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Authors

Tian, Jing

He, Nianpeng

Hale, Lauren

et al.

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Soil organic matter availability and climate drive latitudinal patterns in bacterial diversity from tropical to cold temperate forests

Jing Tian[1]*, Nianpeng He[1]*, Lauren Hale[2], Shuli Niu[1], Guirui Yu[1], Yuan Liu[1], Evgenia Blagodatskaya[3,4], Yakov Kuzyakov[4,5], Qun Gao[2,6], Jizhong Zhou[2,6,7]

[1] Key Laboratory of Ecosystem Network Observation and Modeling, Institute of Geographic Sciences and Natural Resources Research, Chinese Academy of Sciences (CAS), Beijing, China; [2] Department of Microbiology and Plant Biology and School of Civil Engineering and Environmental Sciences, Institute for Environmental Genomics, University of Oklahoma, Norman, OK, USA; [3] Institute of Physicochemical and Biological Problems in Soil Science, Pushchino, Russia; [4] Department of Agricultural Soil Science, University of Göttingen, Göttingen, Germany; [5] Agro-Technology Institute, RUDN University, Moscow, Russia; [6] State Key Joint Laboratory of Environment Simulation and Pollution Control, School of Environment, Tsinghua University, Beijing, China; and [7] Earth and Environmental Sciences, Lawrence Berkeley National Laboratory, Berkeley, CA, USA

Correspondence: Jing Tian Email: tianj@igsnrr.ac.cn; Guirui Yu Email: yugr@igsnrr.ac.cn

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Abstract

Bacteria are one of the most abundant and diverse groups of micro-organisms and mediate many critical terrestrial ecosystem processes. Despite the crucial ecological role of bacteria, our understanding of their large-scale biogeography patterns across forests, and the processes that determine these patterns lags significantly behind that of macroorganisms. Here, we evaluated the geographic distributions of bacterial diversity and their driving factors across nine latitudinal forests along a 3,700-km north–south transect in eastern China, using high-throughput 16S rRNA gene sequencing. Four of 32 phyla detected were dominant: *Acidobacteria*, *Actinobacteria*, *Alphaproteobacteria* and *Chloroflexi* (relative abundance > 5%). Significant increases in bacterial richness and phylogenetic diversity were observed for temperate forests compared with subtropical or tropical forests. The soil organic matter (SOM) mineralisation rate (SOM_{min}, an index of SOM availability) explained the largest significant variations in bacterial richness. Variation partition analysis revealed that the bacterial community structure was closely correlated with environmental variables and geographic distance, which together explained 80.5% of community variation. Among all environmental factors, climatic features (MAT and MAP) were the best predictors of the bacterial community structure, whereas soil pH and SOM_{min} emerged as the most important edaphic drivers of the

bacterial community structure. Plant functional traits (community weighted means of litter N content) and diversity resulted in weak but significant correlations with the bacterial community structure. Our findings provide new evidence of bacterial biogeography patterns from tropical to cold temperate forests. Additionally, the results indicated a close linkage among soil bacterial diversity, climate and SOM decomposition, which is critical for predicting continental-scale responses under future climate change scenarios and promoting sustainable forest ecosystem services.

Keywords: bacterial diversity, climate change, forest ecosystems, soil microbial biogeography, soil organic matter

INTRODUCTION

Microbial diversity and communities play critical roles in regulating multiple ecosystem functions and enhancing ecosystem stability (Fuhrman, 2009; Maestre et al., 2015; Naeem & Li, 1997). Although the geographic distributions of micro-organisms have been recently examined (Chu et al., 2010; Fierer & Jackson, 2006; Ge et al., 2008; Griffiths et al., 2011; Zhou et al., 2016), there are still important gaps in our understanding of the biogeography patterns of microbial diversity at large spatial scales with varying environmental gradients (Bardgett & van der Putten, 2014; Martiny et al., 2006).

The environmental factors that control the distributions of macro-organisms (e.g., plants and animals) have been extensively studied (Willig, Kaufman, & Stevens, 2003). Whether the processes and factors that control the spatial patterns of macro-organisms, such as contemporary environmental conditions and historical contingencies, also apply to micro-organisms remains unclear (Ge et al., 2008; Martiny et al., 2006; Ramette & Tiedje, 2007). Recent studies have documented a wide range of biotic and abiotic factors that influence microbial diversity and community structure such as soil pH (Chu et al., 2010; Fierer & Jackson, 2006; Lauber, Hamady, Knight, & Fierer, 2009), vegetation (Peay, Baraloto, & Fine, 2013), temperature (Zhou et al., 2016), and aridity (Wang et al., 2015). However, as noted by Ge et al. (2008) and de Vries et al. (2012), most studies have documented the variation and potential patterns of microbial diversity under certain ecological variation or gradient. A simultaneous, broad-scale survey of climatic factors, geographic distance, plant and edaphic factors at large spatial scales would be a more effective means of revealing the microbial biogeography patterns and facilitate the evaluation of the relative importance of these factors.

