



# Responses of Soil Bacterial Communities and Enzyme Activities to Straw Return and Potassium Fertilization with Two Soils Under Soil Potassium Balance Condition in Rice–Wheat System

Liang Cheng<sup>1,2</sup> · Huoyan Wang<sup>1,2</sup>

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## Abstract

Long-term imbalance in fertilization has resulted in a serious decline in potassium (K) fertility in the Yangtze River Delta region of China. Understanding of the complex responses of soil microbial communities and enzyme activities to different K fertilizer measures while maintaining soil K balance can provide a scientific basis for rational application of K fertilizer. Two field experiment sites (JY, loam soil with high pH and K fertility; GD, silty loam soil with low pH and K fertility) were selected to study the effects of K fertilizer management on enzyme activities and bacterial communities under soil K balance condition. K fertilizer treatments included no K fertilizer (K0), straw return combined with K fertilizer (SRK), and inorganic K fertilizer only (IK). Soil bacterial communities were examined using MiSeq sequencing. Wheat yield, soil nutrient contents, and enzyme activities were higher in the SRK than in the IK and K0 treatments. Available K was the most important factor affecting wheat yield. The SRK and IK treatments significantly altered bacterial communities and enzyme activities, which in turn affected the cycling of soil nutrients. The positive effect of the SRK treatment on wheat yield, enzyme activities, and potential bacterial functions in silty loam soil was greater than that in loam soil, which was closely related to soil texture, pH, and K fertility. The SRK measure is a promising approach to maintain crop yield and soil fertility and more necessary to be adopted in silty loam soil with low pH and K fertility.

**Keywords** Wheat yield · Soil texture · K fertility · pH

## Abbreviations

JY	Jiangyan	SOM	Soil organic matter
GD	Guangde	MBC	Microbial biomass carbon
K	Potassium	K0	No inorganic K fertilizer and straw incorporation
P	Phosphorus	SRK	Straw return combined with inorganic K fertilizer application
N	Nitrogen	IK	Inorganic K fertilizer only
NH <sub>4</sub> <sup>+</sup> -N	Ammoniac nitrogen	OTU	Operational taxonomic unit
NO <sub>3</sub> <sup>-</sup> -N	Nitrate nitrogen	NGDC	National Genomics Data Center
TN	Total nitrogen	LSD	Least significant difference
AK	Available potassium	NMDS	Non-metric multidimensional scaling
TP	Total phosphorus	PERMANOVA	Permutational multivariate analysis of variance
AP	Available phosphorus	KEGG	Kyoto Encyclopedia of Genes and Genomes

✉ Huoyan Wang  
hywang@issas.ac.cn

<sup>1</sup> State Key Laboratory of Soil and Sustainable Agriculture, Institute of Soil Science, Chinese Academy of Sciences, Nanjing 210008, People's Republic of China

<sup>2</sup> University of Chinese Academy of Sciences, Beijing 100049, People's Republic of China

## 1 Introduction

Soil potassium (K) is an essential nutrient for plant growth (Pettigrew 2008; Song et al. 2020) and can improve the quality and quantity of agricultural products (Zhao et al. 2014). Large amounts of soil K are absorbed by crops, owing to continuous improvement in crop yield, consequently resulting in gradual depletion of soil K (Niu et al. 2013). K deficiency has occurred in many regions of the world and has become a global problem (Zhao et al. 2014). In China, farmers pay more attention to nitrogen (N) fertilizer than to K fertilizer, resulting in K deficiency in many farmlands (Chen et al. 2019). Therefore, soil K deficiency has become an important factor limiting agricultural production in China.

Application of inorganic K fertilizer and crop residues are two effective methods used to alleviate the degradation of soil K fertility (Zhao et al. 2014; Bai et al. 2015). Crop straw is an important organic fertilizer resource that contains many nutrients, of which K is the most abundant one (Bai et al. 2015). China produces large amount of crop straw every year, and approximately 80% of the total K exists in the straw of cereal crops (e.g., rice, wheat, oilseed rape, and maize) (Chen et al. 2019). Straw incorporation can return a large amount of plant K to the soil. It has been reported that China needs to import a large amount of inorganic K fertilizer every year (Yu et al. 2010). Excessive inorganic K fertilizer input cannot lead to further increases in grain yield but will result in resource waste and low use efficiency (Song et al. 2020). Therefore, the effective utilization of crop straw resources is of great practical significance.

Rice–wheat system is one of the most popular cropping systems in the Yangtze River region, which is an important agricultural production region in China (Zhang et al. 2021). However, long-term imbalance in fertilization has resulted in a serious decline in K fertility in the region (Chen et al. 2019). In rice–wheat system, a large amount of crop straw is left in the field after harvest. Farmers typically burn crop straw in the field, which is a convenient and efficient way to remove crop residues (Huang et al. 2021a). However, open burning of crop straw has been reported to pollute the atmospheric environment (Yan et al. 2020) and result in loss of plant's essential nutrients (Kumar et al. 2019). Therefore, local governments prohibit the in situ burning of crop straw and recommend using straw fertilizer resource by returning crop straw to the field.

Many studies have shown that crop straw return can increase soil organic matter, improve soil fertility and yield stability, and reduce inorganic fertilizer application (Li et al. 2010; Zhang et al. 2016; Chen et al. 2017;

Sharma et al. 2021). Soil microorganisms play key roles in straw decomposition and soil nutrient cycling (Zhao et al. 2016; Tian et al. 2019; Xie et al. 2022). Soil bacteria are involved in a series of soil biochemical processes (e.g., decomposition of soil organic matter, transformation of fertilizer, and N fixation). Crop straw return acts as a substitute for inorganic fertilizers and directly or indirectly affects the composition and structure of soil microbial communities by altering soil properties (Zhu et al. 2019; Yan et al. 2020). Ceja-Navarro et al. (2010) found that crop straw return increased the abundance and diversity of beneficial soil bacteria, which is important to soil health and crop growth. In a long-term experiment (> 25 years), Yuan et al. (2013) reported that straw incorporation combined with inorganic K fertilizer increased soil microbial biodiversity compared with the application of inorganic fertilizers alone. Overall, there is a strong interaction between straw decomposition, soil microorganisms, and soil nutrient cycling (Heimann and Reichstein 2008; Zhong et al. 2018).

Soil microorganisms also play a key role in the formation of soil enzymes, which are involved in various biochemical processes in soil ecosystems (Jarosch et al. 2019; Eslaminejad et al. 2020), such as promoting the mineralization of soil organic matter and decomposition of crop residues (Huang et al. 2021a). Previous studies have reported that crop straw return significantly affects soil enzyme activities by changing soil properties and microbial metabolic functions (Zhang et al. 2016; Zhao et al. 2016). For instance, straw return can increase soil organic carbon, which has significant correlation with the activities of sucrase and cellulase (Ge et al. 2010; Huang et al. 2021a). Zhang et al. (2016) showed that the incorporation of maize straw increased soil phosphatase and urease activities, which are closely related to the availability of N and phosphorus (P) in the soil. In addition, Zhang et al. (2016) also found a positive correlation between crop yields and enzyme activities when crop straw was returned to the field.

Long-term neglect of K fertilizer management in rice–wheat cropping system in the Yangtze River Delta region has inhibited further improvement of crop yield and caused negative impact on food security (Chen et al. 2019). In recent years, several studies have reported the effects of using straw K instead of inorganic K fertilizer on soil physical properties (Zhang et al. 2014, 2016), chemical properties (Chen et al. 2017), microbial community (Zhu et al. 2019), and crop yield (Zhang et al. 2021). It has also been reported that long-term straw return without inorganic potassium fertilizer will lead to the continuous loss of soil K, thereby resulting in yield decline (Zhao et al. 2014; Bai et al. 2015). Therefore, ensuring soil K balance (the input of K fertilizer is equal to the aboveground K absorption of crops) is an

effective measure for maintaining soil K fertility and crop yield stability. However, information on the effects of different K fertilizer management measures on soil biological properties (e.g., microbial communities and enzyme activities) under soil K balance condition in rice–wheat system is still limited. Therefore, the most effective K fertilizer application measure for high yield and maintenance of soil fertility need more clarification.

