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Research Article

Effects of tree mycorrhizal type on soil respiration and carbon stock via fine root biomass and litter dynamic in tropical plantations

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Abstract

Tropical forests are among the most productive and vulnerable ecosystems in the planet. Several global forestation programs are aiming to plant millions of trees in tropical regions in the future decade. Mycorrhizal associations are known to largely influence forest soil carbon (C) stocks. However, to date, little is known on whether and how different tree mycorrhizal types affect soil respiration (Rs) and C stocks in tropical forests. In this study, we used a three-decade tropical common garden experiment, with three arbuscular mycorrhizal (AM) and three ectomycorrhizal (EM) monocultures, to investigate the impacts of tree mycorrhizal type on Rs and soil C stocks. Associating biotic (e.g. root biomass, litter dynamic, soil microbes) and abiotic factors (e.g. microclimate) were also measured. Our results showed that AM stands supported significantly higher Rs and soil C stock, litter turnover rate and fine root biomass than EM stands. Further statistical analysis displayed that tree mycorrhizal type was the most important factor in regulating Rs and soil C stock compared with other biotic or abiotic factors. Moreover, we found that mycorrhizal type directly and indirectly affected Rs and soil C stocks via fine root biomass and litter dynamic, i.e. litter production, litter standing crop and litter turnover rate. Our findings highlight important effects of tree mycorrhizal type on forest C cycle, suggesting that planting AM tree species could contribute to promotion of soil C stock in tropical ecosystems.

Keywords mycorrhizal, soil respiration, tropical forests, carbon storage, afforestation

树种菌根类型通过细根生物量和凋落物动态影响土壤呼吸和土壤碳储存

摘要：热带森林是高生产力但同时也是脆弱的生态系统之一。一些全球范围的造林项目计划未来十年在热带地区种植数百万棵树。树种菌根类型影响森林土壤碳储存已成为共识，但在热带地区，树种菌根类型如何影响土壤呼吸(Rs)和碳储存，目前仍知之甚少。为研究树种菌根类型对Rs和土壤碳储存的影响，本实验在一个近30年热带同质园的3种丛枝菌根(AM)树种和3种外生菌根(EM)树种的单种林中，测量了其Rs和表层20 cm的碳含量，以及有关的生物因子(如根生物量、凋落物动态、土壤微生物)和非生物因子(如微气候)。研究结果表明，AM单种林的Rs、土壤碳含量，以及凋落物周转速率和细根生物量显著高于EM单种林。分析表明，与其他生物和非生物因子比较，树种菌根类型对Rs和土壤碳含量的影响最大。进一步分析表明，菌根类型是通过细根生物量和凋落物动态(凋落物产量、凋落物现存量、凋落物周转速率)直接和间接影响Rs和土壤碳含量。本研究结果强调了树种菌根类型对森林碳循环的影响，表明在热带地区种植AM树种可能比种植EM树种更能促进土壤固碳。

关键词：菌根，土壤呼吸，热带森林，碳储存，造林

INTRODUCTION

During the United Nations decade on ecosystem restoration (2021–2030), millions of trees are expected to be planted across the globe, aiming to help mitigating the negative impacts of rising atmospheric CO_2 concentration. In nature, more than 90% of terrestrial plant species on Earth associate with mycorrhizal fungi, which could influence ecosystem structure and functions (Brundrett and Tedersoo 2018). Meanwhile, tree mycorrhizal association is considered an important regulator of soil C stocks from ecosystem to global scales (Averill *et al.* 2014). Therefore, understanding the effect of tree mycorrhizal type on forest C cycle and the associated mechanisms is crucial to predict the feedback between C cycle and global climate change (Terrer *et al.* 2016; Wang *et al.* 2022) or forest managements (Asmelash *et al.* 2016).

Arbuscular (AM) and ectomycorrhizal (EM) fungi are the two dominant types of mycorrhizal fungi that are closely associated with tree species, differing in nutrient use strategies (Phillips *et al.* 2013) and plant traits (Cornelissen *et al.* 2001), which may drive differences in belowground C cycling (Keller and Phillips 2019). For example, EM tree species generally produce leaf litter of relative lower quality (Cornelissen *et al.* 2001) and have less root biomass (Withington *et al.* 2006) than AM trees. It has also been demonstrated that differences in photosynthesis and belowground C allocations between AM and EM tree species may affect C input and then root respiration (Metcalfe *et al.* 2011). Meanwhile, EM fungi can mine nitrogen (N) from soil organic matter (SOM) by producing extracellular enzymes, while AM fungi mainly acquire inorganic N by competing with free-living decomposer microbes in soil (Smith and Smith 2011; Talbot *et al.* 2008), likely affecting soil microbial respiration. These results suggest that tree mycorrhizal type may influence both root and microbial respiration, two key important components of soil respiration (Rs) (Dariah *et al.* 2014; Kuzyakov 2006), and consequently alter ecosystems C cycle.