Soil organic matter (SOM) content is often considered as an overarching edaphic factor dominating soil bacterial diversity at local scales (Fierer, Schimel, & Holden, 2003; Hu et al., 2014). Because most soil micro-organisms rely on organic matter decomposition to obtain energy (Allison, Wallenstein, & Bradford, 2010; Trivedi, Anderson, & Singh, 2013). At the continental or regional scales, climate factors and soil pH rather than SOM

content play important roles in shaping bacterial diversity and communities (Fierer & Jackson, 2006; Liu et al., 2014; Zhou et al., 2016). However, recent studies based on global scale meta-analyses have shown that microbial biomass C peaks in areas with high SOM content (Wieder, Boehnert, & Bonan, 2014; Xu, Thornton, & Post, 2013), and that a geographical gradient of total soil organic carbon (SOC) content is an important driver of the observed bacterial diversity patterns (Delgado-Baquerizo et al., 2016). These studies implied that the SOM content could also underlie community-level diversity and distribution at large spatial scales. However, these global patterns were determined using synthesised data from various studies. These studies provide a foundation for our key hypothesis that these trends could be real and can be demonstrated using original data collected from field soil samples collected across large spatial scales.

Furthermore, a limited number of field studies have considered the role of SOM availability in influencing bacterial diversity and community compositions across large spatial scales (Fierer, Bradford, & Jackson, 2007; Fierer & Jackson, 2006). SOM is not chemically uniform and consists of various pools with varying levels of degradability and turnover rates (Stevenson, 1994; von Lützow et al., 2007). Older, more recalcitrant SOM pools are less decomposable by micro-organisms than younger pools (Blagodatskaya, Yuyukina, Blagodatsky, & Kuzyakov, 2011; Tian, Pausch, Yu, Blagodatskaya, & Kuzyakov, 2016). The availability of SOM is an important factor that influences the abundance of major soil bacterial phyla (Fierer et al., 2007). Therefore, information on soil bacterial biogeography patterns associated with SOM content and quality across large spatial scales should be of considerable importance.

Plants have significant influences on carbon resources and modify the habitat of soil micro-organisms (Kuzyakov, Friedel, & Stahr, 2000; Latz, Eisenhauer, Scheu, & Jousset, 2015). Accumulating evidence indicates that plant diversity is a major driver of soil bacterial diversity and community structure at a range of spatial scales (Peay et al., 2013; Prober et al., 2015; Wardle, 2006). At the ecosystem scale, plant functional traits such as leaf N content and leaf dry matter content explain the variation in SOM content (Lavorel et al., 2011) and rates of litter decomposition (Fortunel et al., 2009). Thus, plant functional traits rather than functional diversity correlate with the nitrification rate (Laughlin, 2011) and the ratio of bacterial to fungal biomass across the landscape scale (de Vries et al., 2012). However, whether plant functional traits (e.g. leaf and litter nutrient contents) can explain bacterial biogeography diversity patterns across a large spatial scale remains unclear.

Forests are home to 80% of the world's terrestrial biodiversity, and these ecosystems are complex webs of organisms that include plants, animals, fungi and bacteria. A detailed analysis of soil microbial diversity and communities across a larger spatial scale would improve our understanding of the functions and services of forest ecosystems. A number of studies have examined bacterial biogeography patterns in forests across large spatial

scales (Shay, Winder, & Trofymow, 2015; Tu et al., 2016; Zhou et al., 2016). However, to date, limited information is available concerning the geographic patterns of soil bacterial diversity in Chinese forests (Ma et al., 2016; Wang, Zheng, et al., 2016; Zhang et al., 2016). In addition, whether total SOM amount and availability and plant functional traits can explain variations in soil bacterial biogeography patterns in different Chinese forests, and their relative importance remains unknown.

The north–south transect of eastern China (NSTEC) used in this study includes a range of forest types from cold temperate to tropical (Zhao et al., 2016). Significant variations in climate, edaphic, plant and geographic distance factors (Tables S1 and S2) occur along this transect, and consequently, the sampling sites reflect large environmental gradients well. This transect therefore provides an ideal set of experimental plots to explore the responses of bacterial diversity and community structure to different environmental gradients at a large spatial scale. The objectives of this study were as follows: (1) to determine and compare bacterial diversity and community structure from tropical to cold temperate forests; (2) to quantitatively assess the relative importance of multiple environmental variables, such as climate, geographic distance, edaphic and plant factors, in shaping bacterial diversity and community structure; (3) to determine whether bacterial diversity and community structure exhibit higher correlations with SOM content and availability than other environmental factors at a large spatial scale, as previously observed in synthesised data analyses (Delgado-Baquerizo et al., 2016); and (4) to connect patterns of plant functional traits to the biogeography of soil bacterial diversity.

MATERIALS AND METHODS

Site description and field sampling

Soil samples were collected from nine typical forests along the NSTEC (108.9°E, 18.7°N to 123.0°E, 51.8°N; Figure S1). The mean annual temperature (MAT) at these sites ranged from −3.67 to 23.15°C, and the mean annual precipitation (MAP) ranged from 473 to 2,266 mm (Table S1). The soil type changed from brown soils to tropical red soils along the transect line, while the vegetation type ranged from cold temperate needle leaf forest to temperate broadleaf deciduous forest, subtropical broadleaf evergreen forest and tropical seasonal rainforests (Table S1).

