Soil Fungal Community Composition Is More Sensitive To Nitrogen And Phosphorus Additions Than The Bacterial Community In A Temperate Meadow Ecosystem

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Abstract

Background and aims Soil microorganisms play key roles in the cyclings of soil nutrient and ecosystem stability, however, the soil microbial community composition often is influenced by nutrient enrichment. Increased nitrogen (N) input endangers plant species diversity and stability of grassland ecosystems, and which should be often affected by the addition of phosphorus (P) addition. However, the resistance of soil microbial community to N and P additions, and their interactions in the N-limited temperate meadow ecosystem remains poorly understood.

Methods We carried out a seven-year experiment with the additions of N and P in a temperate meadow. The community structures of soil bacteria and fungi were examined based on high-throughput sequencing targeting the 16S rRNA and ITS genes, respectively.

Results N addition did not influence on the community composition and species richness of bacteria, while altered soil fungal community composition and increased fungal OTU richness. P addition significantly altered soil fungal and bacterial community compositions and declined the richness of bacterial OTU, increased the OTU richness of fungi. The Proteobacteria (38.5%) and Acidobacteria (22.3%) were the most dominant bacteria. The Ascomycota was the dominant fungi (42.6%) across all samples. The enrichment of soil available P concentration caused by the addition of P played key roles in the bacterial β -diversity, while the β -diversity of soil fungi was mainly influenced by the concentrations of soil N and P, and soil moisture.

Conclusions The sensitivity of soil fungi and bacteria to the addition of P is stronger than N addition, and which is more sensitive than the plant community. Our result highlights the unequal sensitivity of soil fungal and bacterial community composition and structure to the eutrophications of N and P, thereby causing changes in above and belowground communities and influencing ecosystem stability in the studied temperate meadow ecosystem.

Introduction

Soil microbe plays important role in mediating ecosystem stability and multifunctionality (van der Heijden et al., 2008; Delgado-Baquerizo et al., 2016; Bennett et al., 2017). For instance, soil microbe could determine plant diversity and aboveground biomass (Maron et al., 2011; Wang et al., 2019). Soil microbial biodiversity could alter the competition of coexistence species and the relationship between the complexity and productivity of plant species (Wagg et al., 2011; Siefert et al., 2018). Soil microbe could reduce the infection of plant pathogens by increasing soil moisture and nutrient absorption to benefit the growth of the plant. (van der Heijden et al., 2006; Cavagnaro et al., 2015). Besides, soil fungi could alleviate greenhouse gas emissions, such as N₂O and CH₄ (Bender et al., 2014, 2015; Thompson et al., 2016; Storer et al., 2018). The shifts in the soil microbiome could affect soil carbon, nitrogen (N), and phosphorus (P) cyclings because soil microbe could affect litter decomposition (Kang et al., 2020), plants N and P uptakes (Freedman et al., 2013; Mei et al., 2019).

A series of global change factors such as climate warming, N deposition, rising atmospheric carbon dioxide concentration is seriously threatening the ecosystem. The community composition and species diversity of soil microorganisms have been influencing by global change. Several studies have found that the composition of soil microbial communities species could be shifted by N enrichment (Allison et al., 2008). The addition of N reduced bacterial richness in soil but had no impact on fungal biomass in a tropical forest ecosystem in south China (Wang et al., 2018), and decreased bacterial operational taxonomic unit (OTU: Generally, if the similarity of 16S rRNA sequences is higher than 97%, it can be defined as an OTU, and each OTU corresponds to a different 16s rRNA sequence, that is, each OTU corresponds to a microbial specie) richness in a temperate steppe (Zeng et al., 2016), reduced bacterial and fungal richness in a hardwood forest ecosystem (Freedman et al., 2015), but exerted few influences on soil bacterial diversity in the Cedar Creek Ecosystem (Fierer et al., 2012), indicating that the effects of N application on soil microbial communities composition were inconsistent (Ramirez et al., 2012; Leff et al., 2015) but which might be ecosystem-specific. The previous results have demonstrated that the change of soil microbial community could be shaped by the increase in soil available N, the decline of soil pH, and the changes in plant species composition caused by N addition (Rousk et al., 2010; Zeng et al., 2016).