Despite the fact that numerous individual studies had been conducted, how tree mycorrhizal type affects soil C stocks and Rs in forest ecosystems remains controversial. Previous work has suggested that EM forests could store more soil C due to higher soil C/N ratio than AM ones (Averill *et al.* 2014). However, according to a promising framework associating with litter decomposition and soil organic carbon (SOC) stabilization processes (Cotrufo *et al.* 2013), AM forests with more labile litter may accumulate more C than EM ones. A recent study conducted in temperate forests found higher C stock in topsoil of EM-trees-dominating stands but higher C storage in subsoil of AM-trees-dominating stands (Craig *et al.* 2018). Likewise, EM soils were considered to output less C due to N competition between EM fungi and free-living soil microbes than AM ones, which may inhibit SOC decomposition (Averill and Hawkes 2016; Gadgil and Gadgil 1971). However, some field studies reported higher Rs in AM than EM forests (Lang *et al.* 2020; Wang and Wang 2018), and no significant difference between AM and EM forests (Lin *et al.* 2017). Different climate, soil and vegetation conditions in diverse ecosystems may obscure the effect of tree mycorrhizal type on forest C cycle. The common garden experiment may be a better way to

eliminate these confounding effects (Vesterdal *et al.* 2012). Therefore, a long-term experiment at the same place with diverse tree species is necessary to investigate the effects of mycorrhizal type on soil C stocks and Rs in forest ecosystems.

Since most studies have been conducted in temperate forests to examine effects of tree mycorrhizal type on ecosystem C cycle (Craig *et al.* 2018; Lang *et al.* 2020; Wang and Wang 2018), tropical forests are less explored (Lin *et al.* 2017). Tropical forests are an important reservoir of soil C stocks (Townsend *et al.* 2011). Changes in Rs and soil C stock of tropical forests would have profound effects on regional and global climate dynamics. As multispecies afforestation programs have significantly changed the trees species distribution in tropical regions over the past decades (Poorter *et al.* 2015), several global forestation programs are considering to plant millions more trees in tropical regions, potentially altering the tropical C storage potential. As such, understanding the effects of tree mycorrhizal type on Rs and soil C stock will help select appropriate tree species for the tropical afforestation and forestation, and improve model performance in predicting the tropical forest C cycle.

To determine how tree mycorrhizal type affects Rs and soil C stocks, we conducted a 3-year field experiment within a 30-year-old tropical common garden experiment in Xishuangbanna located in Southwest China. Specifically, we aimed to answer the following questions: (i) whether Rs and soil C stock differ between soils beneath AM and EM tree species in tropical monocultures; and (ii) how fine root biomass and litter dynamics interact to determine the mycorrhizal effects.

MATERIALS AND METHODS

Site description and experiment design

This study was conducted in Xishuangbanna Tropical Botanical Garden (XTBG) of Chinese Academy of Science (21.68° N, 101.42° E, 570 m a.s.l.), located in Yunnan Province, southwest China. The climate is tropical monsoon with two contrasted periods (a rainy season from May to October, and a dry season from November to April). The mean annual temperature is 21.6 °C, with monthly air temperature ranging from 16.4°C in January to 25.9°C in May. The mean annual precipitation is 1476.4 mm, of which about 85% occurs in the rainy season, while in the dry season the foggy days compensate for the lower rainfall. Dozens of tropical tree species were planted as monocultures in this common garden area in 1980s, which was established on the same tropical forest land with same original vegetation and soil properties. The area is flat and is about 7 ha. The soil is sandy alluvium, with pH ranging from 4.5 to 5.5 (Zhang *et al.* 2015). The bulk density of the study region ranges from 1.30 to 1.44 g cm^{-3} (Cao *et al.* 2009). Those plantations had similar management history. Organic fertilizers were applied to the stump area when seedlings were planted. After then, nearly no agrochemicals were used (Cao *et al.* 2009). The litter layer on the soil surface was left intact. The canopies were nearly closed, thus almost no understory vegetation existed.

In 2013, three evergreen AM tree species (*Mesua ferrea*, *Metadina trichotoma* and *Artocarpus heterophyllus*) and three evergreen EM ones (*Vatica xishuangbannaensis*, *Parashorea chinensis* and *Dipterocarpus retusus*) were selected, with stand ages from 24 to 32 years, mean diameter at 1.3 m breast height from 18.4 to 24.3 cm (Supplementary Table S1), and tree densities about 1100 trees ha^{-1}. Each tree species had only one monoculture (>400 m^2), which was set as one replicate of the tree mycorrhizal type. Within each monoculture, three 5 m × 5 m plots were selected for Rs measurements and soil sampling. The six monoculture stands were close to each other (maximum distance ca. 250 m). Considering the similar soil substrate, soil texture, climate, stand age and forest management, the differences in soil properties are assumed to be primarily caused by the influence of tree species. This experimental design is comparable to a common garden experiment in spite of the limited number of replicates per tree species (Lin *et al.* 2018; Lorenz and Thiele-Bruhn 2019).

Rs measurements

To measure Rs, two PVC collars (20 cm in inner diameter and 11 cm in height) were installed in each plot in May 2013 (i.e. six collars in each monoculture). Each PVC collar had twenty-four 8-mm holes distributed evenly in the pipe wall below the ground level to allow root growth. Small living plants inside collars were removed manually at least 24 h prior to Rs measurement (Zhou *et al.* 2020). Rs was measured once or twice a month between 9:00 am and 13:00 pm (local time) from August 2013 to August 2016, using the LI-8100 automated soil CO_2 flux system (LI-COR Inc., Lincoln, NE, USA). Soil temperature and water volumetric content at 5 cm

depth were measured adjacent to each collar with auxiliary digital temperature and moisture probes of the LI-8100 system and recorded automatically.

