Li *et al. Annals of Microbiology* (2022) 72:15
https://doi.org/10.1186/s13213-022-01673-9

Annals of Microbiology

UNIVERSITÀ DEGLI STUDI DI MILANO

ORIGINAL ARTICLE

Open Access

# Effects of straw and straw-derived biochar on bacterial diversity in soda saline-alkaline paddy soil

Hongyu Li*, Yuying Xia, Gongliang Zhang, Guiping Zheng, Mingyu Fan and Haicheng Zhao

## Abstract

**Purpose:** In order to provide a scientific basis for the improvement of soda saline-alkaline paddy soil, the pot experiment was performed to explore the effects of rice straw and straw-derived biochar on the diversity of soil bacteria and community structure in soda saline-alkaline soil.

**Methods:** The experiment was four gradients of straw return (3 (RS1), 7.5 (RS2), 12 (RS3), and 16.5 (RS4) t/hm$^2$) and four gradients of biochar return (3 (RB1), 7.5 (RB2), 12 (RB3), and 16.5 (RB4) t/hm$^2$), using 0 t/hm$^2$ as a control (CK). After 5 consecutive years of measuring straw returns, high-throughput sequencing was used to determine the relative abundance, alpha diversity, and changes in the community structure of soil bacteria.

**Result:** Our results demonstrated that straw return significantly increased the relative abundance of Bacteroidetes, Firmicutes, and *Sphingomonas* and significantly reduced the relative abundance of Acidobacteria, Actinobacteria, Gemmatimonadetes, Parcubacteria, *Anaeromyxobacter*, *Pontibacter*, *uncultured_bacterium_f_Draconibacteriaceae*, and *Bryobacter*. Straw-derived biochar return significantly increased the relative abundance of *uncultured_bacterium_f_Draconibacteriaceae* and significantly reduced the relative abundances of Actinobacteria, Gemmatimonadetes, *Thiobacillus*, and *Anaeromyxobacter*, indicating that both straw and its associated biochar return changed the relative abundance of the phyla and genera of some bacteria. Straw return affected bacteria phylum and genus more than straw-derived biochar. With the exception of the 16.5 t/hm$^2$ straw return, which reduced bacterial richness, the treatments did not significantly impact alpha diversity. Compared with straw-derived biochar return, straw return significantly changed the bacterial community structure, and the higher the straw return, the higher the impact on the bacterial community structure. Redundancy analysis (RDA) demonstrated that there was a significant correlation between the physicochemical properties of the soil and the community structure of its bacteria. A Mantel test demonstrated that the content of available phosphorus, available potassium, and organic matter was all important environmental factors affecting community structure.

**Conclusion:** We speculate that straw return regulates the physicochemical properties of the soil, which affects the bacterial community structure.

**Keywords:** Straw, Biochar, Soda saline-alkaline paddy soil, Bacterial diversity

China is a large agricultural country with one of the highest abundances of straw in the world (Zeng et al. 2007). Its straw output is increasing, and the average annual growth rate of straw is approximately 4% over the past few decades (Hong et al. 2016). At present, burning straw is a serious problem because it is a waste of resources,

*Correspondence: ndrice@163.com
College of Agronomy, Heilongjiang Bayi Agricultural University/Key Laboratory of Modern Agricultural Cultivation and Crop Germplasm Improvement of Heilongjiang Province, Daqing 163000, Heilongjiang, China

pollutes the environment, and perturbs the ecological balance of farmland. A major research topic is how to use straw resources to develop new industries that provide ecological and environmental protections, save energy and reduce emissions, and develop sustainable agricultural practices. Straw is rich in carbon, nitrogen, phosphorus, and other nutrients, so straw return can increase the content of soil organic matter, improve soil fertility (Sommer et al. 2011; Turmel et al. 2014), promote the growth of soil microorganisms (Miura et al. 2016), and increase crop yield (Wang et al. 2015). However, excess straw return negatively impacts crop production and soil quality, such as reducing soil nitrogen levels (Shindo and Nishio 2005) and causing problems with disease and weeds (Su et al. 2016; Tardy et al. 2015) In recent years, a new method of straw return has attracted increasing attention: biochar. Biochar is a type of high-carbon solid material with rich pores and an aromatic hydrocarbon structure that can be generated by subjecting agricultural and forestry waste to high temperatures under low oxygen or anaerobic conditions (Cayuela et al. 2013). Due to the unique physical and chemical properties of biochar, it is widely used in pollutant adsorption and soil quality improvement (Gul et al. 2015; Gunes et al. 2016; Xie et al. 2014).

Microorganisms are responsible for maintaining the stability of a soil's ecosystem (Gul et al. 2015) and are sensitive to changes in soil properties. As such, they are used to evaluate soil quality (Marschner et al. 2013). As one of the most abundant microbial species in the soil, bacteria play important functional roles in a variety of soil ecological processes, such as decomposing organic matter and promoting the mineralization of soil nutrients (Hamm et al. 2016). Studies showed that both carbonization and straw return increase the number of bacteria, actinomycetes, and physiological microbe flora in the soil (Gu et al. 2016). Biochar is rich in nutrients, and the unique structure can provide a favorable breeding ground for soil bacteria. This makes biochar conducive to bacterial growth, increasing its relative abundance (Rillig et al. 2010). There are numerous studies assessing the effects of straw and biochar return on soil bacteria; however, its impact on bacterial diversity is often closely related to the raw materials, the return levels of straw and biochar, and the soil type. Bai et al. (2020) found that carbonization return can increase the alpha diversity of soil bacteria in sandy loam soil ($pH = 8.51$), but that straw return has no significant effect on the alpha diversity of soil bacteria. The biochar of wheat and corn stalks return can increase the diversity of weakly acidic rice soil and black soil, respectively (Yao et al. 2017; Zheng et al. 2016). Straw return increased the alpha diversity of bacteria in acidic soil (Bu et al. 2020) but had no significant impact on the alpha diversity of bacteria in alkaline soil (Sun et al. 2015; Yu et al. 2018; Zhang et al. 2021).

The mechanism behind how straw and straw-derived biochar return affect soil bacterial communities is currently unclear. There is little research on how rice straw and carbonization return affect soil bacterial community diversity and structure in soda saline-alkali soil for many years. Therefore, we explored the effects of different amounts of rice straw and its straw-derived biochar on soil bacterial community diversity over several years, providing a theoretical reference for the practical application of straw agriculturalization.

## Materials and methods

### Test site and materials

This experiment was performed in the potted experimental fields (26° 10′ N, 119° 23′ E) of Heilongjiang Bayi Agricultural University, from 2014 to 2018. Daqing City is in the northeast semi-humid, semiarid, grassland-meadow saline area. The soil types are primarily soda-alkalized meadow soil, swampy meadow soil, and soda-salinized meadow alkaline soil. The salinization of this soil is accompanied by an alkalization process. The average annual sunshine is 2726 h, the average annual frost-free period is 166 days, the average annual temperature is 4.2 °C, and the average annual summer temperature is 23.2 °C. The daily temperature difference during the growth and development period of crops exceeds 10 °C, the average annual precipitation is 427.5 mm, and the average annual evaporation is 1635 mm. The tested variety was Kenjiandao 5, which had 12 leaves on the main stem, was 87–90 cm in height, and possessed ≥ 10 °C active accumulated temperature of 2450–2500 °C. The test soil was taken from the 0–15 cm soil layer of soda-salinized meadow alkaline soil in Heilongjiang Bayi Agricultural University in 2014. The physical and chemical properties of the soil are shown in Table 1.

