Assembly processes lead to divergent soil fungal communities within and among twelve forest ecosystems along a latitudinal gradient

Yong Zheng[1,2,#], Liang Chen[3,#], Niu-Niu Ji[3], Yong-Long Wang[3], Cheng Gao[3], Sheng-Sheng Jin[1,2], Hang-Wei Hu[1,2], Zhiqun Huang[1,2], Ji-Zheng He[1,2], Liang-Dong Guo[3,4]*, Jeff R. Powell[5]*

[1] State Key Laboratory for Subtropical Mountain Ecology of the Ministry of Science and Technology and Fujian Province, Fujian Normal University, Fuzhou 350007, China.
[2] School of Geographical Sciences, Fujian Normal University, Fuzhou 350007, China.
[3] State Key Laboratory of Mycology, Institute of Microbiology, Chinese Academy of Sciences, Beijing 100101, China.
[4] University of Chinese Academy of Sciences, Beijing 100049, China.
[5] Hawkesbury Institute for the Environment, Western Sydney University, Locked Bag 1797, Penrith, NSW 2751, Australia.

#These authors contributed equally to this work.
***Corresponding authors:**
Dr./Prof. Liang-Dong Guo
State Key Laboratory of Mycology, Institute of Microbiology, Chinese Academy of Sciences, Beijing 100101, China.
Tel.: +86 10 64807510; *E-mail*: guold@im.ac.cn

Dr./Prof. Jeff R. Powell
Hawkesbury Institute for the Environment, Western Sydney University, Locked Bag 1797, Penrith, NSW 2751, Australia.
Tel.: +61 (0)2 4570 1093; *E-mail*: jeff.powell@westernsydney.edu.au

Type of contribution: Full paper.
Number of text pages: 15 (1.5-line spaced, 481 lines).
The total word count: 4972.
1. ***Abstract*:** 194.
2. ***Introduction*:** 733.
3. ***Materials and Methods*:** 1941.
4. ***Results*:** 888.

5. ***Discussion*****:** 1010 (= 20.3% of the total word count).
6. ***Conclusions*****:** 160.

Number of figures and tables: three figures (three published in colour) and two tables.

Number of figures and tables in Suppl. Infor.: three figures and 10 tables.

Running title: Fungal community assembly in forest soils.

Abstract

Latitudinal gradients provide opportunities to better understand soil fungal community assembly and its relationship with vegetation, climate, soil and ecosystem function. We quantified the relative importance of stochastic and deterministic processes in structuring soil fungal communities using patterns of community dissimilarity observed within and between twelve natural forests. The results revealed that whole fungal communities and communities of arbuscular and ectomycorrhizal fungi consistently exhibited divergent patterns but with less divergence for ectomycorrhizal fungi at most sites. Within those forests, no clear relationships were observed between the degree of divergence within fungal and plant communities. When comparing communities at larger spatial scales, among the twelve forests, we observed distinct separation in all three fungal groups among tropical, subtropical and temperate biomes. Soil fungal β-diversity patterns between forests were greater when comparing forests exhibiting high habitat turnover, with these patterns being driven to a greater extent in each fungal group by temperature, soil pH, soil carbon and plant community composition than by geographic distance. Taken together, although large-scale community turnover could be attributed to specific environmental drivers, strong divergence during community assembly in forest soils at local scales limits the predictability of fungal community assembly outcomes.

Keywords: β-diversity; community assembly; forest; fungi; latitudinal gradient; neutral model

Introduction

Understanding community assembly patterns and the processes resulting in these patterns is critical to gaining a more mechanistic understanding of biodiversity maintenance, community stability and ecosystem functioning (Chase, 2010; Nemergut *et al.*, 2013). Stochastic (neutral) and deterministic (niche-based) processes are both important forces in determining the outcomes of community assembly for macro-organisms (Fargione *et al.*, 2003; Kraft *et al.*, 2008; Ellwood *et al.*, 2009). Neutral theory assumes that many natural community assembly patterns can be generated solely by considering ecological drift, resulting in communities that are dependent on dispersal (immigration) rather than adaptation to their habitats (Hubbell, 2001; Alonso *et al.*, 2006). Niche-based theory, however, states that deterministic factors, such as biotic interactions, species traits, and environmental variables, modulate the local community. When deterministic factors are of greater importance, local communities assembling under similar environmental conditions should converge upon a common composition, while divergence among local communities should be observed when environmental conditions differ (Chase, 2003; Zhou *et al.*, 2013).

Soil microorganisms, especially fungi, have remarkable biodiversity on Earth and play crucial roles in biogeochemical cycling and ecosystem functioning (Philippot *et al.*, 2013; Tedersoo *et al.*, 2014). Mycorrhizal fungi make up important functional guilds within soil fungal communities, forming mutualistic associations with 80% of land plants and obtaining carbon from hosts in exchange for mineral nutrients (Smith & Read, 2008). Therefore, these fungi can greatly affect plant productivity, diversity, and ecosystem processes (van der Heijden et al., 1998, 2015; Mayor et al., 2015). Progress in molecular techniques to efficiently characterise fungal communities had resulted in researchers paying increasing attention to understanding the relative contributions of determinism and neutrality for fungal community assembly (*i.e.*, Gao *et al.*, 2015; Powell *et al.*, 2015; Alzarhani *et al.*, 2019). Previous studies have demonstrated that the relative role of deterministic and stochastic processes in structuring soil fungal community assembly is highly dependent on spatial scale (Caruso *et al.*, 2012; Kivlin *et al.*, 2014; Schroter *et al.*, 2019). However, the relative importance of stochastic and deterministic processes in structuring fungal communities in forest soils at wide-latitude gradient remains largely unknown.

Exploring the fungal community turnover (β-diversity) can lead to insight into the

mechanisms generating and maintaining fungal biodiversity in ecosystems (Ettema & Wardle, 2002; Beck *et al.*, 2015). These studies are typically performed at large geographic scales, but with different environmental foci and differing results. Soil fungal communities across Scotland were more strongly affected by deterministic processes in systems with less disturbance (bogs, moors, and semi-natural grasslands) than in more intensive land use systems such as managed grasslands and arable systems (Powell *et al.*, 2015). Climatic factors, such as mean annual temperature (MAT) and precipitation (MAP) are often observed to be important predictors of soil fungal β-diversity (Tedersoo *et al.*, 2014). Temperature was identified as a key factor influencing soil fungal β-diversity across a large-scale latitudinal gradient in China and USA (*e.g.*, Shi *et al.*, 2014; Zhou *et al.*, 2016). Plants contribute organic carbon to fungi, which benefit the plants via decomposition or direct nutrients supply in symbiotic relationships. However, the relationships between soil fungal β-diversity and that of the vegetation communities were rarely considered.

Using Illumina Miseq high-throughput sequencing, we examined soil fungal communities within twelve Chinese forests situated throughout eastern China, with the largest distance among forests being more than 4 000 km (Fig. S1). To assess the relative importance of stochastic and deterministic factors for soil fungal community assembly in these forests, we employed a null model approach to generate neutral predictions, which were subsequently compared to the observed β-diversity. This analysis reveals three possible outcomes: the observed β-diversity not differing from the neutral prediction (neutral assembly overwhelms determinism) or the observed β-diversity being larger (divergent assembly) or smaller (convergent assembly) than the neutral prediction. Our first aim was to identify these patterns in soil fungal community assembly and their relationships with that of corresponding plant communities. Second, we estimated the relative importance of habitat turnover based on soil, plant, and climatic parameters as drivers of fungal β-diversity (community variation). We hypothesized that the balance between deterministic and stochastic community assembly processes within individual forests and their relationship with assembly processes occurring within plant communities would differ depending on whether entire fungal communities or specific symbiotic guilds were assessed. Finally, we assessed the relative importance of climate, soil and vegetation properties for driving community turnover among soil fungal communities in forests across the entire gradient.

Materials and methods

Sites and sampling

This study was conducted in 12 permanent forest sites established by the Chinese Forest Biodiversity Monitoring Network (CForBio), ranging in latitude from 21.6° N to 50.8° N in China (Fig. S1). The experimental forests were described previously by Ji *et al.* (2019). Briefly, a total of six forest types include tropical rain forest (TRF), subtropical evergreen broad-leaf forest (SEF), mixed evergreen broad-leaved and deciduous broad-leaved forest (MEDF), warm-temperate deciduous broad-leaved forest (WTDF), temperate broad-leaved Korean pine forest (TPF), and cold temperate monsoon coniferous forest (CTCF) were included in this study. During June to October in 2014, 20 plots (20 m × 20 m) each spaced more than 45 m (mean = 247 m) apart were established in each site (Ji *et al.*, 2019). In each plot, ten soil cores (3.5 cm diameter, 10 cm depth) were randomly collected, bulked and subsampled to form a composite sample, which resulted in a total of 240 composite samples in the entire study. Samples were transported on ice to the laboratory and sieved through a 2-mm sieve to remove roots and debris. One portion of the soil samples was stored at −80°C until DNA extraction; another portion was air-dried for analyses of physicochemical soil properties. In the same quadrats that the soil samples were collected from, plant communities were assessed and characterised (Ji *et al.*, 2019), and species were classified as arbuscular mycorrhizal, ectomycorrhizal, ericoid mycorrhizal and non-mycorrhizal based on the published literature (Wang & Qiu, 2006; Maherali *et al.*, 2016). Soil, plant, and climatic variables in each study site are shown in Table S1, Table S2, and Table S3.

Soil and climatic factors

Soil pH, total carbon (TC), total nitrogen (TN), total phosphorus (TP), C:N, and N:P ratios were described in detail by Ji *et al.* (2019). Latitude, longitude, and plant data (basal area, richness and community composition) of the study sites were provided by the CForBio organization (successively obtain from the colleagues as mentioned in the Acknowledgments section around a period in 2015). Mean annual temperature (MAT) and mean annual precipitation (MAP) were obtained from the WorldClim database (www.worldclim.org) with a resolution of 2.5 min (Ji *et al.*, 2019).

Molecular analysis

Molecular analyses were performed independently on the 20 soil samples collected from each of the 12 sites. Genomic DNA was extracted from 0.25 g of frozen soil using a PowerSoil DNA isolation kit (MoBio Laboratories, Inc. USA) according to the manufacturer's instructions. The DNA was diluted 10-fold in water used in all downstream PCR experiments. The primers and PCR conditions for amplifying total fungal (including EM fungi – only 1% of sequence reads belonged to AM fungal; ITS) and AM fungi (18S) DNA are shown in Table S4. A barcode was added to the 5' end of the reverse primers (*i.e.*, ITS2 and AMDGR) to identify samples when demultiplexing sequence reads. The PCR products were purified using a PCR product gel purification kit (Axygen, Union City, CA, USA). The yields of purified PCR products were measured using a TBS 380 Fluorescence Spectrophotometer (Promega, USA), and 50 ng of DNA from each of the 240 purified PCR products were pooled and adjusted to 10 ng μL^{-1}. The pooled DNA was subjected to sequencing on an Illumina MiSeq PE250 platform for sequencing using the paired end (2 × 250 base pair (bp)) option at the Environmental Genome Platform in the Chengdu Institute of Biology, Chinese Academy of Sciences, China.

Sequence processing

Clean sequences were obtained from raw sequences after quality control using 'Quantitative Insights into Microbial Ecology' (QIIME v.1.7.0, Caporaso *et al.*, 2010). Quality control removed low quality reads with no valid primer sequence or barcode sequence, containing ambiguous bases, or with an average quality score < 20. For ITS reads, the ITS1 region of each high-quality sequence was extracted using the fungal ITSx software package (Bengtsson-Palme *et al.*, 2013). Chimeric sequences were detected using the 'chimera.uchime' command in Mothur 1.31.2 (Schloss, 2009) and removed from further analysis. The non-chimeric sequences were grouped into different operational taxonomic units (OTUs) at a 97% similarity level based on the UPARSE pipeline using the USEARCH v8.0 after dereplication and singleton exclusion (Edgar, 2013).