Besides that, N enrichment would break the soil nutrients balance, which is the major determinant for the soil microbial community (Wan et al., 2015). The increasing evidence demonstrated that P deposition is becoming one of the most important P sources at a global scale (Ahn and James, 2001; Vicars et al., 2010). The previous results found that the addition of P increased the abundances of arbuscular mycorrhizal fungi and bacteria in a subalpine meadow (Huang et al., 2016), altered soil fungal and bacterial community structure in an acidic grassland (Rooney and Clipson, 2009), increased abundance of non-AM fungi and altered the soil fungal community structure in a P- deficient woodland (Nielsen et al., 2015). The change of soil available P concentration caused by P addition was a key parameter shifting the diversity and composition of soil microbial community (He et al., 2016; Ling et al., 2017). P addition mitigated the negative influence of N deposition on the functional genes of soil microbe (Su et al., 2015) so that the P addition might migrate the negative impact of N enrichment on the community composition of belowground and aboveground. However, Li et al. (2015) found that P addition could enhance soil microbial biomass, but some studies found that P addition had few influences on the community composition of soil microbe under N enrichment (He et al., 2016; Wang et al., 2018). Up to now, the influences of N and P additions, and their interactive effects on the community composition of soil microbe are still not well understood.

The Songnen grassland is the largest temperate meadow in China, which has been influencing by N deposition and P addition. Our earlier results suggested that the addition of N declined the plant species richness (Zhang et al., 2015, 2016), while P addition did not alter plant species richness but made plant community structure change (Zhao et al., 2019), N addition could enhance the activities of urease and phosphatase (Gong et al., 2015). Moreover, arbuscular mycorrhizal fungi could alleviate P limitation (Mei et al., 2019) and reduce the negative influences of N enrichment on the plant community (Zhang et al., 2016). However, the response of the communities of soil bacteria and fungi to the N and P additions and

the relationship between soil community and plant community remains unclear in the temperate meadow ecosystem.

Consequently, we conducted a 7-year field experiment with N and P additions in the Songnen meadow. This study aimed to clarify the influences of long-term N and P additions, and their interaction on the soil bacterial and fungal communities, and to elucidate the relationship between the communities of belowground and aboveground in a temperate meadow. We hypothesized that: (1) the additions of N and P would influence the community compositions of soil bacteria and fungi because of the decline of soil pH caused by N and P additions; (2) the sensitivities of soil bacteria and fungi would be different; (3) the changes of soil community structure induced by the additions of N and P would be determined by the changes in plant community composition caused by N and P additions.

Material And Methods

Experiment site

This experiment was performed in a temperate meadow (123°45' E, 44° 45' N), in the west of Jilin Province, northeastern China. The annual rainfall in the experiment site is approximately 300-500 mm with a mean of 400 mm (Kang et al., 2020). The annual air temperature on average is 2.4-2.7 °C. In this area, the soil is Chernozem soil, with a higher pH (7.5–9.0) and low organic matter (3–4%) (Zhang et al., 2016). Soil N is limited with a soil N of 1.8 g kg⁻¹ in total and soil available P concentration of 2.5 mg kg⁻¹ (Mei et al., 2019), and which is dominated by *Leymus chinensis*, and with some subordinate species.