### Experimental design

The biochar material, rice straw, was purchased from Liaoning Jinhefu Agricultural Development Co., Ltd. It had pH 9.04 and was comprised of 56.61% carbon, 13.60% nitrogen, and 21.07% ash. We performed a pot experiment, with single factor complete randomized design at nine levels including a control 0 (CK), and annual straw

**Table 1** Nutrient content of experimental soil (2014)

| pH value | Organic matter (g/kg) | Total salt content (%) | Available nitrogen (mg/kg) | Available phosphorus (mg/kg) | Available potassium (mg/kg) |
|---|---|---|---|---|---|
| 8.80 | 18.00 | 0.50 | 98.75 | 12.10 | 193.70 |

return 3.0 (RS1), 7.5 (RS2), 12 (RS3), and 16.5 (RS4) t/hm$^2$, respectively, and annual straw-derived biochar return 3.0 (RB1), 7.5 (RB2), 12 (RB3), and 16.5 (RB4) t/hm$^2$, respectively. The test was performed three times, with four pots each time. In the spring of the first year, the saline-alkaline soil was dried, crushed, and mixed, after which 12 kg of saline-alkaline soil was mixed with quantitative biochar and 5 cm rice straw, respectively. It was then placed into a pot (height 30.5 cm, inner diameter 30 cm), and the base fertilizer was buried 10 cm deep in the soil layer. Water was added until it stabilized, after which it was stirred. There were four hills per hot and three seedlings per hill. Following the annual harvest, the soil from each pot was maintained in its original state. The next spring, it was crushed and mixed with biochar and straw and returned to the original pot. The amount of biochar and straw and the management measures were the same for each treatment every year, and the biochar and straw were continuously returned for 5 years. The fertilizers applied were urea, ammonium sulfate, diammonium phosphate, and potassium sulfate. The application rates of basal fertilizers N, P, and K were 39.6 (N), 69 ($P_2O_5$), and 42 ($K_2O$) kg/hm$^2$, respectively; the application rates of tillering fertilizer and regulating fertilizer N were 28.35 kg/hm$^2$ and 9.35 kg/hm$^2$, respectively, and the application rates of panicle fertilizer N and K were 14.39 (N) and 28.5 ($K_2O$) kg/hm$^2$, respectively.

## Method

### *Soil sampling*

In mid-June (rice tillering period) of 2018 (the 5th year of continuous straw and biochar return), soil samples from the 0–10 cm soil were collected, with three repetitions for each. Each repetition was a mixture of three samples from each pot of the four pots. Each soil sample was divided into two parts. One part was placed into a sterile airtight bag and was immediately placed in liquid nitrogen, brought back to the laboratory, and stored at −80 °C for subsequent analysis of the soil bacteria diversity. Another part of the sample was air-dried in a dark and ventilated place to remove gravel blocks and plant residues and was passed through a 2-mm sieve to determine the soil nutrient content.

### *Extraction of total soil DNA, PCR amplification, and high-throughput sequencing of bacterial 16S rRNA sequences*

We weighed 0.5 g of a soil sample, used a PowerSoil® DNA Isolation Kit to perform DNA extraction, and used agarose gel electrophoresis and a spectrophotometer to detect the purity, concentration, and integrity of the DNA. We then amplified the V3 + V4 region of bacterial 16S rRNA with primers 338F (5′-ACTCCTACG GGAGGCAGCA-3′) and 806R (5′- GGACTACHVGGG TWTCTAAT-3′). The reaction system for the first step of PCR amplification was 50 μl, and the reaction procedure was as follows: 95 °C pre-denaturation 5 min; 15 cycles (including 95 °C, 1 min; 50°C, 1 min; 72°C, 1 min); 72 °C, 7 min; and kept warm at 4 °C. The PCR product was then purified with magnetic beads. The second reaction was 40 μl, and the reaction procedure was as follows: 98 °C pre-denaturation for 30 s; 10 cycles (including 98 °C, 10 s; 65 °C, 30 s; 72 °C, 30 s); and 72 °C, 5 min. The PCR product of the second step was purified with magnetic beads and then quantified by NanoDrop 2000, and the samples were mixed according to a mass ratio of 1:1. The library was sequenced using the Illumina Hiseq 2500 platform (Illumina Corporation, USA) with a 2 × 250 bp paired-end sequencing strategy. The total DNA extraction and sequencing of soil microorganisms were performed by Beijing Biomarker Biotechnology Co., Ltd.

### Data processing

We used the USEARCH method to cluster the effective tags of each sample, and cluster sequences with 97% sequence similarity were used to generate operational taxonomic units (OTUs). We used Mothur (version v.1.30) software to analyze the alpha diversity of the samples. R software was used to perform principal coordinate analysis (PCoA) of OTU abundance and study the differences in community structure between different treatments. We used redundancy analysis (RDA) to study the relationship between the physicochemical properties of the soil and its microbial community structure. Excel 2003 was used for data sorting, and GraphPad prism 6.02 software was used to complete the drawing. DPS v7.05 software was used for variance analysis, and the Duncan's test was applied to identify the significance among the various treatments.

## Results and analysis

### Comparison of the relative abundance of soil bacteria

The top ten bacterial phyla in relative abundance are Proteobacteria, Chloroflexi, Bacteroidetes, Acidobacteria, Actinobacteria, Gemmatimonadetes, Ignavibacteriae, Firmicutes, Verrucomicrobia, and Parcubacteria. In this study, we analyzed the bacteria at the phylum level that significantly differed in relative abundance following straw and straw-derived biochar return (Fig. 1). After straw return, the relative abundance of Bacteroidetes and Firmicutes first increased and then decreased, with both reaching their maximum in the RS3 treatment, which were 42.28% and 318.25% higher than the control. The relative abundance of Acidobacteria, Gemmatimonadetes, and Parcubacteria first decreased and then increased, while the relative abundance of them in RS2

