Applied Soil Ecology 165 (2021) 103982

Contents lists available at ScienceDirect

Applied Soil Ecology

journal homepage: www.elsevier.com/locate/apsoil

Different traits from the paddy soil and upland soil regulate bacterial community and molecular composition under long-term fertilization regimes

Dandan Li [a], Bingzi Zhao [a,*], Jiabao Zhang [a,*], Kailou Liu [b], Qinghai Huang [b]

[a] State Key Laboratory of Soil and Sustainable Agriculture, Institute of Soil Science, Chinese Academy of Sciences, Nanjing, 210008, China
[b] Jiangxi Institute of Red Soil, National Engineering and Technology Research, Center for Red Soil Improvement, Nanchang, 331717, China

ARTICLE INFO

Keywords:
Paddy soil
Upland soil
Long-term fertilization
Bacterial community composition
Molecular composition

ABSTRACT

Fertilization-induced changes in soil properties from the paddy soil and upland soil may directly regulate bacterial community composition, which usually coincide with shifts in molecular composition of soil organic carbon (SOC) in the upland soil. However, systematical comparisons lack on how regulators vary with cropping systems under the same weather conditions and soil parent materials. Here, we simultaneously investigated the changes of soil physicochemical parameters and shifts in the bacterial community and SOC molecular composition in two adjacent rice and maize fields that have received five fertilization regimes for more than 30 years. The separation of the bacterial community composition among the treatments from the paddy soil was mainly determined by soil nitrate-N, and that from the upland soil was mainly determined by soil available P (AP) and pH. The SOC molecular composition from the paddy soil was separated by the treatments with N application or not, with those treatments with N application being enriched with CCH_3 and aromatic C—C, and those without N application being enriched with aromatic C—H. These changed C functional groups showed close association with amorphous Fe_2O_3. For the upland soil, the SOC molecular composition was separated by the treatments with P application or not, with those treatments with P application being enriched with OCH, and those without P application being enriched with CH/CH_2. These changed C functional groups had close association with AP and total P. Our results indicated inconsistent separation patterns and regulators of the bacterial community and SOC molecular composition among the treatments of the paddy soil and upland soil, and suggested that the relatively dominant role of the fertilization-induced changes in soil properties or other soil microbes that controlled the SOC molecular composition over the bacteria measured in the present study.

1. Introduction

Soil bacteria are critical to soil productivity because of their fundamental roles in mediating nutrient cycling, promoting soil structure formation, and maintaining ecological balance in agroecosystems (Zhou et al., 2015; Eo and Park, 2016). Fertilization was one of the major strategies for maintaining soil productivity, which has been proven to effectively modify bacterial community composition via changing soil physicochemical characteristics (Su et al., 2015; Daquiado et al., 2016; Eo and Park, 2016; Samaddar et al., 2019). Previous studies have shown that dominant soil properties governing the bacterial community composition in the paddy soil differed from that in the upland soil (Wang et al., 2015; Li et al., 2018). The redox potential oscillation induced by periodic flooding/drainage in the paddy soil and the near-continuous oxidation in the upland soil triggered series distinct chemical, physical, and biological processes in these two soils (Sahrawat, 2015; Meng et al., 2019). For example, nitrogen to phosphorus (N:P) ratio or nitrate-N was regarded as the most influential factors in the paddy soil following long-term fertilization regimes (Wang et al., 2015; Huang et al., 2019). In the upland soils, the bacterial community composition was predominantly determined by soil organic carbon (SOC) (Li et al., 2017a), pH (Chen et al., 2020), or available P (Li et al., 2018). It is noted that different factors regulating the bacterial community composition in the paddy soil or upland soil were mainly obtained from different experimental sites, which may confuse the comparisons of these two soils, probably because of the confounding influences from different weather

* Corresponding authors.
E-mail addresses: bzhao@issas.ac.cn (B. Zhao), jbzhang@mail.issas.ac.cn (J. Zhang).

https://doi.org/10.1016/j.apsoil.2021.103982
Received 13 June 2020; Received in revised form 2 January 2021; Accepted 28 February 2021
Available online 15 March 2021

conditions (Chen et al., 2017) or soil parent materials (Sheng et al., 2015). Systematically comparative studies conducted under the same conditions are therefore required.

The fertilization-induced changes in the bacterial community composition were usually accompanied by variations in SOC molecular composition in the upland soil, which is one of the key characteristics regulating the sequestration and stabilization of SOC (Grandy and Neff, 2008; Wang et al., 2017; Li et al., 2018). Ng et al. (2014) found that change of aromatic C—C in two agricultural soils amended with different organic manures showed positive correlation with gram-negative bacteria. Wang et al. (2017) found that shift of alkyl C in an alkaline soil under long-term chemical and organic fertilizations exhibited close correlation with gram-positive bacteria. Li et al. (2018) observed that the higher aromatic C—C content induced by long-term P-deficiency showed close associations with Bacillales species in a Calcaric Fluvisol.

The contribution of abiotic processes to the formation of the SOC molecular composition has also been reported on the upland soil rather than on the paddy soil (Randall et al., 1995; Hall et al., 2020; He et al., 2018). Randall et al. (1995) and Hall et al. (2020) emphasized the potential roles of chemical protection of mineral fractions and metal oxides via selectively preserving specific biomolecules in shifting the SOC molecular composition. He et al. (2018) demonstrated that the amount of C input was the most influential factor controlling the SOC molecular composition. More studies are needed to investigate whether the drivers of the SOC molecular composition in the upland soil differed from that in the paddy soil.

Balanced fertilization with concurrent application of nitrogen (N), phosphorus (P), and potassium (K) fertilizers is normally recommended as the best fertilization strategy for crop growth in the paddy soil and upland soil (Johnston, 1997), while imbalanced fertilization lacking one of these major nutrients was still widespread over the past decades (Sheldrick et al., 2003; Zhao et al., 2013). In the present study, two adjacent long-term fertilization experiments, with one growing rice (*Oryza sativa* L.) and another maize (*Zea mays* L.), were used, both of which have received five similar balanced and imbalanced fertilization strategies for more than 30 years. We hypothesized that the bacterial community and SOC molecular composition from the paddy soil and upland soil are regulated by distinct soil traits. The objectives of this study were to (1) detail the fertilization-induced variations in the bacterial community and SOC molecular composition, (2) identify the dominant traits regulating the bacterial community and SOC molecular composition, and (3) assess whether the molecular changes coincide with the shifts in the bacterial community composition.

2. Materials and methods

2.1. Long-term field experiment and soil sampling

Two adjacent long-term field fertilizer experiments were performed in Jiangxi province, China (28°21′N, 116°10′E), with one having an annual cropping system of early rice-late rice and another of early maize-late maize. The rice and maize experiment was initiated in 1981 and 1986, respectively. Each experiment had a completely randomized design with five fertilizer treatments and three replicates. The treatments were: (1) Control, unfertilized control; (2) NPK, balanced fertilization with N, P, and K fertilizers; (3) NP, imbalanced fertilization with only N and P fertilizers; (4) NK, imbalanced fertilization with only N and K fertilizers; and (5) PK, imbalanced fertilization with only P and K fertilizers, which was used in the paddy soil; or P, fertilization with only P fertilizer, which was used in the upland soil. Soil samples used in the present study were collected after the harvest of later rice and later maize in November 2016. More detailed information was provided in supplement materials.

2.2. Soil properties

Soil pH was determined using a glass electrode (FE20, Mettler Toledo, Germany) with a soil:deionized water ratio of 1:5. SOC and total N (TN) contents were measured by dichromate oxidation and Kjeldahl digestion (Page et al., 1982), respectively. Soil total P (TP) and total K (TK) were estimated by digestion with HF-$HClO_4$ (Jackson, 1958), and then, their contents were determined by the molybdenum-blue method (Olsen et al., 1954) and flame photometry (FP640, Huayan, China), respectively. Soil available P (AP) and available K (AK) were extracted with sodium bicarbonate and ammonium acetate (Page et al., 1982), respectively, and their concents were determined by the molybdenum-blue method and flame photometry, respectively. Nitrate (NO_3^--N) and ammonium (NH_4^+-N) were extracted by 2 M KCl, and then, their concentrations were measured with a continuous flow analytical system (Skalar Analytic B.V., De Breda, The Netherlands) (Page et al., 1982). Soil dissolved OC (DOC) content was determined using a total organic carbon analyzer (Multi N/C 3100, Analytik, Jena, Germany) after extraction using a soil:deionized water ratio of 1:5. Free iron oxide (Fe_2O_3) and aluminum oxide (Al_2O_3) were extracted by the dithionite-citrate-bicarbonate method, and amorphous Fe_2O_3 and amorphous Al_2O_3 were extracted by the acidic ammonium oxalate method (Pansu and Gautheyrou, 2006). Extracted iron and aluminum were measured by inductively coupled plasma optical emission spectrometry (Optima 8000, PerkinElmer, USA) and inductively coupled plasma mass spectrometry (NexION 300, PerkinElmer, USA), respectively.