We established a fixed-site field experiment with rice–wheat rotation under soil K balance condition over 6 years (2012–2018) for 12 seasons in JY County (loam soil with high K fertility) and GD County (silty loam soil with low K fertility) in the Yangtze River region. The wheat yield, soil chemical properties, and soil biological properties were investigated during the wheat season of 2018 (last wheat season). We hypothesized that (i) wheat yield, soil nutrient contents, bacterial communities, and enzyme activities would have different responses to long-term straw return and inorganic K fertilizer application under soil K balance condition and (ii) these responses would vary in different soil types. The aim of this work is to provide a scientific basis for rational application of K fertilizer and sustainable development of agricultural ecosystem.

## 2 Materials and Methods

### 2.1 Experimental Sites

The two experimental fields under rice–wheat rotation management were conducted over six growing seasons from 2012 to 2018 in Jiangyan County (JY, 32°26' N, 120°05' E) in Jiangsu Province and Guangde County (GD, 31°03' N, 119°27' E) in Anhui Province. Both sites are typical rice–wheat rotation areas in the Yangtze River Delta region and are characterized by a subtropical monsoon climate. The 30-year (1970–2000, <http://worldclim.org/version2>) mean annual temperature and precipitation for JY and GD were 15.2 °C, 980 mm and 15.8 °C, 1172 mm, respectively. The soil at the experimental site of JY was loam soil (43.8% sand (0.05–2 mm), 47.9% silt (0.002–0.05 mm), and 8.3% clay (<0.002 mm)). At the beginning of the experiment, the topsoil (0–20 cm) had total N of 0.5 g kg<sup>-1</sup>, total P of 1.01 g kg<sup>-1</sup>, available P of 28.6 mg kg<sup>-1</sup>, available K of 75.4 mg kg<sup>-1</sup>, and pH value of 7.66. The GD experimental field had a silty loam soil (24.1% sand (0.05–2 mm), 59.7% silt (0.002–0.05 mm), and 16.2% clay (<0.002 mm)). In the 0–20-cm soil layer, the total N, total P, available P, available K, and pH value were 0.81 g kg<sup>-1</sup>, 0.46 g kg<sup>-1</sup>, 23.6 mg kg<sup>-1</sup>, 55.2 mg kg<sup>-1</sup>, and 5.51, respectively. Local farmers in the two sites were used to transplant rice in late June and harvest rice in early October and sow winter wheat in late October and harvest in early June of the following year.

### 2.2 Experimental Design

The field experiments in both sites began with the rice season in June 2012 and ended with the wheat season in June 2018. In both sites, rice was transplanted by hand at 20×20 cm spacing, and the wheat was sown manually at a rate of 195 kg ha<sup>-1</sup> with a row spacing of 25 cm. The varieties of rice and wheat used throughout the experimental period were Wuyunjing 24 (*Oryza sativa* L.) and Yangmai 20 (*Triticum aestivum* L.), respectively. In the field experiments, the application rates of N and P fertilizers were in accordance with the local practices of farmers in the region, and the application rate of K fertilizer in each treatment was approximately equal to the amount of K absorbed by the aboveground crop (except in the case of the treatment without K fertilizer).

At the JY site, the experiment included three K fertilizer treatments: (a) no inorganic K fertilizer and straw incorporation (K0), (b) straw return combined with inorganic K fertilizer application (SRK), in which, to balance the K levels, the amount of inorganic K fertilizer applied was approximately equal to the amount of K absorbed by the grain in the last crop season, and (c) inorganic K fertilizer only (IK), in which all the aboveground parts produced in the last crop season were completely removed, and, therefore, to balance K levels, the amount of inorganic K fertilizer applied was approximately equal to the amount of K absorbed by the crop aboveground in the last crop season. P and K fertilizers were applied as basal fertilizers during both crop seasons. During the rice season, the application rates of N and P fertilizers were 270 kg N and 90 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>, respectively. The N fertilizer was split applied as basal, tillering, jointing, and panicle fertilizers at ratios of 40, 20, 28, and 12%, respectively. During the wheat season, the application rates of N and P<sub>2</sub>O<sub>5</sub> were 225 and 120 kg ha<sup>-1</sup>, respectively. The N fertilizer was applied as basal, tillering, and jointing fertilizers at ratios of 40, 30, and 30%, respectively. The field trial was arranged in a randomized block design with four replicates, and the plot size was 33 m<sup>2</sup> (4.6×7.18 m).

The field experiment at the GD site was the same experimental design as those at the JY site. P and K fertilizers were applied as basal fertilizers, and N fertilizer was split-applied in two split doses (67% as basal fertilizer and 33% as panicle fertilizer) for both the rice and wheat seasons. The amounts of N and P fertilizers applied were 180 kg N and 120 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>, respectively, for both rice and wheat seasons. All treatments were replicated four times in a randomized block design, with a plot size of 30 m<sup>2</sup> (6×5 m). All plots are separated by cement to prevent nutrient interactions between plots for both JY and GD.

The amount of inorganic fertilizer and straw applied in different treatments in each crop season at both sites from 2012 to 2018 are listed in Table 1. In both sites, inorganic

**Table 1** Annual application rates of inorganic fertilizers and crop straw K for different treatments at Jiangyan (JY) and Guangde (GD)

Sites	Treatments	Inorganic fertilizers in rice season (kg ha <sup>-1</sup> )			Wheat straw K input (kg ha <sup>-1</sup> )	Inorganic fertilizers in wheat season (kg ha <sup>-1</sup> )			Rice straw K input (kg ha <sup>-1</sup> )
		N	P <sub>2</sub> O <sub>5</sub>	K <sub>2</sub> O		N	P <sub>2</sub> O <sub>5</sub>	K <sub>2</sub> O	
JY	K0	270	90	0	0	225	120	0	0
	SRK	270	90	32.9	109	225	120	26.9	235
	IK	270	90	137	0	225	120	253	0
GD	K0	180	120	0	0	180	120	0	0
	SRK	180	120	22.1	67.1	180	120	26.7	259
	IK	180	120	84.6	0	180	120	289	0

*K0* no inorganic K fertilizer and straw incorporation, *SRK* straw return combined with inorganic K fertilizer application, *IK* inorganic K fertilizer only

basal fertilizers were broadcast manually and plowed into the soil by rotary tillage before rice transplanting or wheat sowing. In the K0 and IK treatments, all the crop straws were removed from the plots after harvesting. In the SRK treatment, the crop straw was air-dried and then manually chopped into approximately 5-cm-long pieces after the harvest of rice and wheat. The chopped straw was manually plowed into the 0–20-cm soil layer before rice transplanting or wheat sowing. The inorganic fertilizer sources used in this work for both sites were as follows: N fertilizer (urea, 46% N), P fertilizer (calcium superphosphate, 5.24% P), and K fertilizer (potassium chloride, 49.8% K). All other field management measures, including irrigation and pest control, were consistent with the local practices of farmers in the region.

### 2.3 Sampling and Measurements

At the maturity stage in each crop season, grain yields were determined from the entire plot of each treatment at 14% moisture content.

We collected bulk soil (inter-row without plants) during the wheat filling stage (early May 2018) at both sites. The topsoil (0–15 cm) from the bulk soil was collected using a soil core sampler (5 cm diameter, 0–20 cm depth). Nine soil cores were randomly collected from each plot and combined into a single sample. Fresh soil samples were sieved through a 2-mm mesh to eliminate visible stones and plant residues. Each sample was divided into three subsamples. One subsample was stored at –40 °C for DNA extraction and microbial biomass carbon (MBC) determination. The second subsample was stored at 4 °C for analysis of soil enzyme activities. The third subsample was air-dried in the laboratory for analysis of soil chemical properties.

Soil enzyme activities (urease, sucrase, alkaline phosphatase (for the JY site), acid phosphatase (for the GD site), and cellulase) were determined using an analysis kit (Solarbio Science & Technology Co, Beijing, China), following the protocol described in the manufacturer's instructions.

Soil MBC was determined using the chloroform fumigation extraction method (Song et al. 2022). Both 5 g fumigated and control samples were extracted with 0.5 mol L<sup>-1</sup> K<sub>2</sub>SO<sub>4</sub>, after which the solutions were determined using a C/N analyzer (Multi N/C 2100, Analytik Jena AG, Germany). Soil MBC concentration was determined as the difference in organic C of fumigated and non-fumigated soil samples with a conversion factor of 0.45 (Yan et al. 2021).