Experiment design

Two nutrient factors, N and P additions were included in our experiment, and a completely randomized block was used. There were three treatments: N addition (N), P addition (P), N + P addition (NP), and one control (C), with three replications. The experimental plot size was 2 × 2 m, with buffer zones (2 m in width) between plots. Previous studies reported that the saturation rates of soil N and P additions for plant communities are 10.5 g N m⁻² year⁻¹ and 10 g P m⁻² year⁻¹ in grassland in northern China (Bai et al., 2010; Zhao et al., 2019), respectively. For the additions of N and P, NH₄NO₃ solution (10 g N m⁻² in 10 L water) and Ca(H₂PO₄)₂ solution (10 g P m⁻² in 10 L water) were added to the plot before plants germination, respectively. In the addition of NP treatment, NH₄NO₃ and Ca(H₂PO₄)₂ (10 g N and 10 g P m⁻² in 10 L water) were simultaneously added to the experimental plot. To reduce the water effects caused by N and P additions on the experiment result, the same amount of water (without N and P) as N and P additions was added in the control plot. The experiment started in May 2013.

Sampling and measurements

The soil was sampled in August 2019 for measuring soil pH, the concentrations of soil total N, available N (NH₄-N, NO₃-N), available P, soil moisture, and the community and composition structure of soil

bacteria and fungi. In each plot, five soil cores (5 cm in inner diameter, 20 cm in-depth) were drilled randomly, and then homogenized thoroughly and sieved (2-mm) before chemical analysis. Soil available N and P were determined according to Mei et al. (2019). Soil pH was measured using a glass electrode.

Plant community composition and productivity

To explore the relationship between soil and plant community composition under N and P additions, the plant community composition was studied after seven years of nutrients addition. The plant species in each plot were recorded on 15th July (the peak of the growing season) 2019 according to the methods (Zhao et al., 2019), the plant species number (species number m²) and density were recorded for calculating the Shannon-Wiener index H. We cut randomly two quadrants (50 × 50 cm²) in each plot in September 2019 for measuring the aboveground biomass, which included the dead biomass of the previous year. After cutting the quadrants, the aboveground plants were weighed after drying 48 h at 65 °C.

Soil DNA extraction and high-throughput sequencing

The Power Soil DNA Isolation Kit (MO BIO Laboratories) was used to extract soil DNA. The quantity and quality of DNA were evaluated respectively and then stored the DNA at -80°C before use. The 16S rRNA gene of bacteria (in the V3-V4 region) was amplified by the primers 338-F (5'- ACTCCTACGGGAGGCAGCA-3') and 806-R (5'- GGACTACHVGGGTWTCTAAT-3') (Mori et al., 2014). We amplified the rRNA gene of fungi (ITS1 region) using ITS1-F (5'-CTTGGTCATTTAGAGGAAGTAA-3') and ITS2-R (5'-GCTGCGTTCTTCATCGATGC-3') primers (White et al., 1990, Gardes and Bruns, 1993).

We performed the amplification of PCR in a total volume of 50 µl including the buffer, DNA Polymerase, etc, which should be found in Table S1. The description of the thermal cycling conditions of first step PCR could be found in Table S2. We purified the PCR products of the first step using the DNA Clean Beads (VAHTSTM). The amplification PCR of the second round and conditions of thermal cycling could be found in Table S3 and S4. The sequencings of bacteria and fungi were performed using the platform of Illumina Hiseq 2500 (2×250 paired ends).

Sequence analyses

The paired-end reads of the fungal ITS gene and bacterial 16S rRNA gene were processed, and the ITS region and 16S sequences were screened out for quality control according to the methods by Guo et al. 2019. We clustered all the tags of ≥97% identity into the operational taxonomic units (OTUs). The tags were classified into different taxonomies according to the Silva and UNITE databases for soil bacterial and fungal communities, respectively. There were 17,822 OTUs of soil bacteria and 4986 OTUs of soil fungi after removing those OTUs which did not belong to the soil bacterial and fungal community.