2.3. DNA extraction, 16S rRNA gene amplification and sequencing, and bioinformatics analysis

Soil DNA was extracted from 0.5 g of fresh soil using the Fast DNA SPIN Kit for soil (MP Biomedicals, Santa Ana, CA) following the manufacturer's manual, and the DNA concentration was measured using a NanoDrop 2000 spectrophotometer (Thermo Fisher, USA).

The primers 519F (5′- CAGCMGCCGCGGTAATWC -3′) and 907R (5′- CCGTCAATTCMTTTRAGTTT −3′) were used to amplify the V4–V5 hyper-variable regions of the bacterial 16S rRNA genes. PCR reactions were performed in a 50 μL reaction mix containing 1.25 μM dNTPs, 2 U of Taq DNA polymerase (TaKaRa, Japan), 2 μL (15 μM) of each forward/reverse primer, and 1 μL (50 ng) of genomic community DNA as the template. The thermal program was as follows: initial denaturation at 94 °C for 5 min, 30 cycles of 94 °C for 60 s, 55 °C for 60 s, and 72 °C for 75 s, followed by a final extension step at 72 °C for 10 min. PCR products were purified using a Cycle Pure Kit (OMEGA), and the amount of DNA in each purified sample was measured using a NanoDrop 2000 spectrophotometer (Thermo Fisher, USA). Equal amounts of PCA products were mixed for each sample and then sequenced on the Illumina MiSeq platform (Illumina, Inc., CA, USA). The 16S rRNA gene sequences were submitted to the National Center for Biotechnology Information (NCBI) Sequence Read Archive under the accession number PRJNA591958.

Sequence analysis was performed using the Qualitative Insights into Microbial Ecology (QIIME) pipeline (Caporaso et al., 2010). After trimming the barcodes, primers, low-quality sequences (<Q20), sequences shorter than 200 bp, and singletons, the high-quality sequences obtained were clustered into operational taxonomic units (OTUs) at a threshold of 97% sequence similarity. Taxonomy was assigned using the SILVA 119 database. Nonbacterial sequences (Archaea and chloroplasts) and rare taxa (relative abundance less than 0.001%) were discarded and the remaining sequences of all samples were randomly rarefied to the same sequencing depth (23,466 sequences per sample) for downstream analyses. Principal coordinate analysis (PCoA) based on the weighted UniFrac distance was performed to assess the differences in bacterial community structure among the fertilization treatments in the paddy soil and upland soil.

2.4. Advanced solid-state ^{13}C NMR spectroscopy

All the soil samples were pretreated with 2% (*v*/v) hydrofluoric acid (HF) before NMR analysis to remove paramagnetic compounds and concentrate organic carbon (Skjemstad et al., 1994). The ^{13}C NMR experiments were carried out on a Bruker Avance 400 (Billerica, USA) spectrometer at a ^{13}C frequency of 100 MHz with 4-mm sample rotors in a double-resonance probe head. The quantitative spectra obtained by ^{13}C multiple cross-polarization magic-angle spinning (multiCP MAS) were recorded at a spinning speed of 14 kHz and a contact time of 1 ms, with a relaxation delay of 0.35 s and 90° ^{13}C pulse-length of 4 μs. All the obtained spectra had good signal-to-noise ratios, with minor (< 3%) spinning sidebands and limited overlap with the center bands. Dipolar dephasing multiCP MAS was also conducted to differentiate signals from non-protonated C and mobile C from the total C signal.

The ^{13}C multiCP MAS spectra were subdivided into eight chemical shift regions and were assigned to the following C functional groups: alkyl C (0–44 ppm), OCH_3/NCH (44–64 ppm), O–alkyl C (64–93 ppm), anomeric C (93–113 ppm), aromatic C (113–142 ppm), aromatic C—O (142–162 ppm), COO/N–C=C (162–188 ppm), and ketone/aldehyde C (188–220 ppm). The dipolar dephasing multiCP MAS further separate the rotating CCH_3 and long-chained CH/CH_2 from alkyl C, OCH_3 and NCH from OCH_3/NCH, non-protonated O–alkyl C (OCq) and protonated O–alkyl C (OCH) from O–alkyl C, and the non-protonated aromatic C (aromatic C—C) and protonated aromatic C (aromatic C—H) from aromatic C. The relative abundance of each functional group was obtained by integrating and normalizing the spectral area to the total signal intensity (0–220 ppm) for each spectrum.

2.5. Statistical analyses

One-way analysis of variance (ANOVA) followed by the LSD test was used to identify the statistical differences in soil properties and bacterial taxa among the fertilization treatments. Correlations among the soil properties, molecular compositions, and bacterial taxa were determined by Pearson correlation analysis using IBM SPSS Statistics v19.0 software (Armonk, NY, USA). Significant differences were defined as $p < 0.05$.

Principal component analyses (PCA) were conducted on the soil properties and C functional groups to visualize the distribution of soil properties and SOC molecular composition among the fertilization treatments in both the paddy soil and upland soil. Per-mutational multivariate analysis of variance (PERMANOVA) was carried out to reveal the significant differences in bacterial communities among treatments using the "adonis2" function in the vegan package, and the differential OTUs among treatments were distinguished by using the edgeR package. Key OTUs contributing to the differences between treatments were identified by similarity percentage (SIMPER) analysis (Clarke, 1993). Redundancy analysis (RDA) and multivariate regression tree (MRT) analyses were performed to discern the relationships between bacterial community composition and soil properties (De'ath, 2006). The significance levels of these relationships were evaluated by Monte Carlo test with 999 permutations. In addition, the overall relationship between soil properties and bacterial community composition was analyzed using the Mantel test (999 permutations) in the vegan package. All of these analyses were performed using R statistical software v3.3.3 (R Core Team, 2012).

3. Results

3.1. Soil properties

Overall, the soil properties from the paddy soil significantly differed from the upland soil, regardless of the fertilization treatments, which was mainly reflected by higher SOC and TN contents, while lower Fe/Al oxide contents in the paddy soil than the upland soil ($p < 0.05$, Table 1; Fig. S1). Principal component analysis (PCA) of the soil properties from the paddy soil revealed a distinct separation of the NP plus PK treatments from the NPK plus NK plus Control treatments (Fig. 1a). As expected, the NP and PK treatments had higher TP and AP while lower NO_3^--N and DOC contents, as compared with the other treatments ($p < 0.05$, Table 1). Moreover, the highest amorphous Fe_2O_3 content was observed in the PK treatment and Control ($p < 0.05$, Table 1).

Table 1
Effect of fertilizer treatments on soil pH, and contents of nutrients and free/amorphous Fe- or Al-oxides in the paddy soil and upland soil.

	Paddy soil					Upland soil				
	Control	NPK	NP	NK	PK	Control	NPK	NP	NK	P
pH	5.43 ± 0.22 a	5.39 ± 0.08 a	5.27 ± 0.06 a	5.24 ± 0.35 a	5.28 ± 0.00 a	5.10 ± 0.08 b	4.71 ± 0.10 d	4.94 ± 0.11 c	4.52 ± 0.09 e	5.48 ± 0.05 a
SOC (g/kg)	20.00 ± 0.28 ab	20.50 ± 0.52 a	19.76 ± 0.59 ab	19.89 ± 0.50 ab	19.51 ± 0.27 b	7.82 ± 0.63 c	9.43 ± 0.61 a	8.49 ± 0.52 abc	9.09 ± 0.15 ab	8.39 ± 0.55 bc
TN (g/kg)	2.08 ± 0.05 a	2.05 ± 0.14 a	2.11 ± 0.09 a	2.03 ± 0.06 a	2.12 ± 0.04 a	0.90 ± 0.04 b	1.03 ± 0.03 a	0.99 ± 0.03 ab	1.02 ± 0.05 a	0.98 ± 0.08 ab
TP (g/kg)	0.44 ± 0.02 c	0.57 ± 0.04 b	0.57 ± 0.04 b	0.40 ± 0.02 c	0.68 ± 0.01 a	0.54 ± 0.03 b	0.68 ± 0.12 a	0.72 ± 0.07 a	0.66 ± 0.09 ab	0.69 ± 0.05 a
TK (g/kg)	10.87 ± 0.71 a	11.61 ± 0.41 a	11.48 ± 0.46 a	10.88 ± 0.60 a	11.75 ± 0.11 a	14.44 ± 0.14 a	13.84 ± 0.15 b	14.54 ± 0.03 a	14.22 ± 0.28 ab	14.42 ± 0.43 a
AP (mg/kg)	2.78 ± 0.18 d	3.51 ± 0.07 c	15.68 ± 0.53 b	1.82 ± 0.22 e	19.65 ± 0.27 a	1.36 ± 0.24 e	8.89 ± 0.47 c	9.90 ± 0.69 b	3.93 ± 0.14 d	12.74 ± 0.51 a
AK (mg/kg)	46.04 ± 3.25 b	46.66 ± 7.04 b	48.68 ± 5.34 ab	58.36 ± 7.05 a	54.85 ± 0.23 ab	72.47 ± 10.07 cd	202.17 ± 10.86 b	51.70 ± 2.59 d	254.91 ± 21.23 a	87.96 ± 16.93 c
NO_3^--N (mg/kg)	3.52 ± 0.20 a	3.31 ± 0.23 a	0.44 ± 0.11 b	3.50 ± 0.46 a	0.55 ± 0.11 b	0.63 ± 0.01 d	7.24 ± 0.64 a	1.80 ± 0.76 c	2.93 ± 0.91 b	0.84 ± 0.18 cd
NH_4^+-N (mg/kg)	13.14 ± 3.12 a	15.42 ± 4.73 a	15.23 ± 5.94 a	17.99 ± 4.91 a	14.19 ± 2.36 a	1.82 ± 0.41 c	2.40 ± 0.07 abc	1.91 ± 0.92 bc	2.86 ± 0.12 a	2.75 ± 0.39 ab
DOC (mg/kg)	64.10 ± 3.88 a	75.40 ± 14.28 a	46.61 ± 1.36 b	60.39 ± 11.70 ab	7.94 ± 0.99 c	4.13 ± 0.91 b	7.66 ± 1.37 a	4.53 ± 1.12 b	4.24 ± 0.08 b	4.34 ± 1.66 b
Free Fe_2O_3 (g/kg)	27.34 ± 2.56 b	34.53 ± 0.63 a	33.13 ± 2.39 a	31.99 ± 0.53 a	32.19 ± 0.62 a	45.39 ± 0.62 b	45.82 ± 0.62 ab	45.93 ± 0.69 ab	46.82 ± 0.50 a	46.45 ± 1.10 ab
Amorphous Fe_2O_3 (g/kg)	0.86 ± 0.06 a	0.61 ± 0.03 b	0.69 ± 0.10 b	0.55 ± 0.15 b	0.92 ± 0.02 a	6.10 ± 0.25 a	7.03 ± 0.37 a	7.01 ± 0.19 a	7.17 ± 1.23 a	6.33 ± 0.08 a
Free Al_2O_3 (g/kg)	5.76 ± 0.87 a	6.45 ± 0.77 a	5.58 ± 0.08 a	5.60 ± 0.50 a	5.97 ± 0.35 a	11.06 ± 1.35 ab	11.97 ± 1.04 a	10.82 ± 1.46 ab	11.06 ± 0.55 ab	9.97 ± 0.23 b
Amorphous Al_2O_3 (g/kg)	1.88 ± 0.17 a	1.35 ± 0.17 b	1.53 ± 0.06 b	1.52 ± 0.04 b	1.40 ± 0.10 b	1.83 ± 0.12 b	1.82 ± 0.11 b	1.94 ± 0.24 b	2.34 ± 0.30 a	1.39 ± 0.07 c