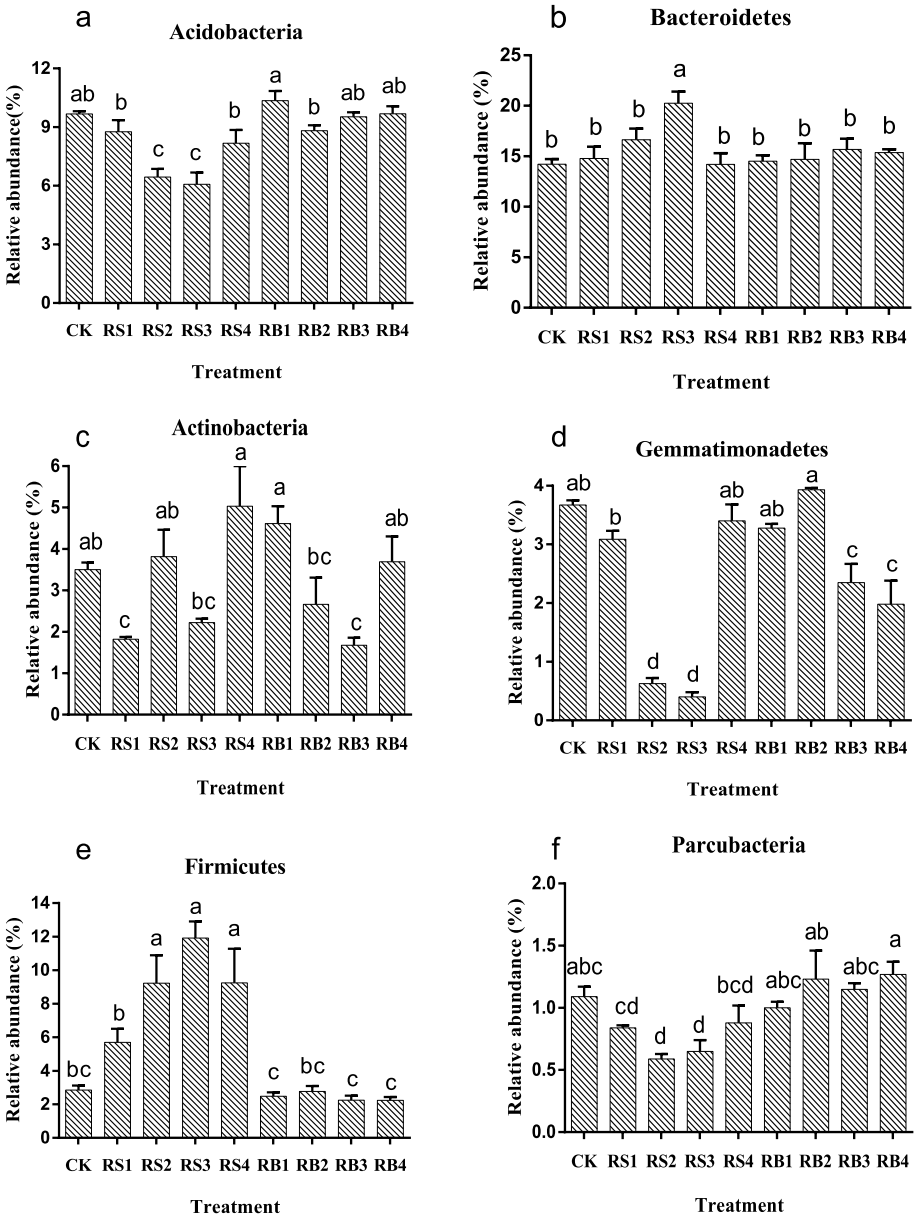


**Fig. 1** Effects of different treatments on the relative abundance of bacterial phyla. Different small letters within one column mean significant difference of Duncan multiple range test among different treatments at 5% level. The same as below

and RS3 treatments was significantly lower than both the control and other treatments. The relative abundance change of Actinobacteria in each treatment was RS4 > RS2 > CK > RS3 > RS1, and that in RS1 treatment was 48% lower than control. Compared with straw return, straw-derived biochar had a smaller effect on the relative abundance of bacterial phyla after returning to the field. The difference in relative abundance between the control and Bacteroidetes, Acidobacteria, Firmicutes, and Parcubacteria was not significant. The relative abundance change of Actinobacteria was RB1 > RB4 > CK > RB2 > RB3, while that in the RB3 treatment was 52% significantly lower than in the control. The relative abundance of Gemmatimonadetes was RB2 > CK > RB1 > RB3 > RB4, where the RB3 and RB4 treatments were 35.97% and 46.05% significantly lower than the control. Based on these analyses, there are differences in the effect of straw and its derived biochar return on the relative abundance of bacterial community at the phylum level.

We further analyzed bacterial genera that were significantly affected by the straw and its derived biochar return (Fig. 2). The top ten bacterial genera in relative abundance are *uncultured bacterium_f_Anaerolineaceae*, *Thiobacillus*, *Anaerolinea*, *Pontibacter*, *Sphingomonas*, *Anaeromyxobacter*, *uncultured_bacterium_f_Draconibacteriaceae*, *Bryobacter*, *uncultured_bacterium_f_Blastocaeraceae_[Subgroup_4]*, and *Geobacter*. As straw return increased, the relative abundance of *Pontibacter*, *Anaeromyxobacter*, *uncultured_bacterium_f_Blastocaeraceae_[Subgroup_4]* and *Bryobacter* decreased, and their relative abundance was lowest in the RS3 and RS4 treatments, which were 91.70%, 45.90%, 64.84%, and 49.38% lower than the control. The relative abundance changes of *Sphingomonas* were RS4 > RS2 > RS3 > CK > RS1. The RS4 treatment was 66.67% higher than the control. As the amount of biochar return increased, the relative abundance of *Thiobacillus* first decreased and then increased and was 24.80% lower in the RB1 treatment than in the control. The relative abundance of *uncultured_bacterium_f_Draconibacteriaceae* was increased, and that in RB3 and RB4 were 42.97% and 66.41% higher