Data analyses

To determine the influence of additions of N and P, and their interactive effects on soil total N and available P concentrations, soil moisture, soil pH, plant species diversity (Shannon-Wiener index, H), aboveground biomass, soil bacterial and fungal α -diversity (the indexes of ACE index, Chao1, Shannon and Simpson), and the dominant (> 1%) bacterial and fungal groups abundances two-way ANOVA was used. The N addition and P additions effects and their interaction on community compositions of soil bacteria and fungi were tested by a PERMANOVA using the vegan package with 104 permutations. We also visualized how taxonomic frequency and abundance responded to N addition and P addition using ternary plots. To explore the variations in soil fungal and bacterial species compositions into the fractions illustrated by plant diversity, productivity, soil N and P concentrations, moisture, and pH, we performed redundancy analysis based on the distance (dbRDA) according to Wang et al. (2018).

Results

Plant and soil parameters

N addition remarkably enhanced soil total N concentration, but no influence on the concentration of soil total P (Fig. 1a, 1b). P addition both improved the concentrations of total N ($P < 0.05$) and available P ($P < 0.05$) in soil (Fig. 1a and b). In contrast to the control, the soil N and P concentrations were higher in the NP addition treatment, but the difference in soil N concentration was noticed between the treatments of N and NP additions is not significant (Fig. 1a), and had no obvious difference in soil P concentration between the additions of P and NP (Fig. 1b). N addition and P addition interactively affected soil N concentration ($P < 0.01$). The addition of N decreased plant species diversity ($P < 0.05$, Fig. 1c), but significantly increased aboveground biomass (Fig. 1d). P addition significantly increased plant species diversity (Fig. 1d), but no effect on aboveground biomass. NP addition increased plant species diversity compared to the N addition ($P < 0.05$, Fig. 1c). However, no difference in aboveground biomass between the treatments of N and NP addition was detected (Fig. 1d).

OTU richness and α -diversity

Rarefaction analysis was performed to compare the levels of bacterial and fungal diversities in terms of the total number of OTU and the Chao 1 index, and the rarefaction results are presented in Supplementary Data Fig. S1. N addition had no impact on the soil bacterial OTU richness ($P > 0.05$), but which significantly reduced in the treatments of P ($P < 0.05$) and NP ($P < 0.05$) additions (Fig. 2a). N and P additions had no interactive effect on the bacterial OTU richness ($P > 0.05$). Both of the N ($P < 0.05$) and P ($P < 0.05$) additions increased the soil fungal OTU richness (Fig. 2b). The interactions between N and P additions affected the fungal OTU richness ($P < 0.01$), significantly.

The additions of N and P both remarkably declined the Chao1 index of soil bacterial (Fig. 2c). The Chao1 index in the treatment of NP addition was highly lower than those in N addition, P addition, and the control, significant differences were detected among all four treatments (Fig. 2c). A significant primary effect of P addition on the Chao1 index of bacteria was noticed. The additions of N and P remarkably

enhanced the Chao1 index of soil fungal, no obvious difference was detected between NP addition and the additions of N and P (Fig. 2d).

Soil bacterial and fungal community compositions

Proteobacteria (38.5%), Acidobacteria (22.4%), Actinobacteria (14.8%), and Gemmatimonadetes (8.3%) were dominated the soil bacterial community across all the treatments (Fig. 3a). P addition reduced the abundances of Proteobacteria ($P < 0.05$) and Gemmatimonadetes ($P < 0.05$), increased Acidobacteria ($P < 0.01$) and Actinobacteria abundances ($P < 0.05$), while neither the addition of N nor the addition of NP had influence on the dominant bacterial taxa abundances ($P > 0.05$). For soil bacteria, Proteobacteria and Gemmatimonadetes were closely tied to P addition (Fig. S2a), while no phyla were closely tied to N addition or control (Fig. S2a), several families were shared by N and P additions.

Ascomycota (42.7%), Basidiomycota (11.8%), Gemmatimonadetes (8.3%), and Chloroflexi (7.3%) dominated the soil fungal community across all samples (Fig. 3b). N addition increased Ascomycota ($P < 0.05$) and Basidiomycota ($P < 0.05$) abundances. The addition of P reduced the abundances of Basidiomycota ($P < 0.05$) and Chytridiomycota ($P < 0.05$). For soil fungal composition, Ascomycota was closely tied to P addition (Fig. S2b), while no phyla were closely tied to N addition (Fig. S2b).