Soil pH, organic carbon and microbial community measurement

Top mineral soil cores (0–10 cm; 7 cm diameter) were collected by five-point method (at the center and four corners to avoid the tree stump area) in each plot in May 2014. Five soil cores were pooled together and treated as one sample, and there were three samples for each monoculture. About 1 kg soil were quartered from each sample and were sieved (2 mm) after removing roots and rocks. About 800 g of each soil sample were air-dried. Subsamples were ground to determine pH and SOC content. Soil pH was measured by 1:2.5 soil:water ratio with Acidity Detector (PHS-3C, INESA Scientific Instrument Co., Ltd, Shanghai, China). SOC was determined by dichromate oxidation method.

Soil microbial community composition was determined by phospholipid fatty acids (PLFAs) method. Specifically, subsamples of about 200 g from each soil sample were transported in ice boxes to the laboratory within 4 h, freeze-dried at −60 °C after removing plant detritus, and then stored at −20 °C for further PLFA determination. All samples were dried and then analyzed by gas chromatography (GC) following trans-esterification for quantitative analysis relative to an internal standard. The GC conditions were set by the MIDI Sherlock program (MIDI, Inc. Newark, DE). Bacterial fatty acid standards and the software SHERLOCK version 6.2 were used to identify the peaks. The areas measured by gas chromatography-flame ionization detector (GC-FID) were used to calculate the abundance of PLFA markers (in nmol PLFA g^{-1} dry soil). Total lipid abundance was calculated as the sum of lipids with chain length from C10 to C20. This value was used as an indicator of total microbial biomass (e.g. Huang *et al.* 2014). Specifically, the terminal-branched saturated PLFA peaks 15:0, 17:0, i14:0, i15:0, i16:0, i17:0, a15:0, a17:0, 16:1ω 7c, 16:1ω 9c, 18:1ω 5c, 18:1ω 7c, cy17:0, cy19:0, 15:0 3OH and 16:1 2OH were used as markers for bacterial (Wang *et al.* 2016). The PLFAs 18:2ω6, 9 and C18:1ω9 were used as an indicator of fungi (Kaiser *et al.* 2010).

Litter dynamic and fine root biomass measurements

Three 0.5 m × 0.5 m traps with 1.0 mm size in each plot were established for litterfall measurement, which were collected twice a month in rainy season and once a month in dry season from August 2013 to August 2016. The litter traps with 1.0 mm mesh gauze net were uniformly fixed at 1.0 m height above the ground to evenly reduce the rainfall. Litterfall was further sorted into leaves, twigs and reproductive parts (i.e. flowers, fruits and seeds), and then oven-dried at 75 °C for 48 h to reach a constant mass and weighed. Litter standing crop on forest floor was collected in three 1 m × 1 m subplots randomly set in each plot per 3 months from August 2013 to August 2016. Litter was then sorted into flowers, fruits and seeds and were oven-dried as litterfall. We estimated litter turnover rate (k), using k = annual litter production/litter standing crop on forest floor (Tang *et al.* 2010).

Fine root biomass (<2 mm diameters) was determined using soil core method in May 2014 when Rs was measured. Soil cores (n = 5) were randomly collected in each plot, approximately 1.5 m apart from the nearest tree using a soil corer (inner diameter 7 cm) at depths of 0–20 cm. Visible fine roots were hand-picked and all attached residues, including soil, dead roots, stem materials and litter fractions, were carefully removed with tweezers. The fine root was oven-dried at 65 °C for 48 h to reach a constant mass and weighed.

Data analysis

The normality of distribution of all data was examined with Shapiro–Wilk's test, while homogeneity of variance was tested with Bartlett test. Field measurement data of Rs were logarithmic transformed to achieve linearity and homoscedasticity. The linear mixed model in *R* package *lmerTest* was used to examine the effects of mycorrhizal type, monitoring year and their interaction on Rs, soil temperature and water content, with tree species set as the random effect.

To estimate annual Rs of each tree monoculture based on the filed measurements, linear mixed models were used to develop empirical models (Supplementary Table S3), which related Rs to soil temperature and water content, with collars within each plot set as the random effect (Wang *et al.* 2006). Annual Rs of each monoculture was calculated using the following equation:

$$\ln \mathrm{Rs} = a + b \times \mathrm{Ts} + c \times \mathrm{Ws} + d \times \mathrm{Ts} \times \mathrm{Ws} \quad (1)$$

where ln is natural logarithm; Ts and Ws are soil temperature and soil volumetric water content at 5 cm depth, respectively; parameters a, b, c and d

are regression coefficients. Insignificant terms were removed by a backward elimination procedure. Since only the daily dataset in 2014 could be accessed (Supplementary Fig. S1), annual Rs was only estimated for 2014.