The sequence number per sample was normalized to that sample with the fewest

reads (5290 and 1509 for the ITS1 and 18S sequences, respectively) using the 'sub.sample' command in Mothur to eliminate the influence of different read numbers on the analysis of fungal communities. Representative sequences from OTUs were selected through the command of 'get.oturep' and were identified by a basic local alignment search tool (BLAST) search against the international nucleotide sequence databases collaboration (INSDc) and UNITE database (Kõljalg *et al.*, 2013) for ITS1 sequences and the Maarj*AM* 18S rRNA gene database (Öpik *et al.*, 2010) for 18S sequences (accessed on Apr. 9th, 2016 and on Sept. 28th, 2016, respectively), using an E value less than $1e^{-50}$ as a threshold. Furthermore, the ITS1 OTUs were assigned to functional guilds following the methods of Tedersoo *et al.* (2014) and using the information in FunGuild (Nguyen *et al.*, 2016; accessed on Nov. 24th, 2016). The results of ITS sequences were described in detail by Ji *et al.* (2019). Briefly, of the obtained 14 911 ITS OTUs (1 267 912 reads), a subset of 9 883 ITS OTUs (reads ≥ 3) was extracted to form the total fungal community, and all identified 2 709 EM fungal OTUs (431 138 reads, 34.0% of all fungal sequences) were used for subsequent analyses in the current study. We obtained a total of 4 517 918 high-quality 18S rRNA gene sequences, which were clustered into 638 18S OTUs. After normalization, we obtained 592 18S OTUs (357 633 reads) which all matched with known AM fungal sequences from the Maarj*AM* database. Three (*i.e.*, total, AM and EM) fungal community matrices (Table S5) were used in all analyses, including generation of null models based on the neutral model: total fungi (all ITS1 OTUs), EM fungi (ITS1 OTUs assigned to EM taxa) and AM fungi (18S OTUs assigned to Glomeromycota). The DNA sequences of the ITS and 18S rRNA gene amplicons were deposited in the National Center for Biotechnology Information under accession nos. LT986405–LT998319 and MK352490–MK353081, respectively. The raw sequencing data (fastq files) have been submitted to the Environmental Genomic Cloud (http://egcloud.cib.cn) with the sample nos. XX to XX under the project number XX.

Neutral model simulation and comparison analysis

The approach described in Etienne (2007) was used to estimate immigration and diversity parameters from a neutral model, and these estimates were used to simulate communities assembled under only neutral processes. First, the fungal (total, AM, and EM) sample-OTU data obtained from each site was used to estimate the theta (Θ, diversity index) and immigration rate (I) parameters of the neutral model. The formula was used

for multiple samples to estimate neutral parameters using the PARI/GP code included in Etienne (2007). Parameters were estimated for the total, AM, and EM fungal communities separately from each of the 12 forest sites. Using the estimated parameters, the PARI/GP function 'urn2.gp' was performed to create 100 communities of matching size. The detailed processes conducted for simulating the neutral community were described by Maaß *et al.* (2014). The β-diversity of a real set of local assemblages (observed fungal communities in the sample-OTU matrices) were compared to the frequency distribution of β-diversities obtained from 100 simulations of the corresponding fungal community under the neutral model.

The PARI/GP output files were imported into R (R Core Team, 2015) to calculate pairwise Sørensen community dissimilarities (β-diversity) among all communities within each simulation (Powell *et al.*, 2015). The potential importance of stochasticity was evaluated using standardized effect size (SES) calculated as the difference in observed statistics (median, interquantile range [IQR] and interdecile range [IDR]) associated with dissimilarity distributions and corresponding expectations produced by the neutral models using the following formula: ($estimate_{obs.}$ – mean of $estimate_{exp.}$) / standard deviation of the $estimate_{exp}$, where $estimate_{obs.}$ denotes the estimate (median/IQR/IDR) associated with the observed distribution of Sørensen dissimilarities and $estimate_{exp.}$ denotes the estimate (median/IQR/IDR) associated with the distribution of Sørensen dissimilarities for each of the individual simulated communities. If the SES value was not significantly ($P < 0.05$) different from zero, the community assembly was interpreted as being driven primarily by neutral process; otherwise, the community assembly was regarded as deterministic. In the case of a significant difference in the SES from zero, a positive effect size indicated that the estimate was higher than predicted (divergence), while a negative effect size indicated that the estimate was lower than predicted (convergence) under the neutral hypothesis. The SES values were compared amongst the three fungal groups within each site and the mean values of all 12 sites. The same analyses were conducted for the total plant community, as well as the AM and EM plant communities.

Estimation of community and habitat turnover

By applying the approach described in Ranjard *et al.* (2013), we estimated relationships between fungal β-diversity along the latitudinal gradient and in relation to habitat turnover among forests. Briefly, pairwise fungal β-diversities (as Sørensen index) were

calculated using the 'labdsv' package (Roberts, 2016) in R. To compare with latitude, β-diversity was estimated as the median Sørensen dissimilarity among local communities within each of the 12 forests. To compare with habitat turnover among forests, β-diversity was estimated as the Sørensen dissimilarity between each pair of forests after aggregating each OTU-sample matrix so that each sample represented the sum of OTU frequencies in each of the 12 forests. Habitat turnover (environmental dissimilarity, *Ed*) was calculated from the Euclidean distance between sites ('dist' function in R) based on the site-level soil, plant, and climatic factors using the formula $Ed = 1 - Euc_d / Euc_{max}$, in which Euc_d is the Euclidean distance and Euc_{max} is the maximum distance between sites in the integrated data matrix including soil, plant, and climate properties.

Statistical analysis

Multiple comparisons of group means among the 12 forest sites were carried out with Tukey's honestly significant difference (HSD) test after one-way analysis of variance (ANOVA) or pairwise comparisons after the nonparametric Kruskal–Wallis test where data did not satisfy homogeneity of variance, at $P < 0.05$, which indicated a significant effect of site on environmental factors (soil, plant, and climate), the observed β-diversity, and SES values. Similar analyses of one-way ANOVA followed by multiple comparisons were also conducted for the observed β-diversity amongst different fungal groups and different climate zones. Fungal community compositions were ordinated using nonmetric multidimensional scaling (NMDS) with dissimilarity matrices using the 'metaMDS' function in the 'vegan' package (Oksanen *et al.*, 2013). We used the 'lmodel2' package (Legendre, 2011) to perform type II linear regression (ordinary least squares) on the relationships between the observed fungal β-diversity and latitude and habitat turnover (*Ed*) after estimating these at the site level. To evaluate the effects of climate zone (tropical, subtropical, and temperate) and site (both all data and separate data sets of tropical, subtropical, and temperate) on total, AM, and EM fungal β-diversities, permutational multivariate analysis of variance (PerMANOVA) was conducted based on distance matrices (Sørensen dissimilarity) using the 'adonis' function in the 'vegan' package with 999 permutations. To explore the independent influence of dispersal limitation on the fungal community assembly, partial Mantel tests were further performed to analyze the correlation between fungal communities and geographic distance after excluding the effects of significant abiotic (soil and climate related) and biotic (plant

related) factors using the 'ecodist' package (Goslee & Urban, 2007). In turn, partial Mantel tests were carried out to explore the relationships between the fungal community and abiotic/biotic factors after excluding the influence of geographic distance. All analyses were conducted in R 3.0.2 (R Core Team, 2015).

Results

Divergence dominated fungal and plant community assembly outcomes within forests

Distributions of observed community dissimilarities (β-diversity) among the total, AM, and EM fungal communities and among total, AM and EM plant communities are presented as probability densities (Fig. S2). Their distributions indicate that substantial variation was observed among communities for all groups across many sites, with some of the distributions being bimodal (indicating convergence among some samples within a site). SES values for all these fungal groups were consistently greater than zero (Table S6), indicating significant divergence. The highest fungal SES values within a site were generally associated with total or AM fungal communities, and the lowest SES values were always observed for the EM fungal communities except for site Baotianman (BTM, Table S6). Estimates of *Theta* and *I* are provided in Table S7.

The results of null model analyses indicated that there was strong evidence for divergent assembly among total, AM, and EM fungal communities in each of the three forest biomes (tropical, subtropical, and temperate zones) based on deviations in observed dispersion (Fig. 1b, c) and/or average dissimilarity (Fig. 1a) from the 100 simulations under the null model. For the plant communities, not all SES estimates associated with average dissimilarity differed from zero (Fig. 1a). However, dispersion among observed communities relative to those simulated assuming was substantial in all cases (Fig. 1b, c), reflecting the large range of dissimilarities observed at most sites (Fig. S2) and providing evidence for divergent assembly.

We did not observe any correlations between SESs for fungal and plant communities when comparing responses at individual sites (Table S6) or among biomes (Fig. 1). The most important landscape-level environmental variables for explaining variation in SES values for total fungi were temperature (P = 0.009), latitude (P = 0.012) and total plant species richness (P = 0.034), with plant community composition (P = 0.053) and

precipitation (P = 0.051) being marginally nonsignificant (Table 1). For AM fungi, SES values were significantly correlated with soil C:N ratio (P = 0.020) and marginally nonsignificantly correlated with pH (P = 0.065) and plant community composition (P = 0.079) (Table 1). No environmental factors were correlated with EM fungal SES values (Table 1). SES values for plant communities tended to be correlated more closely with plant community variables, including plant species richness (all and AM plants), basal area (AM plants), and community composition (EM plants).

Environmental drivers of fungal β-diversity across forests

The mean observed β-diversity of EM fungi was consistently and significantly greater than that of total and AM fungi ($P < 0.0001$), but no significant β-diversity difference was detected among the three plant groups (P = 0.179, Table S8). Fungal β-diversity was significantly greater within temperate forests, followed by tropical forests and then subtropical forests ($P < 0.0001$, Table S9). There was no significant difference in plant β-diversity across tropical, subtropical, and temperate forests (P = 0.764, Table S9). Both fungal and plant compositions were structured by climate (Figure 2, Table S10) and site (Table S10), regardless of whether whole communities or AM/EM subsets were assessed. We observed larger effects of climate zone and site on AM fungi (R^2 = 0.280) than on total (R^2 = 0.095) and EM (R^2 = 0.069) fungi, based on larger R^2 values in PerMANOVA analyses (Table S10). Among all groups of total, AM, and EM fungi, the effects of site on β-diversities were larger (*i.e.*, greater R^2 values) in the subtropical (R^2, 0.222, 0.403, 0.172, respectively) zone as compared with in temperate (R^2, 0.183, 0.247, 0.156, respectively) and tropical (R^2, 0.086, 0.266, 0.064, respectively) zones (Table S10). Variation explained by climate zone and type was similar when comparing the total, AM, and EM plant groups (Table S10).

At the sampling site level, we found that the observed AM fungal β-diversity was positively correlated with latitude ($P < 0.0001$, Fig. 3a), which is the opposite pattern as was observed for AM plant β-diversity (P = 0.025, Fig. 3c). The increase in AM fungal β-diversity with latitude was mirrored by a decline in AM fungal richness with latitude ($P < 0.001$, Fig. S3). No significant correlation with latitude was observed for β-diversity within the other fungal groups (Fig. 3a) despite total (P = 0.004) and EM (P = 0.023) plant β-diversity exhibiting negative relationships with latitude (Fig. 3c). We observed the

highest β-diversity in soil EM fungal communities followed by total fungal communities and then AM fungal communities (Fig. 3b). β-diversity in all fungal groups increased with greater habitat turnover (*Ed*; P values < 0.05 for total and EM fungi, $P = 0.063$ for AM fungi; Fig. 3b). The β-diversity for all plant groups were not significantly associated with habitat turnover, which may be due to the limited overlap in plant composition among plots (Fig. 3d).