Field sampling was performed in July 2013. To reduce the influence of anthropogenic disturbances, all sampling plots were located in well-protected national nature reserves, where vegetation and soil were representative for the given forest type. Four randomly chosen experimental plots (30 m × 40 m) were established in each forest site. All plots were located 100–400 m apart from each other. At each plot, we collected 40–50 surface soil scores (0–10 cm deep). These soil samples were composited and sieved through a 2-mm mesh to thoroughly homogenise and the roots and visible organic debris were removed manually. A portion of each soil sample

was collected in a 50-ml centrifuge tube, which was then placed in an ice-box and transferred to the laboratory. The tubes were stored at −80°C for soil DNA extraction. The remaining soil was used to measure chemical characteristics.

Climate, soil and plant attributes

The climatic variables, including MAT and MAP, were extracted from the meteorological database of CERN.

Soil pH was measured using a pH meter (1:2.5 w/v). The SOC and total nitrogen (TN) contents were determined by a Vario EL III Elemental Analyser (Elementar, Germany). The concentrations of dissolved organic carbon (DOC) and dissolved organic nitrogen (DON) were measured by the method recommended by Jones and Willett (2006). Soil total P concentration (mg/kg) was measured by the ammonium molybdate method using an AutoAnalyser3 continuous-flow analyser (Bran Luebbe, Germany) after H_2SO_4–H_2O_2–HF digestion. The average values of soil chemical properties in each sampled forest are presented in Table S2

The potential SOM mineralisation rate was used to represent the SOM availability (Fierer & Jackson, 2006; Fierer et al., 2007). The rate was estimated by measuring the rates of CO_2 production over the course of a 14-day incubation period at 25°C after adjusting all soils to 60% of their water-holding capacity.

In each plot, information on tree species was surveyed, and the Shannon–Weaver index was then used to describe tree diversity (Table S2). The leaves and litter of the dominant and common plant species were collected according to a unified protocol (Zhao et al., 2016). The leaf C and N contents (Leaf C, Leaf N) were obtained from Zhao et al. (2016), and were transformed to community weighted means (Table S2). The litter C and N contents (Litter C, Litter N) were transformed to community weighted means too (Table S2).

Soil DNA extraction

Soil DNA was extracted from each sample using the PowerSoil kit (MoBio Laboratories, Carlsbad, CA, USA) according to the manufacturer's instructions. The quality of the purified DNA was assessed based on the 260/280 nm and 260/230 nm absorbance ratios obtained, using a NanoDrop ND-1000 spectrophotometer (NanoDrop Technologies Inc., Wilmington, DE, USA). The DNA was stored at −80°C until use.

16S rRNA gene amplification and sequencing

An aliquot of the extracted DNA from each sample was used as a template for amplification. The V3–V4 hypervariable regions of bacterial 16S rRNA genes were amplified using the primers 338F 5′-barcode-ACTCCTACGGGAGGCAGCAG-3′ and 806R 5′-GGACTACHVGGGTWTCTAAT-3′. PCR reactions were performed in triplicate with a 20 μl mixture containing 4

μl of 5× FastPfu Buffer, 2 μl of 2.5 mM dNTPs, 0.8 μl of each primer (5 μM), 0.4 μl of FastPfu Polymerase, and 10 ng of template DNA. The following thermal programme was used for amplification: 95°C for 3 min, followed by 27 cycles at 95°C for 30 s, 55°C for 30 s, and 72°C for 45 s and a final extension at 72°C for 10 min. PCR amplicons were extracted from 2% agarose gels and purified using an AxyPrep DNA Gel Extraction Kit (Axygen Biosciences, Union City, CA, USA) according to the manufacturer's instructions and quantified using QuantiFluor™ -ST (Promega, USA). The purified amplicons from all samples were pooled in equimolar concentrations. Sequencing was conducted on an Illumina MiSeq platform at Majorbio BioPharm Technology Co., Ltd. (Shanghai, China).

Processing of sequencing data

Raw sequences >200 bp with an average quality score >20 and without ambiguous base calls were quality processed, using the Quantitative Insights into Microbial Ecology (QIIME) pipeline (version 1.17). Operational taxonomic units (OTUs) were clustered with a 97% similarity cutoff using UPARSE (Edgar, Haas, Clemente, Quince, & Knight, 2011) (version 7.1 http://drive5.com/uparse/). The taxonomic assignment was performed using the Ribosomal Database Project (RDP) classifier (http://rdp.cme.msu.edu/). To correct for sampling effort (number of analysed sequences per sample), we used a randomly selected subset of 19,460 sequences per sample for subsequent analysis.

Statistical analysis

The phylogenetic diversity was estimated using Faith's index, which incorporates the phylogenetic breadth across taxonomic levels (Faith, 1992; Faith, Lozupone, Nipperess, & Knight, 2009). The number of OTUs indicates the phylotype richness. The relationships between bacterial diversity and richness and environmental factors were assessed with linear regression analyses using SPSS 16.0 for Windows. Stepwise multiple regressions were applied to identify the most influential environmental variables on bacterial diversity and richness due to collinearity among environmental factors.

Non-metric multidimensional scaling (NMDS) and detrended correspondence analysis (DCA) were used to assess changes in the bacterial community. Analyses including Anosim, MRPP and Adonis were further performed to confirm significant changes in community structures in any pair of samples. NMDS, DCA and statistical analyses were performed in R v.3.2.1 with the VEGAN package.