Considering that most of the variables were heteroscedastic, the Kruskal–Wallis test were used to test the effects of tree mycorrhizal type on concerned variables at species level using package '*agricolae*' in *R*. Relationships among variables were analyzed with Pearson correlation and Spearman's rank correlation. The relative importance of each variable was quantified with variable importance of projection calculated from partial least squared regression (PLSR) using '*pls*' package in *R* (Chong and Jun 2005). PLSR can manage the multicollinearity among independent variables and can be applied with relative large number of independent variables in small datasets (Ekblad *et al.* 2005; Trogisch *et al.* 2016). The hypothesized models of tree mycorrhizal type effects on Rs and SOC were analyzed using the piecewise structural equation model (PSEM) in *R* package '*piecewiseSEM*', which can be used with hierarchical data and small datasets (Lefcheck 2016; Shipley 2000). All statistical analysis were conducted with a significance level of 0.05 using *R* statistical software (R Core Team 2020).

RESULTS

Effects of tree mycorrhizal type on microclimate

Soil temperature at 5 cm depth showed significant seasonal variations for all six monocultures, with the higher values in summer but the lower ones in winter (Fig. 1b). Similarly, the temporal dynamics of water volumetric content also illustrated distinct seasonal patterns, with the high values in rainy season and low ones in dry season (Fig. 1c). However, no significant difference was found for soil temperature and water volumetric content between AM and EM monocultures (Table 1).

Tree mycorrhizal type drives carbon cycling in long-term tropical plantations

Tree mycorrhizal type significantly affected Rs in these plantations (Table 1). Specifically, the annual mean Rs of AM monocultures (141.4 mol m^{-2} yr^{-1}) was significantly higher than that of EM ones (78.1 mol m^{-2} yr^{-1}, $P < 0.05$, Fig. 2b). Among three AM monocultures, the Rs was the highest under *A. heterophyllus* stand, with 21.2% and 33.0% greater than *M. trichotoma* and *M. ferrea*, respectively (Fig. 2a). However, there was no significant difference for annual Rs among three EM monocultures (Fig. 2a). In addition, neither stand age (Supplementary Table S2) nor plot basal area (Supplementary Table S4) had significant effect on Rs. Seasonal patterns of Rs were also detected over the experimental period, with the high values in summer and the low values in winter, which were associated with changes in soil temperature (Fig. 1).

Similarly, litter dynamic and soil properties were also significantly influenced by tree mycorrhizal type in these plantations. Our results showed that both litter production and litter standing crop of EM monocultures were significantly higher than those of AM ones (Fig. 3a and b). However, the litter turnover rate and fine root biomass of AM monocultures were greater than those of EM ones (Fig. 3c and d). AM stands tended to have a higher SOC, with 34.2% greater than those in EM ones (Fig. 3e). Similarly, soil pH and microbial biomass in AM stands were 20.5% and 103% greater than those in EM stands, respectively (Fig. 3f and g). However, no significant difference was found for fungi/bacteria ratio between EM and AM stands (Fig. 3h).

SOC, soil pH and microbial biomass displayed significant positive correlations with litter turnover, while a negative correlation was found between fungi/bacteria and litter turnover rate (Fig. 4). Similarly, SOC, soil pH and microbial biomass were positively correlated with fine root biomass, but there was no significant correlation between fungi/bacteria ratio and fine root biomass. Fine root biomass and litter turnover rate in all stands were positively correlated with Rs (Fig. 5). However, both litter production and litter standing crop exhibited significantly negative correlations with Rs. Meanwhile, Rs was significantly correlated with SOC, pH and microbial biomass, but there was no correlation between Rs and fungi/bacteria (Fig. 5; Supplementary Table S5).

Linking litter dynamic, fine root biomass to the effects of tree mycorhizal type on Rs and C stocks

Our analysis showed that tree mycorrhizal type was the most important factor in accounting for the variance of Rs compared with microbial biomass, SOC, fine root biomass, litter dynamics and soil pH (Fig. 6a; Supplementary Fig. S2). The PSEM analysis further showed that AM stands had a larger direct effect on Rs (β = 0.63, standardized coefficient), and comparative indirect effects through fine root biomass (β = 0.41) and litter dynamics (β = 0.21 for