Soil pH was determined using a pH meter (Mettler Toledo FE28, Switzerland) with a 1:2.5 ratio of soil to distilled water. Soil ammonium nitrogen (NH<sub>4</sub><sup>+</sup>-N) and nitrate nitrogen (NO<sub>3</sub><sup>-</sup>-N) were extracted using 2 M KCl solution at a soil/water ratio of 1:8, and their contents were determined using a continuous-discrete analyzer (SmartChem 200; Westco Scientific Instruments, Brookfield, CT, USA) after shaking for 1 h. Available phosphorus (AP) was extracted with 0.5 M NaHCO<sub>3</sub> (pH=8.5) and determined using the molybdenum blue method. Available K (AK) was determined via a flame photometer after extraction with 1 M NH<sub>4</sub>OAc.

Air-dried soil samples were passed through a 0.15-mm sieve to measure soil organic matter (SOM), total nitrogen (TN), and total phosphorus (TP). SOM content was measured using the potassium dichromate method and determined using the titration method. TN content was measured using the Kjeldahl method. TP content was extracted after H<sub>2</sub>SO<sub>4</sub>–HClO<sub>4</sub> digestion and determined using the molybdenum blue method.

### 2.4 Soil DNA Extraction and Illumina MiSeq High-Throughput Sequencing

DNA was extracted from 0.5 g soil samples using the FastDNA SPIN Kit for Soil (MP Biomedicals, Santa Ana, CA) according to the manufacturer's instructions. The quality of the genomic DNA was checked on 1% agarose gels, and the concentration and purity were determined using a NanoDrop ND-2000 spectrophotometer (Thermo Scientific, Wilmington, USA). The bacterial 16S rRNA gene

V4-V5 hypervariable region was amplified using the primer pair 515F and 907R (Hu et al. 2021). The qualified DNA samples were delivered to Shanghai Majorbio Bio-Pharm Technology Co. Ltd. for high-throughput sequencing of the 16S rRNA gene on the Illumina MiSeq PE 300 platform (Illumina, San Diego, CA, USA). PCR reaction for each DNA sample was performed in 20- $\mu$ L reaction mixtures containing 4  $\mu$ L 5 $\times$  FastPfu buffer, 2  $\mu$ L dNTPs (2.5  $\mu$ M), 0.8  $\mu$ L Forward (5  $\mu$ M) and Reverse (5  $\mu$ M) Primers, 0.4  $\mu$ L FastPfu polymerase, 0.2  $\mu$ L BSA, 10 ng DNA template, and double distilled water (final volume up to 20  $\mu$ L). The PCR condition was as follows: initial denaturation at 95 °C for 3 min, amplified for 30 cycles (denaturation at 95 °C for 30 s, annealing at 55 °C for 30 s, and extension at 72 °C for 45 s), then a final extension at 72 °C for 10 min. The products were checked using a 2% agarose gel and then purified using an agarose gel DNA purification kit (AxQPrepDNA). Sequences obtained in this study were submitted in the National Genomics Data Center (NGDC) Genome Sequence Archive (<https://bigd.big.ac.cn/gsub/>) with accession number CRA006805.

## 2.5 Processing of Illumina Sequencing Data

The raw FASTAQ data were merged with FLASH software (Magoc and Salzberg 2011) and then processed using Quantitative Insights into Microbial Ecology (QIIME, v. 1.9.1) (Caporaso et al. 2010). Sequences with low quality (below the average quality score of 25) and those <300 bp were discarded (Bao et al. 2019). A total of 885,979 high-quality bacterial reads were obtained in the present study. The chimeras were detected and filtered by the UCHIME algorithm (Edgar et al. 2011). The qualified sequences for bacterial reads were clustered into operational taxonomic units (OTUs; >97% sequence identity) using the UCLUST method. The most abundant sequence within each cluster was selected as a representative sequence for that OTU. SILVA 132 database (<http://www.arb-silva.de/download/archive/qiime/>) was used to assign taxonomic identities for bacterial OTUs. Representative sequences were aligned using PyNAST. Non-bacterial sequences (containing <0.001% of total sequences) were discarded, and the remaining sequences of all samples were then rarefied to the same sequencing depth of 18,000 (the lowest sequence read depth among samples) for beta ( $\beta$ ) diversity comparisons (Xiang et al. 2018).

## 2.6 Statistical Analysis

One-way analysis of variance (ANOVA) followed by the least significant difference (LSD) test ( $P < 0.05$ ) was performed to test the effects of different K fertilizer treatments on crop yield, soil chemical properties, enzyme activities, bacterial

$\alpha$ -diversity indices, and bacterial potential functions using the SPSS software (version 22.0; IBM Software, Chicago, IL, USA). Non-metric multidimensional scaling (NMDS) ordination plots based on Bray–Curtis dissimilarities were used to display the differences in bacterial community composition. The permutational multivariate analysis of variance (PERMANOVA) was used to determine the significant differences in bacterial communities among treatments using the “Adonis” function. The correlation between the soil properties and bacterial communities was calculated using the Mantel test. These statistical analyses (NMDS, PERMANOVA, and Mantel test) were implemented using the “vegan” package (Version 2.4–2) in the R environment (version 4.0.2, <http://cran.r-project.org>). The relationships between soil nutrients and enzyme activities were determined using Pearson correlation analysis in R version 4.0.2. In addition, Pearson correlation heatmaps were drawn with the “pheatmap” package (Version 1.0.12) in R version 4.0.2. Automatic linear modeling was performed to predict the importance of soil properties on wheat yield using the SPSS software (version 22.0) (Sun et al. 2015). The potential bacterial functions were generated using PICRUSt2 program based on Kyoto Encyclopedia of Genes and Genomes (KEGG) database (Chen et al. 2020; Wu et al. 2022).

Sustainable yield index (SYI) of wheat yield was evaluated according to the method outlined by Zhang et al. (2021).

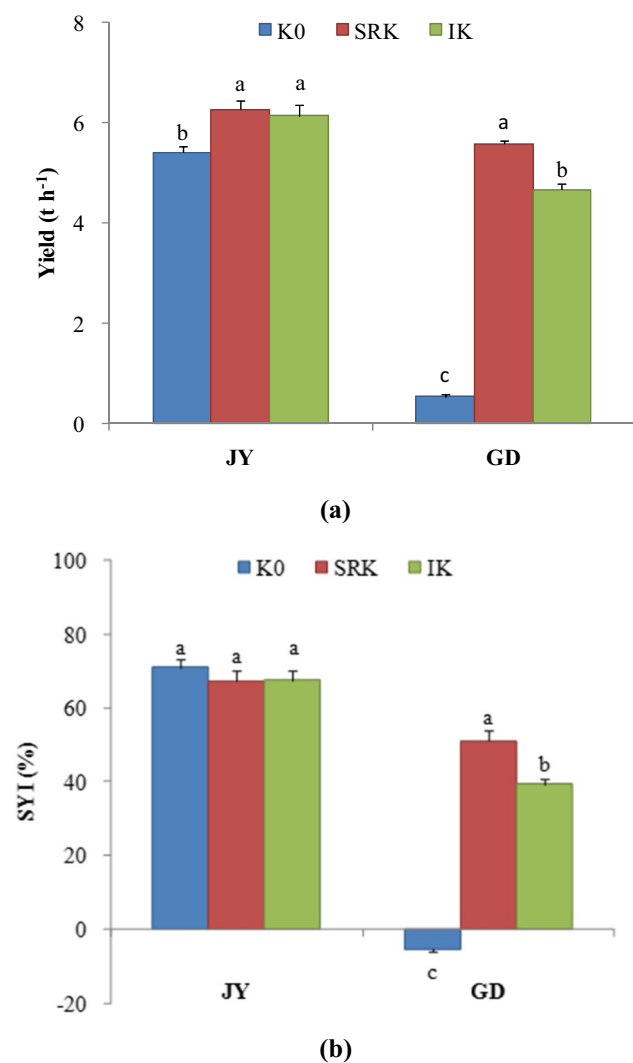
$$SYI = ((Y_{\text{mean}} - Y_{\text{sd}})/Y_{\text{max}}) \times 100\%$$

where  $Y_{\text{mean}}$  is the mean value of wheat yield during 2012–2018 for each treatment,  $Y_{\text{sd}}$  is the standard deviation of wheat yield, and  $Y_{\text{max}}$  is the maximum wheat yield over 2012–2018 for each treatment.