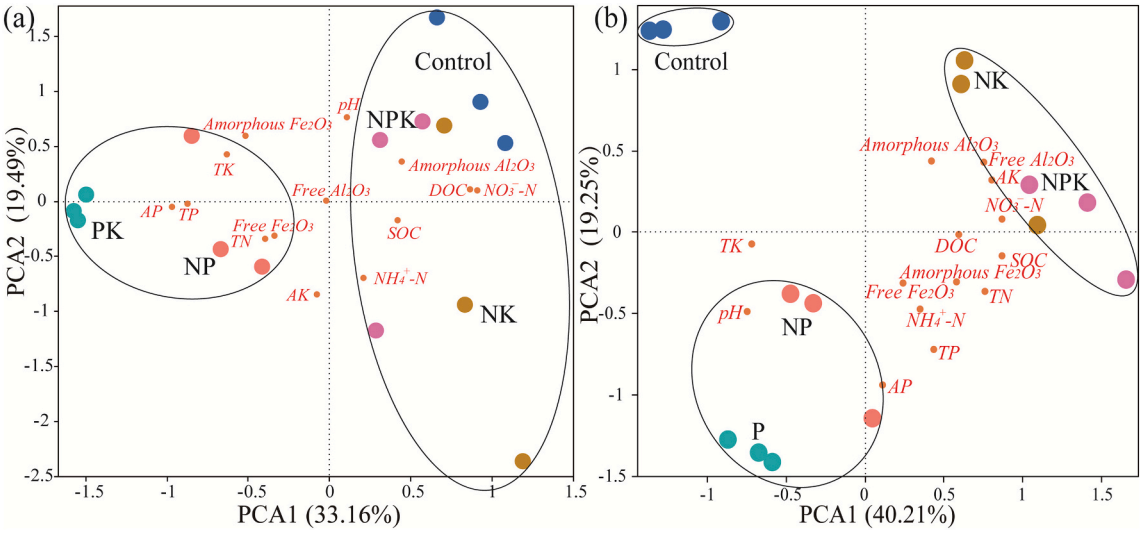

Fig. 1. Principal component analysis (PCA) plots based on soil properties depicting their distributions among the five fertilizer treatments from the paddy soil (a) and upland soil (b).

The soil properties from the upland soil could be divided into three distinct groups, namely, NP plus P, NPK plus NK treatments, and the Control (Fig. 1b). The differences among the three groups were mainly attributed to the higher AP content in the NP and P treatments, the lower pH and higher NO_3^--N and AK contents in the NPK and NK treatments, and the lower SOC, TP, and AP contents in the Control ($p < 0.05$, Table 1; Fig. 1b).

3.2. Bacterial community composition

Chloroflexi (19.9–34.0%), Proteobacteria (11.9–22.7%), and Acidobacteria (18.1–27.1%) were the dominant phyla across the treatments of the both soils (Tables S1 and S2). As expected, the paddy soil exhibited contrasting bacterial communities from the upland soil (Fig. S2), and they responded differently to fertilization regimes (Fig. 2). The PCoA with weighted distance showed that the bacterial community composition from the paddy soil was clearly separated into two groups, namely, the NP plus PK treatments and the NPK plus NK plus Control treatments (Fig. 2a). However, three distinct groups were found for the upland soil, that is, the NP plus P, NPK plus NK, and Control (Fig. 2b). The PERMANOVA results further confirmed the significant differences between the groups ($p < 0.05$, Table S3).

The SIMPER and edgeR analyses showed that the differences between the two groups of the paddy soil were mostly attributed to the enriched members of Acidobacteriaceae subgroup 1 in the NP plus PK group, and the higher abundances of Ktedonobacterales members observed in the NPK plus NK plus Control group (Fig. 3a; Table S4). The distinctions among the three groups of the upland soil were largely ascribed to the abundant Gemmatimonadaceae species in the NP plus P group; the enriched Thermogemmatisporales and Acidobacteria subgroup 2 members in the NPK plus NK group; and the lower abundance of Acidobacteriaceae subgroup 1 in the Control (Fig. 3b; Table S5).

3.3. The SOC molecular composition

The paddy soil and upland soil had significantly different SOC molecular composition, which was mainly reflected by higher CH/CH_2 content, while lower aromatic C—C and aromatic C—H contents in the paddy soil than the upland soil (Table 2; Fig. S3). The PCA analysis showed that the SOC molecular composition from the paddy soil were clearly separated by the treatments with N application or not, that is, the treatments with N application (i.e., NPK, NP, and NK) had similar molecular composition, which was clearly differed from those without N application (i.e., PK and Control) along axis 1 (Fig. 4a), with the former

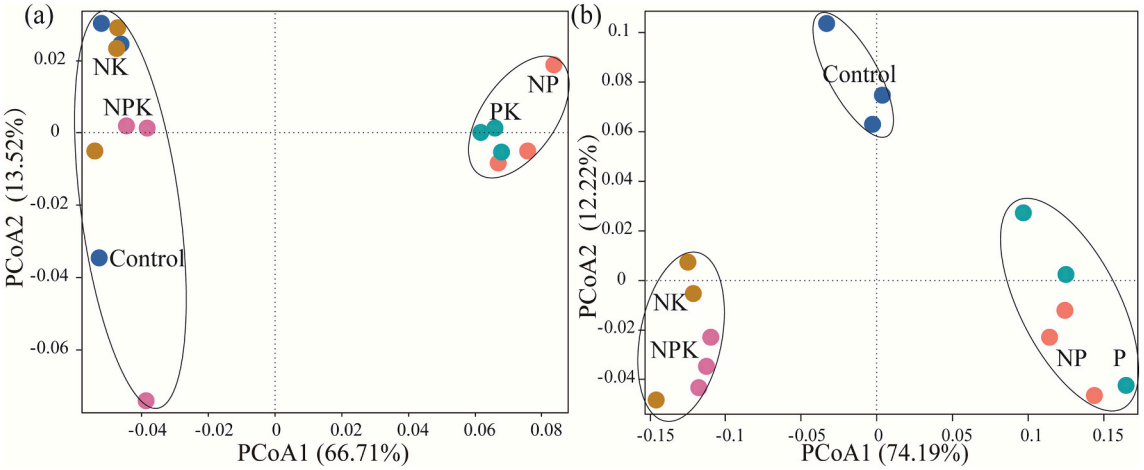

Fig. 2. Principal coordinate analysis (PCoA) plots based on the weighted UniFrac distances showing the variations of bacterial community composition among the five fertilizer treatments from the paddy soil (a) and upland soil (b).