Finally, we performed partial Mantels test to reveal relationships among geographic distance and individual environmental factors on fungal community composition. Pure geographic distance was not significantly correlated with any fungal community groupings (Table 2). Soil pH and total C were significant variables explaining variation in all fungal groups after accounting for geographic distance, plants and climatic factors, while temperature and plant community composition also explained variation in all fungal groups after accounting for other variables (Table 2). Variation in AM fungal community composition was further explained by variation in AM plant basal area (Table 2).

Discussion

Divergence-dominated fungal community assembly

The relative importance of stochastic and deterministic processes in structuring a microbial community is currently being debated (Zhou & Ning, 2017). Previous studies documented that the assembly of microbial communities depends largely on the deterministic process (niche-based) driven by contemporary environmental changes, such as pH, temperature, precipitation, salinity and nutrients (Fierer & Jackson, 2006; Lozupone & Knight, 2007; Wang *et al.*, 2016; Zhou *et al.*, 2016; Zhang *et al.*, 2019; Zheng *et al.*, 2020). Microbial communities can also be driven by stochastic processes such as geographical separation and dispersal limitation (Wang *et al.*, 2013). Compared with most cases focusing on bacteria from the available microbial community studies, the importance of stochastic processes in generating and maintaining fungal biodiversity is rarely appreciated, and a few studies have addressed the importance of neutrality for these communities in soil and roots (Caruso *et al.*, 2012; Schroter *et al.*, 2019).

Here, we observed evidence for assembly processes leading to strong divergence among soil fungal communities within forests each distributed along a large environmental gradient. This was the case for whole fungal communities and for subsets

consisting of AM and EM fungi, although divergence was less among communities of the latter group. The degree of divergence at whole fungal communities was greater in more diverse forests in warmer climates at lower latitudes, which is consistent with how each of the forest biomes was ranked in terms of degree of divergence (tropical > subtropical > temperate). Interpreting these differences is challenging due to the possibility of fungal community shifts leading to differences in abundance of functional guilds among these communities, so we mainly discuss the results of the AM and EM fungal community subsets.

These results indicating a greater tendency toward divergence are consistent with expectations under two scenarios: (1) environmental heterogeneity (*e.g.*, in soil properties, vegetation and/or microclimate) within forests leads to strong niche-based assembly within soil fungal communities or (2) strong biotic interactions lead to communities diverging more than expected under similar environmental conditions. Powell and Bennett (2016) observed similar results for AM fungal communities sampled in a global survey (Davison *et al.*, 2015). In that analysis, divergence and neutrality were much more frequent outcomes than convergence when trying to compare local communities sampled from similar environments (roots of a single host species collected from multiple plants within a single plot). Here, we observed greater dispersion among AM fungal communities within sites in the higher latitude forests (greater NMDS area of temperate and subtropical than that tropical forest sites; Fig. 2b), but this pattern was confounded by the reduced AM fungal richness at higher latitudes. Similarly, the value associated with the null model approach was revealed in that it detected stronger divergence, on average, in temperate and subtropical forests than in tropical forests (Fig. 1a), although this was independent of other attributes of those plant communities. Veresoglou *et al.* (2019) also reported that latitudinal differences in AM fungal β-diversity were driven to a greater extent by forest types than by latitude. We also observed stronger divergence in soils with higher C-to-N ratios, where antagonistic interactions with decomposer fungi might be more prevalent (Bunn *et al.*, 2019).

Less divergence was observed among EM fungal communities in the current study than what was observed for AM fungi and whole fungal communities, which might be attributed to relatively high host specificity for EM fungi compared with AM fungi (Tedersoo *et al.*, 2008; Davison *et al.*, 2015; Wang *et al.*, 2019). If this was the case we might expect a positive relationship between the degree of divergence and variation in

EM fungal host communities. However, we did not observe such a relationship. We did not observe much difference in the degree of divergence among the three forest types either. This result may reflect the limitations associated with trying to interpret assembly processes in EM fungal communities from soil samples instead of root samples, where in the latter there might be stronger evidence of niche-based assembly. Regardless, there is some validity to interpreting these data as these samples do reflect the composition of EM fungal communities available to colonise root samples in these environments.

Drivers of soil fungal β-diversity in forests at large scales

In addition to a greater prevalence of divergent community assembly for soil fungi within each forest, we observed high levels of divergence among soil fungal communities across the forests. This divergence could be partially explained by climate, vegetation and soil properties. For the most part, the environmental drivers associated with fungal community turnover were consistent among the three fungal groups, with soil C, plant community composition, MAT and soil pH being the most important variables. These results are consistent with other large scale studies of soil fungal diversity (*e.g.*, Tedersoo *et al.*, 2014; Zhou *et al.*, 2016; Guo *et al.*, 2018).

However, we did observe that AM fungal β-diversity under these forests was lower than the other groups and its relationship with habitat turnover overall was not as strong. It is difficult to draw conclusions from these particular results given the possibility that this is determined, in part or as a whole, by greater variation in the ITS region (used to sample total fungi and EM fungi) than that of the 18S rRNA gene (used to sample AM fungi). That said, when comparing compositional shifts in soil fungal communities among the 12 forests, strengths of relationships with some individual variables tended to be greater for AM fungi than for the other two groups. This difference was particularly large for soil C, which is intriguing given speculation on the role that AM fungal-derived proteins (*i.e.*, glomalin) play in determining the recalcitrance of soil C stocks (Rillig *et al.*, 2003). The AM fungal subset was also the only fungal group to exhibit a significant relationship between composition and the basal area of probable host trees. This may reflect the more generalist nature of AM fungal and plant associations, where frequency of host plants may be a greater driver of composition than host identity, than in more specialist EM fungal and plant associations.

Conclusions

Here we were able to demonstrate substantial divergence among soil fungal communities, including mycorrhizal fungal communities both within and among forests at a large geographic scale (> 4 000 km). Within forests, fungal communities were consistently more divergent than expected under neutrality and exhibited patterns that appeared to indicate that the relative strength of deterministic assembly processes differed among fungi and plants in these forests. We did observe that the degree of divergence was related, in part, to environmental variation when looking at whole fungal communities or those of AM fungi. However, we were unable to detect patterns to explain the degree of divergence among EM fungal communities despite the inclusion of variables that were related to compositional shifts at large scales. Taken together, although soil fungal community turnover at large scales could be attributed to specific environmental drivers, strong divergence during fungal community assembly in forest soils at local scales limits the predictability of fungal community assembly outcomes.

Acknowledgments

We are grateful to staff of the CForBio for soil sample collection. We appreciate Professors Ke-Ping Ma, Xiao-Jun Du, Wei-Guo Sang, Zhan-Qing Hao, Ming-Xi Jiang, Wan-Hui Ye, Min Cao, Xian-Kun Li, Guang-Ze Jin, Xiao-Yong Chen, and Bu-Hang Li for providing the plant information. This study was financially supported by the Strategic Priority Research Program (B) of the Chinese Academy of Sciences (XDB31000000), National Natural Science Foundation of China (31971447 and 31210103910) and the Australian Research Council (DP130102501 and FT190100590). We thank Dr. Jianjun Wang from Nanjing Institute of Geography and Limnology, Chinese Academy of Sciences for the helpful comments on the early version of the manuscript.

Author Contributions

Y.Z., L.C. and L.-D.G. designed the experiments and analysed the data in consultation with C.G., S.-S.J., H.-W.H., Z.H., J.-Z.H., and J.R.P. The field surveys, soil samplings, and molecular work were conducted by Y.Z., N.-N.J., L.C., Y.-L.W., and C.G. The

manuscript was written by Y.Z. and J.R.P. with contributions from all co-authors.

Data Availability

The data that support the findings of this study are openly available on figshare at https://www.doi.org/10.6084/m9.figshare.13543046.

References

Alonso D, Etienne RS, McKane AJ. 2006. The merits of neutral theory. *Trends in Ecology & Evolution* **21**: 451–457.

Alzarhani AK, Clark DR, Underwood GJC, Ford H, Cotton TEA, Dumbrell AJ. 2019. Are drivers of root-associated fungal community structure context specific? *The ISME Journal* **13**: 1330–1344.

Beck S, Powell JR, Drigo B, Cairney JWG, Anderson IC. 2015. The role of stochasticity differs in the assembly of soil- and root-associated fungal communities. *Soil Biology & Biochemistry* **80**: 18–25.

Bengtsson-Palme J, Ryberg M, Hartmann M, Branco S, Wang Z, Godhe A, De Wit P, Sánchez-García M, Ebersberger I, de Sousa F *et al.* 2013. Improved software detection and extraction of ITS1 and ITS2 from ribosomal ITS sequences of fungi and other eukaryotes for analysis of environmental sequencing data. *Methods in Ecology and Evolution* **4**: 914–919.

Bunn RA, Simpson DT, Bullington LS, Lekberg Y, Janos DP. 2019. Revisiting the 'direct mineral cycling' hypothesis: arbuscular mycorrhizal fungi colonize leaf litter, but why? *The ISME Journal* **13**: 1891–1898.

Caporaso JG, Kuczynski J, Stombaugh J, Bittinger K, Bushman FD, Costello EK, Fierer N, Peña AG, Goodrich JK, Gordon JI *et al.* 2010. QIIME allows analysis of high-throughput community sequencing data. *Nature Methods* **7**: 335–336.

Caruso T, Hempel S, Powell JR, Barto EK, Rillig MC. 2012. Compositional divergence and convergence in arbuscular mycorrhizal fungal communities. *Ecology* **93**: 1115–1124.

Chase JM. 2003. Community assembly: when should history matter? *Oecologia* **136**: 489–498.

Chase JM. 2010. Stochastic community assembly causes higher biodiversity in more productive environments. *Science* **328**: 1388–1391.

Davison J, Moora M, Öpik M, Adholeya A, Ainsaar L, Bâ A, Burla S, Diedhiou AG, Hiiesalu I, Jairus T *et al.* 2015. Global assessment of arbuscular mycorrhizal fungus diversity reveals very low endemism. *Science* **349**: 970–973.

Edgar RC. 2013. UPARSE: highly accurate OTU sequences from microbial amplicon reads. *Nature Methods* **10**: 996–998.

Ellwood MDF, Manica A, Foster WA. 2009. Stochastic and deterministic processes jointly structure tropical arthropod communities. *Ecology Letters* **12**: 277–284.

Etienne RS. 2007. A neutral sampling formula for multiple samples and an 'exact' test of neutrality. *Ecology Letters* **10**: 608–618.

Ettema CH, Wardle DA. 2002. Spatial soil ecology. *Trends in Ecology & Evolution* **17**: 177–183.

Fargione J, Brown CS, Tilman D. 2003. Community assembly and invasion: an experimental test of neutral versus niche processes. *Proceedings of the National Academy of Sciences of the United States of America* **100**: 8916–8920.

Fierer N, Jackson RB. 2006. The diversity and biogeography of soil bacterial communities. *Proceedings of the National Academy of Sciences of the United States of America* **103**: 626–631.

Gao C, Zhang Y, Shi NN, Zheng Y, Chen L, Wubet T, Bruelheide H, Both S, Buscot F, Ding Q *et al.* 2015. Community assembly of ectomycorrhizal fungi along a subtropical secondary forest succession. *New Phytologist* **205**: 771–785.

Gardes M, Bruns TD. 1993. ITS primers with enhanced specificity for Basidiomycetes - application to the identification of mycorrhizae and rusts. *Molecular Ecology* **2**: 113–118.