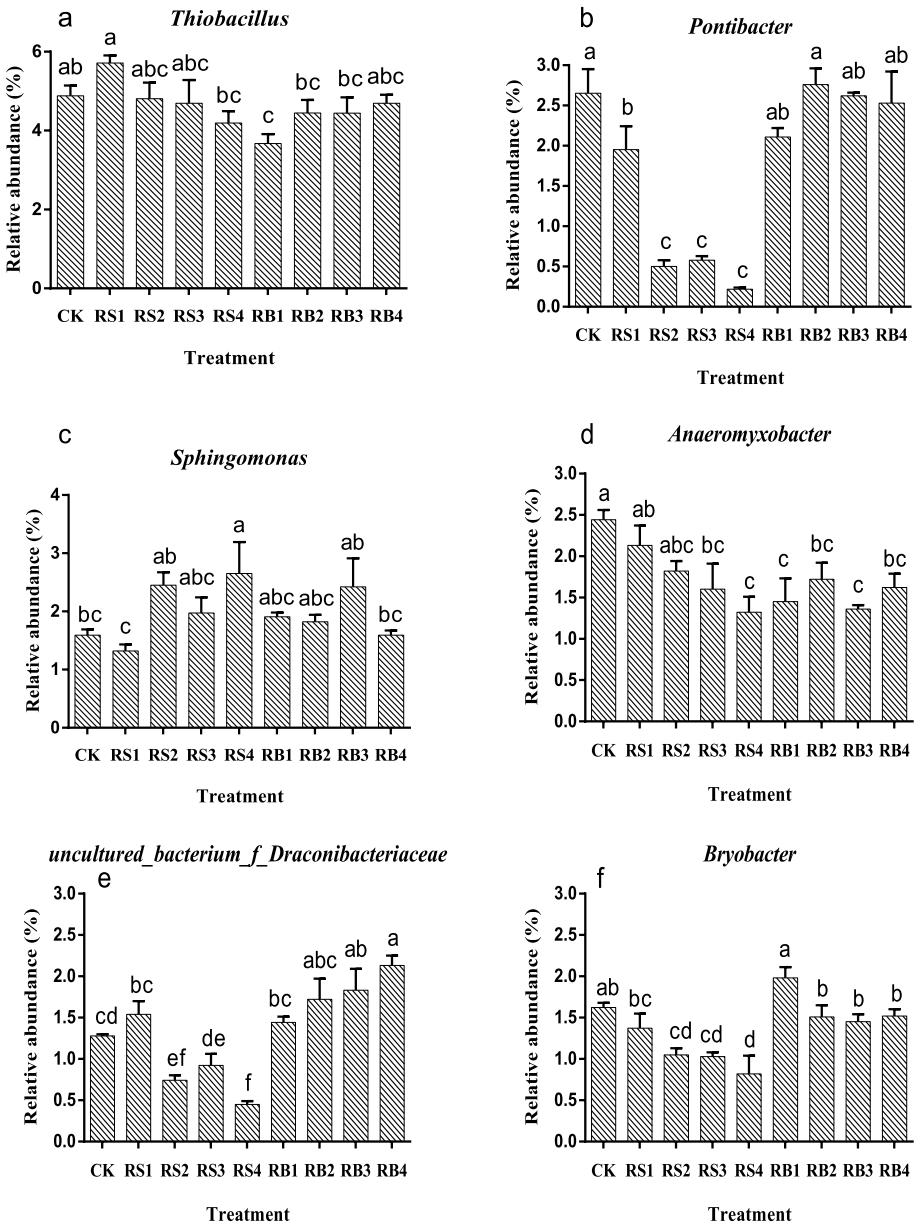


**Fig. 2** Effects of different treatments on the relative abundance of bacterial genera

than in the control. Biochar return significantly reduced the relative abundance of *Anaeromyxobacter*. Our results demonstrate that straw return impacts the relative abundance of bacteria at the phylum and genus level more than biochar does. RS2 and RS3 treatments have a greater impact on the bacteria at the phylum level, RS4 treatment has a greater impact on bacteria at genus level, and the effects of RB3 and RB4 treatments on bacteria at the phylum and genus levels were relatively large.

### Comparison of alpha diversity of soil bacterial community

The statistical results of the OTU number and alpha diversity of each treatment at 97% similarity are shown in Table 2. The sample sequencing depth index values all exceed 99.5%, indicating that the sequencing depth includes most types of bacteria in the sample, and the amount of sequencing data was adequate. The number of bacterial OTUs was between 1676 and 1771. The number of OTUs in the RS3 and RS4 treatments was 4.11% and 4.23% lower than in the control, and the difference between the other treatments and the control was not significant. The Chao1 and ACE index were both used to measure the species abundance. The number of species in the RS4 treatment was significantly lower than in the control, though the differences between the other treatments and the control were not significant. We used Simpson and Shannon indexes to measure species diversity. The Shannon index value is inversely proportional to the Simpson index value, indicating higher species diversity in the sample. In this study, the differences between the control and the Simpson and Shannon indexes in each treatment did not reach a significant level, indicating that straw return and carbonization have no significant effect on bacterial alpha diversity.

### Comparison of soil bacterial community structure

In order to study the changes of the bacterial community structure after continuous straw and straw-derived biochar return, PCoA analysis was performed based on the OTU level, and the principal coordinate combination with the largest contribution rate was selected (Fig. 3). The first and second principal axes accounted for 44.54% and 9.31%, respectively, of the variation of the bacterial community structure. The distances of the biochar return treatments were closer to the control, indicating that the community structure is similar. Compared with the biochar return treatments, straw return treatments were clearly distinguishable from the control. As the straw return amount increased, the projection distance of each treatment on the PC1 axis from the control increased, indicating that adding straw changed the soil bacterial community structure, and that high level had a more significant impact.

### Redundant analysis of soil physicochemical properties and bacterial community structure

The soil physicochemical properties and related bacterial genera were analyzed using RDA (Fig. 4), which further clarified the environmental factors that affect the changes in community structure. The relationship between the rays in the figure is represented by the angle: an obtuse angle represents a negative correlation, and an acute angle represents a positive correlation. RDA analysis demonstrated that the eigenvalues of the two main axes were 28.43% and 9.45%, respectively. *Sphingomonas*, *Hydrogenispora*, and *Lentimicrobium* are negatively correlated with TN; *Clostridium_sensu_stricto_1* is negatively correlated with TN and AN and positively correlated with other environmental factors; *Pontibacter*, *Bryobacter*, and *Anaeromyxobacter* are positively correlated with TN and are negatively correlated with other environmental factors; *Thiobacillus* is positively correlated to pH and negatively correlated with other environmental factors; and *Anaerolinea* and *Geobacter* are positively correlated to AP, TK, TP, and PH and negatively correlated with other environmental factors.

**Table 2** Soil bacterial community alpha index in different treatments

| Treatment | OTU | ACE index | Chao1 index | Simpson index | Shannon index | Coverage |
|---|---|---|---|---|---|---|
| CK | 1750 ± 11ab | 1790.16 ± 6.35ab | 1804.77 ± 2.85abc | 0.0058 ± 0.0006ab | 6.37 ± 0.50ab | 99.74% |
| RS1 | 1771 ± 10a | 1822.23 ± 9.14a | 1832.24 ± 12.43a | 0.0065 ± 0.0003a | 6.33 ± 0.02ab | 99.74% |
| RS2 | 1751 ± 17ab | 1812.34 ± 7.01ab | 1829.04 ± 10.53a | 0.0052 ± 0.0005ab | 6.41 ± 0.01ab | 99.66% |
| RS3 | 1678 ± 17c | 1764.29 ± 16.87b | 1779.22 ± 19.14cd | 0.0063 ± 0.0001a | 6.32 ± 0.06ab | 99.73% |
| RS4 | 1676 ± 16c | 1702.90 ± 33.23c | 1751.11 ± 10.74d | 0.0064 ± 0.0003a | 6.38 ± 0.07ab | 99.71% |
| RB1 | 1755 ± 9ab | 1793.87 ± 7.51ab | 1815.98 ± 7.83ab | 0.0045 ± 0.0003b | 6.48 ± 0.02a | 99.70% |
| RB2 | 1769 ± 4a | 1799.27 ± 16.25ab | 1833.53 ± 5.13a | 0.0056 ± 0.0006ab | 6.37 ± 0.05ab | 99.52% |
| RB3 | 1730 ± 5b | 1773.13 ± 6.93ab | 1791.52 ± 15.01bc | 0.0062 ± 0.0007a | 6.30 ± 0.07b | 99.52% |
| RB4 | 1729 ± 6b | 1774.09 ± 2.08ab | 1790.19 ± 4.54bc | 0.0059 ± 0.0004ab | 6.33 ± 0.05ab | 99.57% |