The partial Mantel test was used to evaluate the linkages between the bacterial microbial community structure and the measured environmental variables. To determine the relative importance of geographic distance, climate, edaphic and plant factors in shaping bacterial community structure, a canonical correspondence analysis-based variation partitioning analysis (VPA) was implemented. The spatial decomposition method, principal

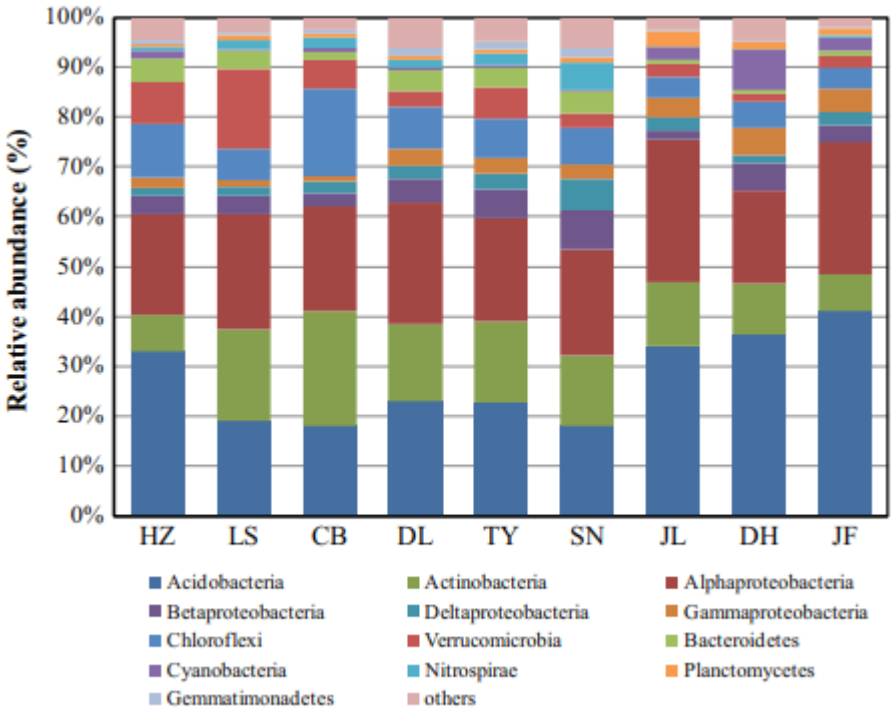
coordinates of neighbour matrices (PCNM) was applied to the geographic coordinates of the samples to separate them into spatial variables (Ramette & Tiedje, 2007). Partial Mantel and VPA analyses were carried out in R v.3.2.1 with the VEGAN package. Matrices of the pairwise taxonomic distance between bacterial communities (Bray–Curtis) and Euclidean distance of environmental variables were also constructed in R v.3.2.1 using the VEGAN package.

RESULTS

Distribution of taxa and phylotypes

Across all soils, a total of 1,035,979 quality sequences were obtained, and 19,460–37,633 sequences were identified per sample (average of 28,777). These sequences were grouped into 65,982 OTUs at the 97% similarity level. When all samples were compared at an equivalent sequencing depth of 19,460 per sample, the dominant phyla across all samples were *Acidobacteria*, *Actinobacteria*, *Alphaproteobacteria* and *Chloroflexi* (relative abundance > 5%), which accounted for more than 67% of the bacterial sequences (Figure 1). In addition, *Betaproteobacteria*, *Deltaproteobacteria*, *Gammaproteobacteria*, *Verrucomicrobia*, *Bacteroidetes*, *Cyanobacteria*, *Nitrospirae*, *Planctomycetes* and *Gemmatimonadetes* were also present in almost all soils at relatively low abundances, and 19 other rarer phyla were identified.

Figure 1. Relative abundances of the dominant bacteria groups in nine forest ecosystems along a north–south transect in eastern China. The relative abundances are based on the proportional frequencies of the DNA sequences that could be classified



Bacterial α diversity across the nine forest ecosystems

The bacterial α diversity, as estimated by phylotype richness (OTU numbers) and Faith's phylogenetic diversity, varied significantly across the nine forest ecosystems (Figure 2). The observed bacterial richness ranged from 852 to 1,659 and was the lowest in subtropical (JL and DH) and tropical forests (JF) at low latitudes ($p < .05$, Figure 2). Proportionally, these forests also shared less overlapped phylotypes compared to the other six forests (Table S3).

The bacterial richness in these forest soils exhibited the largest correlation with SOM_{min} among all environmental factors assayed here (Table 1, $r = .626$, $p < .0001$). In addition to soil pH ($r = .672$, $p < .0001$), SOM_{min} was the best predictor of phylogenetic diversity (Table 1, $r = .618$, $p < .0001$). The SOC content exhibited a weak correlation with bacterial richness ($r = .334$, $p < .05$) (Table 1). Significant correlations were also noted between phylotype richness and phylogenetic diversity and plant functional diversity (tree richness) and traits (leaf N, leaf N and leaf C/N) (Table 1).