Goslee SC, Urban DL. 2007. The ecodist package for dissimilarity-based analysis of

ecological data. *Journal of Statistical Software* **22**: 1–19.
Guo X, Feng JJ, Shi Z, Zhou XS, Yuan MT, Tao XY, Hale L, Yuan T, Wang JJ, Qin YJ *et al.* 2018. Climate warming leads to divergent succession of grassland microbial communities. *Nature Climate Change* **8**: 813–818.
Hubbell SP. 2001. *The unified neutral theory of biodiversity and biogeography*. Princeton, USA: Princeton University Press.
Ji NN, Gao C, Sandel B, Zheng Y, Chen L, Wu BW, Li XC, Wang YL, Lü PP, Sun X *et al.* 2019. Late Quaternary climate change explains soil fungal community composition rather than fungal richness in forest ecosystems. *Ecology and Evolution* **9**: 6678–6692.
Kivlin SN, Winston GC, Goulden ML, Treseder KK. 2014. Environmental filtering affects soil fungal community composition more than dispersal limitation at regional scales. *Fungal Ecology* **12**: 14–25.
Kõljalg U, Nilsson RH, Abarenkov K, Tedersoo L, Taylor AFS, Bahram M, Bates ST, Bruns TD, Bengtsson-Palme J, Callaghan TM *et al.* 2013. Towards a unified paradigm for sequence-based identification of fungi. *Molecular Ecology* **22**: 5271–5277.
Kraft NJB, Valencia R, Ackerly DD. 2008. Functional traits and niche-based tree community assembly in an Amazonian forest. *Science* **322**: 580–582.
Lee J, Lee S, Young JPW. 2008. Improved PCR primers for the detection and identification of arbuscular mycorrhizal fungi. *FEMS Microbiology Ecology* **65**: 339–349.
Legendre P. 2011. *lmodel2: Model II Regression. R package version 1.7-2.* URL https://CRAN.R-project.org/package=lmodel2.
Lozupone CA, Knight R. 2007. Global patterns in bacterial diversity. *Proceedings of the National Academy of Sciences of the United States of America* **104**: 11436–11440.
Maaß S, Migliorini M, Rillig MC, Caruso T. 2014. Disturbance, neutral theory, and patterns of beta diversity in soil communities. *Ecology and Evolution* **4**: 4766–4774.
Maherali H, Oberle B, Stevens PF, Cornwell WK, McGlinn DJ. 2016. Mutualism persistence and abandonment during the evolution of the mycorrhizal symbiosis. *American Naturalist* **188**: E113–E125.
Mayor J, Bahram M, Henkel T, Buegger F, Pritsch K, Tedersoo L. 2015. Ectomycorrhizal impacts on plant nitrogen nutrition: emerging isotopic patterns, latitudinal variation and hidden mechanisms. *Ecology Letters* **18**: 96–107.
Nemergut DR, Schmidt SK, Fukami T, O'Neill SP, Bilinski TM, Stanish LF, Knelman JE, Darcy JL, Lynch RC, Wickey P *et al.* 2013. Patterns and processes of microbial community assembly. *Microbiology and Molecular Biology Reviews* **77**: 342–356.
Nguyen NH, Song ZW, Bates ST, Branco S, Tedersoo L, Menke J, Schilling JS, Kennedy PG. 2016. FUNGuild: An open annotation tool for parsing fungal community datasets by ecological guild. *Fungal Ecology* **20**: 241–248.
Oksanen J, Blanchet FG, Kindt R, Legendre P, Minchin PR, O'Hara RB, Simpson GL, Solymos P, Stevens MHH, Wagner H. 2013. Vegan: community ecology package. R package version 2.0-10. Available at: http://CRAN.R-project.org/package=vegan.
Öpik M, Vanatoa A, Vanatoa E, Moora M, Davison J, Kalwij JM, Reier Ü, Zobel M. 2010. The online database Maarj*AM* reveals global and ecosystemic distribution patterns in arbuscular mycorrhizal fungi (Glomeromycota). *New Phytologist* **188**: 223–241.
Philippot L, Raaijmakers JM, Lemanceau P, van der Putten WH. 2013. Going back to the roots: the microbial ecology of the rhizosphere. *Nature Reviews Microbiology* **11**: 789–799.
Powell JR, Bennett AE. 2016. Unpredictable assembly of arbuscular mycorrhizal fungal communities. *Pedobiologia* **59**, 11–15.
Powell JR, Karunaratne S, Campbell CD, Yao HY, Robinson L, Singh BK. 2015. Deterministic processes vary during community assembly for ecologically dissimilar taxa. *Nature Communications* **6**: 8444.

R Core Team. 2015. *R: a language and environment for statistical computing*. URL http://www.R-project.org/.

Ranjard L, Dequiedt S, Chemidlin Prévost-Bouré N, Thioulouse J, Saby NPA, Lelievre M, Maron PA, Morin FER, Bispo A, Jolivet C, Arrouays D, Lemanceau P 2013. Turnover of soil bacterial diversity driven by wide-scale environmental heterogeneity. *Nature Communications* **4**: 1434.

Rillig MC, Ramsey PW, Morris S, Paul EA. 2003. Glomalin, an arbuscular-mycorrhizal fungal soil protein, responds to land-use change. *Plant and Soil* **253**: 293–299.

Roberts DW. 2016. *labdsv: Ordination and Multivariate Analysis for Ecology. R package version 1.8-0*. URL https://CRAN.R-project.org/package=labdsv.

Sato K, Suyama Y, Saito M, Sugawara K. 2005. A new primer for discrimination of arbuscular mycorrhizal fungi with polymerase chain reaction-denature gradient gel electrophoresis. *Grassland Science* **51**: 179–181.

Schloss PD, Westcott SL, Ryabin T, Hall JR, Hartmann M, Hollister EB, Lesniewski RA, Oakley BB, Parks DH, Robinson CJ *et al.* 2009. Introducing mothur: open-source, platform-independent, community-supported software for describing and comparing microbial communities. *Applied and Environmental Microbiology* **75**: 7537–7541.

Schroter K, Wemheuer B, Pena R, Schoning I, Ehbrecht M, Schall P, Ammer C, Daniel R, Polle A. 2019. Assembly processes of trophic guilds in the root mycobiome of temperate forests. *Molecular Ecology* **28**: 348–364.

Schwarzott D, Schüßler A. 2001. A simple and reliable method for SSU rRNA gene DNA extraction, amplification, and cloning from single AM fungal spores. *Mycorrhiza* **10**: 203–207.

Shi LL, Mortimer PE, Slik JWF, Zou XM, Xu J, Feng WT, Qiao L. 2014. Variation in forest soil fungal diversity along a latitudinal gradient. *Fungal Diversity* **64**: 305–315.

Simon L, Lalonde M, Bruns TD. 1992. Specific amplification of 18S fungal ribosomal genes from vesicular-arbuscular endomycorrhizal fungi colonizing roots. *Applied and Environmental Microbiology* **58**: 291–295.

Smith SE, Read DJ. 2008. *Mycorrhizal symbiosis*. Cambridge, UK: Academic Press.

Tedersoo L, Bahram M, Põlme S, Kõljalg U, Yorou NS, Wijesundera R, Ruiz LV, Vasco-Palacios AM, Thu PQ, Suija A *et al.* 2014. Global diversity and geography of soil fungi. *Science* **346**: 1256688.

Tedersoo L, Jairus T, Horton BM, Abarenkov K, Suvi T, Saar I, Kõljalg U. 2008. Strong host preference of ectomycorrhizal fungi in a Tasmanian wet sclerophyll forest as revealed by DNA barcoding and taxon-specific primers. *New Phytologist* **180**: 479–490.

van der Heijden MGA, Klironomos JN, Ursic M, Moutoglis P, Streitwolf-Engel R, Boller T, Wiemken A, Sanders IR. 1998. Mycorrhizal fungal diversity determines plant biodiversity, ecosystem variability and productivity. *Nature* **396**: 69–72.

van der Heijden MGA, Martin FM, Selosse MA, Sanders IR. 2015. Mycorrhizal ecology and evolution: the past, the present, and the future. *New Phytologist* **205**: 1406–1423.

Veresoglou SD, Liu L, Xu T, Rillig MC, Wang M, Wang J, Chen Y, Hu Y, Hao Z, Chen B. 2019. Biogeographical constraints in Glomeromycotinan distribution across forest habitats in China. *Journal of Ecology* **107**: 684–695.

Wang B, Qiu Y-L. 2006. Phylogenetic distribution and evolution of mycorrhizas in land plants. *Mycorrhiza* **16**: 299–363.

Wang JJ, Pan FY, Soininen J, Heino J, Shen J. 2016. Nutrient enrichment modifies temperature-biodiversity relationships in large-scale field experiments. *Nature Communications* **7**: 13960.

Wang JJ, Shen J, Wu YC, Tu C, Soininen J, Stegen JC, He JZ, Liu XQ, Zhang L, Zhang EL. 2013. Phylogenetic beta diversity in bacterial assemblages across ecosystems:

deterministic versus stochastic processes. *The ISME Journal* **7**: 1310–1321.
Wang YL, Gao C, Chen L, Ji NN, Wu BW, Li XC, Lü PP, Zheng Y, Guo LD. 2019. Host plant phylogeny and geographic distance strongly structure Betulaceae-associated ectomycorrhizal fungal communities in Chinese secondary forest ecosystems. *FEMS Microbiology Ecology* **95**: fiz037.
White TJ, Bruns T, Lee S, Taylor J. 1990. Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. *In*: Innis MA, Gelfand DH, Sninsky JJ, White TJ, eds. *PCR protocols: a guide to methods and applications*. San Diego, USA: Academic Press, 315–322.
Zhang KP, Shi Y, Cui XQ, Yue P, Li KH, Liu XJ, Tripathi BM, Chu HY. 2019. Salinity is a key determinant for soil microbial communities in a desert ecosystem. *mSystems* **4**: e00225-18.
Zheng Y, Ji NN, Wu BW, Wang JT, Hu HW, Guo LD, He JZ. 2020. Climatic factors have unexpectedly strong impacts on soil bacterial β-diversity in 12 forest ecosystems. *Soil Biology & Biochemistry* **142**: 107699.
Zhou JZ, Deng Y, Shen LN, Wen CQ, Yan QY, Ning DL, Qin YJ, Xue K, Wu LY, He ZL *et al.* 2016. Temperature mediates continental-scale diversity of microbes in forest soils. *Nature Communications* **7**: 12083.
Zhou JZ, Liu WZ, Deng Y, Jiang YH, Xue K, He ZL, Van Nostrand JD, Wu LY, Yang YF, Wang AJ. 2013. Stochastic assembly leads to alternative communities with distinct functions in a bioreactor microbial community. *mBio* **4**: e00584-12.
Zhou JZ, Ning DL. 2017. Stochastic community assembly: does it matter in microbial ecology? *Microbiology and Molecular Biology Reviews* **81**: e00002-17.

Figure legends

Figure 1. The co-occurrence (X and Y axes represent plant and fungi, respectively) of standardized effect size (SES) for observed community dissimilarities relative to 100 simulations based on a null model of neutral community assembly. Estimates were calculated in relation to central tendencies, from the median **(a)**, and dispersion from the interquartile range (IQR, middle 50% of observations; **b**) and interdecile range (IDR, middle 80% of observations; **c**). Analyses were performed separately for total, arbuscular (AM) and ecto- (EM) mycorrhizal fungi and plants. The median (points) and 95% confidence interval (lines) of the estimates are presented for communities sampled from tropical, subtropical and temperate forest ecosystems. A reduction or increase in the central tendency or dispersion is evidence of community convergence or divergence from a common composition, respectively. Central tendency estimates **(a)** focus on shifts in community composition that tend to occur across all communities while dispersion estimates **(b, c)** focus on extreme shifts between pairs of communities.

Figure 2. Non-metric multidimensional scaling (NMDS) plot showing shifts in community composition for total **(a)**, arbuscular mycorrhizal (AM, **b**), and ectomycorrhizal (EM, **c**) fungi, as well as total **(d)**, AM **(e)**, and EM **(f)** plants, across tropical, subtropical, and temperate forest ecosystems. Solid lines indicate the two-dimensional space that contains all observations within a forest ecosystem type.