## 3 Results

### 3.1 Wheat Yield

Wheat yield after 6 consecutive years is shown in Fig. 1a. The result showed that the effect of different K treatments on wheat yield was very significant ( $P < 0.05$ ). In the JY site, the wheat yields under the SRK and IK treatments were 16.0% and 13.6% higher than those under the K0 treatment, respectively. The wheat yield in the SRK treatment was 2.17% higher than that in the IK treatment. In the GD site, wheat yield differed significantly in the order as follows: K0 < IK < SRK after 6 consecutive years. Wheat yield was significantly ( $P < 0.05$ ) higher in the SRK and IK treatments than in the K0 treatment, with increases of 942% and 770%, respectively. In addition, the results in Fig. 1b showed that there was no significant difference ( $P > 0.05$ ) in SYI among different treatments in the JY site. However, SYI in the K0



**Fig. 1** Effects of different K fertilizer treatments on the wheat yield (a) and sustainable yield index (b) after 6 consecutive years in a rice–wheat cropping system. Vertical bars are standard error ( $n=4$ ). Different lowercase letters above the bars indicate significant differences among fertilization treatments ( $P < 0.05$ ). K0, no inorganic K fertilizer and straw incorporation; SRK, straw return combined with inorganic K fertilizer application; IK, inorganic K fertilizer only. JY, Jiangyan; GD, Guangde

treatment was less than zero, and SYI in the SRK treatment was significantly higher ( $P < 0.05$ ) than that in the IK treatment for GD.

### 3.2 Soil Chemical Properties

The soil chemical properties of the different K fertilizer treatments are presented in Table 2. In the JY site, soil  $\text{NO}_3^-$ -N showed no significant difference among all treatments ( $P > 0.05$ ). The SRK and IK treatments increased soil  $\text{NH}_4^+$ -N by 90.6% and 41.0%, TN by 23.3% and 9.1%, AK by 105% and 95.3%, TP by 6.9% and 0.6%, AP by 18.5% and

5.8%, and SOM by 13.2% and 0.5%, respectively, compared with the K0 treatment. IK treatment significantly ( $P < 0.05$ ) increased  $\text{NH}_4^+$ -N and AK compared with K0 treatment, while TN, TP, AP, and SOM showed no significant differences between the IK and K0 treatments ( $P > 0.05$ ). In the GD site, soil  $\text{NH}_4^+$ -N and TP showed no significant differences among all treatments ( $P > 0.05$ ). The SRK and IK treatments increased soil  $\text{NO}_3^-$ -N, TN, AK, and SOM compared with the K0 treatment, with increases of 32.1% and 8.5%, 11.3% and 1.9%, 48.6% and 22.8%, and 17.6% and 4.5%, respectively. The SRK treatment significantly ( $P < 0.05$ ) increased soil AP by 27.1% compared with K0 treatment. The improving effect of the SRK treatment on soil fertility was better than that of the IK treatment. In addition, the SRK treatments increased pH, while IK treatment decreased pH compared to K0 treatment in both sites.

### 3.3 Soil Biological Properties

Compared with K0, the SRK and IK treatments showed the potential to increase enzyme activities and MBC. In the JY site (Fig. 2a), the enzyme activity levels were ranked in the order as follows:  $\text{K0} < \text{IK} < \text{SRK}$ . The activities of urease, sucrase, alkaline phosphatase, and cellulase increased by 14.7, 47.9, 40.9, and 235%, respectively, in the SRK treatment and 2.9, 21.9, 20.0, and 58.2%, respectively, in the IK treatment compared with those in the K0 treatment. Compared with the IK treatment, the SRK treatment increased urease activity by 11.4%, sucrase activity by 21.4%, alkaline phosphatase activity by 17.4%, and cellulase activity by 111%. The SRK and IK treatments increased soil MBC by 38.3% and 21.5% compared with the K0 treatment. In the GD site (Fig. 2b), the enzyme activity levels were ranked in the order as follows:  $\text{K0} < \text{IK} < \text{SRK}$ . Compared to those in the K0 treatment, the activities of urease, sucrase, acid phosphatase, and cellulase increased by 22.1, 248, 27.3, and 227%, respectively, in the SRK treatment and 7.21, 13.6, 5.83, and 36.2%, respectively in the IK treatment. Compared with the IK treatment, the SRK treatment increased urease activity by 14.1%, sucrase activity by 207%, alkaline phosphatase activity by 20.3%, and cellulase activity by 140%. Soil MBC was higher in the SRK and IK treatments than in the K0 treatment, with increases of 16.5% and 8.46%, respectively.

### 3.4 Soil Bacterial Community and Potential Functions

The bacterial communities among different K fertilizer treatments were evaluated at the OTU level. PERMANOVA analysis indicated that fertilization treatments had significant effects on bacterial communities (JY:  $R^2=0.1338$ ,  $P=0.011$ ; GD:  $R^2=0.2517$ ,  $P=0.002$ ) (Fig. 3a and b). NMDS analyses

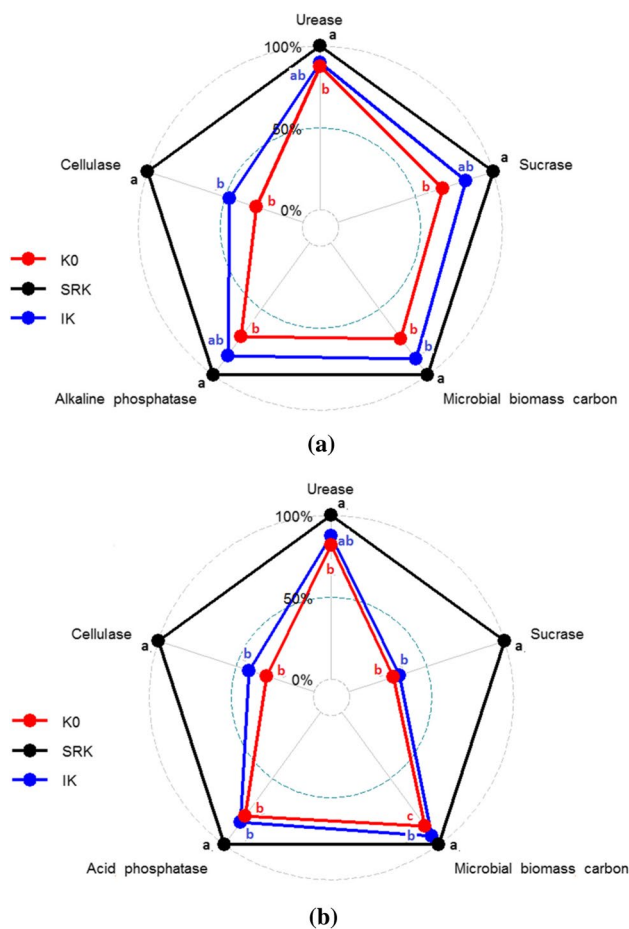
**Table 2** Soil chemical properties at 0–15 cm depth after 6 consecutive years under different fertilization treatments at Jiangyan (JY) and Guangde (GD)

Sites	Treatments	pH	NH <sub>4</sub> <sup>+</sup> -N (mg kg <sup>-1</sup> )	NO <sub>3</sub> <sup>-</sup> -N (mg kg <sup>-1</sup> )	TN (g kg <sup>-1</sup> )	AK (mg kg <sup>-1</sup> )	TP (g kg <sup>-1</sup> )	AP (mg kg <sup>-1</sup> )	SOM (g kg <sup>-1</sup> )
JY	K0	7.22 ± 0.04ab	2.78 ± 0.26c	17.3 ± 0.8a	0.70 ± 0.03b	44.6 ± 2.6b	1.16 ± 0.03b	37.2 ± 2.4b	2.04 ± 0.05b
	SRK	7.33 ± 0.05a	5.30 ± 0.35a	18.0 ± 0.6a	0.86 ± 0.03a	91.4 ± 3.1a	1.24 ± 0.02a	44.2 ± 1.7a	2.31 ± 0.08a
	IK	7.11 ± 0.03b	3.92 ± 0.06b	18.0 ± 0.7a	0.76 ± 0.02b	87.0 ± 2.7a	1.17 ± 0.02b	39.7 ± 1.9ab	2.05 ± 0.04b
GD	K0	5.28 ± 0.07b	13.6 ± 0.4a	52.8 ± 4.3b	0.87 ± 0.04b	35.5 ± 0.8c	0.55 ± 0.02a	26.5 ± 2.2b	2.14 ± 0.08b
	SRK	5.45 ± 0.07a	15.3 ± 1.6a	69.8 ± 4.8a	0.97 ± 0.04a	52.8 ± 2.1a	0.60 ± 0.04a	33.6 ± 2.8a	2.52 ± 0.05a
	IK	5.21 ± 0.04b	15.0 ± 0.9a	57.3 ± 3.5b	0.89 ± 0.04b	43.6 ± 2.6b	0.57 ± 0.03a	26.3 ± 0.6b	2.24 ± 0.11b