The dbRDA results showed that significant shifts in the community compositions of bacterial (Fig. 4a) and fungal (Fig. 4b) were detected. The variation in the community of bacteria was mainly interpreted by the content of soil available P ($P < 0.01$) and soil moisture ($P < 0.05$). Soil available P content ($P < 0.01$), soil total N content ($P < 0.05$), and soil moisture ($P < 0.05$) were regarded as the better vectors to illustrate the shift in the community composition of soil fungi. The additions P ($P < 0.05$) and NP ($P < 0.05$) strongly altered the community composition of bacteria, while the addition of N had no effect ($P > 0.05$; Fig. 5a). Significant main effects of N ($P < 0.01$) and P additions ($P < 0.01$), and interactive effect of P addition $\times$ N addition ($P < 0.01$) on soil fungal community composition were observed (Fig. 5b).

Discussion

Influences of N and P additions on soil bacterial diversity

The present study supplies some new insight into the impacts of N addition and P addition on the communities of bacteria and fungi in a temperate meadow. N addition highly enhanced soil N concentration and aboveground biomass, but reduced plant species diversity. Several results found that N enrichment reduced soil bacteria richness in the tropical forest, temperate steppe, and arctic tundra ecosystem (Campbell et al., 2010; Zeng et al., 2016; Wang et al., 2018; Yan et al., 2018), however, one earlier result found that N addition had few effects on the soil bacterial diversity at Cedar Creek (Ramirez et al., 2010). The addition of N also had no influence on soil bacterial OTU richness and α -diversity in the current study, (Fig. 2), suggesting that the response of soil bacterial species richness to N addition was very strong in Songnen meadow, which is inconsistent with our first hypothesis. It might be explained partly by the background nutrient availability or soil pH because many previous results have

demonstrated that the diversity of soil bacteria is frequently affected by soil pH, the decline of soil pH decreased the soil bacteria richness (Fierer and Jackson, 2006; Wang et al., 2018). The soil pH in our experiment site was much higher than the above-studied ecosystem, and N addition had no influence on soil pH, suggesting that N addition had few effects on soil bacterial α -diversity because no significant changes in soil pH resulted from N addition in the highly alkalized N-limited temperate meadow (Fig. S3a). Moreover, soil microbial diversity is likely to increase with the enhancement of plant diversity (van der Heijden et al., 2008). N addition highly decreased plant species diversity, but no impact on soil bacterial richness, suggesting that the sensitivity of plant species to N addition might be faster than soil bacteria. The present results also suggest that the controls on species diversities of plant and soil bacteria may not be the same (Fierer et al., 2012).

P addition significantly reduced soil bacterial OTU richness and α -diversity which is not in agreement with the earlier results that the P addition did not affect soil bacterial richness (Eo and Park, 2016), and the soil bacterial diversity is enhanced with P addition in an agricultural ecosystem (Tan et al., 2013). However, the reduction in the soil bacterial diversity might have no negative influence on soil functionality because of the functional redundancies in soil bacteria community (Pan et al., 2014). A significant negative correlation between soil bacterial richness and soil P concentration was detected (Fig. S3b). The present result also indicates that P addition might increase the competition for nutrients between soil bacteria and plants, and then reduce soil bacterial richness (Zhang et al., 2014). Moreover, P addition increased soil available P concentration which might reduce phosphate solubilizing bacteria, and then decrease soil bacterial richness, indirectly. However, P addition had no influence on plant species diversity and aboveground biomass, suggesting that P is not the most important limited nutrients.