Figure 3. Relationships of observed fungal **(a, b)** and plant **(c, d)** community dissimilarity (β-diversity) with latitude **(a, c)** and habitat turnover (environmental dissimilarity, *Ed*; **b, d)**. To compare with latitude, β-diversity was estimated as the median Sørensen dissimilarity among local communities within each of the 12 forests. To compare with habitat turnover among forests, β-diversity was estimated as the Sørensen dissimilarity between each pair of forests after aggregating each OTU/species-sample matrix within each forest. Pairwise environmental dissimilarities were calculated based on Euclidean distances among site-level environmental variables. Solid lines indicate that the predicted relationship is significant ($P < 0.05$) or marginally nonsignificant ($0.05 \leq P < 0.10$) and dashed lines indicate that the relationship is not significant ($P \geq 0.10$). Relationships within each group are based on type II linear regression estimated using ordinary least squares and *P*-values are as follows: between total ($P = 0.573$), AM ($P < 0.0001$) and EM ($P = 0.372$) fungal β-diversity and latitude **(a)**; between total ($P = 0.034$), AM ($P = 0.063$) and EM ($P = 0.043$) fungal β-diversity and *Ed* **(b)**; between total ($P = 0.004$), AM ($P = 0.025$) and EM ($P = 0.023$) plant β-diversity and latitude **(c)**; between total ($P = 0.220$), AM ($P = 0.346$) and EM ($P = 0.387$) plant β-diversity and *Ed* **(d)**. AM: arbuscular mycorrhizal; EM: ectomycorrhizal.

Fig. 1

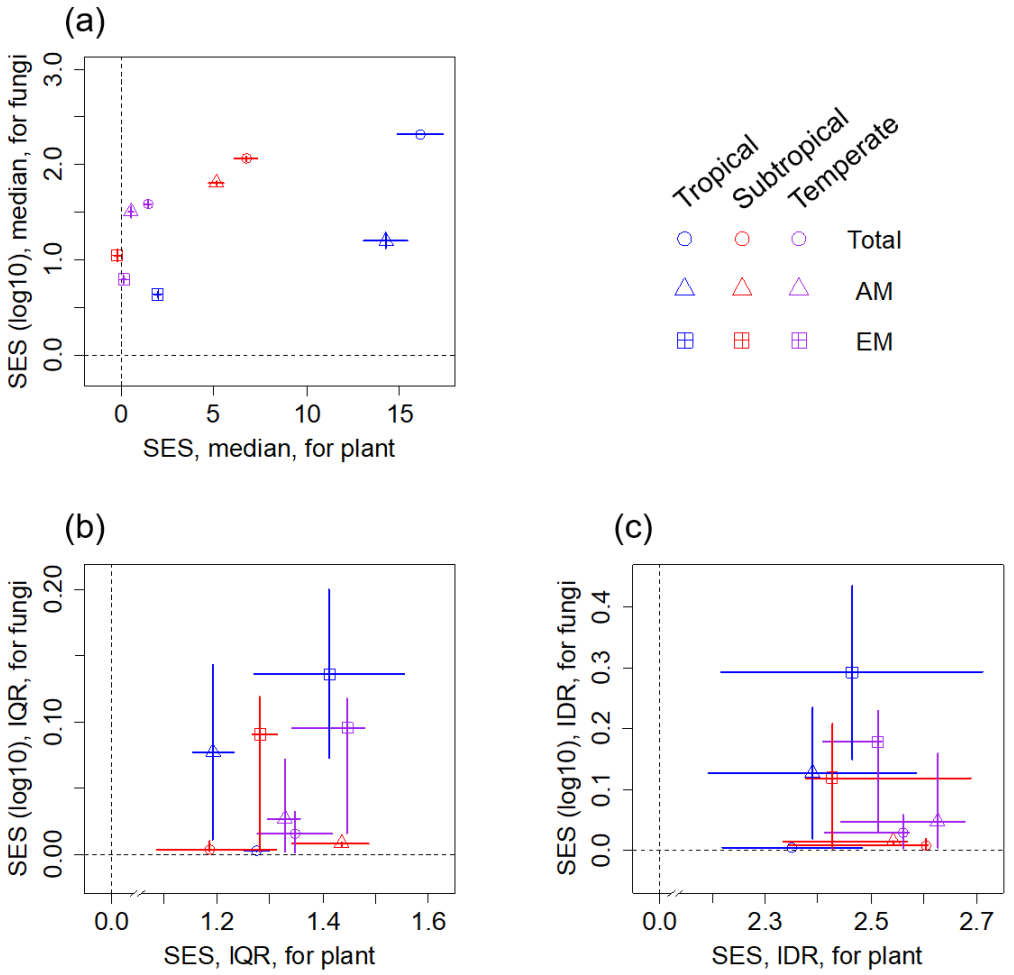

Figure 1. The co-occurrence (X and Y axes represent plant and fungi, respectively) of standardized effect size (SES) for observed community dissimilarities relative to 100 simulations based on a null model of neutral community assembly. Estimates were calculated in relation to central tendencies, from the median **(a)**, and dispersion from the interquartile range (IQR, middle 50% of observations; **b**) and interdecile range (IDR, middle 80% of observations; **c**). Analyses were performed separately for total, arbuscular (AM) and ecto- (EM) mycorrhizal fungi and plants. The median (points) and 95% confidence interval (lines) of the estimates are presented for communities sampled from tropical, subtropical and temperate forest ecosystems. A reduction or increase in the central tendency or dispersion is evidence of community convergence or divergence from a common composition, respectively. Central tendency estimates **(a)** focus on shifts in community composition that tend to occur across all communities while dispersion estimates **(b, c)** focus on extreme shifts between pairs of communities.

Fig. 2

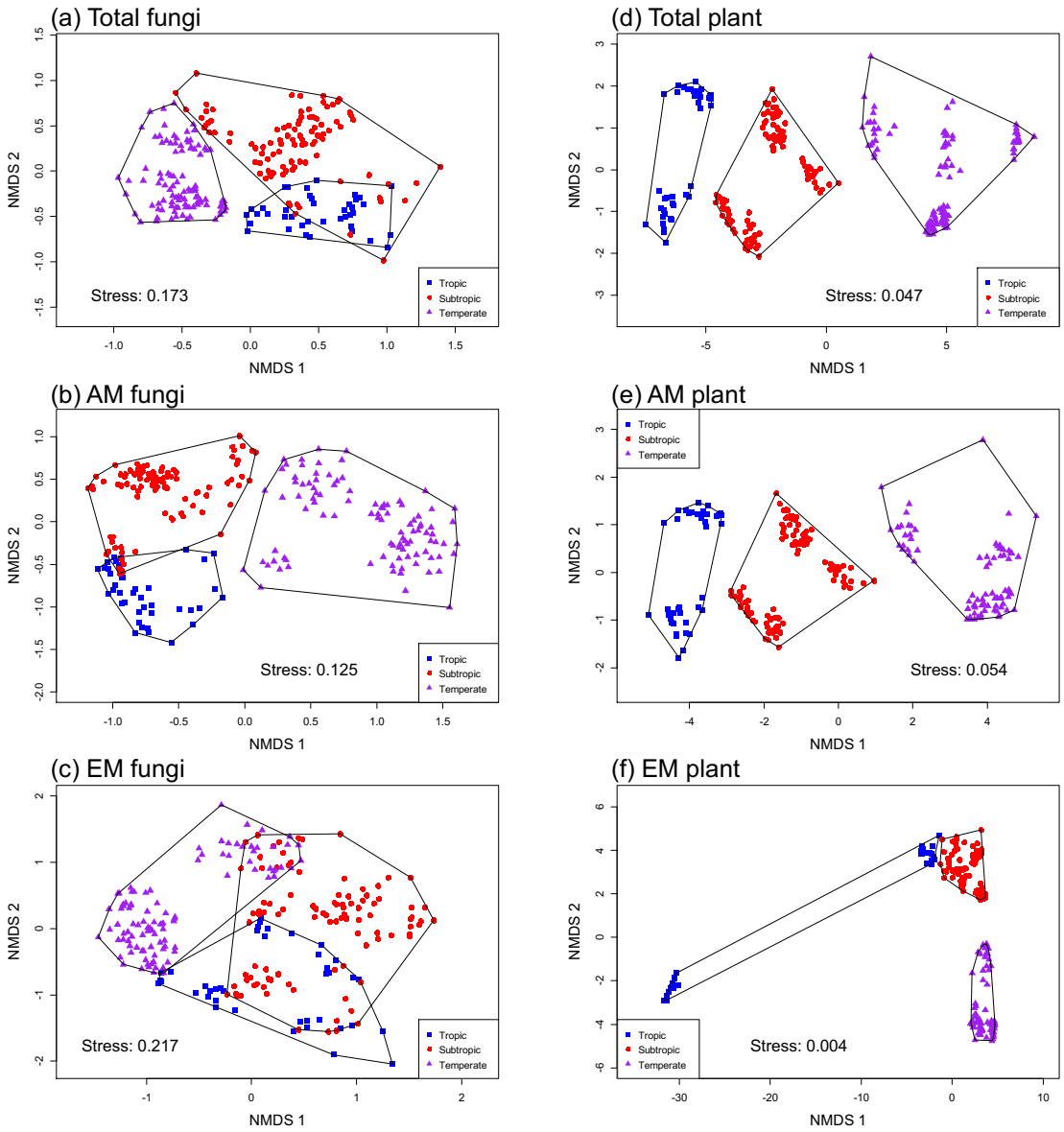

Figure 2. Non-metric multidimensional scaling (NMDS) plot showing shifts in community composition for total **(a)**, arbuscular mycorrhizal (AM, **b**), and ectomycorrhizal (EM, **c**) fungi, as well as total **(d)**, AM **(e)**, and EM **(f)** plants, across tropical, subtropical, and temperate forest ecosystems. Solid lines indicate the two-dimensional space that contains all observations within a forest ecosystem type.