Different letters in the same column indicate significant differences among fertilization treatments according to one-way ANOVA (LSD,  $P < 0.05$ )

TN total nitrogen, AK available potassium, TP total phosphorus, AP available phosphorus, SOM soil organic matter

K0 no inorganic K fertilizer and straw incorporation, SRK straw return combined with inorganic K fertilizer application, IK inorganic K fertilizer only

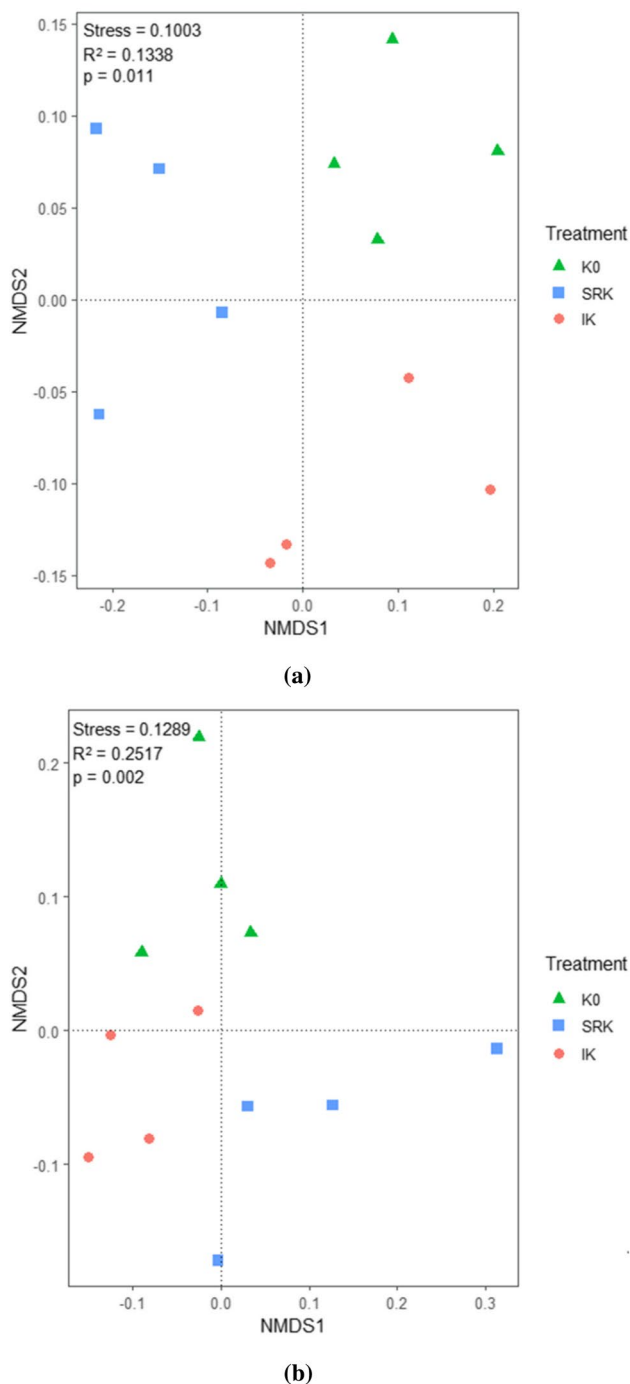


**Fig. 2** Radar chart showing the responses of enzyme activities and microbial biomass carbon (MBC) to different fertilization treatments after 6 consecutive years at Jiangyan (a) and Guangde (b). Different lowercase letters associated with the datapoints on the same perpendicular line indicate significant differences among fertilization treatments ( $P < 0.05$ ). K0, no inorganic K fertilizer and straw incorporation; SRK, straw return combined with inorganic K fertilizer application; IK, inorganic K fertilizer only

based on Bray–Curtis distance also revealed that samples from different treatments were clearly separated, and samples from the same treatment could be gathered together.

The bacterial functions (N-cycling and P-cycling) were predicted using PICRUST2 program. Through comparing with Kyoto Encyclopedia of Genes and Genomes (KEGG) database, a total of 7992 functional genes were obtained, including 30 N-cycling and 4 P-cycling gene families. For JY (Table 3), the SRK and IK treatments increased the putative abundances of *nxrA* and *nxrB*, involved in nitrification; *nirK*, *norB*, and *nosZ* involved in denitrification; *nasB*, *nasA*, *narB*, and *nirA* involved in assimilatory nitrate reduction; *narI* involved in dissimilatory nitrate reduction; and *glnA* involved in Glutamine synthesis compared with the K0 treatment.

For GD (Table 4), the SRK treatment increased the putative abundances of *narI*, *nirK*, *norB*, *norC*, and *nosZ* involved in denitrification; *nasB*, *nasA*, *narB*, and *nirA* involved in assimilatory nitrate reduction; *nirB* involved in dissimilatory nitrate reduction; *gdh2* and *cynS* involved in organic N decomposition; *glnA* involved in glutamine synthesis; and *ppa* involved in P-cycling compared with the K0 treatment. The IK treatment increased the putative abundance of *nirS* involved in denitrification compared with the K0 treatment. In addition, the differences in the total abundances of gene families in the same function were compared in Table S1. The relative total abundances of gene families involved in N-cycling and P-cycling in GD were less than those in JY. In the JY site, the SRK treatment increased the putative gene abundance of nitrification by 6.88%, denitrification by 0.31%, assimilatory nitrate reduction by 1.67%, dissimilatory nitrate reduction by 0.21%, and P-cycling by 0.17% compared with the IK treatment. In the GD site, the SRK treatment increased the putative gene abundance of nitrification by 2.90%,



**Fig. 3** Non-metric multidimensional scaling (NMDS) analysis based on Bray–Curtis dissimilarity display differences in the bacterial community compositions for Jiangyan (**a**, JY) and Guangde (**b**, GD). Different symbols represent the samples under different treatments. Differences of bacterial community compositions among treatments were tested by Adonis analysis. K0, no inorganic K fertilizer and straw incorporation; SRK, straw return combined with inorganic K fertilizer application; IK, inorganic K fertilizer only

**Table 3** Normalized reads of the gene families involved in N-cycling and P-cycling pathways under different fertilization treatments at Jiangyan (JY) site

Pathway	Gene	K0	SRK	IK
Nitrification	<i>pmoA</i>	269a	249a	254a
	<i>pmoB</i>	272a	251a	257a
	<i>pmoC</i>	602a	579a	577a
	<i>hao</i>	158a	150a	159a
	<i>nxrA</i>	1300b	1499a	1362b
Denitrification	<i>nxrB</i>	1482b	1690a	1525b
	<i>narI</i>	704b	887a	844ab
	<i>napA</i>	84a	81a	87a
	<i>napB</i>	84a	80a	86a
	<i>nirS</i>	73b	66b	102a
Assimilatory nitrate reduction	<i>nirK</i>	1192b	1569a	1525a
	<i>norB</i>	860b	1205a	1182a
	<i>norC</i>	189a	190a	210a
	<i>nosZ</i>	602b	838a	865a
	<i>nasB</i>	588b	824a	768a
Dissimilatory nitrate reduction	<i>nasA</i>	8765b	9661a	9546a
	<i>narB</i>	389b	583a	613a
	<i>nirA</i>	585b	689a	637ab
	<i>narI</i>	704b	887a	844ab
	<i>napA</i>	84a	81a	87a
Organic N decomposition	<i>napB</i>	84a	80a	86a
	<i>nirB</i>	4728a	4974a	5218a
	<i>nirD</i>	7419a	7822a	7525a
	<i>nrfA</i>	368a	284b	308b
	<i>nrfH</i>	308a	266b	294ab
Glutamine synthesis	<i>gudB</i>	293b	373a	262b
	<i>gdhA</i>	10,791a	10,475a	10,533a
	<i>gdh2</i>	4202a	4652a	4772a
P-cycling	<i>cynS</i>	879a	892a	959a
	<i>glnA</i>	33,022b	34,314a	34,344a
	<i>plsC</i>	30,320a	30,680a	30,493a
	<i>ppa</i>	12,346a	12,752a	12,742a
P-cycling	<i>pstB</i>	18,833a	18,501a	18,515a
	<i>pstC</i>	18,406a	18,075a	18,123a

Different letters in the same row indicate significant differences among fertilization treatments according to one-way ANOVA (LSD,  $P < 0.05$ )

K0 no inorganic K fertilizer and straw incorporation, SRK straw return combined with inorganic K fertilizer application, IK inorganic K fertilizer only

denitrification by 22.4%, assimilatory nitrate reduction by 33.0%, dissimilatory nitrate reduction by 10.1%, and P-cycling by 1.42% compared with the IK treatment.