When the climate, plant and edaphic factors were entered into a stepwise regression, bacterial richness significantly correlated with SOM_{min} (42.1%), followed by MAP (20.2%) and pH (15.9%) (Table S4). Plant functional traits (leaf C/N) resulted in a weak but significant contribution (4.85%) to describe

patterns in bacterial richness (Table S4). MAP explained the largest significant variation in phylogenetic diversity (56.6%), whereas pH (15.9%) and SOM_{min} (7.6%) collectively contributed to 23.5% of the variation (Table S4).

Figure 2. Bacterial phylotype richness and phylogenetic diversity of the nine forest ecosystems along a north–south transect in eastern China. Different lower-case letters indicate significant differences. The error bars represent the *SEs* of the *M* (*n* = 4)

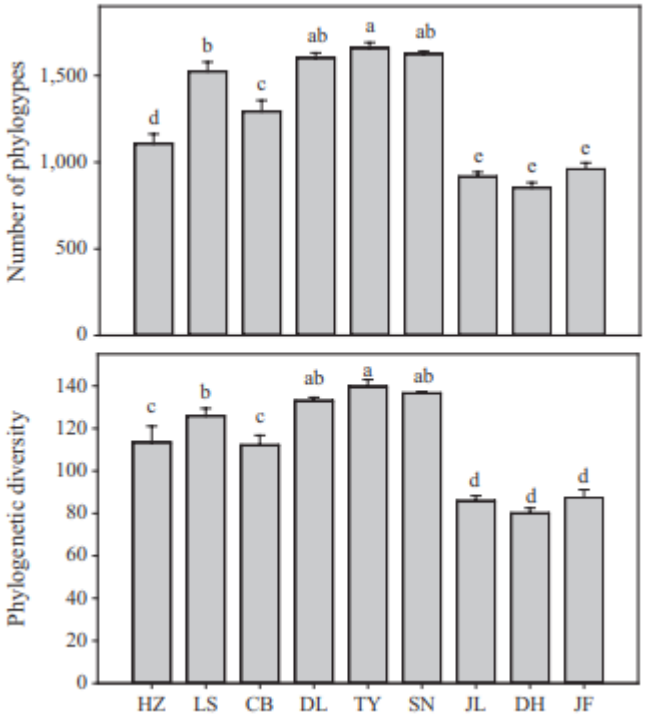


Table 1. Pearson correlations between bacterial phylotype richness and phylogenetic diversity and plant and soil characteristics

r	Richness	Diversity
TD	−.305	−.473**
TR	−.367*	−.467**
Leaf C	−.264	−.216
Leaf N	.443**	.406*
Litter C	−.004	.047
Litter N	−.113	−.011
Leaf C/N	−.416**	−.388*
Litter C/N	.205	.149
SOC	.334*	.318
TN	.371*	.360*
C/N	−.284	−.292
TP	.287	.332*
pH	.590**	.672**
DOC	−.611**	−.619**
DON	−.234	−.336*
SOM_{min}	.626**	.618**

TD, Tree diversity (Shannon); TR, tree richness; Tree C/N, Tree C/Tree N; Litter C/N, Litter C/Litter N; SOC, soil organic carbon; TN, total nitrogen; C/N, SOC/TN; TP, total phosphorus; DOC, dissolved organic carbon; DON, dissolved organic nitrogen; SOM_{min}, potential SOM decomposition rate. Asterisks represent significant of correlation (*$p < .05$, *$^{*}p < .01$).

Bacterial community structure across the nine forest ecosystems

The soil samples from the nine forests were clearly separated from each other in both the NMDS and DCA plots, indicating that the bacterial community structure differed across the nine forests (Figure 3, Figure S2). The results of MRPP, ADONIS and ANOSIM analyses further confirmed the significant differences among each paired forests ($p < .05$; Table S5). The soil samples from three low-latitude forests (JL, DH and JF) were separated from those from the other six forests in NMDS2 and DCA2 (Figure 3, Figure S2). This finding was reinforced by a cluster analysis based on a Bray–Curtis distance matrix of all samples (Figure S3).

Linking bacterial community structure to geographic distance and environmental factors

The soil bacterial community dissimilarity was significantly correlated with the historical factor of geographic distance (Figure 4a), which revealed that spatial isolation affected community structure, presumably via dispersal limitation. Strong relationships were also observed between the microbial community structure and all environmental variables (including all plant, edaphic and climate factors) (Figure 4b).

Figure 3. Non-metric multidimensional scaling (NMDS) ordination of the soil bacterial community structure in nine forest ecosystems along a north–south transect in eastern China. The compositional variation is represented by the Bray–Curtis distance matrix based on the abundance of OTUs