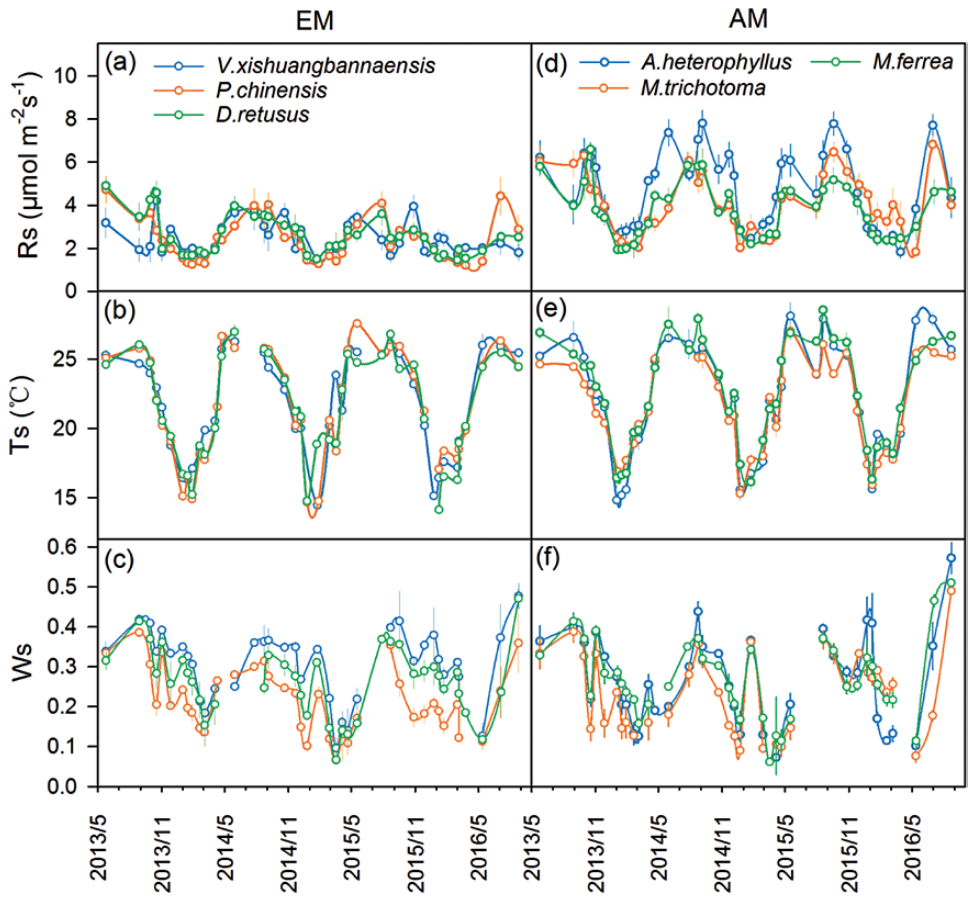

Figure 1: Field-measured Rs (**a**, **d**), soil temperature (Ts, **b**, **e**) and soil volumetric water content (Ws, **c**, **f**) between AM and EM monocultures in 3 years. Error bars denote standard deviations. Notes that six tree species include *Vatica xishuangbannaensis*, *Parashorea chinensis*, *Dipterocarpus retusus*, *Mesua ferrea*, *Metadina trichotoma* and *Artocarpus heterophyllus*.

Table 1: Effects of tree mycorrhizal type (Myc), monitoring year (Year) and their interactions (Myc × Year) on Rs, soil temperature (Ts) and water content (Ws) in 3 years (2013/5–2014/4, 2014/5–2015/4, 2015/5–2016/4) among tree species monocultures

	lnRs		Ts		Ws	
	Estimate ± SE	***P***	**Estimate ± SE**	***P***	**Estimate ± SE**	***P***
Intercept	1.32 ± 0.07	<0.001	21.35 ± 0.45	<0.001	0.27 ± 0.02	<0.001
Myc_EM	−0.51 ± 0.09	<0.001	−1.18 ± 0.63	0.07	0.02 ± 0.03	0.47
Year	0.01 ± 0.02	0.51	0.20 ± 0.20	0.31	−0.002 ± 0.01	0.62
Myc × Year	−0.03 ± 0.03	0.32	0.17 ± 0.27	0.53	−0.01 ± 0.01	0.07

Tested by linear mixed models, with tree species as the replicates.

litter turnover and $\beta = 0.18$ for forest floor, Fig. 6b). Tree mycorrhizal type also directly and indirectly affected SOC through litter turnover and fine root biomass (Supplementary Fig. S3). It was worth noting that SOC was excluded from the best PSEM model, indicating fine root biomass, litter dynamics and tree mycorrhizal type were more primary driving factors for Rs. Taking together, a schematic diagram took shape and showed that litter dynamics, fine root biomass, soil C stocks and Rs were significantly different between AM and EM stands in tropical forests (Fig. 7). AM trees generally produce high-quality litter with fast turnover and large amounts of fine roots, leading to high soil microbial biomass, SOM content and Rs. In contrast, EM trees tend to have larger amount but low-quality litter with

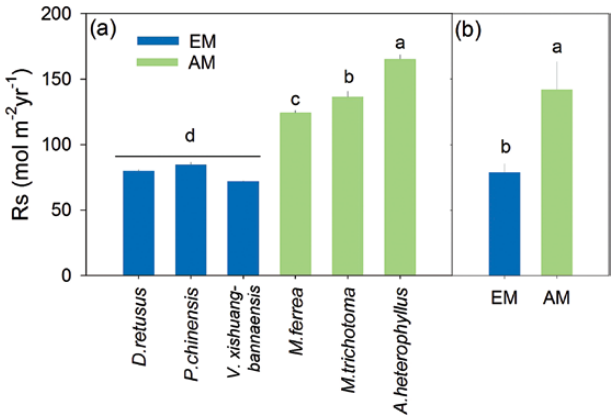

Figure 2: Annual Rs of six monocultures (**a**) and grouped by the mycorrhizal type (**b**). Error bars denote standard deviations. Symbols a, b, c and d represent the significant differences of Rs on species (a) or mycorrhizal group levels (b). Notes that six tree species include *Vatica xishuangbannaensis*, *Parashorea chinensis*, *Dipterocarpus retusus*, *Mesua ferrea*, *Metadina trichotoma* and *Artocarpus heterophyllus*.

slow turnover and less fine root biomass, leading to relatively less soil microbial biomass, SOM content and smaller Rs than AM trees.