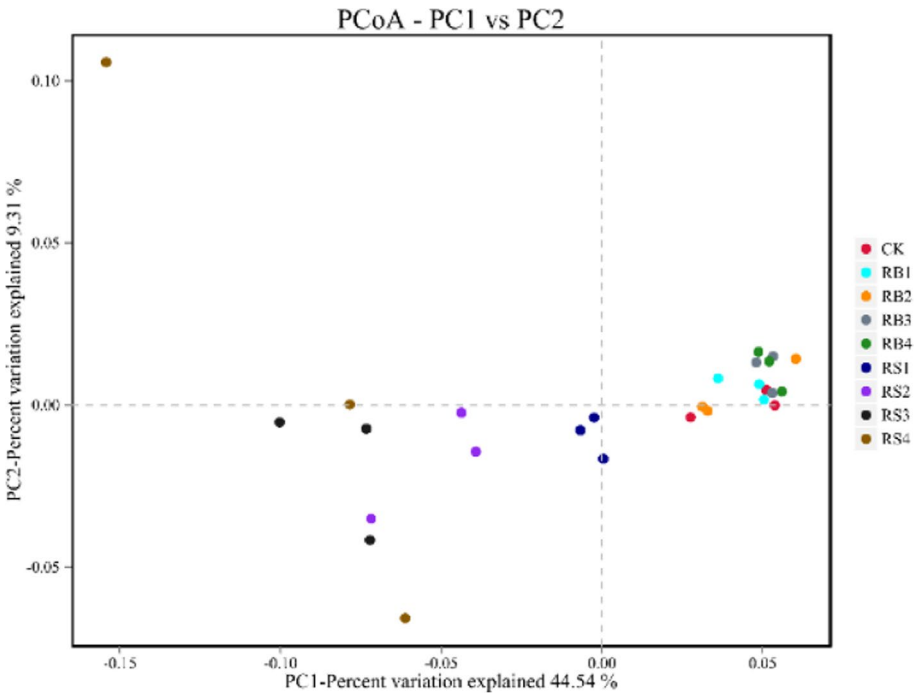


**Fig. 3** Principal coordinate analysis of different treatments

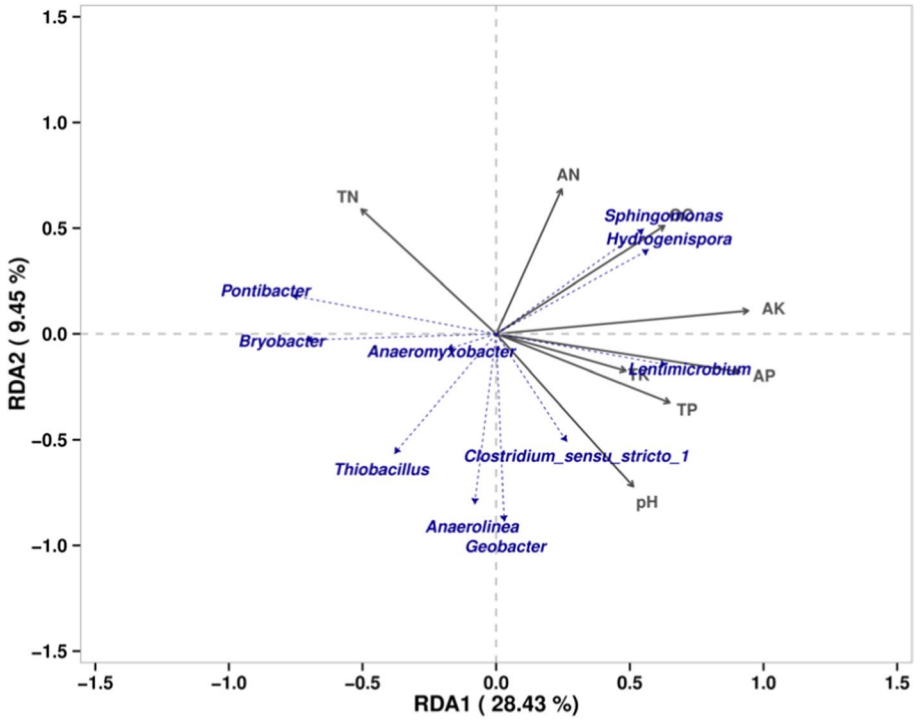


**Fig. 4** Redundancy analysis of bacteria communities and environmental factors of soil. TP, total phosphorus; AK, available potassium; OC, organic matter; AN, available nitrogen; TN, total nitrogen; TK, total potassium; AP, available phosphorus

A Mantel test performed on the soil bacterial flora and soil physical and chemical indicators demonstrated that the relationship between the content of OC, AP, AK, and bacterial community structure was reached significant and extremely significant level (Table 3). Of these, the content of AP and AK had the greatest impact on community structure.

## Discussion

### The influence of straw and straw-derived biochar return on the relative abundance of soil bacterial groups

Straw crops are rich in nutrients such as nitrogen, phosphorus, and potassium, which are released after returning to the field. This can improve soil fertility and nutrient cycling (Yin et al. 2018), make the soil loose and porous, and improve microbial and enzymatic activities in the soil (Eagle et al. 2000). Similarly, biochar also has a positive impact on the physical, chemical (Peng et al. 2011), and biological properties of the soil (Van et al. 2010), all of which improve soil quality. Our results demonstrate that straw and straw-derived biochar return changed the relative abundance of some bacteria at phylum and genus level. The relative abundance of Proteobacteria was highest for both control and treatments. This is consistent with previously published results, which found Proteobacteria to be the dominant phylum in soil bacteria (Bai et al. 2020; Bu et al. 2020; Song et al. 2019; Su et al. 2020). The relative abundance of Bacteroidetes and Firmicutes increased following straw return. Previous studies demonstrated that Bacteroidetes are important decomposers of hemicellulose and xylan (Maarastawi et al. 2018), which positively affects carbon circulation in the soil. Firmicutes also play an important role in the decomposition of organic matter, working as a carbon cycle-promoting bacteria to promote cellulose degradation (Xu et al. 2019). Combined with our previous research (Li et al. 2021), it shows that straw return can accelerate the decomposition of organic matter and the release of nutrients, effectively strengthening the soil carbon cycle of paddy fields. Previous studies demonstrated that adding biochar reduced the relative abundance of Actinobacteria and Gemmatimonadetes (Su et al. 2020; Sun et al. 2016; Zheng et al. 2016). In this study, straw and carbonization return both reduced the relative abundance of Actinobacteria and Gemmatimonadetes. The relative abundance of Actinobacteria and Gemmatimonadetes in 12.0 t/hm$^2$ treatment of biochar was significantly lower than that in the control. Our previous studies demonstrated that the pH of the 12.0 t/hm$^2$ biochar return treatment was significantly higher compared to the control and other treatments, and that straw return had the same trend (Li et al. 2020, 2021). Straw return reduced the relative abundance of Acidobacteria and Parcubacteria. Acidobacteria mostly belongs to the oligotrophic group (Bergmann et al. 2010), is conducive to growth in low pH soil environments, and is very sensitive to increases in pH (Mao et al. 2012; Wu et al. 2019). Therefore, we speculate that straw and biochar return increased soil pH, inhibiting the growth of some bacterial growth.