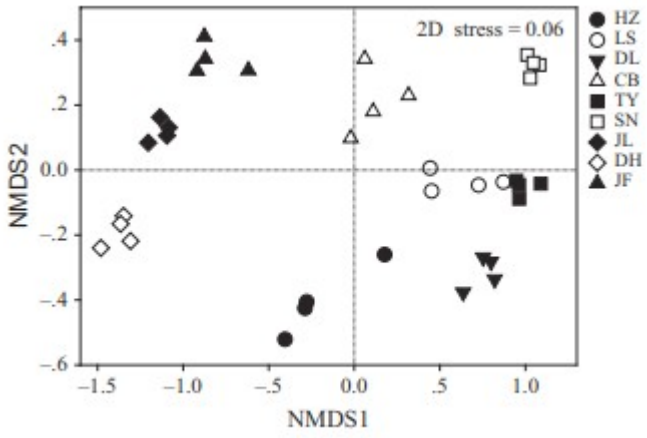
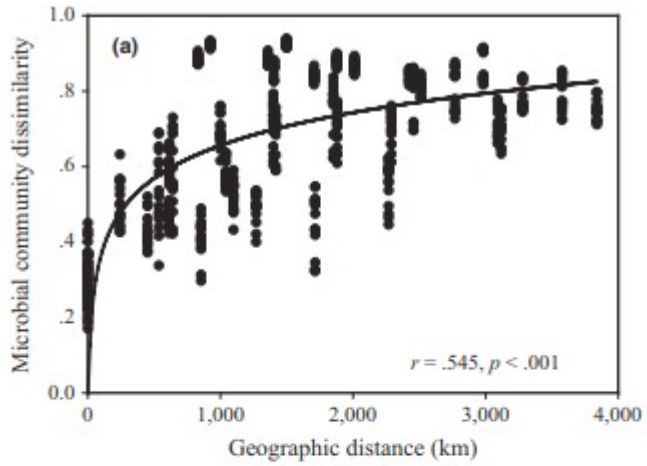
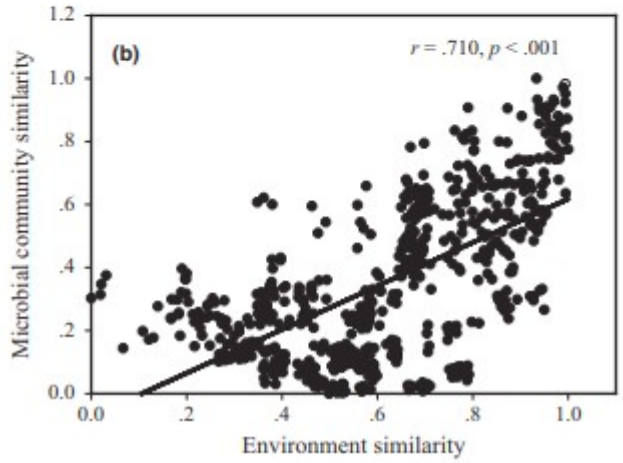


Figure 4. Correlations between the bacterial community dissimilarities (similarity) and geographic distances and environment similarity





A partial Mantel test was performed to reveal the major environmental variables shaping microbial community structure. MAT ($r_M = .583$, $p < .001$), MAP ($r_M = .490$, $p < .001$), pH ($r_M = .430$, $p < .001$), SOM_{min} ($r_M = .279$, $p < .001$) and DOC ($r_M = .287$, $p < .001$) were the most important factors that independently contributed to variations in soil bacterial community structure (Table 2). Plant functional diversity and traits (TD, TR and litter N) resulted in weak but significant correlations ($r_M = .110$, $r_M = .146$ and $r_M = .085$, respectively, $p < .05$) (Table 2).

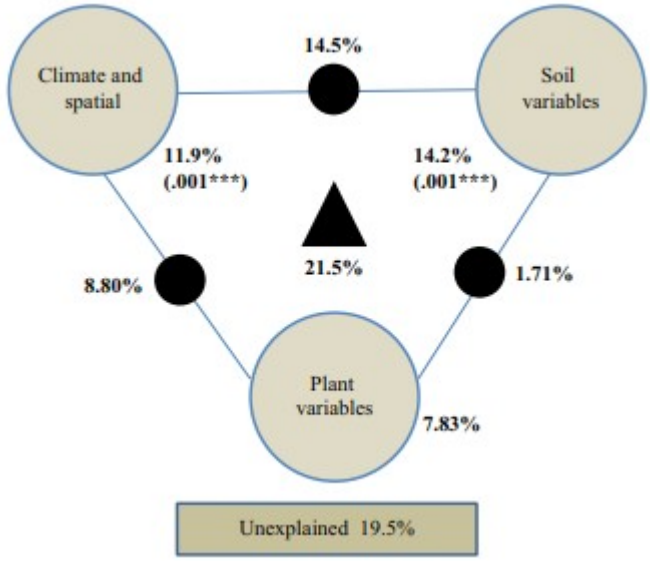
Table 2. Partial mantel test of soil bacterial community structure with environmental characteristics

Environmental characteristics	r	p
MAT	.583	.001
MAP	.491	.001
TD	.111	.012
TR	.146	.007
Leaf C	−.025	.709
Leaf N	−.039	.835
Litter C	−.125	1.000
Litter N	.085	.041
Leaf C/N	−.014	.600
Litter C/N	.036	.190
SOC	−.142	1.000
TN	−.180	1.000
C/N	−.056	.959
TP	−.074	.962
pH	.431	.001
DOC	.288	.001
DON	.125	.012
SOM_{min}	.279	.001

TD, Tree diversity (Shannon); Tree C/N, Tree C/Tree N; Litter C/N, Litter C/Litter N; SOC, soil organic carbon; TN, total nitrogen; C/N, SOC/TN; DOC, dissolved organic carbon; DON, dissolved organic nitrogen; SOM_{min}, potential SOM decomposition rate. The environmental variables that used for CCA model were selected here. The correlation (r) and significance (p) were determined based on 9,999 permutations between community structure (Bray–Curits distance) and environmental variables (Euclidean distance).