DISCUSSION

Understanding the effect of tree mycorrhizal type on C cycling and the associated potential mechanisms is crucial to predict the feedback between C cycle and climate change (Averill and Hawkes 2016; Zhou *et al.* 2017). In this study, we found AM tree monocultures stored higher SOC than EM ones in tropical plantations (Fig. 3e). Our result was consistent with a meta-analysis which found higher mineral soil C concentration in AM than EM stands in tropical forests and suggests that the pattern might largely result from the differences of litter quality, decomposition dynamics and the conversion of floor C into mineral soil C (Lin *et al.* 2017). In contrast, Averill *et al.* (2014) observed that EM forests could contain more soil C than AM ones at the global scale due to higher soil carbon/nitrogen ratio (C/N) in EM than AM forests. However, a latter synthesis study in temperate forests found no significant differences in top-layer soil C between AM and EM forests but higher soil N content in AM than EM forests (Zhu *et al.* 2018), suggesting that the higher soil C/N in EM-dominated temperate forests was primary driven by lower N content rather than larger C stocks. A field study further showed that EM-trees-dominating stands contained more C in topsoil, while AM-trees-dominating stands had

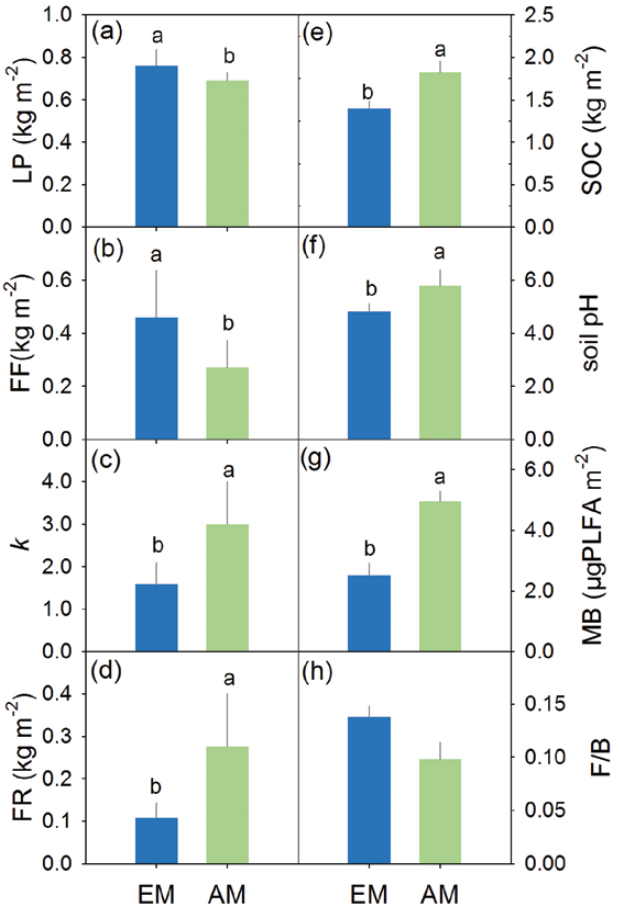

Figure 3: Litter production (LP, **a**), forest floor (FF, **b**), litter turnover rate (*k*, **c**) and fine root biomass (FR, **d**), SOC content (**e**), soil pH (**f**), soil microbial biomass (MB, **g**) and fungi/bacterial ratios (F/B, **h**) at top 20 cm depth in AM and EM monocultures. Error bars denote standard deviations. Symbols a and b represent significant difference of corresponding factors between AM and EM stands.

more C in subsoil by accelerating the production of microbial residues in three temperate forests (Craig *et al.* 2018), indicating that the soil C stock pattern may vary among biomes.

Our further analysis showed that SOC was driven by the tree mycorrhizal type through litter turnover rate and fine root biomass (Supplementary Fig. S3). Previous studies found that fast turnover of fine roots induced higher C input in soil (Luke McCormack *et al.* 2012), and greater root biomass enhanced root exudation of organic compounds into soil (Wurzburger *et al.* 2017). Root exudation and the glomalin-related soil-protein produced by AM fungi promote the formation of soil microaggregates, which contributes to soil C storage (Leifheit *et al.* 2014; Rillig 2004). All these findings support the microbial efficiency-matrix stabilization framework (Cotrufo *et al.* 2013) that, in fast decomposition systems, SOM