Influences of N and P additions on the soil fungal diversity

Previous studies showed that the influences of N addition or P addition on soil fungal diversity are controversial. One previous result demonstrated that N enrichment decreased soil fungal diversity which depended on the types of ecosystem and the dose of nutrient addition (Zhou et al., 2016), but other results found that N fertilization highly enhanced soil fungal richness in N-limited ecosystem (Weber et al., 2013; Mueller et al., 2014). In the present study, the addition of N increased soil fungal richness which is in agreement with the above later results which might suggest that some of the fungal species are more adaptable and tolerant to N addition, and soil N and P concentrations had significant positive influences on soil fungal richness (Fig. S3c, S3d). Therefore, a small amount of nitrogen addition might contribute to the growth of soil fungi because that can improve plant growth and reduce the N competition of soil fungi indirectly.

The previous study reported that P fertilization reduced the soil fungi species richness (He et al., 2016). P addition significantly enhanced the soil fungal richness in our present result, which is inconsistent with the above results. The possible reason is that the addition of P could mitigate the P deficiency which improves the biomass of soil fungi (Liu et al., 2012). Moreover, in the studied meadow soil available P was usually severely limited (Zhao et al., 2019), P addition increased fungal diversity which suggests that

the P deficiency might be used for predicting the positive impacts of P addition on soil fungal richness directly.

Influences of N and P additions on the soil bacterial community composition

Several earlier results suggested that the addition of N shaped the community composition of soil bacteria, for instance, increased Proteobacteria abundance (Ramirez et al., 2012) but decreased the abundance of Chloroflexi and Acidobacteria (Zeng et al., 2016). Nevertheless, the N addition influences on soil bacterial community composition are inconsistent in grassland across the globe (Leff et al., 2015). Our results showed that N addition had no impact on the community composition of soil bacteria, which confirmed the consistency of N enrichment impacts on the bacterial community in soil. The possible reason is that N addition did not decline soil pH in the present study because many previous results reported that the decline of pH is a crucial factor that changes the bacterial community (Wang et al., 2018; Guo et al., 2019). Moreover, the db-RDA result indicates that the changes in bacterial community were mediated mainly by soil available P concentration, suggesting that the small changes of soil N addition caused by N input could not directly affect soil bacterial community composition by declining pH and affect soil C/N ratio. Although the addition of N significantly affected the structure of plant community and productivity in this studied ecosystem (Zhao et al., 2019), the changes of plant community structure and ANPP had no significant impact on the soil bacteria community, suggesting that the responses of soil bacteria and plant community might be dissimilar.

A previous study showed that P addition played a crucial role in soil bacterial community structure in tropical forests (Wang et al., 2018), however, another result suggested that P addition did not manipulate soil bacterial community composition in a temperate steppe (Ling et al., 2017). In the current research, the addition of P significantly altered soil bacterial community structure, increased Actinobacteria and Acidobacteria abundances, while declined the abundances of Proteobacteria and Gemmatimonadetes. Several studies have shown that changes in pH caused by P addition play a vital role in influencing the community composition of soil bacteria (Ling et al., 2017; Wang et al., 2018). In the current study, P addition had few effects on soil pH, however, the soil bacterial community was strongly determined by soil P concentration in the additions of P and NP, suggesting that the increase in soil P availability and nutrient balance might play a critical role in shaping the soil bacterial community composition. Besides, the increase of Acidobacteria and Actinobacteria might improve plant growth because Acidobacteria has highly metabolically activities in rhizosphere soil (Lee et al., 2008) and Actinobacteria could improve plant growth by alleviating soil disease suppression (Palaniyandi et al., 2013), which would increase C input from plant to soil with the enhancement of soil bacterial activities (Li et al., 2015). However, the influence of C/N/P ratios caused by the additions of N and P on the community of soil bacteria needs further study.

Influences of N and P additions on the community composition soil fungi

Previous results have demonstrated that the addition of N could alter the community structure of soil fungi (Leff et al., 2015; Yan et al., 2018; Wu et al., 2021). In the current present study, we found N addition

highly increased Ascomycota abundance, but decreased Basidiomycota abundances. Ascomycota is saprotrophic fungi that can accelerate soil C decomposition (Xiong et al., 2014), the increase of Ascomycota induced by N addition would increase soil C decomposition and speed up the C cycle. The present result might be used to explain the reason that the increased litter decomposition of *Leymus chinensis* caused by N addition in the studied ecosystem (Gong et al., 2015). N addition declined Basidiomycota abundance, which might reduce the pressures of competition on Ascomycota for resources (Weber et al., 2013). Furthermore, Glomeromycota abundance remarkably declined under the N addition suggesting that N addition would reduce plant species diversity because most of the species are mycorrhizal plants.