Variance partition analysis was subsequently performed to dissect the relative contributions of geographic distance and environmental factors to soil bacterial community structure (Figure 5). A total of 80.5% of the community variations could be explained by all measured variables. Geographic distance and climate, soil and plant variables independently explained 11.9%, 14.2% and 7.83%, respectively.

Figure 5. Variation partitioning analysis of the bacterial community explained by climate and spatial (MAT, MAP and geographic distance) variables, soil variables (including SOC, SOM_{min}, TN, TP, pH, DOC, DON and soil C/N) and plant variables (TD, TR, leaf C, leaf N, litter C, litter N, leaf C/N, litter C/N) and their interactions. The values in parentheses are *p* values



DISCUSSION

Effects of SOM content and availability in shaping bacterial α diversity and community structure

Our main hypothesis that bacterial diversity differs across forests and that SOM availability significantly shapes diversity patterns was only partially verified. The importance of SOM content as a driver of soil latitudinal bacterial α diversity at the global scale was recently highlighted by a meta-analysis (Delgado-Baquerizo et al., 2016). In this study, SOM availability, determined by SOM mineralisation, explained 42% of the variation in bacterial biodiversity. In addition, SOM availability correlated with bacterial richness; however, it was the second most important edaphic driver of phylogenetic diversity and community structure (Tables 1 and 2; Table S3). This finding supports the work of Siciliano et al. (2014), who found that soil fertility (e.g., SOM) was the most important edaphic property associated with microbial richness, whereas pH was primarily associated with bacterial phylogenetic diversity and β diversity (variation in community composition). Compared with higher latitudes, the higher temperatures and precipitation at low latitudes may result in higher organic matter decomposition and leaching, thus leading to lower SOM content (Wieder et al., 2014; Xu et al., 2013) with higher biochemical recalcitrance (Wang, He, et al., 2016; Wang, Zheng, et al., 2016). It is conceivable that the higher SOM availability in higher latitude soils provides more nutritive properties, allowing a variety of appropriately adapted bacterial species within a community to thrive. However, the initial community from which the relatively fast-growing

species are drawn is determined by the pH of the environment (Siciliano et al., 2014). Moreover, we found that climate and spatial variables and soil variables together contributed an extra 14.5% to the variance of the bacterial community structure, indicating considerable interactions between those variables (Figure 5).

The different bacterial communities across the SOM availability gradient (Table 2) were clearly related to shifts in the relative abundances of bacterial taxa at the phylum level (Figure S5). In accordance with findings presented by Fierer et al. (2007), there was a significant positive relationship between SOM_{min} and the relative abundance of *Bacteroidetes*, whereas a negative relationship was observed with the relative abundance of *Acidobacteria*. *Bacteroidetes* can be categorised as copiotrophic and are the initial metabolisers of labile C and are thus more abundant in soils with increased C availability (Fierer et al., 2007; Padmanabhan et al., 2003). In contrast to copiotrophic groups, *Acidobacteria* exhibit oligotrophic attributes that are negatively correlated with soil C availability (Fierer et al., 2007; Trivedi et al., 2013). In contrast to results of Fierer et al. (2007), we observed a negative relationship between SOM_{min} and the relative abundance of *Gammaproteobacteria*, but not *Betaproteobacteria*. In addition, we also observed significant positive relationships between SOM_{min} and the relative abundances of *Nitrospirae*, *Deltaproteobacteria*, *Verrucomicrobia* and *Gemmatimonadetes*. Thus, although the bacterial community composition was clearly influenced by SOM_{min}, some variations were observed in the relationships between the relative abundances of specific phyla and SOM_{min} under different habitats.

Our results provide evidence that SOM availability explains a significant proportion of the observed latitudinal bacterial α and β diversity across different forest ecosystems (Tables 1 and 2; Table S4). The overall positive relationship between SOM availability and bacterial diversity observed in this study has important implications for understanding the mechanisms underlying forest ecosystem functions. For instance, increased bacterial α diversity was observed in SOM-rich forests with low net primary productivity, such as forests located in northeast China (Figure 2). These results imply that the reduction in net primary productivity and SOM associated with climatic events and human management in these forest ecosystems may have negative impacts on soil bacterial diversity and, consequently, ecosystem functions.

Our results also reinforce prior research demonstrating the importance of soil pH as an important edaphic factor influencing bacterial diversity and composition across a large spatial scale (Chu et al., 2010; Fierer & Jackson, 2006; Griffiths et al., 2011; Rousk et al., 2010). In addition to SOM and soil pH, previous studies have reported that some other edaphic factors (e.g., soil texture, available P content) also played important roles in influencing soil bacterial diversity (Bergkemper et al., 2016; de Vries et al., 2012). Therefore,

the relative importance of multiple edaphic variables in shaping bacterial diversity and community structure should be investigated in future studies.