**Table 4** Normalized reads of the gene families involved in N-cycling and P-cycling pathways under different fertilization treatments at Guangde (GD) site

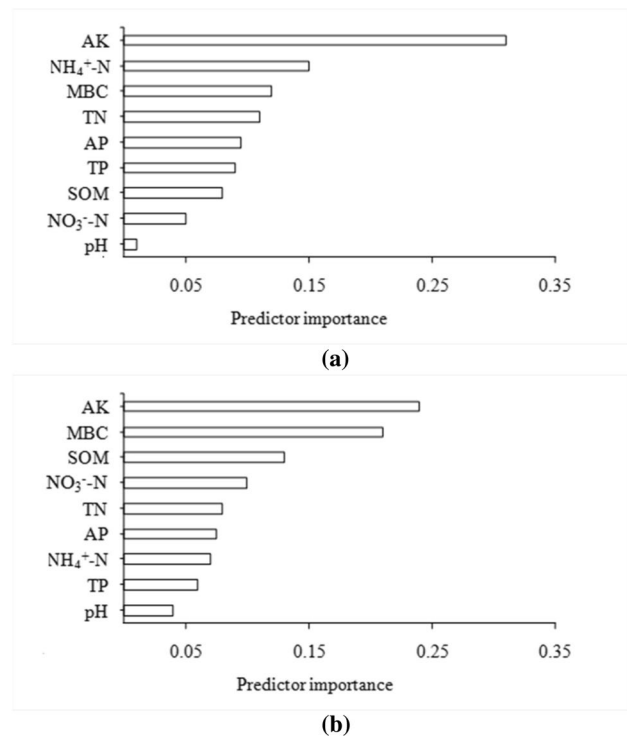
Pathway	Gene	K0	SRK	IK
Nitrification	<i>pmoA</i>	162a	187a	189a
	<i>pmoB</i>	164a	188a	189a
	<i>pmoC</i>	392a	456a	421a
	<i>hao</i>	86a	95a	108a
	<i>nxrA</i>	1001a	1069a	1002a
	<i>nxrB</i>	1099a	1111a	1109a
Denitrification	<i>narI</i>	468b	585a	508ab
	<i>napA</i>	175ab	210a	150b
	<i>napB</i>	173ab	209a	149b
	<i>nirS</i>	110b	120ab	148a
	<i>nirK</i>	699b	1053a	842ab
	<i>norB</i>	648b	918a	800ab
	<i>norC</i>	127b	173a	121b
	<i>nosZ</i>	388b	661a	492b
Assimilatory nitrate reduction	<i>nasB</i>	125b	198a	111b
	<i>nasA</i>	5990b	7607a	6031b
	<i>narB</i>	211b	516a	150b
	<i>nirA</i>	1161b	1395a	1013b
Dissimilatory nitrate reduction	<i>narI</i>	468a	585a	508a
	<i>napA</i>	175a	210a	150a
	<i>napB</i>	173a	209a	149a
	<i>nirB</i>	4163b	5156a	4356b
	<i>nirD</i>	5098a	5549a	5209a
	<i>nrfA</i>	649a	604a	700a
Organic N decomposition	<i>nrfH</i>	516a	459a	525a
	<i>gudB</i>	217a	138b	135b
	<i>gdhA</i>	11,941a	11,249a	11,847a
	<i>gdh2</i>	3214b	4370a	3513b
Glutamine synthesis	<i>cynS</i>	1075b	1388a	993b
	<i>glnA</i>	29,799b	30,892a	29,753b
P-cycling	<i>plsC</i>	27,800a	27,758a	27,273a
P-cycling	<i>ppa</i>	10,217b	11,590a	10,656b
P-cycling	<i>pstB</i>	17,950a	17,800a	18,046a
P-cycling	<i>pstC</i>	17,348a	17,281a	17,414a

Different letters in the same raw indicate significant differences among fertilization treatments according to one-way ANOVA (LSD,  $P < 0.05$ )

K0 no inorganic K fertilizer and straw incorporation, SRK straw return combined with inorganic K fertilizer application, IK inorganic K fertilizer only

### 3.5 Correlation Analysis of Bacterial Communities Against Soil Properties and Wheat Yield

The results of automatic linear modeling displayed apparent differences in the importance of soil properties to wheat yield after 6 consecutive years (Fig. 4). Predictive importance of each soil property to



**Fig. 4** Predictive importance of soil properties to wheat yield after 6 consecutive years for Jiangyan (a, JY) and Guangde (b, GD) determined by automatic linear modeling. TN, total nitrogen; AK, available potassium; TP, total phosphorus; AP, available phosphorus; SOM, soil organic matter; MBC, microbial biomass carbon

wheat yield in JY was ranked in the order as follows:  $\text{pH} < \text{NO}_3^- \text{-N} < \text{SOM} < \text{TP} < \text{AP} < \text{TN} < \text{MBC} < \text{NH}_4^+ \text{-N} < \text{AK}$ . In the GD site, the predictive importance of each soil property to wheat yield was in the following order:  $\text{pH} < \text{TP} < \text{NH}_4^+ \text{-N} < \text{AP} < \text{TN} < \text{NO}_3^- \text{-N} < \text{SOM} < \text{MBC} < \text{AK}$ .

Results of the Mantel test showed the correlation between the bacterial community structure and soil properties (Table 5). The highest to the lowest Pearson's correlation scores of the Mantel test in the JY site are listed as follows: cellulase activity, SOM, urease activity,  $\text{NH}_4^+ \text{-N}$ , alkaline phosphatase activity, TP, AK, AP, MBC, TN, sucrose activity, pH, and  $\text{NO}_3^- \text{-N}$ . The bacterial community structure was significantly ( $P < 0.05$ ) correlated with soil properties (except for  $\text{NO}_3^- \text{-N}$ ). In the GD site, the bacterial community structure had a significant ( $P < 0.05$ ) correlation with soil cellulase activity, MBC, urease activity, sucrose activity, AK, pH,  $\text{NO}_3^- \text{-N}$ , acid phosphatase activity, AP, SOM, and TN.

Pearson's correlation analysis showed the relationship between soil nutrients and enzyme activities after 6 consecutive years. In the JY site (Fig. 5a), SOM, MBC, and TP were significantly positively correlated with urease activity. pH, TN, SOM, MBC,  $\text{NH}_4^+ \text{-N}$ , AK, and AP had a significant positive correlation ( $P < 0.05$ ) with cellulase activity. TN,

**Table 5** Pearson's correlations ( $r$ ) between soil properties and bacterial community composition determined by Mantel test

Soil properties	JY		GD	
	$r$	$P^\dagger$	$r$	$P$
pH	0.240	0.046*	0.440	0.006**
NH <sub>4</sub> <sup>+</sup> -N	0.377	0.007**	0.235	0.139
NO <sub>3</sub> <sup>-</sup> -N	0.038	0.339	0.432	0.007**
TN	0.293	0.026*	0.286	0.036*
AK	0.331	0.018*	0.457	0.001***
TP	0.350	0.013*	0.280	0.087
AP	0.304	0.021*	0.350	0.050*
SOM	0.507	0.001***	0.349	0.009**
MBC	0.300	0.025*	0.512	0.001***
Urease activity	0.482	0.001***	0.502	0.001***
Sucrase activity	0.256	0.038*	0.471	0.001***
Alkaline phosphatase activity	0.359	0.013*	—	—
Acid phosphatase activity	—	—	0.400	0.004**
Cellulase activity	0.526	0.001***	0.550	0.001***