Fig. 3

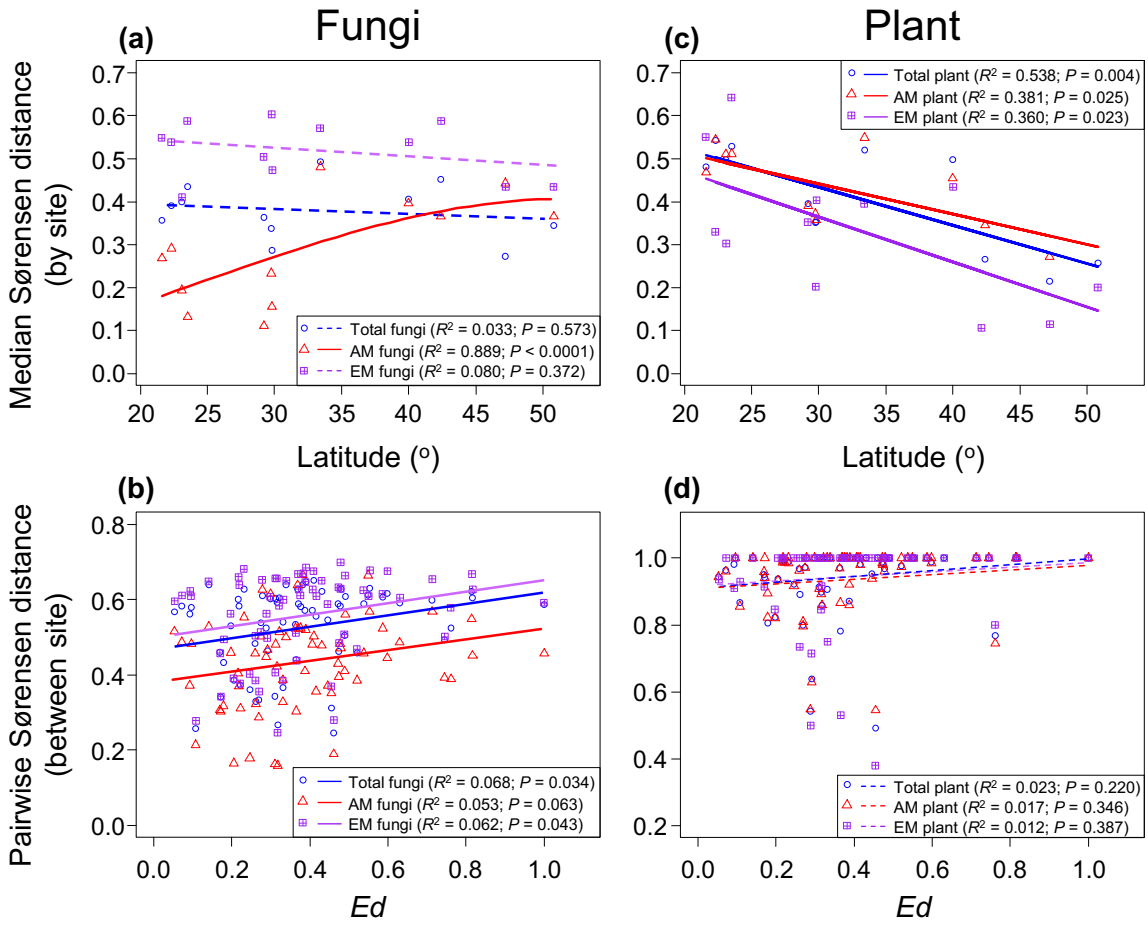

Figure 3. Relationships of observed fungal **(a, b)** and plant **(c, d)** community dissimilarity (β-diversity) with latitude **(a, c)** and habitat turnover (environmental dissimilarity, *Ed*; **b, d)**. To compare with latitude, β-diversity was estimated as the median Sørensen dissimilarity among local communities within each of the 12 forests. To compare with habitat turnover among forests, β-diversity was estimated as the Sørensen dissimilarity between each pair of forests after aggregating each OTU/species-sample matrix within each forest. Pairwise environmental dissimilarities were calculated based on Euclidean distances among site-level environmental variables. Solid lines indicate that the predicted relationship is significant ($P < 0.05$) or marginally nonsignificant ($0.05 \leq P < 0.10$) and dashed lines indicate that the relationship is not significant ($P \geq 0.10$). Relationships within each group are based on type II linear regression estimated using ordinary least squares and *P*-values are as follows: between total ($P = 0.573$), AM ($P < 0.0001$) and EM ($P = 0.372$) fungal β-diversity and latitude **(a)**; between total ($P = 0.034$), AM ($P = 0.063$) and EM ($P = 0.043$) fungal β-diversity and *Ed* **(b)**; between total ($P = 0.004$), AM ($P = 0.025$) and EM ($P = 0.023$) plant β-diversity and latitude **(c)**; between total ($P = 0.220$), AM ($P = 0.346$) and EM ($P = 0.387$) plant β-diversity and *Ed* **(d)**. AM: arbuscular mycorrhizal; EM: ectomycorrhizal.

Table 1

Results of Pearson's correlation analyses between site-level environmental variables and the standardized effect sizes (SESs) associated with shifts in median community composition relative to predictions assuming neutral assembly. Separate analyses were performed for total, arbuscular mycorrhizal (AM), and ectomycorrhizal (EM) fungal and plant communities (results for plant communities are presented within parentheses) groups and environmental factors.

Environmental parameters	**Total fungi (plant)**		**AM fungi (plant)**		**EM fungi (plant)**	
	r value	*P* value	*r* value	*P* value	*r* value	*P* value
pH	-0.327 (-0.324)	0.300 (0.305)	-0.548 (-0.330)	**0.065** (0.321)	-0.432 (0.302)	0.161 (0.429)
TC	-0.269 (-0.120)	0.398 (0.711)	0.227 (-0.235)	0.478 (0.488)	-0.149 (-0.574)	0.645 (0.106)
TN	-0.141 (-0.054)	0.662 (0.867)	-0.165 (-0.232)	0.609 (0.493)	-0.300 (-0.587)	0.343 (0.096)
TP	-0.357 (-0.239)	0.255 (0.454)	-0.336 (-0.293)	0.286 (0.383)	-0.486 (-0.427)	0.109 (0.252)
C:N	-0.358 (-0.284)	0.254 (0.371)	0.659 (-0.246)	**0.020** (0.465)	0.165 (-0.254)	0.608 (0.510)
N:P	0.147 (-0.070)	0.649 (0.828)	0.336 (-0.071)	0.286 (0.835)	0.173 (0.111)	0.591 (0.776)
Plant basal area	-0.199 (0.016)	0.536 (0.960)	-0.028 (0.665)	0.931 (**0.025**)	0.021 (-0.272)	0.949 (0.478)
Plant richness	0.612 (0.563)	**0.034** (**0.057**)	-0.050 (0.670)	0.877 (**0.024**)	-0.129 (-0.325)	0.688 (0.393)
Plant comm. comp.	0.570 (0.106)	**0.053** (0.743)	-0.551 (0.082)	**0.079** (0.810)	0.130 (0.696)	0.688 (**0.037**)
Plant SES	0.483 (/)	0.112 (/)	0.117 (/)	0.732 (/)	0.183 (/)	0.638 (/)
Latitude	-0.693 (-0.464)	**0.012** (0.129)	0.238 (-0.533)	0.457 (0.091)	-0.216 (-0.293)	0.500 (0.444)
MAT	0.716 (0.463)	**0.009** (0.129)	-0.280 (0.525)	0.378 (0.098)	0.279 (0.330)	0.381 (0.385)
MAP	0.574 (0.287)	**0.051** (0.365)	0.090 (0.270)	0.781 (0.422)	0.191 (-0.125)	0.553 (0.749)

Table 2

Results of partial Mantel tests (*r* and *P* values) between each fungal community matrix and matrices of environmental parameters associated with geographic distance, soil properties, corresponding plant parameters, and climatic factors.

Factors	**Total fungi**		**AM fungi**		**EM fungi**	
	r value	*P* value	*r* value	*P* value	*r* value	*P* value
Geographic distance	-0.006	0.692	0.014	0.101	0.0001	0.497
pH	**0.073*****[b]	0.001	**0.076*****	0.001	**0.044*****	0.001
TC[c]	**0.184*****	0.001	**0.321*****	0.001	**0.149*****	0.001
TN	-0.034	0.982	0.009	0.253	-0.017	0.923
TP	0.009	0.272	-0.028	0.994	0.001	0.454
C:N	-0.093	1.000	0.025	0.053	-0.076	1.000
N:P	-0.083	1.000	-0.081	1.000	-0.084	1.000
Plant basal area (Tot., AM, EM, respectively)	-0.029	0.961	**0.083*****	0.001	-0.005	0.598
Plant richness (Tot., AM, EM, respectively)	-0.039	0.998	0.016	0.121	0.019	0.063
Plant community composition	**0.270*****	0.001	**0.213*****	0.001	**0.199*****	0.001
MAT	**0.072*****	0.001	**0.108*****	0.001	**0.032****	0.008
MAP	-0.092	1.000	-0.176	1.000	-0.075	1.000

The relationships which were found to be significant are indicated in bold, **, $P \leq 0.01$; ***, $P \leq 0.001$. TC, soil total carbon; TN, total nitrogen; TP, total phosphorus; C:N, ratio of TC to TN; N:P, ratio of TN to TP; BSA, plant basal area; MAT, mean annual temperature; MAP, mean annual precipitation. AM, arbuscular mycorrhizal; EM, ectomycorrhizal.

Supplementary Information

Title:

Assembly processes lead to divergent soil fungal communities within and among twelve forest ecosystems along a latitudinal gradient

Names of authors:

Yong Zheng[1,2,#], Liang Chen[3,#], Niu-Niu Ji[3], Yong-Long Wang[3], Cheng Gao[3], Sheng-Sheng Jin[1,2], Hang-Wei Hu[1,2], Zhiqun Huang[1,2], Ji-Zheng He[1,2], Liang-Dong Guo[3,4]*, Jeff R. Powell[5]*

Complete postal addresses or affiliations:

[1] State Key Laboratory for Subtropical Mountain Ecology of the Ministry of Science and Technology and Fujian Province, Fujian Normal University, Fuzhou 350007, China.

[2] School of Geographical Sciences, Fujian Normal University, Fuzhou 350007, China.

[3] State Key Laboratory of Mycology, Institute of Microbiology, Chinese Academy of Sciences, Beijing 100101, China.

[4] University of Chinese Academy of Sciences, Beijing 100049, China.

[5] Hawkesbury Institute for the Environment, Western Sydney University, Locked Bag 1797, Penrith, NSW 2751, Australia.

#These authors contributed equally to this work.

***Corresponding authors:**

Dr./Prof. Liang-Dong Guo

State Key Laboratory of Mycology, Institute of Microbiology, Chinese Academy of Sciences, Beijing 100101, China.

Tel.: +86 10 64807510; *E-mail*: guold@im.ac.cn

Dr./Prof. Jeff R. Powell

Hawkesbury Institute for the Environment, Western Sydney University, Locked Bag 1797, Penrith, NSW 2751, Australia.

Tel.: +61 (0)2 4570 1093; *E-mail*: jeff.powell@westernsydney.edu.au

Running title: Fungal community assembly in forest soils.

Figure legends

Figure S1. Geographic distribution of sampling sites in a map of China, which was slightly modified with an inset map based on Ji et al. (2019). Temperate forests include Genhe (GH), Liangshui (LS), Changbaishan (CBS), Donglingshan (DLS), and Baotianman (BTM); subtropical forests include Gutianshan (GTS), Badagongshan (BDGS), Tiantongshan (TTS), Heishiding (HSD), and Dinghushan (DHS); tropical forests include Nonggang (NG) and Xishuangbanna (XSBN). The inset color map shows the distribution of 20 quadrats in the 24-ha plot of GTS.

Figure S2. Distributions of Sørensen dissimilarities (β-diversity) of the observed communities of total fungi **(a)**, AM (arbuscular mycorrhizal) fungi **(b)**, EM (ectomycorrhizal) fungi **(c)**, total plant **(d)**, AM plant **(e)**, and EM plant **(f)** across twelve forests.

Figure S3. A significantly relationship was observed between the arbuscular mycorrhizal (AM) fungal diversity (OTU richness) and latitude. The parameters of this strong relationships were: AIC = 118.7; $R^2_{adj} = 0.7631$; $F = 18.72$; $P < 0.001$.

Fig. S1

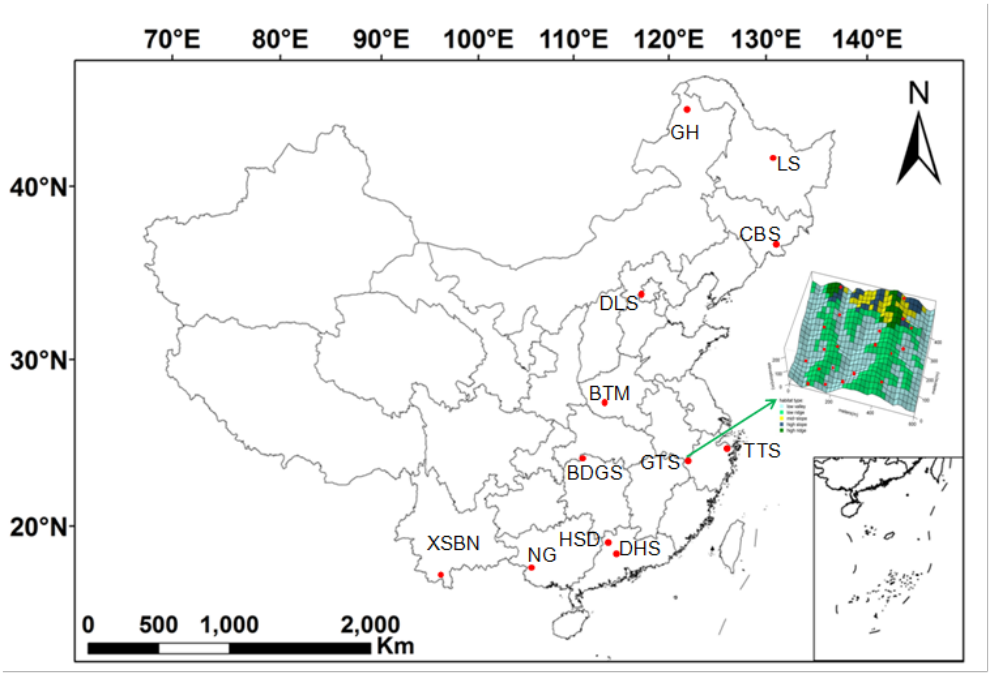

Figure S1. Geographic distribution of sampling sites in a map of China, which was slightly modified with an inset map based on Ji et al. (2019). Temperate forests include Genhe (GH), Liangshui (LS), Changbaishan (CBS), Donglingshan (DLS), and Baotianman (BTM); subtropical forests include Gutianshan (GTS), Badagongshan (BDGS), Tiantongshan (TTS), Heishiding (HSD), and Dinghushan (DHS); tropical forests include Nonggang (NG) and Xishuangbanna (XSBN). The inset color map shows the distribution of 20 quadrats in the 24-ha plot of GTS.

