by a dry grinding process, which reduced the protective effects of lignin and cellulose. The easily decomposed substances in the peanut shell were released into the soil, providing more energy for microbial growth and increasing the contribution of microbial activity to soil respiration [25].

This study also found that applying biochar prepared from peanut shell can improve soil basal respiration to a certain extent: soil basal respiration was higher in all treatments than in controls and decreased with biochar dosage. This is because the organic matter in biochar can be decomposed by soil microorganisms, releasing soluble carbon and improving the effectiveness of micronutrients, thus increasing the soil respiration rate. In addition, the application of peanut shell biochar to red soil also increased soil pH, which promoted crop growth and enhanced soil respiration. Rehman et al. [26] also showed that the application of corncob biochar, cotton stalk biochar, and rice straw biochar could significantly improve the soil respiration rate, with the application of 3% biochar enhancing soil basal respiration better than 1.5% biochar. After biochar application, the content of easily oxidized organic carbon in the soil carbon pool increases and the active organic carbon can be readily used by microorganisms, thus increasing soil respiration [27]. However, in the present study, SBR decreased with increasing biochar dosage. This may be because the biochar was prepared via high-temperature anaerobic pyrolysis and contained toxic substances such as polycyclic aromatic hydrocarbons and phenols. When applied excessively to soil, biochar increases the soil's inorganic salts and concentration of toxic substances, which can affect the normal growth of microorganisms or inhibit some microbial activity, thus reducing soil respiration [28].

As an important indicator of soil microbial activity, soil MBC is a key factor in soil organic matter decomposition and nutrient transformation. It is often used to evaluate the biological characteristics of soil quality [44]. The present study showed that soil MBC content increased with peanut shell dosage, which is consistent with the results of Zhao et al. [29]. This can be attributed to the fact that peanut shell provides an abundant source of carbon and energy for microbial growth and reproduction, promotes the processes of microbial mineralization and fixation, stimulates microbial growth, and promotes the accumulation of soil MBC [30]. During the growth period of sweet potatoes, soil fertility and hydrothermal conditions are essential environmental influences on the soil's biological characteristics [31]. Suitable hydrothermal conditions enhance microbial metabolism, improve the microbial decomposition of crop straw, and generate large amounts of active organic matter, which have a cumulative effect on soil MBC content. Application of peanut shell biochar also increased soil MBC content to a certain extent. This can be attributed to its high specific surface area, loose porous structure, and strong absorbability, which allow it to store enough water and nutrients and provide a good microenvironment for microbial processes [32,33]. Moreover, because of the strong acidity of red soil, the application of biochar can improve soil pH by providing a suitable alkaline microenvironment that promotes the growth and reproduction of microorganisms [43]. Soil MBC content increased and then decreased with the biochar dosage, which is consistent with the results of Muhammad et al. [34]. It may be that biochar application introduces toxic components that, beyond a certain dosage, limit soil microbial metabolism and decrease soil MBC [35]. Therefore, the addition of excessive biochar has a negative effect on soil MBC content [36].

A *soil microbial community* refers to a biological community composed of soil bacteria and other microorganisms. Changes in the soil microbial community determine the function of a soil ecosystem [5]. PLFAs are commonly used to quantitatively characterize active soil microbial communities in complex soil environments [37]. The results of the present study show that peanut shell application increased the PLFA contents of total soil microorganisms and different soil microbial groups to certain extents. These PLFA contents increased and then decreased with increasing peanut shell dosage, being higher in all treatments than in controls. This may be because peanut shell is a source of the nutrients and organic matter needed for microbial physiological metabolism and thereby increases the soil microbial biomass. This result also indicates that peanut shell application can have significant biological effects, making it a useful way to optimize soil environments. The PLFA contents of total soil microorganisms and different soil microbial groups had different responses to biochar application. In the growth period of sweet potatoes, biochar application increased the PLFA content of total soil microorganisms, while in the harvest period, it decreased the PLFA content of different soil microbial groups. It may be that the unique structure and strong adsorption function of biochar provide a better physiological and biochemical environment for the survival of microorganisms. This may reduce competition and facilitate the reproduction of microorganisms such as actinomycetes and gram-positive bacteria [38]. At the same time, biochar can improve soil aeration conditions while its micropores can store water and soluble organic matter, thereby providing the nutrients needed by soil microbes and enhancing their physiological and metabolic activities [39]. Lehmann et al. [40] suggested that the application of biochar to soil can lead to changes in fungal communities. This may be because the lignin it contains promotes the fungal metabolism related to biomass degradation, which is conducive to the growth of certain fungal phyla. In addition to providing a suitable habitat, biochar addition also increases soil microbial biomass by promoting the metabolism of active organic compounds on the surface of biochar, which provides nutrients for microorganisms (Zhang et al., 2015). With increasing biochar dosage, the PLFA contents of total soil microorganisms and different groups of microorganisms increased first and then decreased. This may be because the structure of biochar is complex and contains polycyclic aromatic hydrocarbons, which give the biochar a high bio-inertia. At high dosages, it was difficult for the biochar to be utilized by soil microorganisms, which inhibited increases in soil microbial PLFAs.

6. Conclusions

The application of peanut shell and its biochar increased soil basal respiration and soil MBC content, and the PLFA contents of total soil microorganisms and different soil microbial groups to certain extents. It enhanced these PLFA contents most significantly at moderate dosages, while high dosages reduced them, thus changing the soil microbial community structure. The present study is based on a single year of field experiments and provided promising results. However, to better understand the long-term effects of returning

peanut shell and its biochar to the field and to determine the optimal fertilization mode for dryland red soil, longer experiments using these materials and their combinations are needed.

Declarations

Ethics approval

Not applicable.

Consent to participate

Not applicable.

Consent for publication

The manuscript has not been published before and all authors agree to publish it on Heliyon.

Availability of data and materials

All data generated or analyzed during this study are included in this published article [and its supplementary information files].

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Competing 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.

Author contribution statement

JHZ, SBZ, and XPL - Conceived and designed the experiments; JPF and JHZ - Performed the experiments; XPL, LPS, and SQX - Analyzed and interpreted the data; JHZ and SBZ - Contributed reagents, materials, analysis tools or data; WYH, JPF, and JHZ - Wrote the paper.

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