In the present result, P addition significantly affected the community composition of fungi in the soil, which is in agreement with the earlier results (Liu et al., 2012; Nielsen et al., 2015; He et al., 2016). The abundances of Ascomycota and Mortierellomycota significantly increased, but Basidiomycota and Chytridiomycota abundances declined significantly under P addition. Moreover, the db-RDA results showed that N and P addition effects on soil fungal community composition were possibly determined by soil P concentration (Liu et al., 2018). The current results support the above results, and the changes in soil fungal community structure would alter the ecological functions of soil. For instance, Mortierellomycota could defend against soil degradation (Li et al., 2019). Additionally, the changes in fungi community structure also supported our previous results that P addition could mitigate the negative impact of N addition on plant community structure (Zhao et al., 2019). The current results indicate that the changes in the community composition of soil fungi might be a good indicator that could be used to explain the reason that P addition affects plant community structure. However, how these increased or decreased soil fungi affect the growth of different functional groups needs further studies.

Conclusion

In the present study, soil bacterial and fungal diversities differently responded to the additions of N and P in a salinized meadow. The addition of N had few influences on the richness of soil bacteria but had a positive impact on soil fungal richness. P addition reduced the soil bacterial richness and increased soil fungal richness. N addition did not influence bacterial community composition but affected the fungi in soil. Additionally, both the communities of soil bacteria and fungi shifted due to P addition. The changes in soil P availability induced by N and P additions mainly determined the β -diversity of soil bacteria and fungi. Our findings indicate that the responses of the soil microbial communities of bacteria and fungi to seven-year N and P additions are not consistent in the studied ecosystem. Our results highlight that soil fungal community might be better used as a good indicator that can indicate response of soil microbial community to N and P enrichments in the temperate meadow ecosystem.

Declarations

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Compliance with ethical standards

Conflict of interest The authors declare no conflicts of interest.

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Figures

Figure 1

Effects of the additions of nitrogen (N) and phosphorus (P) on the concentrations soil N (a) and P (b), plant species diversity (Shannon index, c) and aboveground biomass (d). C represents control, N represents N addition, P represents P addition, NP represents nitrogen plus phosphorus addition. Lowercase letters indicate significant difference at the level of 0.05. * indicates $P < 0.05$; ** indicates $P < 0.01$; *** indicates $P < 0.001$; ns indicates no significant.

Figure 2

Effects of the additions of N and P on soil bacterial (a) and fungal (b) OUTs number, the Chao1 index of soil bacterial (c) and fungal (d) communities. C represents control, N represents nitrogen addition, P represents P addition, NP represents nitrogen plus phosphorus addition. Lowercase letters represent significant difference at the level of 0.05. * indicates $P < 0.05$; ** indicates $P < 0.01$; *** indicates $P < 0.001$; ns indicates no significant.

Figure 3

Effect of N and P additions on the relative abundances of the soil bacterial (a) and fungal (b) groups. C represents the control, N represents N addition, P represents P addition, NP represents nitrogen plus phosphorus addition. The dominant groups are shown (the relative abundances > 1%), while the rare groups (the relative abundances < 1%) are integrated into “other”.

Figure 4

Soil bacterial (a) and fungal (b) community compositions and their variation partition ordination plots of db-RDA under additions of N and P in a temperate meadow in northeastern China.

Figure 5

Results of PERMANOVA testing the effects of the additions of N and P on the bacterial (a) and fungal (b) communities in soil. Different letters in the pairwise comparisons indicate the significant differences at the level of 0.05.

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