Effects of plant functional traits, climate variables and geographic distance in shaping bacterial α diversity and community structure

In addition to edaphic factors, we also noted significant correlations between bacterial diversity and other measured variables (plant functional traits, climatic variables and geographic distance) as well as significant interactions between variables (Tables 1 and 2; Figures 4 and 5). These results suggest that the observed latitudinal diversity gradient patterns of bacterial diversity in forests across a large spatial scale are a result of multiple complicated ecological processes.

Plant functional traits (leaf C/N and litter N) exhibited weak but significant contributions to bacterial richness and community structure (Table 2; Table S4). This finding partly supported our second hypothesis that plant functional traits independently explain some variations in bacterial α diversity and communities. Bacterial phylotype richness increased with reduced community-weighted means of leaf C/N (Table 1; Table S4). Generally, leaves with higher C/N ratios are decomposed more slowly than those with a low C/N ratio, likely due to reduced nitrogen availability and, consequently, lower bacterial diversification (Kelly et al., 2010) or the promotion of resource partitioning among the component soil organisms (Wardle, 2006). In line with finding, leaf C/N was significantly positively correlated with the relative abundance of *Acidobacteria*, which can adapt to low nutrient availability (Table S6). Tree functional diversity explained variations in bacterial community structure but not bacterial richness and phylogenetic diversity (Table 2; Table S4), indicating that plant diversity drives soil bacterial community structures at a range of spatial scales (Peay et al., 2013; Wardle, 2006). In a previous study, plant diversity was correlated with composition of soil bacterial and fungal microbial communities, but not patterns in their α diversity across a global range of temperate grasslands (Prober et al., 2015). In contrast, the bacterial β diversity coupled with the diversity of herbs rather than threes was observed across latitudinal forest ecosystems (Wang, He, et al., 2016; Wang, Zheng, et al., 2016). This indicated that the influences of the diversity of herbs or shrubs on below-ground biodiversity in forest ecosystems should also be considered in future studies.

Climate can influence the bacterial community structure, but the influence can range from significant (Garcia-Pichel, Loza, Marusenko, Mateo, & Potrafka, 2013; Zhou et al., 2016) to marginal (Angel, Soares, Ungar, & Gillor, 2010; Fierer & Jackson, 2006). Our findings indicate important roles of MAT and MAP in explaining the soil bacterial community structure (Table 2). These roles can be attributed to the direct and indirect effects of temperature and precipitation on bacterial communities. Although not directly evidenced in this study, the MAT was previously shown to directly affect microbial metabolic rates (Zhou et al., 2016), which may strongly

influence complex ecological processes such as speciation, competition and dispersal of microbial communities. Temperature or precipitation might also indirectly affect bacterial communities by mediating other environmental factors, including plant leaf N content (Reich & Oleksyn, 2004) or nutrient availability (Cross, Hood, Benstead, Huryn, & Nelson, 2015). For instance, leaf N increased from the tropics to the cooler and drier mid-latitudes because of temperature-related plant physiological stoichiometry and biogeographical gradients with soil substrate age (Reich & Oleksyn, 2004). Warmer temperatures and higher MAP in tropical forests can lead to higher SOM decomposition and leaching, resulting in lower SOM availability. Our study supports these linkages based on the close correlations observed between climate (MAT and MAP) and leaf N content and SOM availability ($r = -.445$ to $-.616$, $p < .05$).

The significant correlation between bacterial community dissimilarity and geographical distance (Figure 4) suggests that dispersal limitation is also an important driver of changes in bacterial communities across this 3,700-km transect line. Some surveys have suggested that bacterial community similarities are more closely related to environmental factors than differences in geographic distance (Chu et al., 2010; Hollister et al., 2010); others have reported that dispersal limitation is an important driver of changes in bacterial communities (Cho & Tiedje, 2000; Wang et al., 2015; Xiong et al., 2012). These inconsistent observations may be attributable to difference in the scales at which the studies were conducted as the relative importance of environmental and historical factors depends on the survey scale (Ge et al., 2008; Hendershot, Read, Henning, Sanders, & Classen, 2017; Ricklefs, 2004). Our result agreed with a previous study that described the influences of historical contingencies on soil bacterial biogeography are most likely to be detected at intermediate scales (10–3,000 km) (Martiny et al., 2006).

CONCLUSIONS

In conclusion, this study represents one of the largest attempts to comprehensively investigate the regulation of soil bacterial diversity and community structure under multiple environmental gradients along a north–south transect in eastern China. We identified a combination of abiotic and biotic predictors of soil bacterial α diversity and community structure. Our study is the first to show that SOM availability is the most important factor influencing bacterial α diversity (OTU richness) from tropical to cold temperate forests. In addition, SOM availability and soil pH played equally important roles in shaping bacterial β diversity (variations in community structure). Climate features (MAT and MAP) exhibited the highest correlations with bacterial β diversity. Plant functional trait (leaf C/N ratio) is vital to bacterial richness, whereas plant diversity is an important factor influencing bacterial β diversity. These results provide a better understanding of the link between soil bacterial diversity and forest ecosystem services, and provide a framework to predict microbial

continental-scale responses under future climate change. Future studies are necessary to investigate and compare the community turnover and interaction network of bacteria in small-spatial scales within each forest and among different forests.

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DATA ACCESSIBILITY

The raw sequence data from this study have been deposited in SRA at the NCBI database with the assigned study SRP080891 and BioSamples SAMN05239525-SAMN05439560.

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