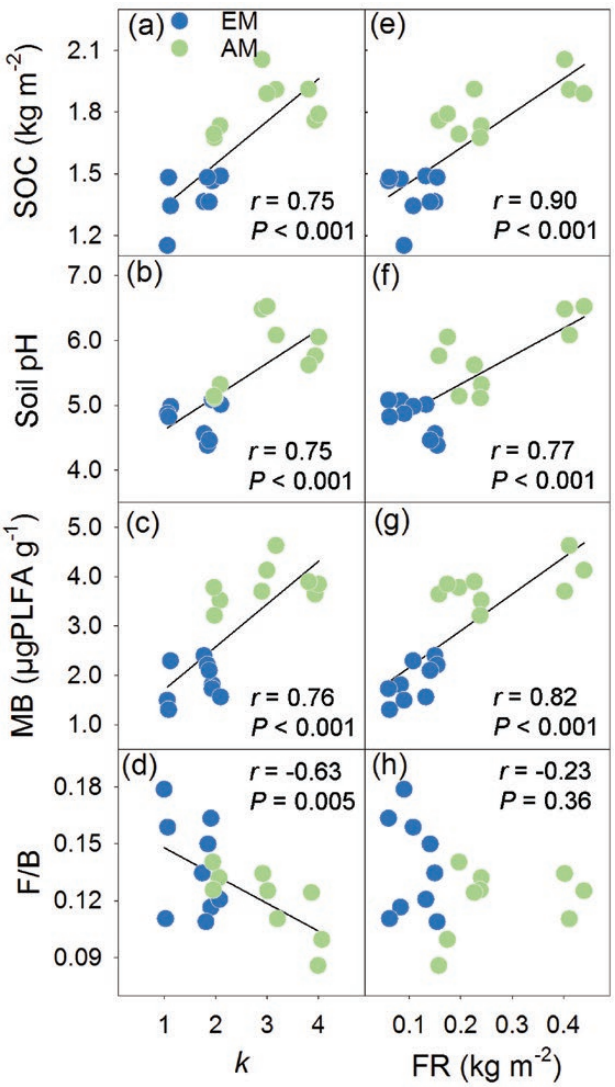

Figure 4: Relationships between SOC (**a**, **e**), pH (**b**, **f**), microbial biomass (MB, **c**, **g**), fungi/bacterial ratio (F/B, **d**, **h**) and litter turnover (*k*) and fine root biomass (FR).

can be stabilized more by the production of microbial residues (Fig. 3c). In contrast, the competition between EM plants and free-living soil microbes may inhibit the decomposition of SOC and thus benefit soil C storage (Averill *et al.* 2014). However, soil C stock can also be diminished in EM stands since EM fungi are able to mine nitrogen from SOM by producing extracellular enzymes (Lindahl and Tunlid 2014). Hence, the effect of tree mycorrhizal type on soil C stocks may associate with a suite of physiological traits of plants and mycorrhizal fungi, which deserves to explore in future experiments.

The tree mycorrhizal type would also affect soil C efflux (Metcalfe *et al.* 2011). Our results displayed that Rs in AM monocultures was higher than those of EM ones (Fig. 2). A meta-analysis detected no overall significant difference of Rs between AM- and EM-trees-dominating stands across biomes but found Rs increased with the dominating of AM tree species (Lin *et al.* 2017). Large variations of climate, soil properties and vegetation conditions (e.g. stand age, community structure) at the global scale could confound the mycorrhizal effect on Rs (Wang and Wang 2018). Several studies had tried to reduce the confounding effects. A microcosm experiment with litter and soils collected from AM and EM stands (Taylor *et al.* 2016) and a mesocosms experiment with seedlings (Wurzburger *et al.* 2017) both reported higher Rs in AM than EM treatments. A recent survey study in northern hardwood forest and a common garden experiment with five 12-year-old monocultures also found that Rs was greater in AM- than EM-trees-dominated stands, although the different soil matrix might confound the impacts of tree mycorrhizal type to some degree (Lang *et al.* 2020; Wang and Wang 2018). Our common garden experiment with mature monocultures showed no significant differences in microclimate (Table 1), which confirmed the pattern that AM stands held higher Rs than EM ones.

Similar to SOC storage, the effect of tree mycorrhizal type on Rs was primarily driven by fine root biomass and litter dynamic rather than soil properties and soil microbes (Fig. 6b), since soil properties and microbes were regulated by fine roots and litter dynamic (Supplementary Fig. S3). Previous frameworks had linked plant traits with soil C process (Metcalfe *et al.* 2011; Wardle *et al.* 2004). Greater biomass and faster turnover of fine root, and higher-quality litter provide more substrate and stimulate soil microbes, thus promoting Rs (Figs 3 and 5; Luo and Zhou 2006). Our results showed strong linkages between mycorrhizal type and plant traits. AM forests are well known to produce litter with higher quality than EM forests (Fig. 3c; Lin *et al.* 2017) and harbor greater fine root biomass (Fig. 3d), turnover (Withington *et al.* 2006) and/or surface area (Lang *et al.* 2020). Therefore, a linkage between the tree mycorrhizal type and soil C process may emerge. Furthermore, the effect of mycorrhizal type on Rs via plant traits may result from the contrasting nutrient economy between AM and EM trees (Phillips *et al.* 2013). In short, since AM fungi usually lacks saprotrophic abilities but has well-developed hyphae that can rapidly take up inorganic nitrogen (Smith and Smith 2011), AM trees may primarily utilize inorganic nitrogen forms, accompanying with high-quality litter and fast turnover that leads to relative larger Rs. In contrast, EM trees tend to an organic nutrient economy, since EM fungi can excavate nitrogen and