Fig. S2

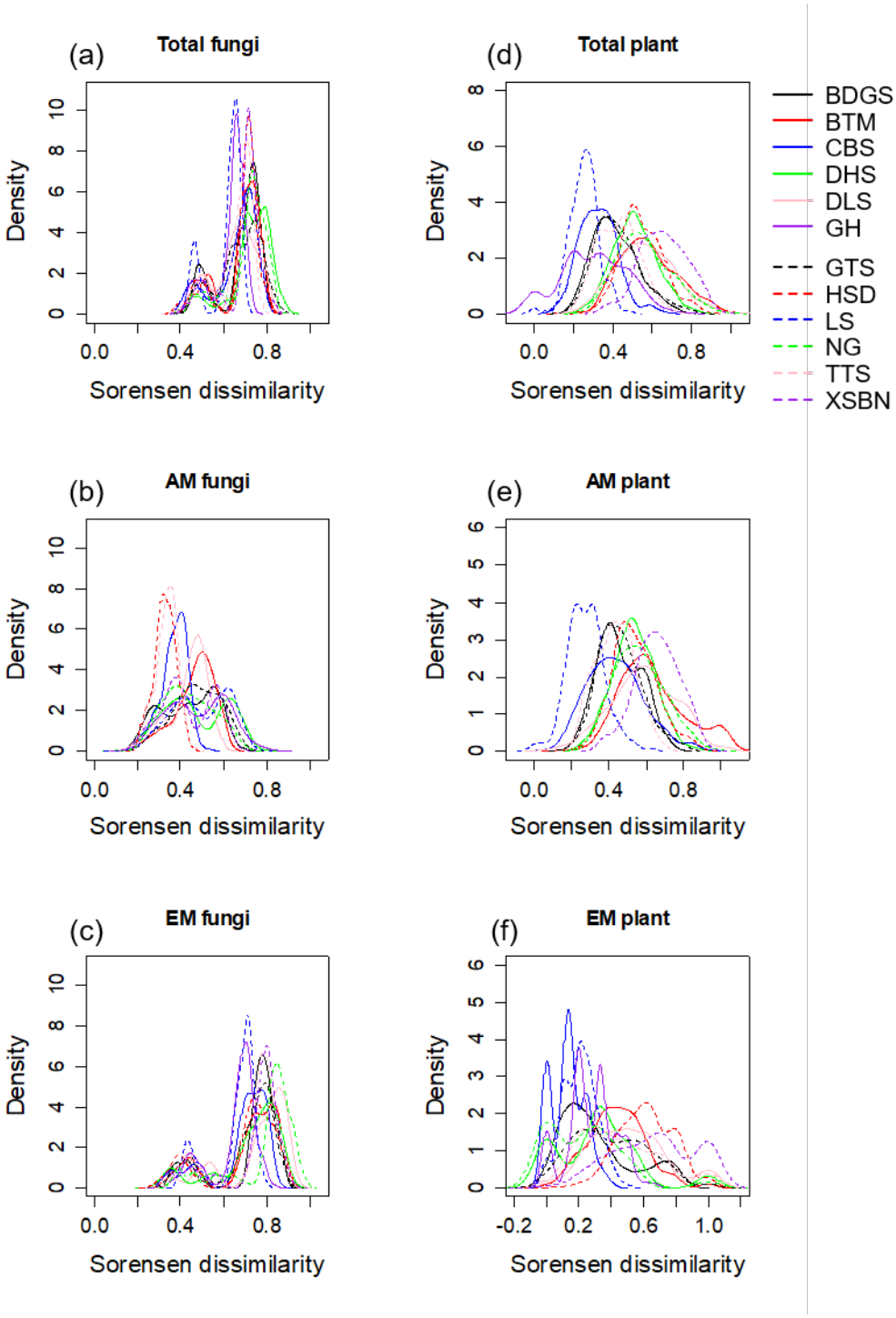

Figure S2. Distributions of Sørensen dissimilarities (β-diversity) of the observed communities of total fungi **(a)**, AM (arbuscular mycorrhizal) fungi **(b)**, EM (ectomycorrhizal) fungi **(c)**, total plant **(d)**, AM plant **(e)**, and EM plant **(f)** across twelve forests.

Fig. S3

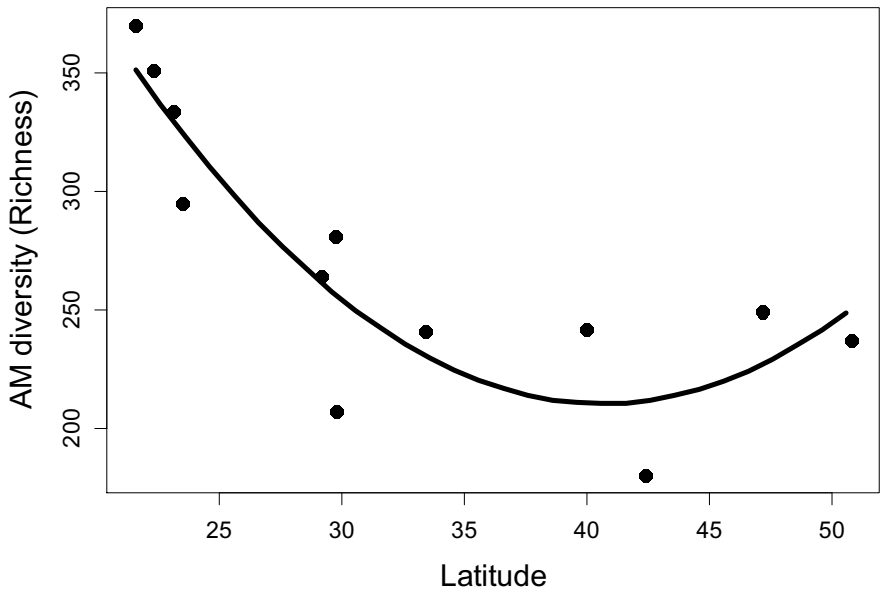

Figure S3. A significantly relationship was observed between the arbuscular mycorrhizal (AM) fungal diversity (OTU richness) and latitude. The parameters of this strong relationships were: AIC = 118.7; $R^2_{adj} = 0.7631$; $F = 18.72$; $P < 0.001$.

Table S1.

The basic soil, plant, and climatic information of 12 forest sites (content supplement based on Table 1 in Zheng *et al.* 2020).

Site	Soil factors						Plant factors			Climatic factors		
	pH	TC[b] (g kg^{-1})	TN (g kg^{-1})	TP (g kg^{-1})	C:N	N:P	Forest types[c]	Basal area (×1000 cm^2)	Richness	Climate zone	MAT (^{o}C)	MAP (mm)
BDGS	4.21±0.04f [a]	11.39±0.422a	0.796±0.019a	0.778±0.046cd	14.28±0.329def	1.080±0.058c	SEF	20.48±1.243ab	45.35±1.390a	Subtropic	11.5	2105
BTM	4.88±0.07cd	6.233±0.508cd	0.444±0.041d	0.536±0.050de	14.44±0.384de	0.900±0.117cd	WTDF	15.52±0.876bcd	24.10±2.531de	Temperate	15.1	886
CBS	5.48±0.05b	7.950±0.529bc	0.652±0.044abc	1.162±0.053ab	12.26±0.188fg	0.553±0.022d	TPF	17.71±1.169ab	11.40±0.600f	Temperate	3.6	700
DHS	3.78±0.01g	4.078±0.248def	0.290±0.015e	0.156±0.007f	13.98±0.272defg	1.894±0.102b	SEF	10.58±0.645cde	26.65±2.045cd	Subtropic	20.9	1929
DLS	6.71±0.05a	7.104±0.581c	0.536±0.042bcd	0.838±0.052c	13.26±0.213defg	0.633±0.023cd	WTDF	9.770±0.521de	10.10±0.680fg	Temperate	4.8	550
GH	4.93±0.07c	11.93±1.364a	0.507±0.051cd	0.921±0.047bc	23.50±0.830a	0.572±0.070d	CTCF	14.43±1.692bcde	3.150±0.274g	Temperate	-5.3	450
GTS	4.68±0.04de	4.034±0.254def	0.208±0.016e	0.136±0.018f	19.79±0.450b	1.948±0.224b	SEF	17.10±1.326bc	36.55±2.222b	Subtropic	15.3	1964
HSD	4.12±0.03f	3.056±0.188ef	0.231±0.010e	0.078±0.003f	13.08±0.353efg	3.014±0.114a	SEF	16.30±1.328bcd	45.80±2.022a	Subtropic	19.6	1744
LS	5.64±0.08b	11.80±0.561a	0.703±0.038a	1.026±0.060abc	17.24±0.799c	0.702±0.031cd	TPF	24.07±3.426a	17.10±0.695ef	Temperate	-0.3	676
NG	6.54±0.05a	5.327±0.174cde	0.549±0.014bcd	1.210±0.166a	9.696±0.136h	0.575±0.059d	TRF	8.552±0.429e	32.60±1.645bc	Tropic	22	1500
TTS	3.78±0.05g	10.37±0.964ab	0.667±0.047ab	0.446±0.022e	15.25±0.367cd	1.545±0.121b	MEDF	14.58±0.833bcde	30.90±1.174bcd	Subtropic	16.2	1375
XSBN	4.51±0.04e	2.285±0.099f	0.187±0.005e	0.252±0.013ef	12.19±0.278g	0.782±0.049cd	TRF	15.13±1.343bcde	47.65±2.833a	Tropic	21.8	1493

[a] Values (mean ± SE) in the same column without shared letters denote significant difference at $P < 0.05$, as indicated by Tukey's HSD test. [b] TC, total carbon content; TN, total nitrogen content; TP, total phosphorus content; C:N, ratio of TC to TN; N:P, ratio of TN to TP; MAT, mean annual temperature; MAP, mean annual precipitation. [c] Forest types: TRF, tropical rain forests (XSBN and NG); SEF, subtropical evergreen broad-leaf forests (DHS, HSD, GTS, and BDGS); MEDF, mixed evergreen broad-leaved and deciduous broad-leaved forest (TTS); WTDF, warm-temperate deciduous broad-leaved forests (BTM and DLS); TPF, temperate broad-leaved Korean pine forests (CBS and LS); CTCF, cold temperate monsoon coniferous forest (GH).

Table S2.

The plant community composition data (available at the link below).

https://www.doi.org/10.6084/m9.figshare.13543046

Table S3.

The raw data of environmental factors including soil, plant basal area and richness, climatic parameters, and sampling time information (available at the link below).

https://www.doi.org/10.6084/m9.figshare.13543046

Table S4.

Primer sets and PCR conditions used to amplify fungal DNA prior to Illumina sequencing.