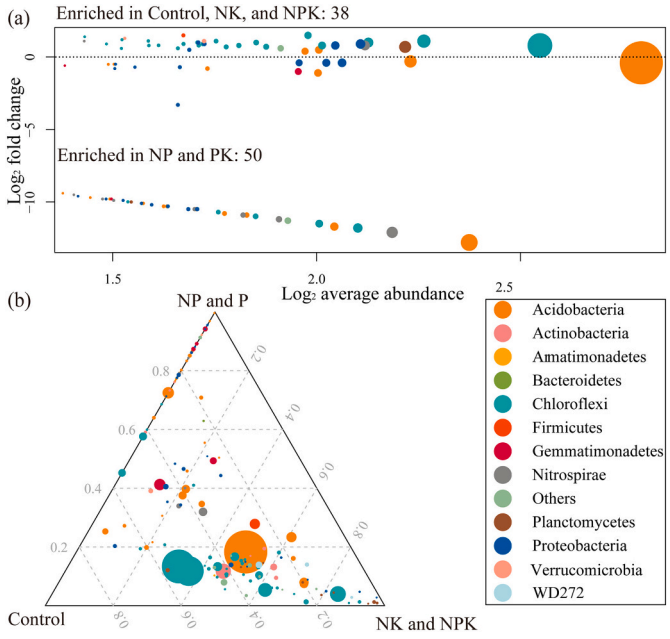

Fig. 3. Bacterial OTUs that show significantly different abundance between the groups in the paddy soil (a) and upland soil (b). Only the OTUs containing more than 0.1% of the total sequence were considered. Circle size represents the average abundance of each OTU. Circle color represents the corresponding OTUs assigned to major phyla. The OTU location in the ternary plot (b) represents their proportional abundances in the three groups.

being characterized by higher CCH_3 and aromatic C—C contents, and the later by higher aromatic C—H content (Table 2; Fig. 4a).

The SOC molecular composition from the upland soil was clearly separated by the treatments with P application or not, that is, the treatments with P application (i.e., NPK, NP, and P) had similar molecular composition, which was clearly distinct from those without P application (i.e., NK and Control) along axis 1 (Fig. 4b), with the former being characterized by higher OCH content, and the later by higher CH/CH_2 content (Table 2; Fig. 4b).

3.4. *Correlations between the soil properties and changed molecular compositions, and determinants of the bacterial community composition*

Correlation analysis showed that in the paddy soil, the amorphous Fe_2O_3 content had positive correlation with aromatic C—H content which was enriched in the PK plus Control group, while it had negative correlations with CCH_3 and aromatic C—C contents which were enriched in the NPK plus NP plus NK group (Table 3; Fig. 4). In the upland soil, the TP and AP contents showed significant positive associations with OCH content which was enriched in the NPK plus NP plus P group, and they had negative associations with CH/CH_2 content which was enriched in the NK plus Control group (Table 3; Fig. 4).

The bacterial community composition and soil properties shared the same cluster patterns for each soil type (Figs. 1 and 2), and their significant and positive correlations for the paddy soil (Mantel $r = 0.557$, $p = 0.001$) and upland soil (Mantel $r = 0.486$, $p = 0.002$) were confirmed by the Mantel analysis. The RDA and MRT analyses were carried out to further explore the relationships between soil properties and bacterial community composition (Fig. 5). Bacterial communities of the paddy soil were distinctly separated from the upland soil under the dominant influence of SOC content (Fig. S4).

In the paddy soil, the NO_3^--N, AP, DOC, TK, and TP significantly correlated with the bacterial community composition ($p < 0.05$, Fig. 5a; Table S6), and these five soil properties accounted for 91.5% of the total variance in the community composition, of which 88.86% was explained

Table 2
Percentages of total spectral area (%) assigned to different functional groups resolved by ^{13}C multiCP MAS nuclear magnetic resonance and dipolar dephased multiCP MAS.

	Treatment	220–188 ppm	188–162 ppm	162–142 ppm	142–113 ppm			113–93 ppm	93–64 ppm		64–44 ppm			44–0 ppm		
		Ketone/aldehyde C	COO/N–C=C	Aromatic C–O	Aromatic C			Anomeric C	O–alkyl C		OCH_3/NCH			Alkyl C		
					Total	Arom. C–C	Arom. C–H		Total	OC_q	OCH	Total	OCH_3	NCH	Total	CCH_3	CH/CH_2
Paddy soil	Control	1.81	9.25	3.04	8.19	3.40	4.79	7.73	28.50	1.50	27.00	13.70	1.04	12.66	27.78	5.23	22.56
	NPK	0.80	9.14	2.32	7.42	4.58	2.84	7.94	28.77	2.23	26.54	14.27	1.77	12.49	29.35	7.17	22.17
	NP	1.39	9.58	2.30	7.36	4.08	3.29	7.99	28.24	1.94	26.31	14.07	1.89	12.18	29.06	7.12	21.95
	NK	1.62	9.51	2.65	7.44	3.98	3.46	7.87	28.77	1.74	27.04	13.73	1.52	12.20	28.42	7.25	21.17
	PK	1.53	9.15	2.55	7.28	2.86	4.42	7.88	29.35	1.52	27.83	13.88	1.49	12.39	28.38	5.93	22.45
Upland soil	Control	0.77	9.19	4.26	14.96	8.83	6.13	8.13	26.82	2.18	24.64	13.86	2.84	11.02	22.01	6.08	15.93
	NPK	1.71	8.92	3.74	16.68	9.49	7.19	8.44	28.36	1.87	26.49	11.88	2.35	9.53	20.27	5.91	14.36
	NP	1.97	9.96	4.54	13.42	6.90	6.52	9.34	29.25	2.15	27.10	12.32	1.67	10.66	19.19	5.33	13.86
	NK	2.34	9.90	4.05	14.32	8.27	6.05	8.75	27.38	2.28	25.10	12.05	2.08	9.98	21.21	4.96	16.25
	P	2.03	9.56	4.72	15.26	8.25	7.01	8.29	27.65	2.12	25.52	12.32	2.22	10.10	20.17	5.86	14.31

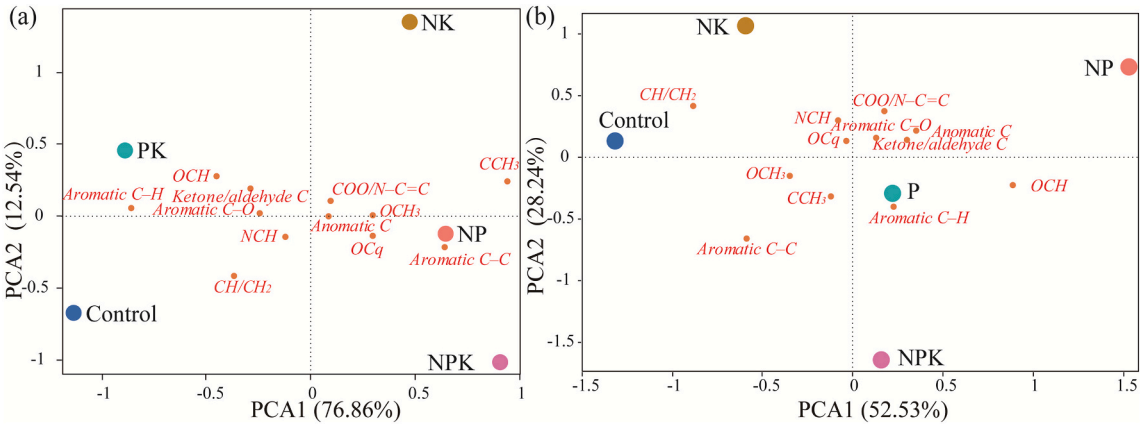

Fig. 4. Principal component analysis (PCA) plots based on the relative contents of C function groups depicting the distributions of molecular composition among the five fertilizer treatments from the paddy soil (a) and upland soil (b).

Table 3
Correlation coefficients between soil properties and SOC molecular compositions in the paddy soil and upland soil.

	Paddy soil			Upland soil	
	Aromatic C–C	Aromatic C–H	CCH_3	OCH	CH/CH_2
pH	*ns*	*ns*	*ns*	*ns*	*ns*
SOC	0.520*	*ns*	*ns*	*ns*	*ns*
TN	*ns*	*ns*	*ns*	*ns*	*ns*
TP	*ns*	*ns*	*ns*	0.569*	−0.525*
TK	*ns*	*ns*	*ns*	*ns*	*ns*
AP	*ns*	*ns*	*ns*	0.652**	−0.877**
AK	*ns*	*ns*	*ns*	*ns*	*ns*
NO_3^--N	*ns*	*ns*	*ns*	*ns*	*ns*
NH_4^+-N	*ns*	*ns*	*ns*	*ns*	*ns*
DOC	0.758**	ns	ns	*ns*	*ns*
Free Fe_2O_3	ns	−0.747**	0.731**	*ns*	*ns*
Amorphous Fe_2O_3	−0.779**	0.761**	−0.787**	*ns*	*ns*
Free Al_2O_3	*ns*	*ns*	*ns*	*ns*	*ns*
Amorphous Al_2O_3	*ns*	0.579*	−0.582*	*ns*	*ns*

by RDA 1 and RDA 2 (Fig. 5a). The NP plus PK group was associated with higher AP and TP contents, while NPK plus NK plus Control group was associated with higher NO_3^--N and DOC contents. The MRT results showed that separation of the NP plus PK group from NPK plus NK plus Control group mainly relied on NO_3^--N content, which explained 85.2% of the total variation (Fig. 5b).