$^\dagger$ : \*, \*\*, and \*\*\* indicate statistical significance at  $P < 0.05$ ,  $< 0.01$ , and  $< 0.001$  levels, respectively, tested by the Mantel test

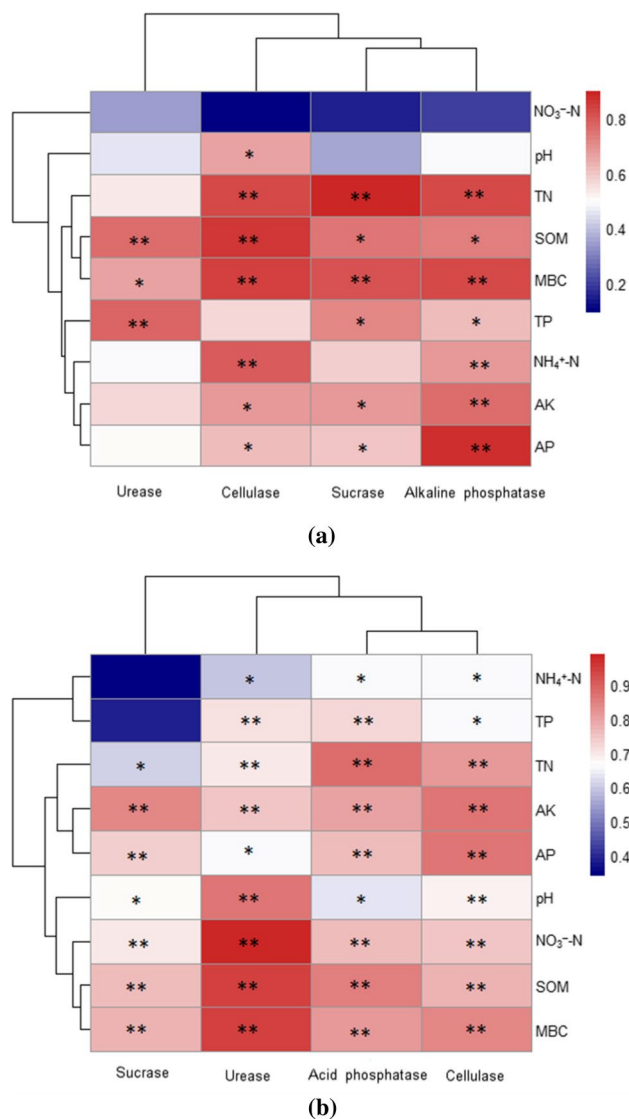
TN total nitrogen, AK available potassium, TP total phosphorus, AP available phosphorus, SOM soil organic matter, MBC microbial biomass carbon

JY Jiangyan, GD Guangde

SOM, MBC, TP, AK, and AP had a significant positive correlation ( $P < 0.05$ ) with sucrase activity. TN, SOM, MBC, TP, NH<sub>4</sub><sup>+</sup>-N, AK, and AP had a significant positive correlation ( $P < 0.05$ ) with alkaline phosphatase activity. In the GD site (Fig. 5b), TN, AK, AP, pH, NO<sub>3</sub><sup>-</sup>-N, SOM, and MBC showed significant positive correlations ( $P < 0.05$ ) with sucrase activity. Urease, cellulase, and acid phosphatase activities were significantly ( $P < 0.05$ ) correlated with all soil properties.

## 4 Discussion

Fertilization is an important method to maintain soil nutrient balance and improve soil fertility (Zhao et al. 2019). In the present study, K fertilization strategies significantly affected most soil properties after a period of 6-year field experiments (Table 2). Previous studies reported that long-term inorganic fertilizer alone significantly reduce soil pH (Ge et al. 2010; Li et al. 2021), which is consistent with this study. Soil acidity caused by inorganic fertilizers, especially N fertilizer, is mainly a result of nitrification, which leads to the formation of nitric acid. Moreover, plant roots will release H<sup>+</sup> to the soil when they absorb cations, which will also lead to soil acidification. Although the amount of N and P fertilizer applied in IK and K0 treatments were the same,



**Fig. 5** A heatmap showing the Pearson's correlations between soil nutrients and enzyme activities for Jiangyan (**a**, JY) and Guangde (**b**, GD). Correlation coefficient ( $r$ ) is represented on the right side of the legend with different colors. \* $P < 0.05$  and \*\* $P < 0.01$ . TN, total nitrogen; AK, available potassium; TP, total phosphorus; AP, available phosphorus; SOM, soil organic matter; MBC, microbial biomass carbon

the soil pH in the IK treatment was lower than that in the K0 treatment. This might be because the IK treatment produced more roots and absorbed more cations from soil compared with the K0 treatment. Huang et al. (2021b) reported that large amounts of organic acids can be generated during straw decomposition, which could accelerate soil acidification. However, we found that the SRK treatment increased soil pH compared to the K0 and IK treatments. This might be because a large number of base cations were returned to the soil after straw decomposition, thus increasing soil pH. Han et al. (2020) found that straw return increased soil pH for a

maize–rice rotation system compared with no straw return. Chen et al. (2021) reported that mulching with straw addition could alleviate soil acidification.

Many studies have shown that straw return is an important way to increase the level of soil nutrients and finally improve crop yield (Su et al. 2019; Li et al. 2021). In our experiments, the soil N content (soil TN,  $\text{NH}_4^+\text{-N}$ , and  $\text{NO}_3^-\text{-N}$ ) was higher in the SRK treatment than in the K0 and IK treatments, indicating that N input was greater than N output in the SRK treatment, which is consistent with previous reports that straw return is beneficial for the accumulation of N in the soil (Sharma et al. 2021; Liu et al. 2022). To our knowledge, crop straw has a high C:N. The N of crop straw itself can be returned to the soil to increase the input of soil N. Straw return also had a positive effect on the growth of soil microorganisms. The fertilizer-N immobilization by soil microorganisms could be strengthened to promote the transformation of soil N into slowly available N sources, thereby reducing N loss and improving N-use efficiency (Zhang et al. 2016). SRK treatment increased soil TP content compared with K0 and IK treatments, probably because the P in straw was returned to the soil, thereby increasing the soil P pool. The significant increase of AP content in the SRK treatment might be related to the increase of soil phosphatase activity by straw return (Han et al. 2020). The soil AK content was higher in the SRK and IK treatments than that in the K0 treatment, corroborating the result of previous research (Zhao et al. 2014) and indicating that inorganic K fertilizer and straw return both alleviated soil K depletion. In addition, the SRK treatment increased AK content compared with the IK treatment, which was consistent with previous studies (Zhao et al. 2014; Bai et al. 2015). In contrast to soil N, soil P and K are more stable and are not lost in the form of gas. Excessive K fertilizer could be converted into non-exchangeable K and fixed in the soil (Zhao et al. 2014). Straw return acted as slowly released K fertilizer, which could reduce the fixation of K by soil. Organic acids produced by straw decomposition might promote the release of soil non-exchangeable K (Bai et al. 2015). Moreover, we found that the AK content of the SRK treatment was close to that of the IK treatment in JY. In contrast, the SRK treatment significantly increased the AK content compared with the IK treatment in GD. This might be because the finer-textured (GD, silty loam) soil fixed more K nutrients, thus reducing the soil AK content in the IK treatment.

In agricultural soils, increasing soil carbon storage is of great significance. Crop straw is a substantial C source and plays an important role in increasing soil organic matter content (Su et al. 2019). Although straw return can increase soil respiration and C consumption by improving the activity of soil microorganism group, most previous studies have reported the positive effects of straw return on soil organic carbon (Chen et al. 2021; Huang et al. 2021b; Li et al. 2021). The release of carbon from decomposed straw increased the

input of soil organic carbon. Zhao et al. (2014) reported that crop straw retention enhanced soil organic carbon and it increased with increasing straw inputs. In the present study, we also found that the SRK treatment significantly improved SOM compared with K0 and IK treatments.