### Effects of straw and straw-derived biochar return on soil bacterial alpha diversity

The soil bacterial community plays a key role in the process of soil regulation, and the biomass and composition of soil bacteria determine the sustainability of agricultural soils (Segal et al. 2016). Several studies have been conducted on the effects of long-term straw return and carbonization return on the alpha diversity of soil bacteria, but no consensus has yet been reached. Some studies have demonstrated that straw return increased the bacterial alpha diversity of acidic soil (Bu et al. 2020), but did not have a significant impact on alkaline soil diversity (Sun et al. 2015; Yu et al. 2018; Zhang et al. 2021). Returning crop straw biochar to the field increased the bacterial alpha diversity of weakly acidic soils (Yao et al. 2017; Zheng et al. 2016) and reduced the alpha diversity of weakly alkaline soils (Liu et al. 2019). Yin et al. (2021) found that applying peanut shell biochar had no significant effect on the bacterial alpha diversity of tobacco cinnamon soil ($pH = 7.10$). Bai et al. (2020) found that straw carbonization can increase the alpha diversity of soil bacteria in sandy loam soil ($pH = 8.51$); however, straw return did not significantly impact it. Additional analysis demonstrated that the primary reasons for the different test results were the use of different raw materials and the return level of straw and biochar, as well as the soil type. The abundance and diversity of microbial communities largely depend on the pH and nutritional status of the soil (Mao et al. 2012; Tao et al. 2017). Bacteria are very sensitive to pH changes; for example, the diversity and richness of bacteria in desert soils ($pH > 8.0$) and in temperate and tropical forest soils ($pH < 4.5$) are both lower than that of grassland soil in Minnesota ($pH = 6.1$) (Lauber et al. 2009). With the exception of the 16.5 t/hm$^2$

**Table 3** Analysis of Mantel test

| | pH | OC | TN | TP | TK | AN | AP | AK |
|---|---|---|---|---|---|---|---|---|
| *r*-value | 0.145 | 0.466 | 0.054 | 0.157 | 0.081 | 0.089 | 0.489 | 0.655 |
| *p*-value | 0.183 | 0.011 | 0.297 | 0.102 | 0.208 | 0.193 | 0.002 | 0.001 |

straw return treatment, which reduced bacterial abundance, the effects of other treatments on alpha diversity were not significant. This is consistent with the results of Zhang et al. (2021) and Sun et al. (2015). One reason is that the soda saline-alkaline paddy soil used in this study has a relatively high pH. After adding straw and biochar, the pH exceeds 8.0, which inhibits some bacterial growth. The second reason is that adding large amounts of straw and biochar increases the C/N ratio in the soil, meaning there is not enough nitrogen for bacterial activity. This inhibits the growth of some microorganisms in the soil.

### Effects of straw and straw-derived biochar return on soil bacterial community structure

Changes in microbial diversity or community structure could have dramatic impacts on ecosystem processes (Prosser 2002). Straw provides energy and nutrients for bacterial growth (Bai et al. 2018) and can redistribute bacterial community composition (Chen et al. 2017). Previous studies demonstrated that long-term straw return can significantly change bacterial community structure (Bu et al. 2020; Navarro-Noya et al. 2013; Yu et al. 2018). We reached similar conclusions in this study: the straw return significantly changed the bacterial community structure of soda saline-alkaline soil over 5 consecutive years, and the level of influence increased commensurate with application increases. Compared with straw return, straw carbonization return had a smaller impact on bacterial community structure, which was similar to the results of previous studies (Jing et al. 2016; Pan et al. 2016). Some studies demonstrated that the primary factor affecting soil bacterial community structure is soil type (Song et al. 2019). Different soil types respond differently to the composition and structure of soil bacterial communities when adding biochar (Liu et al. 2019). The application of biochar primarily affects the microbial community composition of both acidic and sandy soils (Han et al. 2017; Wang et al. 2016). Previous studies have found that adding organic matter can benefit soil microbial biomass, activity, and community structure (Bronick and Lal 2005). After the straw is returned to the field for a long time, the AK, TOC, and AP contents of the soil all affect the bacterial community (Su et al. 2020). In this study, we found that available phosphorus, available potassium, and organic matter content are important environmental factors affecting the structure of the bacterial community. Our previous research demonstrated that the content of available phosphorus, available potassium, and organic matter after straw return is also important environmental factors that affect the structure of the fungal community (Li et al. 2021). Along with previous studies, we can conclude that straw return changes the physicochemical properties of the soil, affects the living environment of soil microorganisms, and induces changes in the structure of the soil microbial community.

## Conclusion

(1) Five consecutive years of straw return and straw-derived biochar return have changed the relative abundance of some bacteria at phylum and genus level in soda saline-alkaline paddy soil. The effect of straw return is higher than that of straw-derived biochar return.
(2) Five consecutive years of straw return and straw-derived biochar return have not significantly increased bacterial alpha diversity. Compared with biochar return, straw return significantly changed the bacterial community structure. Available phosphorus, available potassium, and organic matter content are important environmental factors affecting the differences in soil bacterial community structure.
(3) Based on the above analysis, we conclude that straw return regulates the physicochemical properties of the soil, thereby affecting the bacterial community structure.

**Acknowledgements**
Not applicable.

**Authors' contributions**
This work was carried out in collaboration between all authors. Authors YX and GZ performed the experimental investigation. Authors MF and HZ performed the data curation and the analysis. Corresponding and first author HL designed the study, performed the supervision, the writing—review and the editing, and funding acquisition. Another author GZ performed the writing—review and the editing and the project administration. The author(s) read and approved the final manuscript.

**Funding**
This research was funded by National Key R&D Program of China (2018YFD0300104)

**Availability of data and materials**
The datasets used and analyzed during the current study are available from the corresponding author on reasonable request.

## Declarations

**Ethics approval and consent to participate**
Not applicable

**Consent for publication**
All the authors have approved the manuscript that is enclosed.

**Competing interests**
The authors declare that they have no competing interests.

Received: 9 December 2021 Accepted: 10 March 2022
Published online: 03 April 2022

**References**


Bai N, Zhang H, Zhou S, Sun H, Lv W (2020) Long-term effects of straw return and straw-derived biochar amendment on bacterial communities in soil aggregates. Sci Rep 10(1):7891. https://doi.org/10.1038/s41598-020-64857-w

Bai ZG, Thomas C, Ruiperez GM, Batjes NH, Paul M, Bünemann Else K (2018) Effects of agricultural management practices on soil quality: a review of long-term experiments for Europe and China. Agric Ecosyst Environ 265:1–7. https://doi.org/10.1016/j.agee.2018.05.028

Bergmann MJ, Driscoll DC, Emerson DT (2010) Indium gallium nitride-based ohmic contact layers for gallium nitride-based devices. US, US7943924 B2.