Pearson correlation analysis revealed that Acidobacteriaceae subgroup 1 species enriched in the NP plus PK group showed negative associations with the NO_3^--N and DOC contents and positive associations with the TP and AP contents. Members of the Ktedonobacterales, which were particularly abundant in the NPK plus NK plus Control group, were negatively associated with the TP and AP contents and positively associated with the NO_3^--N and DOC contents (Table S7).

In the upland soil, significant correlations between the contents of AP, SOC, AK, NO_3^--N, TN, and pH and the bacterial community composition were found ($p < 0.05$, Fig. 5c; Table S6), and these six soil properties explained 83.2% of the total variance in the community composition, of which 75.1% was explained by RDA 1 and RDA 2 (Fig. 5c). The MRT analysis revealed that the community composition was separated into two main groups based on the AP content by the first split, which explained 54.3% of the total variation. Higher AP content was associated with the NP plus P group. Further division of the remaining groups was determined by soil pH, which explained 20.5% of the variation in the community composition. Lower pH was associated with the NPK plus NK group (Fig. 5d).

Pearson correlation analysis revealed that, in the upland soil, the abundant Gemmatimonadaceae species in the NP and P treatments showed positive associations with the AP content and pH. For the species significantly enriched in the NPK and NK treatments, both the Thermogemmatisporales and unclassified Acidobacteria subgroup 2 members were negatively correlated with the soil pH. In addition, the Acidobacteriaceae subgroup 1, which was significantly depleted in the Control, showed negative correlations with the SOC and NO_3^--N contents (Table S8).

4. Discussion

4.1. Fertilization-induced variation in bacterial community composition show associations with different traits from the paddy soil and upland soil

In the paddy soil, the NO_3^--N was considered as the main factor determining the bacterial community composition, which clearly separated the NP plus PK treatments from the NPK plus NK plus Control treatments (Fig. 5a). Fluctuating redox potential evoked by periodic flooding/drainage creates an active environment for denitrification and nitrification (Jin et al., 2020). As a substrate of denitrification, the NO_3^--N has been consistently recognized as a determinant factor shaping bacterial composition in paddy soils (Wang et al., 2015; Chen et al., 2016; Yi et al., 2019). In the present study, the NP and PK treatments had higher AP and TP contents (Table 1), which may beneficial for inducing denitrification potential. Wei et al. (2017a) reported that P addition increased gaseous N loss in P-limited paddy soils through stimulating denitrification. In addition, the NP plus PK treatments were enriched with Acidobacteriaceae subgroup 1 members (Fig. 3a; Table S4), which have been found to be involved in denitrification under oxygen-limited conditions, facilitating the reduction of NO_3^--N (Cheng et al., 2017; Rasigraf et al., 2017). This was consistent with the findings that the Acidobacteriaceae subgroup 1 members negatively correlated with NO_3^--N (Table S7), and that the NP plus PK treatments had lower NO_3^--N content (Table 1).

In the upland soil, AP and pH were regarded as the main factors determining the bacterial community composition, and they partitioned the community composition into three distinct groups: NP plus P, NPK plus NK, and Control (Fig. 5b). Soil P availability is a major limiting factor determining microbial growth and activity (Zhong and Cai, 2007; He et al., 2008; Turner et al., 2013), and AP content have also been reported as important property determining bacterial composition (Chu et al., 2007; Li et al., 2018; Shen et al., 2018). Meanwhile, variation of soil pH caused by urea fertilization and its considerable effect on bacterial composition in upland soils have been well documented (Francioli et al., 2016; Ling et al., 2016; Yang et al., 2017). In the present study, the

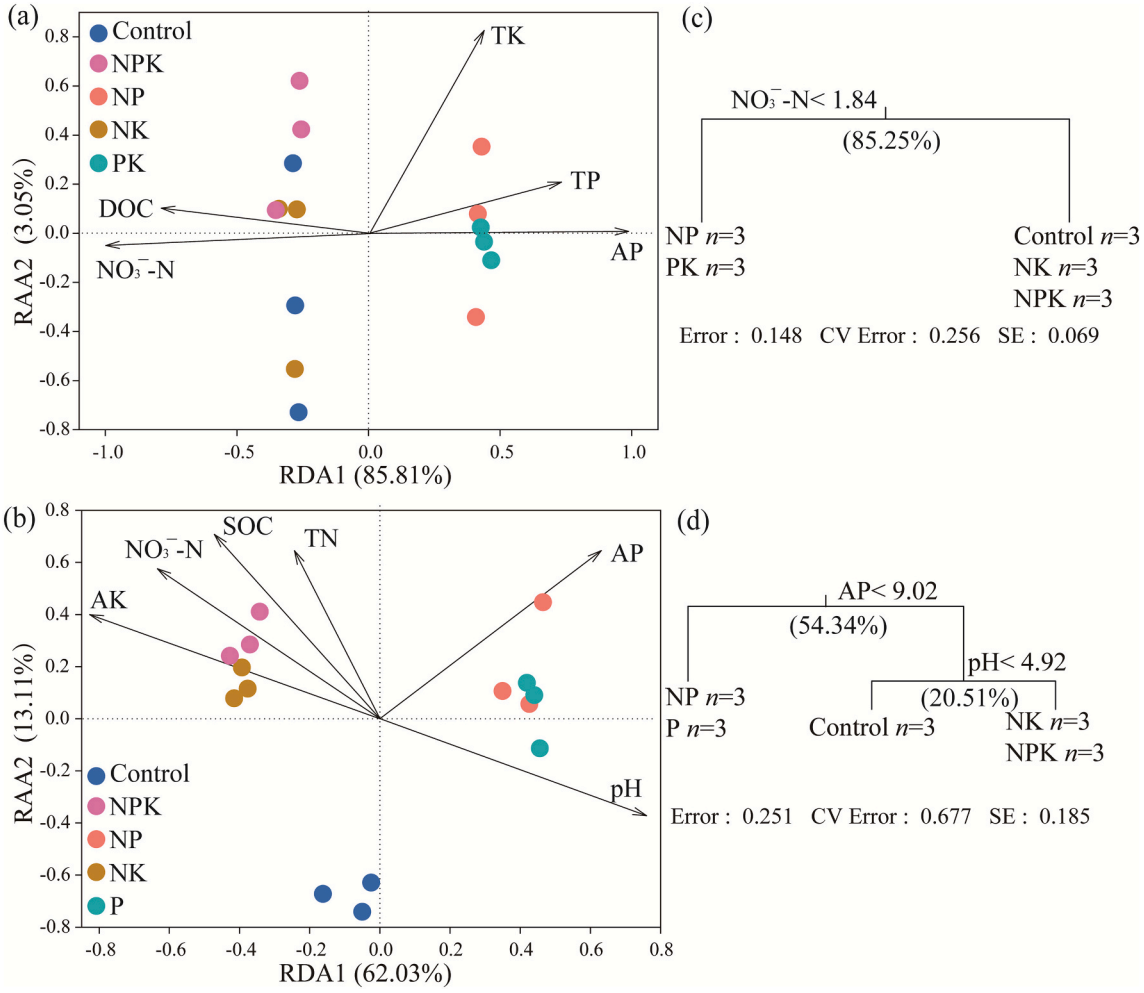

Fig. 5. Redundancy analysis (RDA) (a, b) and Multivariate regression tree (MRT) (c, d) of the bacterial communities and soil properties in the paddy soil (a, c) and upland soil (b, d) under the five fertilizer treatments. (a, b) RDA correlation plots exhibiting variances in bacterial community composition explained by the soil properties. (c, d) Numbers under the crosses of each split indicate percentages of variation explained by the split. CV Error represent cross-validation error, SE represent standard error of the tree. Specific treatment regime and the numbers of samples included in the analysis are shown in each split.

NP plus P treatments had higher AP content, accompanied by higher abundances of Gemmatimonadaceae species (Table 1; Fig. 3). Gemmatimonadaceae species have been reported to be polyphosphate-accumulating microorganisms (Zhang et al., 2003), which are more responsive to higher levels of P availability (Su et al., 2015). This is consistent with the positive relationship between Gemmatimonadaceae and AP content (Table S8). The NPK plus NK treatments had lower pH and AP contents, and their bacterial community composition was characterized by the enrichments of Thermogemmatisporales and Acidobacteria subgroup 2 species. The observed negative relationships between the soil pH and abundances of Thermogemmatisporales and Acidobacteria subgroup 2 (Table S8) are consistent with previous results showing that lower pH favored for the growth of both Thermogemmatisporales and Acidobacteria subgroup 2 (Rousk et al., 2010; Lin et al., 2019). The Control had significantly lower abundance of unclassified Acidobacteriaceae subgroup 1 members (Fig. 3), which have been described as nitrification-related microorganisms (Cheng et al., 2017), consisting with our observed results that the lower level of Acidobacteriaceae subgroup 1 in the Control treatment was accompanied by the lower level of soil NO_3^--N (Table 1; Fig. 3).