Crop yield is directly related to soil properties. In this work, the SRK treatment was better than the IK treatment in increasing wheat yield at both sites. These findings are in agreement with those of previous studies (Zhao et al. 2014; Bai et al. 2015). This increase was probably associated with SRK treatment, which returned a large part of the nutrients back into the soil and improved the soil physical, chemical, and biological properties (Zhao et al. 2016). Interestingly, the response of wheat yield to different K fertilizer treatments varied in different sites after 6 consecutive years. In JY, the wheat yield in the K0 treatment was  $5.41\text{ t ha}^{-1}$ , whereas that in GD was  $0.54\text{ t ha}^{-1}$ . This result showed that the application of N and P fertilizers without K fertilizer could not meet the growth of crop, and soil K nutrient was the main limiting factor for wheat yield in the K0 treatment in GD. There is a dynamic balance between AK and non-exchangeable K in soil, and part of non-exchangeable K could be converted into AK when the soil AK concentration was low (Li et al. 2021). Our previous research (Li et al. 2020) reported that due to the low basic K fertility of GD soil, soil K nutrient was gradually depleted with the increasing year and thus resulted in inadequate K supply for crop growth in the K0 treatment. By contrast, the non-exchangeable K in JY soil was transformed into exchangeable K in the K0 treatment. Therefore, the response of wheat yield in the GD site to K depletion was much greater than that in the JY site.

In general, the degradation of straw in soil will produce a large quantity of labile organic compounds (Zhao et al. 2016), which provide nutrients and energy for the growth of soil microorganisms. Moreover, straw return can reduce soil bulk density, increase soil aeration, and improve soil hydrothermal conditions (Zhang et al. 2016), which created favorable conditions for the growth of soil microorganisms. The results obtained in our study also indicated that long-term straw return increased soil MBC. In addition, the MBC in the IK treatment were found to be higher than that in the K0 treatment for both sites. This result might be related to the higher root residues of IK treatment.

Soil enzymes play a vital role in soil system and regulate many important biochemical processes in soil (Li et al. 2019; Liu et al. 2021). The close relations between soil microbes and soil enzyme activities are well known. Most soil enzymes are produced by soil microorganisms (Zhu et al. 2019). Previous studies have shown that straw return can increase microbial biomass, thus providing energy and a favorable environment for the increase of soil enzyme (Gianfreda et al. 2005; Sharma et al. 2021). Soil sucrase and cellulase decompose sucrose and cellulose in straw into

monosaccharides, thus providing available C nutrients for soil microorganisms, which in turn stimulates the enhancement of other soil enzyme activities. Soil urease and phosphatase participate in the cycling of soil N and P, respectively. Zhu et al. (2019) and Huang et al. (2021b) reported that long-term straw return treatment increases the soil enzyme activities of cellulase, sucrase, phosphatase, and urease, which promoted the decomposition of straw and nutrient availability compared to inorganic fertilizers. In the present study, there was a significant positive correlation between soil enzyme activities and soil nutrient contents (Fig. 5). The highest enzyme activities of urease, sucrase, phosphatase, and cellulase were observed in SRK, followed by IK and K0, corroborating previous studies. The promoting effect of straw return on cellulase activity was far greater than that of straw return on the activity of other enzymes. This result might be related to the large amount of cellulose in crop straw.

Numerous studies have shown that fertilization plays an important role in shaping soil bacterial community structure and metabolic function through influencing the living environment (Geisseler and Scow 2014; Navarro-Noya et al. 2013; Wang et al. 2018). The changes of bacterial communities are of great significance to the decomposition of soil organic matter and nutrient dynamics and thus are essential for maintaining soil fertility and productivity in agricultural ecosystems (Li et al. 2014; Luo et al. 2019). Soil bacteria has preferences for specific ecological niches and thereby is very sensitive to changes in soil properties (Dangi et al. 2020). A study by Sun et al. (2015) showed that soil pH is the most important factor to explain the changes of soil bacterial community structure under different fertilization measures. This is because pH is the regulator of vital movement. Zhu et al. (2019) reported that soil AK plays an important role in shaping bacterial community in a double-rice cropping system after 9 years of straw incorporation and reduced inorganic fertilizer. Zhao et al. (2019) indicated that the change in soil available N may result in a significant change in the bacterial community after straw addition in different long-term fertilization soils. Su et al. (2019) indicated that soil AK, total organic carbon, and AP contents are important factors affecting soil bacterial community after long-term straw return. Due to the supplemental inputs of nutrients from straw return, soil organic carbon is the key property regulating soil microbial community structure (Chen et al. 2021). In the present study, the soil bacterial communities were significantly correlated with soil pH, TN, AK, TP, and SOM for both JY and GD, indicating that long-term application of inorganic K fertilizer and straw return altered the soil bacterial distribution.

Many studies have shown that changes in soil properties caused by fertilization can drive the shifts in soil bacterial functional profiles (Yuan et al. 2019; Zhang et al. 2019).

It is generally believed that the abundance of functional gene is related to soil nutrient content (Huang et al. 2019). Huang et al. (2021b) indicated a positive relationship between soil microbial carbon metabolism ability and SOM content. In the present study, higher soil permeability of JY soil increased the degree of aeration and accelerated the decomposition of organic compounds compared with GD soil. For this reason, GD soil had a stronger C retention capacity than JY soil. Higher content of soil inorganic N content can increase the substrates for nitrification and denitrification, thus resulting in greater abundance of relevant N transformation genes (Zhang et al. 2019). In this study, the abundances of gene families involved in nitrification, denitrification, assimilatory nitrate reduction, dissimilatory nitrate reduction, organic N decomposition, and glutamine synthesis were enhanced by straw return. We also found that the N-preserving capacity of GD soil was stronger than that of JY soil (Table 2). The possible reasons for this result might be the following: (i) the abundances of gene families involved in N-cycling in GD were less than the corresponding abundances in JY; (ii) compared with that of JY, GD has a higher cation exchange capacity and negative surface charge, thus having a stronger  $\text{NH}_4^+$  adsorption capacity, which decreased the level of bioavailable  $\text{NH}_4^+$  in GD soil; and (iii) neutral soil is generally considered to be conducive to the growth of soil bacteria, and the soil pH of JY was slightly alkaline, whereas the soil pH of GD was acidic, which resulted in the potential N-cycling and P-cycling functions of JY being higher than those of GD. In addition, the promoting effect of SRK on the potential N-cycling and P-cycling functions in GD were greater than those in JY, indicating that straw return appeared to be more effective for improving soil biological properties in GD soil.

Previous studies have shown that plant has a strong impact on soil bacterial communities (Zhang et al. 2019; Yan et al. 2020). In the present study, the highest wheat yield was observed in the SRK treatment; thus, it had more litter, root exudates, and root residues, which promoted the activity of soil microorganisms. The activities of soil microbes and enzyme in the IK treatment were higher than those in the K0 treatment, which confirmed the importance of plants in determining the soil microbial community. In the present study, soil AK content was the most important factor affecting wheat yield. However, under the condition of extreme K deficiency in the K0 treatment, especially the K0 treatment in GD (wheat yield was almost zero), soil bacteria and enzymes still maintained a measurable level of activity, indicating that the stability of soil bacteria in changing environments was higher than that of plants (Huang et al. 2021b). Soil AK might indirectly affect soil bacterial community through plants, which was one of the reasons that the positive effect of straw return on enzyme activities and potential N-cycling and P-cycling functions in GD was greater than that in JY.

## 5 Conclusions

The wheat yield, soil chemical properties, soil bacterial communities and enzyme activities were investigated after 6 consecutive years under soil potassium (K) balance condition in rice–wheat system. Wheat yield with low soil K fertility was more sensitive to K fertilizer application than that with high soil K fertility. Soil available potassium (AK) was the most important factor affecting wheat yield. Soil texture, AK content, and pH had a significant impact on the soil bacterial community, thus affecting changes in enzyme activities and nutrient cycling, as well as ultimately affecting crop yield. The positive effects of straw return on wheat yield, soil nutrients, and microorganisms in silty loam soil with low pH and K fertility were greater than those in loam soil with high pH and K fertility. Overall, straw return combined with inorganic K fertilizer is a promising approach to maintain crop yield and soil fertility and more necessary to be adopted in the silty loam soil with low pH and K fertility.

**Supplementary Information** The online version contains supplementary material available at <https://doi.org/10.1007/s42729-023-01142-5>.

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**Data Availability** The data that support the findings of this study are available from the corresponding author upon reasonable request.

## Declarations

**Competing Interests** The authors declare no competing interests.

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