Bronick CJ, Lal R (2005) Soil structure and management: a review. Geoderma 124:3–22. https://doi.org/10.1016/j.geoderma.2004.03.005

Bu RY, Ren T, Lei MJ, Liu B, Li XK, Cong RH, Zhang YY, Lu JW (2020) Tillage and straw-returning practices effect on soil dissolved organic matter, aggregate fraction and bacteria community under rice-rice-rapeseed rotation system. Agric Ecosyst Environ 287. https://doi.org/10.1016/j.agee.2019.106681

Cayuela ML, Sánchez-Monedero MA, Roig A, Hanley K, Enders A, Lehmann J (2013) Biochar and denitrification in soils: when, how much and why does biochar reduce $N_2O$ emissions? Sci Rep 3:1732. https://doi.org/10.1038/srep01732

Chen Z, Wang H, Liu X, Zhao X, Li C (2017) Changes in soil microbial community and organic carbon fractions under short-term straw return in a rice-wheat cropping system. Soil Tillage Res 165:121–127. https://doi.org/10.1016/j.still.2016.07.018

Eagle AJ, Bird JA, Horwath WR, Linquist BA, Brouder SM, Hill JE, Chris VK (2000) Rice yield and nitrogen utilization efficiency under alternative straw management practices. Agron J 92:1096–1103. https://doi.org/10.2134/agronj2000.9261096x

Gu MY, Tang GM, Ge CH, Ma HG, Zhang ZD, Xu WL (2016) Effects of straw incorporation modes on microbial activity and functional diversity in sandy soil. Chin J Eco-Agric 24(04):489–498. https://doi.org/10.13930/j.cnki.cjea.150882

Gul S, Whalen JK, Thomas BW, Sachdeva V, Deng HY (2015) Physico-chemical properties and microbial responses in biochar-amended soils: mechanisms and future directions. Agr Ecosyst Environ 206:46–59. https://doi.org/10.1016/j.agee.2015.03.015

Gunes A, Inal A, Sahin O, Taskin MB, Atakol O, Yilmaz N, Nicholson F (2016) Variations in mineral element concentrations of poultry manure biochar obtained at different pyrolysis temperatures, and their effects on crop growth and mineral nutrition. Soil Use Manag 31(4):429–437. https://doi.org/10.1111/sum.12205

Hamm AC, Tenutea M, Krayse DO, Ominski KH, Tkachuk VL, Flaten DN (2016) Bacterial communities of an agricultural soil amended with solid pig and dairy manures, and urea fertilizer. Appl Soil Ecol 103:61–71. https://doi.org/10.1016/j.apsoil.2016.02.015

Han G, Lan J, Chen Q, Yu C, Bie S (2017) Response of soil microbial community to application of biochar in cotton soils with different continuous cropping years. Sci Rep 7:10184. https://doi.org/10.1038/s41598-017-10427-6

Hong JL, Ren LJ, Hong JM, Xu CQ (2016) Environmental impact assessment of corn straw utilization in China. J Clean Prod 112:1700–1708. https://doi.org/10.1016/j.jclepro.2015.02.081

Jing T, Wang J, Dippold M, Yang G, Blagodatskaya E, Kuzyakov Y (2016) Biochar affects soil organic matter cycling and microbial functions but does not alter microbial community structure in a paddy soil. Sci Total Environ 556:89–97. https://doi.org/10.1016/j.scitotenv.2016.03.010

Lauber CL, Hamady M, Knight R, Fierer N (2009) Pyrosequencing-based assessment of soil pH as a predictor of soil bacterial community structure at the continental scale. Appl Environ Microbiol 75:5111–5120. https://doi.org/10.1128/AEM.00335-09

Li H, Wang Z, Fan M, Liu M, Lv Y, Liu L (2021) Effects of continuous straw returning on nutrients of soda saline-alkaline paddy soil and fungal community structure. Agric Res Arid Areas 2021(02):15–23 http://kns.cnki.net/kcms/detail/61.1088.S.20210426.1730.003.html

Li H, Zhang G, Fan M, Zheng G, Lv Y (2020) Effects of continuous biochar returning on nutrients of soda saline-alkali paddy soil and fungal community structure. J Soil Water Conserv 34(06):345–351+360

Liu H, Wang Y, Jiang DH, Sun DY, Yang F (2019) Insight into the correlation between biochar amendment and shifts in bacterial community 4 years after a single incorporation in soybean-and maize-planted soils in northeastern China. Can J Microbiol 65. https://doi.org/10.1139/cjm-2018-0366

Maarastawi SA, Katharina F, Marius L, Claudia K (2018) Crop rotation and straw application impact microbial communities in Italian and Philippine soils and the rhizosphere of Zea mays. Front Microbiol 9:1295. https://doi.org/10.3389/fmicb.2018.01295

Mao JD, Johnson RL, Lehmann J, Olk DC, Neves EG, Thompson ML, Schmidt-Rohr K (2012) Abundant and stable char residues in soils: implications for soil fertility and carbon sequestration. Environ Sci Technol 46:9571–9576. https://doi.org/10.1021/es301107c

Marschner P, Kandeler E, Marschner B (2013) Structure and function of the soil microbial community in a long-term fertilizer experiment. Soil Biol Biochem 35:453–461. https://doi.org/10.1016/S0038-0717(02)00297-3

Miura T, Niswati A, Swibawa IG, Haryan S, Gunit H, Arai M, Yamada K, Shimano S, Kaneko N, Fujie K (2016) Shifts in the composition and potential functions of soil microbial communities responding to a no-tillage practice and bagasse mulching on a sugarcane plantation. Biol Fertil Soils 52(3):307–322. https://doi.org/10.1007/s00374-015-1077-1

Navarro-Noya YE, Gómez-Acata S, Montoya-Ciriaco N, Rojas-Valdez A, Dendooven L (2013) Relative impacts of tillage, residue management and crop-rotation on soil bacterial communities in a semi-arid agroecosystem. Soil Biol Biochem 65:86–95. https://doi.org/10.1016/j.soilbio.2013.05.009

Pan F, Li Y, Chapman SJ, Khan S, Yao H (2016) Microbial utilization of rice straw and its derived biochar in a paddy soil. Sci Total Environ 559:15–23. https://doi.org/10.1016/j.scitotenv.2016.03.122

Peng X, Ye LL, Wang CH, Zhou H, Sun B (2011) Temperature-and duration-dependent rice straw-derived biochar: characteristics and its effects on soil properties of an Ultisol in southern China. Soil Tillage Res 112(2):159–166. https://doi.org/10.1016/j.stll.2011.01.002

Prosser JI (2002) Molecular and functional diversity in soil micro-organisms. Plant Soil 244:9–17. https://doi.org/10.1023/A:1020208100281

Rillig MC, Wagner M, Salem M, Antunes PM, George C, Ramke HG, Titirici MM, Antonietti M (2010) Material derived from hydrothermal carbonization: effects on plant growth and arbuscular mycorrhiza. App Soil Ecol 45(3):238–242. https://doi.org/10.1016/j.apsoil.2010.04.011