4.2. Fertilization-induced changes in SOC molecular composition show associations with different traits from the paddy soil and upland soil

In the paddy soil, the molecular composition of SOC was separated by the treatments with N application or not (Fig. 4a). The treatments with N application (NPK plus NP plus NK) were characterized by higher CCH_3 and aromatic C—C contents, and those without N application (PK plus Control) by higher aromatic C—H content (Table 2). In addition, we found that the higher amorphous Fe_2O_3 content in PK plus Control group showed positive association with aromatic C—H content and negative associations with CCH_3 and aromatic C—C contents (Table 3), implying that changes in Fe_2O_3 content in the paddy soil may be one of the potential roles in mediating the variations in molecular composition in this soil. Chemical stabilization of SOC by binding to Fe/Al oxides is a prevailing mechanism controlling SOC sequestration in paddy soil (Zhou et al., 2009; Wei et al., 2017b; Yang et al., 2018). Some studies have shown that amorphous Fe-oxide was the deterministic factor for SOC sequestration and stabilization due to its higher sorption and chelation capacities considering its small particle size, large surface area, and charge characteristics (Cui et al., 2014; Das et al., 2019). Amorphous Fe_2O_3 tended to selectively preserve plant-derived SOC from biological attack (Mikutta et al., 2005; Hall et al., 2018). Wen et al. (2019) and Xue et al. (2020) observed that amorphous Fe_2O_3 would preferentially stabilize aromatic compounds. Aromatic C—H content appeared to be

plant-derived compound (Zhang et al., 2015). We therefore inferred that aromatic C—H enriched in the PK plus Control treatments resulted most likely from the protection of higher amorphous Fe_2O_3 by enhancing surface area and complexation capacity (Berhe et al., 2012). On the contrary, the NPK plus NP plus NK treatments with lower amorphous Fe_2O_3 content may tend to stabilize more C originating from microbial processes. Zhang et al. (2015) reported that the non-protonated aromatic C (aromatic C—C) was possibly a microbially processed product, and the higher rotating CCH_3 group in the treatments with N application may be derived from microbial degradation of long-chain polymethylene structures (Zhang et al., 2015). However, the significant correlations between amorphous Fe_2O_3 and SOC molecular structures were obtained from extreme fertilization treatments, conclusive evidence for their relationships would require further study with gradual fertilization management.

In the upland soil, molecular composition was separated by the treatments with P application or not (Fig. 4b). The treatments with P application (NPK plus NP plus P) were characterized by higher OCH content, and those without P application (NK plus Control) by higher CH/CH_2 content. The specific P-shaped molecular composition of the upland soil was consistent with some previous studies (Xu et al., 2017a; Li et al., 2018; Shen et al., 2018). P-deficiency has been shown to reduce the microbial biomass and enzymatic activity more significantly than N- or K-deficiency in upland soils (Chu et al., 2007; Shen et al., 2018). This discrepancy may result in the relative accumulation of CH/CH_2 groups in the NK plus Control treatments that suffered from long-term P-deficiency, since the long-chain CH/CH_2 in aliphatic structure was suggested as the most biologically recalcitrant fraction of SOC and has been found to be selectively preserved because its intrinsic molecular properties incur greater energy cost for microbial decomposition (Baldock et al., 1992; Zech et al., 1997; Xu et al., 2017b). The negative associations between CH/CH_2 and TP and AP content have also been observed in P-limited soils (Gressel et al., 1996; Mathers and Xu, 2003). On the contrary, the NPK plus NP plus P treatments had higher OCH content (Table 2), which is considered as easily degradable fraction in the O–alkyl C that derived from plant polysaccharides (Kögel-Knabner, 2002; Zhang et al., 2015; Li et al., 2020). We thus inferred that the higher OCH content in the treatments with P application may be attributed to the larger roots and higher amount of exudates from greater maize productivity following P application.

4.3. *Fertilization-induced changes in the SOC molecular composition are independent of the bacterial community composition*

Inconsistent distributions of the soil bacterial community and SOC molecular composition among the long-term fertilization strategies were observed in both the paddy soil and upland soil (Figs. 2 and 4), which were contrary to some previous studies emphasizing their close relationships (Ng et al., 2014; Wang et al., 2017; Li et al., 2018). This inconsistencies suggested that soil bacterial community might not be the dominant driver structuring SOC molecular composition under long-term imbalanced fertilization, even though bacterial species were reported as the predominant microbes responsible for SOC decomposition and mineralization in acidic paddy and upland soils (Murase et al., 2006; Qiu et al., 2018; Zhan et al., 2018). Similarly, in a neutral-acid soil (pH 5.99–7.66), Chen et al. (2020) found that that shifts of SOC molecular composition were not accompanied by changes in bacterial community composition after subjected to long-term fertilization. The molecular composition of SOC could be influenced by various factors other than the bacteria, such as the chemical and physical stabilization processes (Grandy and Neff, 2008; Zhao et al., 2020) and fungal-mediated transformations (Li et al., 2017b; Ng et al., 2014). Our results indicated that the differences in chemical stabilization processes carried out by amorphous Fe_2O_3 may play a more pronounced role in driving structural divergence of SOC after long-term imbalanced fertilization in the paddy soil. Whereas the typical P-shaped SOC molecular composition in the upland soil might result from the influence of fungal community composition, since fungi play a substantial role in organic matter degradation in upland soils (Zhan et al., 2018). More particularly, Li et al. (2017b) found that the SOC molecular changes of acidic forest soils were tightly related to shifts in soil fungal community composition, and Li et al. (2019) also reported that the SOC molecular composition of an acidic grassland was more tightly linked to fungi than bacteria.

The paddy soil and upland soil exhibited distinct patterns of both bacterial community and SOC molecular composition (Fig. 2; Table 2). This differentiation may be derived from their contrasting redox potential, which could trigger a series of distinct biological and chemical processes (Sahrawat, 2015; Meng et al., 2019). Further studies are needed to confirm. Another point should be mentioned was that our findings were obtained from the long-term imbalanced fertilization experiments with extreme treatments, and additional efforts should be made to gather more comprehensive assessments of the effects of imbalanced fertilization.

5. Conclusion

Using two adjacent long-term field experiments performed on paddy soil and upland soil for more than 30 years in subtropical China, we found that NO_3^--N was the most influential factor in regulating the bacterial community composition in the paddy soil, whereas AP and pH became the important factors in the upland soil. It was observed that the SOC molecular composition from the paddy soil was separated by treatments with N application or not, in which treatments with N application were enriched with aromatic C—H and CCH_3 and treatments without N application were enriched with aromatic C—H, and all these changed C functional groups had close associations with soil amorphous Fe_2O_3. However, the SOC molecular composition from the upland soil was separated by treatments with P application or not, in which treatments with and without P application were characterized by enrichment of OCH and CH/CH_2, respectively, and both of them showed close association with soil AP and TP. Our work detailed the profound influence of long-term imbalanced fertilization on the bacterial community and SOC molecular composition in the paddy soil and upland soil. Further studies are required by integrating more real field situations to ensure these results are consistent.

Lowercase letters refer to the comparison within the five treatments of each soil parameter for each soil type at $p < 0.05$.

ns: non-significant at $p = 0.05$; * and ** marks: significant at $p < 0.05$ and $p < 0.01$, respectively.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgements

This work was financially supported by the National Natural Science Foundation of China (41977102), the Earmarked Fund for China Agriculture Research System (CARS-03), and the Natural Science Foundation of Jiangxi Province (20192BAB203022). We greatly thank the anonymous Reviewers and the Editor for their constructive comments.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.apsoil.2021.103982.

References

Baldock, J.A., Oades, J.M., Waters, A.G., Peng, X., Vassallo, A.M., Wilson, M.A., 1992. Aspects of the chemical structure of soil organic materials as revealed by soil-state ^{13}C NMR spectroscopy. Biogeochemistry 16, 1–42.

Berhe, A.A., Suttle, K.B., Burton, S.D., Banfield, J.F., 2012. Contingency in the direction and mechanics of soil organic matter responses to increased rainfall. Plant Soil 358, 371–383.

Caporaso, J.G., Kuczynski, J., Stombaugh, J., Bittinger, K., Bushman, F.D., Costello, E.K., Fierer, N., Peña, A.G., Goodrich, J.K., Gordon, J.I., Huttley, G.A., Kelley, S.T., Knights, D., Koenig, J.E., Ley, R.E., Lozupone, C.A., McDonald, D., Muegge, B.D., Pirrung, M., Reeder, J., Sevinsky, J.R., Tumbaugh, P.J., Walters, W.A., Widmann, J., Yatsunenko, T., Zaneveld, J., Knight, R., 2010. QIIME allows analysis of high-throughput community sequencing data. Nat. Methods 7, 335–336.

Chen, C., Zhang, J.N., Lu, M., Qin, C., Chen, Y.H., Yang, L., Huang, Q.W., Wang, J.C., Shen, Z.G., Shen, Q.R., 2016. Microbial communities of an arable soil treated for 8 years with organic and inorganic fertilizers. Biol. Fertil. Soils 52, 455–467.