Target	Nested PCR	Primer	Sequence (5'–3')	Thermal condition	Reference
Fungi (incl. EM fungi)	First PCR	ITS1F	CTTGGTCATTTAGAGGAAGTAA	95°C for 5 min, 30 cycles of at 94°C for 50 s, 52°C for 1 min, 68°C for 1 min.	White et al., 1990
		ITS4	TCCTCCGCTTATTGATATGC		Gardes & Bruns, 1993
	Second PCR	ITS5	GGAAGTAAAAGTCGTAACAAGG	95°C for 3 min, 30 cycles of at 94°C for 50 s, 52°C for 1 min, 68°C for 1 min.	White et al., 1990
		ITS2	GCTGCGTTCTTCATCGATGC		White et al., 1990
AM fungi	First PCR	GeoA2	CCAGTAGTCATATGCTTGTCTC	95°C for 5 min, 30 cycles of at 94°C for 1 min, 58°C for 50 s, 68°C for 1 min.	Schwarzott & Schüβler, 2001
		AML2	GAACCCAAACACTTTGGTTTCC		Lee et al., 2008
	Second PCR	NS31	TTGGAGGGCAAGTCTGGTGCC	95°C for 3 min, 30 cycles of at 94°C for 1 min, 58°C for 50 s, 68°C for 1 min.	Simon et al., 1992
		AMDGR	CCCAACTATCCCTATTAATCAT		Sato et al., 2005

Note: AM, arbuscular mycorrhizal; EM, ectomycorrhizal.

Table S5.

The fungal community composition data (available at the link below).

https://www.doi.org/10.6084/m9.figshare.13543046

Table S6.

The values of standardized effect size (SES) associated with median community dissimilarities for the observed total, arbuscular mycorrhizal (AM), and ectomycorrhizal (EM) fungal and plant (in parentheses). β-diversities are presented as the mean of the estimate relative to those of 100 simulated communities for each of the 12 locations.

Zone	**Site**	**SES value**[a]			
		Total fungi (plant)	AM fungi (plant)	EM fungi (plant)	Comparison (plant)
Subtropic	BDGS	227.2 (8.513)[b]	27.87 (5.660)	5.373 (-1.621)	H-M-L (h-m-l)[c]
Temperate	BTM	295.9 (2.787)	5.592 (1.816)	36.18 (-0.052)	H-L-M (h-m-l)
Temperate	CBS	7.881 (0.023)	23.35 (0.152)	1.090 (-1.210)	M-H-L (m-h-l)
Subtropic	DHS	260.2 (1.557)	95.31 (1.139)	61.30 (n.a.)	H-M-L (h-l-n.a.)
Temperate	DLS	34.38 (0.345)	7.183 (0.364)	4.680 (1.812)	H-M-L (l-m-h)
Temperate	GH	19.27 (-0.245)	197.4 (n.a.)	6.519 (-0.506)	M-H-L (h-n.a.-l)
Subtropic	GTS	49.28 (2.063)	87.38 (2.320)	5.739 (0.048)	M-H-L (m-h-l)
Subtropic	HSD	53.85 (1.959)	67.43 (3.141)	3.232 (0.846)	M-H-L (m-h-l)
Temperate	LS	56.00 (4.164)	185.8 (-0.337)	6.412 (0.516)	M-H-L (h-l-m)
Tropic	NG	217.0 (7.319)	4.617 (5.976)	2.859 (n.a.)	H-M-L (h-l-n.a.)
Subtropic	TTS	131.8 (19.56)	68.66 (13.32)	7.148 (n.a.)	H-M-L (h-l-n.a.)
Tropic	XSBN	197.7 (24.93)	55.34 (22.55)	7.174 (1.954)	H-M-L (h-m-l)
Fungi	ANOVA[d]	Df = 2; F = 7.922; P = 0.002			
	Mean[e]	129.2±30.17A	68.83±18.89AB	12.31±5.177B	H-M-L
Plant	Kruskal-Wallis test[d]	χ^2 = 8.530, P = 0.014			
	Mean[e]	6.082±2.339A	5.099±2.105AB	0.199±0.410B	h-m-l

[a] The SES values were calculated according to the formula as '(observed estimate−mean of the simulated estimates) / standard deviation of the simulated estimates' using the Sørensen dissimilarities of total, AM, and EM fungal and plant community data.

[b] Consistently positive effect size (SES value > 0) indicates that the estimate is higher than predicted (divergence) under the neutral hypothesis.

[c] Letters of 'H (h)', 'M (m)', and 'L (l)' denote the SES values is high, medium, and low, respectively, following the order of total, AM, and EM fungi and plants in the same row.

[d] The outcomes of one-way ANOVA or nonparametric Kruskal-Wallis test analyses on the effect of site on the SES values amongst different fungal and plant groups.

[e] Values (mean ± SE) in that row without shared capital letters denote significant difference amongst fungal and plant groups at $P < 0.05$, as indicated by Tukey's HSD test.

Table S7.

The estimated parameters associated with the neutral model of biodiversity fit at the level of Chinese forest soil for total, AM, and EM fungal and plant (within parentheses) communities.

Site	**Theta (Θ)**			***I* (median)**		
	Total fungi (plant)	AM fungi (plant)[a]	EM fungi (plant)	Total fungi (plant)	AM fungi (plant)	EM fungi (plant)
BDGS	16.72 (22.83)	13.76 (18.35)	12.57 (3.39)	4.12E+12 (128)	672 (110.3)	18 (3.40)
BTM	20.40 (16.51)	22.70 (15.38)	7.52 (4.14)	3.64E+12 (29)	26 (20.9)	21377 (3.91)
CBS	50.06 (6.19)	12.33 (4.66)	29.57 (1.68)	89 (15.5)	191 (6.80)	13 (6.72)
DHS	12.85 (23.42)	9.08 (21.84)	4.29 (n.a.)	5.11E+12 (30.5)	4537 (24.1)	2.6E+12 (n.a.)
DLS	34.85 (8.59)	19.29 (4.58)	14.24 (1.91)	501 (4.53)	46 (3.10)	26.5 (6.41)
GH	35.56 (1.80)	6.46 (n.a.)[b]	13.93 (1.31)	275 (1.46)	2.5E+12 (n.a.)	46.5 (1.37)
GTS	25.08 (24.16)	6.86 (16.57)	12.39 (3.13)	8189 (40.25)	3E+12 (33.9)	24 (3.47)
HSD	32.31 (47.12)	8.97 (35.20)	16.76 (9.91)	868 (48.44)	3346 (51.6)	14 (4.46)
LS	31.29 (6.65)	8.31 (7.07)	15.46 (1.50)	1016 (56.23)	10764 (13.8)	51 (54.5)
NG	16.72 (21.13)	20.46 (20.79)	16.98 (n.a.)	3.80E+12 (40.59)	108 (37.5)	23.5 (n.a.)
TTS	16.26 (12.72)	10.37 (10.34)	10.88 (n.a.)	4.69E+12 (491)	1727 (313)	48.5 (n.a.)
XSBN	20.40 (19.14)	11.78 (16.98)	12.82 (3.03)	3.1E+12 (5.4E+11)	2251 (4.55E+11)	26 (4.02)

[a] AM, arbuscular mycorrhizal; EM, ectomycorrhizal.

[b] n.a., Result was not available because of the limited plant community data in these sites.

Table S8.

The observed β-diversity (Sørensen index) of the total, arbuscular mycorrhizal fungi (AM), and ectomycorrhizal (EM) fungal and plant (within parentheses) communities at each of 12 forest sites.

Sequence	Sites	Total fungi (plant)	AM fungi (plant)	EM fungi (plant)
1	BDGS	0.338 (0.352)	0.233 (0.371)	0.603 (0.203)
2	BTM	0.493 (0.520)	0.480 (0.552)	0.571 (0.396)
3	CBS	0.453 (0.266)	0.365 (0.346)	0.588 (0.107)
4	DHS	0.399 (0.499)	0.194 (0.510)	0.410 (0.303)
5	DLS	0.405 (0.499)	0.396 (0.454)	0.538 (0.434)
6	GH	0.345 (0.258)	0.365 (n.a.)	0.435 (0.201)
7	GTS	0.364 (0.396)	0.112 (0.390)	0.505 (0.352)
8	HSD	0.434 (0.530)	0.132 (0.512)	0.588 (0.642)
9	LS	0.272 (0.215)	0.443 (0.272)	0.434 (0.115)
10	NG	0.390 (0.543)	0.291 (0.545)	0.539 (0.330)
11	TTS	0.287 (0.354)	0.156 (0.357)	0.474 (0.403)
12	XSBN	0.357 (0.482)	0.269 (0.468)	0.548 (0.549)
Fungi	ANOVA[a]	Df = 2; F = 20.62; P < 0.0001		
	Mean	0.378±0.019B[b]	0.286±0.036C	0.519±0.019A
Plant	ANOVA[a]	Df = 2; F = 1.815; P = 0.179		
	Mean	0.410±0.034A	0.434±0.028A	0.336±0.047A

[a] The outcomes of one-way ANOVA analyses on the effect of site on the observed β-diversity amongst different fungal and plant groups.

[b] Values (mean ± SE) in that row without shared capital letters denote significant difference in observed β-diversity amongst fungal and plant groups at $P < 0.05$, as indicated by Tukey's HSD test.

Table S9.

The observed β-diversity (Sørensen index) of the total, arbuscular mycorrhizal (AM), and ectomycorrhizal (EM) fungal and plant communities across tropic, subtropic, and temperate climate zones.

Type		**Tropic**		**Subtropic**		**Temperate**	
		Mean[a]	Median[a]	Mean	Median	Mean	Median
Fungi	Total	0.712	0.734	0.463	0.437	0.770	0.818
	AM	0.754	0.774	0.526	0.574	0.792	0.826
	EM	0.716	0.744	0.514	0.533	0.765	0.807
	ANOVA[b]	Df = 2; F = 115; P < 0.0001					
	Mean	0.739 ± 0.010B[c]		0.508 ± 0.020C		0.796 ± 0.010A	
Plant	Total	0.800	0.930	0.828	0.921	0.824	0.961
	AM	0.801	0.927	0.830	0.922	0.799	0.875
	EM	0.757	0.990	0.854	1.000	0.783	1.000
	ANOVA[b]	Df = 2; F = 0.281; P = 0.764					
	Mean	0.786 ± 0.015A[c]		0.837 ± 0.008A		0.802 ± 0.012A	

[a] The values were shown as the mean (median) of the Sørensen index within each climate zone for each plant group.

[b] The outcomes of one-way ANOVA analyses on the fungal and plant observed β-diversity amongst three different climate zones.

[c] Values (mean ± SE) in that row with shared capital letters denote non-significant difference amongst three climate zones at P < 0.05, as indicated by Tukey's HSD test.

Table S10.

Detailed PerMANOVA outcomes using all data focusing on the effects (*i.e.*, predictors) of climate zone (n = 240) and site (n = 240) and re-analyzed to reveal the site effect for each three climate zones of tropic (n = 40), subtropic (n = 100), and temperate regions (n = 100). The analyses were conducted for total, arbuscular mycorrhizal (AM), and ectomycorrhizal (EM) fungi and plant communities individually.

Effect	**Df**	**Total fungi (plant)**		**AM fungi (plant)[a]**		**EM fungi (plant)[a]**	
		R^2	*P* value	R^2	*P* value	R^2	*P* value
Climate zone	2	0.095 (0.143)	0.001 (0.001)	0.280 (0.157)	0.001 (0.001)	0.069 (0.130)	0.001 (0.001)
Site (All data)	11	0.262 (0.572)	0.001 (0.001)	0.490 (0.540)	0.001 (0.001)	0.207 (0.598)	0.001 (0.001)
Site (Tropic)	1	0.086 (0.359)	0.001 (0.001)	0.266 (0.361)	0.001 (0.001)	0.064 (0.327)	0.003 (0.001)
Site (Subtropic)	4	0.222 (0.492)	0.001 (0.001)	0.403 (0.480)	0.001 (0.001)	0.172 (0.528)	0.001 (0.001)
Site (Temperate)	4	0.183 (0.562)	0.001 (0.001)	0.247 (0.465)	0.001 (0.001)	0.156 (0.620)	0.001 (0.001)