Segal LM, Miller DN, Mcghee RP, Loecke TD, Cook KL, Shapiro CA, Drijber RA (2016) Bacterial and archaeal ammonia oxidizers respond differently to long-term tillage and fertilizer management at a continuous maize site. Soil Till Res 168:110–117. https://doi.org/10.1016/j.still.2016.12.014

Shindo H, Nishio T (2005) Immobilization and remineralization of N following addition of wheat straw into soil: determination of gross N transformation rates by 15N-ammonium isotope dilution technique. Soil Biol Biochem 37:425–432. https://doi.org/10.1016/j.soilbio.2004.07.027

Sommer R, Ryan J, Masri S, Singh M, Diekmann J (2011) Effect of shallow tillage, moldboard plowing, straw management and compost addition on soil organic matter and nitrogen in a dryland barley/wheat-vetch rotation. Soil Till Res 115:39–46. https://doi.org/10.1016/j.still.2011.06.003

Song Y, Li X, Xu M, Jiao W, Bian Y, Yang X, Gu C, Wang F, Jiang X (2019) Does biochar induce similar successions of microbial community structures among different soils? Bull Environ Contam Toxicol 103(4):642–650

Su P, Brookes PC, He Y, Wu JJ, Xu JM (2016) An evaluation of a microbial inoculums in promoting organic C decomposition in a paddy soil following straw incorporation. J Soil Sediment 16(6):1776–1786. https://doi.org/10.1007/s11368-015-1340-y

Su Y, Lv JL, Yu M, Ma ZH, Xi H, Kou CL, He ZC, Shen AL (2020) Long-term decomposed straw return positively affects the soil microbial community. J Appl Microbiol 128(1). https://doi.org/10.1111/jam.14435

Sun DQ, Meng J, Xu EG, Chen WF (2016) Microbial community structure and predicted bacterial metabolic functions in biochar pellets aged in soil after 34 months. Appl Soil Ecol 100:135–143. https://doi.org/10.1016/j.apsoil.2015.12.012

Sun RB, Zhang XX, Guo XS, Wang DG, Chu HY (2015) Bacterial diversity in soils subjected to long-term chemical fertilization can be more stably maintained with the addition of livestock manure than wheat straw. Soil Bio Biochem 88:9–18. https://doi.org/10.1016/j.soilbio.2015.05.007

Tao JM, Liu XD, Liang YL, Niu JJ, Xiao YH, Gu YB, Ma LY, Meng DL, Zhang YG, Huang WK, Peng DL, Yin HQ (2017) Maize growth responses to soil microbes and soil properties after fertilization with different green manures. Appl Microbiol Biot 101:1289–1299. https://doi.org/10.1007/s00253-016-7938-1

Tardy V, Chabbi A, Charrier X, Berranger CD, Maron PA (2015) Land use history shifts in situ fungal and bacterial successions following wheat straw input into the soil. PLoS One 10(6):1–17. https://doi.org/10.1371/journal.pone.0130672

Turmel MS, Speratti A, Baudron F, Verhulst N, Govaets B (2014) Crop residue management and soil health: a systems analysis. Agr Syst 134:6–16. https://doi.org/10.1016/j.agsy.2014.05.009

Van ZL, Kimber S, Morris S, Chan KY, Downie A, Rust J, Joseph S, Cowie A (2010) Effects of biochar from slow pyrolysis of papermill waste on agronomic performance and soil fertility. Plant Soil 327(1/2):235–246. https://doi.org/10.1007/s11104-009-0050-x

Wang J, Xiong Z, Kuzyakov Y (2016) Biochar stability in soil: meta-analysis of decomposition and priming effects. GCB Bioenergy 8:512–523. https://doi.org/10.1111/gcbb.12266

Wang XH, Yang HS, Liu J, Wu JS, Chen WP, Wu J, Zhu LQ, Bian XM (2015) Effects of ditch-buried straw return on soil organic carbon and rice yields in a rice-wheat rotation system. Catena 127:56–63. https://doi.org/10.1016/j.catena.2014.10.012

Wu C, Shi LZ, Xue SG, Li WC, Jiang XX, Rajendran M, Qian ZY (2019) Effect of sulfur-iron modified biochar on the available cadmium and bacterial community structure in contaminated soils. Sci Total Environ 647:1158–1168. https://doi.org/10.1016/j.scitotenv.2018.08.087

Xie T, Reddy KR, Wang C, Yargicoglu E, Spokas K (2014) Characteristics and applications of biochar for environmental remediation: a review. Crit Rev Environ Sci Technol 45(9):939–969. https://doi.org/10.1080/10643389.2014.924180

Xu M, Xian Y, Wu J, Gu Y, Yang G, Zhang X, Peng H, Yu X, Xiao Y, Li L (2019) Effect of biogas slurry addition on soil properties, yields, and bacterial composition in the rice-rape rotation ecosystem over 3 years. J Soil Sediment 19(5):2534–2542. https://doi.org/10.1007/s11368-019-02258-x

Yao Q, Liu JJ, Yu ZH, Li YS, Jin J, Liu XB, Wang GH (2017) Changes of bacterial community compositions after three years of biochar application in a black soil of northeast China. Appl Soil Ecol 113:11–21. https://doi.org/10.1016/j.apsoil.2017.01.007

Yin HJ, Zhao WQ, Li T, Cheng X, Liu Q (2018) Balancing straw returning and chemical fertilizers in China: role of straw nutrient resources. Renew Sust Energ Rev 81:2695–2702. https://doi.org/10.1016/j.rser.2017.06.076

Yin QY, Liu JH, Liu GS, Yang XL, Li XF, Zhang YL, Li Y, Ye HC (2021) Effect of biochar application for four consecutive years on microbial community structure of tobacco cinnamon soil. J Agr Sci Tech-Iran 2(01):176–185. https://doi.org/10.13304/j.nykjdb.2019.0505

Yu DL, Wen ZG, Li XM, Song XJ, Wu HJ, Yang P, Lin WX (2018) Effects of straw return on bacterial communities in a wheat-maize rotation system in the North China Plain. PLoS One 13(6):e0198087. https://doi.org/10.1371/journal.pone.0198087

Zeng XY, Ma YT, Ma LR (2016) Utilization of straw in biomass energy in China. Renew Sustain Energ Rev 11(5):976–987. https://doi.org/10.1016/j.rser.2005.10.003

Zhang HL, Bai L, Zheng XQ, Li SX, Zhang JQ, Zhang HY, Zhou S, Sun HF, Lv WG (2021) Effects of straw returning and fertilization on soil bacterial and fungal community structures and diversities in rice-wheat rotation soil. Chin J Eco-Agric 29(3):531–539. https://doi.org/10.13930/j.cnki.cjea.200502

Zheng JF, Chen JH, Pan GX, Liu XY, Zhang XH, Li LQ, Bian RJ, Cheng K, Zheng JW (2016) Biochar decreased microbial metabolic quotient and shifted community composition four years after a single incorporation in a slightly acid rice paddy from southwest China. Sci Total Environ 571:206–217. https://doi.org/10.1016/j.scitotenv.2016.07.135

## Publisher's Note