Chen, L., Li, F., Li, W., Ning, Q., Li, J., Zhang, J., Ma, D., Zhang, C., 2020. Organic amendment mitigates the negative impacts of mineral fertilization on bacterial communities in Shajiang black soil. Appl. Soil Ecol. 150, 103457.

Chen, R.R., Zhong, L.H., Jing, Z.W., Guo, Z.Y., Li, Z.P., Lin, X.G., Feng, Y.Z., 2017. Fertilization decreases compositional variation of paddy bacterial community across geographical gradient. Soil Biol. Biochem. 114, 181–188.

Cheng, J.B., Chen, Y.C., He, T.B., Liao, R.J., Liu, R.L., Yi, M., Huang, L., Yang, Z.M., Fu, T. L., Li, X.Y., 2017. Soil nitrogen leaching decreases as biogas slurry DOC/N ratio increases. Appl. Soil Ecol. 111, 105–113.

Chu, H.Y., Lin, X.G., Fujii, T., Morimoto, S., Yagi, K., Hu, J.L., Zhang, J.B., 2007. Soil microbial biomass, dehydrogenase activity, bacterial community structure in response to long-term fertilizer management. Soil Biol. Biochem. 39, 2971–2976.

Clarke, K.R., 1993. Nonparametric multivariate analyses of changes in community structure. Aust. J. Ecol. 18, 117–143.

Cui, J., Li, Z., Liu, Z., Ge, B., Fang, C., Zhou, C., Tang, B., 2014. Physical and chemical stabilization of soil organic carbon along a 500-year cultived soil chronosequence originating from estuarine wetlands: temporal patterns and land use effects. Agric. Ecosyst. Environ. 196, 10–20.

Daquiado, A.R., Kuppusamy, S., Kim, S.Y., Kim, J.H., Yoon, Y.E., Kim, P.J., Oh, S.H., Kwak, Y.S., Lee, Y.B., 2016. Pyrosequencing analysis of bacterial community diversity in long-term fertilized paddy field soil. Appl. Soil Ecol. 108, 84–91.

Das, R., Purakayastha, T.J., Das, D., Ahmed, N., Kumar, R., Biswas, S., Walia, S.S., Singh, R., Shukla, V.K., Yadava, M.S., Ravisankar, N., Datta, S.C., 2019. Long-term fertilization and manuring with different organics alter stability of carbon in colloidal organo-mineral fraction in soils of varying clay mineralogy. Sci. Total Environ. 684, 682–693.

De'ath, G., 2006. Mvpart: multivariate partitioning. R package version 1.2–4.

Eo, J., Park, K.C., 2016. Long-term effects of imbalanced fertilization on the composition and diversity of soil bacterial community. Agric. Ecosyst. Environ. 231, 176–182.

Francioli, D., Schulz, E., Lentendu, G., Wubet, T., Buscot, F., Reitz, T., 2016. Mineral vs. organic amendments: microbial community structure, activity and abundance of agriculturally relevant microbes are driven by long-term fertilization strategies. Front. Microbiol. 7, 1446.

Grandy, A.S., Neff, J.C., 2008. Molecular C dynamics downstream: the biochemical decomposition sequence and its impact on soil organic matter structure and function. Sci. Total Environ. 404, 297–307.

Gressel, N., Mccoll, J.G., Preston, C.M., Newman, R.H., Powers, R.F., 1996. Linkages between phosphorus transformations and carbon decomposition in a forest soil. Biogeochemistry 33, 97–123.

Hall, S.J., Berhe, A.A., Thompson, A., 2018. Order from disorder: do soil organic matter composition and turnover co-vary with iron phase crystallinity? Biogeochemistry 140, 93–110.

Hall, S.J., Ye, C.L., Weintraub, S.R., Hockaday, W.C., 2020. Molecular trade-offs in soil organic carbon composition at continental scale. Nat. Geosci. 13, 687–692.

He, J.Z., Zheng, Y., Chen, C.R., He, Y.Q., Zhang, L.M., 2008. Microbial composition and diversity of an upland red soil under long-term fertilization treatments as revealed by culture-dependent and culture-independent approaches. J. Soils Sediments 8, 349–358.

He, Y.T., He, X.H., Xu, M.G., Zhang, W.J., Yang, X.Y., Huang, S.M., 2018. Long-term fertilization increases soil organic carbon and alters its chemical composition in three wheat-maize cropping sites across central and South China. Soil Tillage Res. 177, 79–87.

Huang, Q., Wang, J.L., Wang, C., Wang, Q., 2019. The 19-years inorganic fertilization increased bacterial diversity and altered bacterial community composition and potential functions in a paddy soil. Appl. Soil Ecol. 144, 60–67.

Jackson, M.L., 1958. Soil Chemical Analysis. Englewood Cliffs, New Jersey.

Jin, W., Cao, W., Liang, F., Wen, Y., Wang, F., Dong, Z., Song, H., 2020. Water management impact on denitrifier community and denitrification activity in a paddy soil at different growth stages of rice. Agr. Water Manage. 241, 106354.

Johnston, A.E., 1997. Food security in the WANA region, the essential need for fertilizers. Proceedings of the Regional Workshop of the International Potash Institute. Izmir, Turkey, pp. 11–30.

Kögel-Knabner, I., 2002. The macromolecular organic composition of plant and microbial residues as inputs to soil organic matter. Soil Biol. Biochem. 34, 139–162.

Li, D.D., Chen, L., Xu, J.S., Ma, L., Olk, D.C., Zhao, B.Z., Zhang, J.B., Xin, X.L., 2018. Chemical nature of soil organic carbon under different long-term fertilization regimes is coupled with changes in the bacterial community composition in a Calcaric Fluvisol. Biol. Fertil. Soils 54, 999–1012.

Li, D.D., Zhao, B.Z., Olk, D.C., Zhang, J.B., 2020. Soil texture and straw type modulate the chemical structure of residues during four-year decomposition by regulating bacterial and fungal communities. Appl. Soil Ecol. 155, 103664.

Li, F., Chen, L., Zhang, J.B., Yin, J., Huang, S.M., 2017a. Bacterial community structure after long-term organic and inorganic fertilization reveals important associations between soil nutrients and specific taxa involved in nutrient transformations. Front. Microbiol. 8, 187.

Li, Y., Li, Y., Chang, S.X., Liang, X., Qin, H., Chen, J., Xu, Q., 2017b. Linking soil fungal community structure and function to soil organic carbon chemical composition in intensively managed subtropical bamboo forests. Soil Biol. Biochem. 107, 19–31.

Li, Y., Nie, C., Liu, Y.H., Du, W., He, P., 2019. Soil microbial community composition closely associates with specific enzyme activities and soil carbon chemistry in a long-term nitrogen fertilized grassland. Sci. Total Environ. 654, 264–274.

Lin, Y., Ye, G., Kuzyakov, Y., Liu, D., Fan, J., Ding, W., 2019. Long-term manure application increases soil organic matter and aggregation, and alters microbial community structure and keystone taxa. Soil Biol. Biochem. 134, 187–196.

Ling, N., Zhu, C., Xue, C., Chen, H., Duan, Y.H., Peng, C., Guo, S.W., Shen, Q.R., 2016. Insight into how organic amendments can shape the soil microbiome in long-term field experiments as revealed by network analysis. Soil Biol. Biochem. 99, 137–149.

Mathers, N.J., Xu, Z., 2003. Solid-state ^{13}C NMR spectroscopy: characterization of soil organic matter under two contrasting residue management regimes in a 2-year-old pine plantation of subtropical Australia. Geoderma 114, 19–31.

Meng, D., Li, J., Liu, T., Liu, Y., Yan, M., Hu, J., Li, X., Liu, X., Liang, Y., Liu, H., Yin, H., 2019. Effects of redox potential on soil cadmium solubility: insight into microbial community. J. Environ. Sci.-China 75, 224–232.

Mikutta, R., Kleber, M., Jahn, R., 2005. Poorly crystalline minerals protect organic carbon in clay subfractions from acid subsoil horizons. Geoderma 128, 106–115.

Murase, J., Matsui, Y., Katoh, M., Sugimoto, A., Kimura, M., 2006. Incorporation of ^{13}C-labeled rice-straw-derived carbon into microbial communities in submerged rice field soil and percolating water. Soil Biol. Biochem. 38, 3483–3491.

Ng, E.L., Patti, A.F., Rose, M.T., Schefe, C.R., Wilkinson, K., Smernik, R.J., Cavagnaro, T. R., 2014. Does the chemical nature of soil carbon drive the structure and functioning of soil microbial communities? Soil biol. Biochem. 70, 54–61.

Olsen, S.R., Cole, C.V., Watanabe, F.S., Dean, L.A., 1954. Estimation of Available Phosphorus in Soils by Extraction With Sodium Bicarbonate. Washington DC.

Page, A.L., Miller, R.H., Keeney, D.R., 1982. Methods of soil analysis. Part 2, Chemical and Microbiological Properties. Madison, WI.

Pansu, M., Gautheyrou, J., 2006. Handbook of Soil Analysis. Mineralogical, Organic and Inorganic Methods. Springer, Berlin Heidelberg, Berlin.

Qiu, H.S., Ge, T.D., Liu, J.Y., Chen, X.B., Hu, Y.J., Wu, J.S., Su, Y.R., Kuzyakov, Y., 2018. Effects of biotic and abiotic factors on soil organic matter mineralization: experiments and structural modeling analysis. Eur. J. Soil Biol. 84, 27–34.

R Core Team, 2012. R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna.

Randall, E.W., Mahieu, N., Powlson, D.S., Christensen, B.T., 1995. Fertilization effects on organic matter in physically fractionated soils as studied by ^{13}C NMR: results from two long-term field experiments. Eur. J. Soil Sci. 46, 557–565.

Rasigraf, O., Schmitt, J., Jetten, M.S.M., Lüke, C., 2017. Metagenomic potential for and diversity of N-cycle driving microorganisms in the Bothnian Sea sediment. Microbiologyopen 6.

Rousk, J., Bååth, E., Brookes, P.C., Lauber, C.L., Lozupone, C., Caporaso, J.G., Knight, R., Fierer, N., 2010. Soil bacterial and fungal communities across a pH gradient in an arable soil. The ISME Journal 4, 1340–1351.

Sahrawat, K.L., 2015. Redox potential and pH as major drivers of fertility in submerged rice soils: a conceptual framework for management. Commun. Soil Sci. Plant. 46, 1597–1606.

Samaddar, S., Chatterjee, P., Truu, J., Anandham, R., Kim, S., Sa, T., 2019. Long-term phosphorus limitation changes the bacterial community structure and functioning in paddy soils. Appl. Soil Ecol. 134, 111–115.

Sheldrick, W.F., Syers, J.K., Lingard, J., 2003. Soil nutrient audits for China to estimate nutrient balances and output/input relationships. Agric. Ecosyst. Environ. 94, 341–354.

Shen, D.Y., Ye, C.L., Hu, Z.K., Chen, X.Y., Guo, H., Li, J.Y., Du, G.Z., Adl, S., Liu, M.Q., 2018. Increased chemical stability but decreased physical protection of soil organic carbon in response to nutrient amendment in a Tibetan alpine meadow. Soil Biol. Biochem. 126, 11–21.

Sheng, R., Qin, H., O'Donnell, A.G., Huang, S., Wu, J., Wei, W., 2015. Bacterial succession in paddy soils derived from different parent materials. J. Soils Sediments 15, 982–992.

Skjemstad, J.O., Clarke, P., Taylor, J.A., Oades, J.M., Newman, R.H., 1994. The removal of magnetic materials from surface soil. A soild state ^{13}C CP/MAS NMR study. Aust. J. Soil Res. 32, 1215–1229.

Su, J.Q., Ding, L.J., Xue, K., Yao, H.Y., Quensen, J., Bai, S.J., Wei, W.X., Wu, J.S., Zhou, J. Z., Tiedje, J.M., Zhu, Y.G., 2015. Long-term balanced fertilization increases the soil microbial functional diversity in a phosphorus-limited paddy soil. Mol. Ecol. 24, 136–150.

Turner, B.L., Lambers, H., Condron, L.M., Cramer, M.D., Leake, J.R., Richardson, A.E., Smith, S.E., 2013. Soil microbial biomass and the fate of phosphorus during long-term ecosystem development. Plant Soil 367, 225–234.

Wang, H.Y., Nie, Y., Butterly, C.R., Wang, L., Chen, Q.H., Tian, W., Song, B.B., Xi, Y.G., Wang, Y., 2017. Fertilization alters microbial community composition and functional patterns by changing the chemical nature of soil organic carbon: a field study in a halosol. Geoderma 292, 17–24.

Wang, N., Ding, L.J., Xu, H.J., Li, H.B., Su, J.Q., Zhu, Y.G., 2015. Variability in responses of bacterial communities and nitrogen oxide emission to urea fertilization among various flooded paddy soils. FEMS Microbiol. Ecol. 91, 3.

Wei, X.M., Hu, Y.J., Peng, P.Q., Zhu, Z.K., Atere, C.T., O'Donnell, A.G., Wu, J.H., Ge, T. D., 2017a. Effect of P stoichiometry on the abundance of nitrogen-cycle genes in phosphorus-limited paddy soil. Biol. Fertil. Soils 53, 767–776.
Wei, Z., Ji, J., Li, Z., Yan, X., 2017b. Changes in organic carbon content and its physical and chemical distribution in paddy soils cultivated under different fertilisation practices. J. Soils Sediments 17, 2011–2018.
Wen, Y.L., Liu, W.J., Deng, W.B., He, X.H., Yu, G.H., 2019. Impact of agricultural fertilization practices on organo-mineral associations in four long-term field experiments: implications for soil C sequestration. Sci. Total Environ. 651, 591–600.
Xu, J.S., Zhao, B.Z., Chu, W.Y., Mao, J.D., Olk, D.C., Zhang, J.B., Wei, W.X., 2017a. Evidence from nuclear magnetic resonance spectroscopy of the processes of soil organic carbon accumulation under long-term fertilizer management. Eur. J. Soil Sci. 68, 703–715.
Xu, Y.H., Fan, J.L., Ding, W.X., Gunina, A., Chen, Z.M., Bol, R., Luo, J.F., Bolan, N., 2017b. Characterization of organic carbon in decomposing litter exposed to nitrogen and sulfur additions: links to microbial community composition and activity. Geoderma 286, 116–124.
Xue, B., Huang, L., Huang, Y., Ali Kubar, K., Li, X., Lu, J., 2020. Straw management influences the stabilization of organic carbon by Fe (oxyhydr)oxides in soil aggregates. Geoderma 358, 113987.
Yang, F., Tian, J., Meersmans, J., Fang, H.J., Yang, H., Lou, Y.L., Li, Z.F., Liu, K.L., Zhou, Y., Blagodatskaya, E., Kuzyakov, Y., 2018. Functional soil organic matter fractions in response to long-term fertilization in upland and paddy systems in South China. Catena 162, 270–277.
Yang, Y.R., Li, X.G., Liu, J.G., Zhou, Z.G., Zhang, T.L., Wang, X.X., 2017. Bacterial diversity as affected by application of manure in red soils of subtropical China. Biol. Fertil. Soils 53, 639–649.
Yi, X., Yi, K., Fang, K., Gao, H., Dai, W., Cao, L., 2019. Microbial community structures and important associations between soil nutrients and the responses of specific taxa to rice-frog cultivation. Front. Microbiol. 10, 1752.
Zech, W., Senesi, N., Guggenberger, G., Kaiser, K., Lehmann, J., Miano, T.M., Miltner, A., Schroth, G., 1997. Factors controlling humification and mineralization of soil organic matter in the tropics. Geoderma 79, 117–161.
Zhan, Y.S., Liu, W.J., Bao, Y.Y., Zhang, J.W., Petropoulos, E., Li, Z.P., Lin, X.G., Feng, Y. Z., 2018. Fertilization shapes a well-organized community of bacterial decomposers for accelerated paddy straw degradation. Sci. Rep. 8, 7981.
Zhang, H., Sekiguchi, Y., Hanada, S., Hugenholtz, P., Kim, H., Kamagata, Y., Nakamura, K., 2003. *Gemmatimonas aurantiaca* gen. Nov., sp. nov., a gram-negative, aerobic, polyphosphate-accumulating micro-organism, the first cultured representative of the new bacterial phylum *Gemmatimonadetes* phyl. Nov. Int. J. Syst. Evol. Microbiol. 53, 1155–1163.
Zhang, Y.L., Yao, S.H., Mao, J.D., Olk, D.C., Cao, X.Y., Zhang, B., 2015. Chemical composition of organic matter in a deep soil changed with a positive priming effect due to glucose addition as investigated by ^{13}C NMR spectroscopy. Soil Biol. Biochem. 85, 137–144.
Zhao, B.Z., Chen, J., Zhang, J.B., Xin, X.L., Hao, X.Y., 2013. How different long-term fertilization strategies influence crop yield and soil properties in a maize field in the North China plain. J. Plant Nutr. Soil Sci. 176, 99–109.
Zhao, Q., Callister, S.J., Thompson, A.M., Kukkadapu, R.K., Tfaily, M.M., Bramer, L.M., Qafoku, N.P., Bell, S.L., Hobbie, S.E., Seabloom, E.W., Borer, E.T., Hofmockel, K.S., 2020. Strong mineralogic control of soil organic matter composition in response to nutrient addition across diverse grassland sites. Sci. Total Environ. 736, 137839.
Zhong, W.H., Cai, Z.C., 2007. Long-term effects of inorganic fertilizers on microbial biomass and community functional diversity in a paddy soil derived from quaternary red clay. Appl. Soil Ecol. 36, 84–91.
Zhou, J., Guan, D., Zhou, B., Zhao, B., Ma, M., Qin, J., Jiang, X., Chen, S., Cao, F., Shen, D., Li, J., 2015. Influence of 34-years of fertilization on bacterial communities in an intensively cultivated black soil in northeast China. Soil Biol. Biochem. 90, 42–51.
Zhou, P., Song, G., Pan, G., Li, L., Zhang, X., 2009. Role of chemical protection by binding to oxyhydrates in SOC sequestration in three typical paddy soils under long-term agro-ecosystem experiments from South China. Geoderma 153, 52–60.