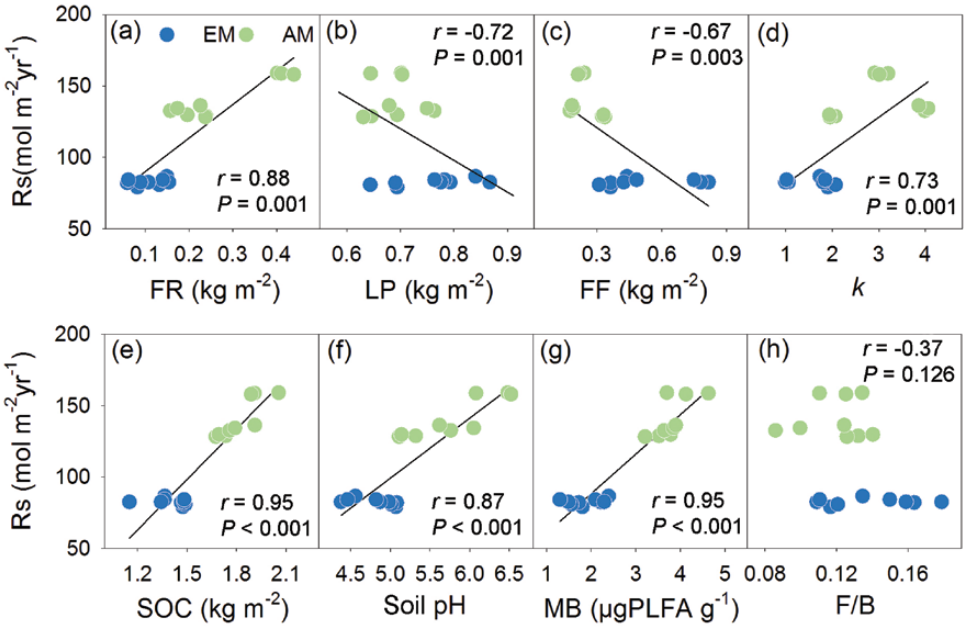

Figure 5: Relationships between Rs and fine root biomass (FR, **a**), litter production (LP, **b**), forest floor (FF, **c**), litter turnover (*k*, **d**), SOC concentration (**e**), soil pH (**f**), soil microbial biomass (MB, **g**) and fungi/bacterial ratio (F/B, **h**).

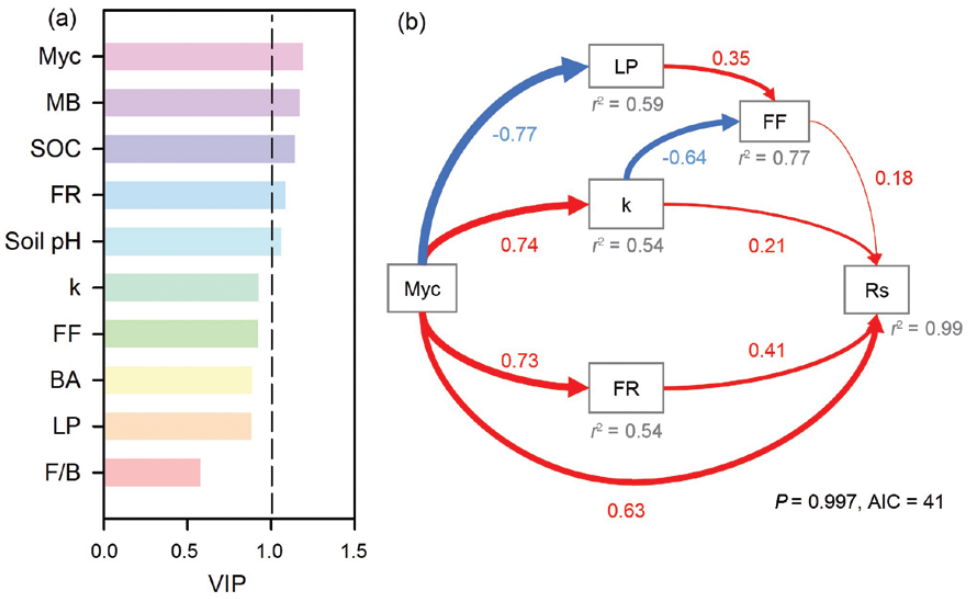

Figure 6: (**a**) Variable importance for projection (VIP) of all measured factors for Rs obtained by PLSR model (R^2 = 0.96). VIP > 1 indicates that the corresponding factor significantly influenced Rs. (**b**) Direct and indirect effects of tree mycorrhizal type (Myc) on Rs through fine root biomass (FR) and litter dynamic (including litter production [LP], forest floor [FF] and litter turnover [*k*]) illustrated by the final PSEM. Significant positive and negative relationships between nodes in PSEM are shown in red and blue, respectively. Positive paths from Myc in PSEM indicate a significant greater effect of arbuscular- over ectomycorrhizal trees. Standardized path coefficients in PSEM are labeled near the arrows, and their magnitudes are indicated by the thickness of arrows. Abbreviations: BA = plot basal area, F/B = fungi/bacterial ratio, MB = microbial biomass.

phosphorus from organic matter (Lindahl and Tunlid 2014), which matches the relatively low-quality litter and slow turnover that contribute to relatively small Rs. However, the linkage between the nutrient economy of mycorrhizal fungi and forest C cycle needs more research.

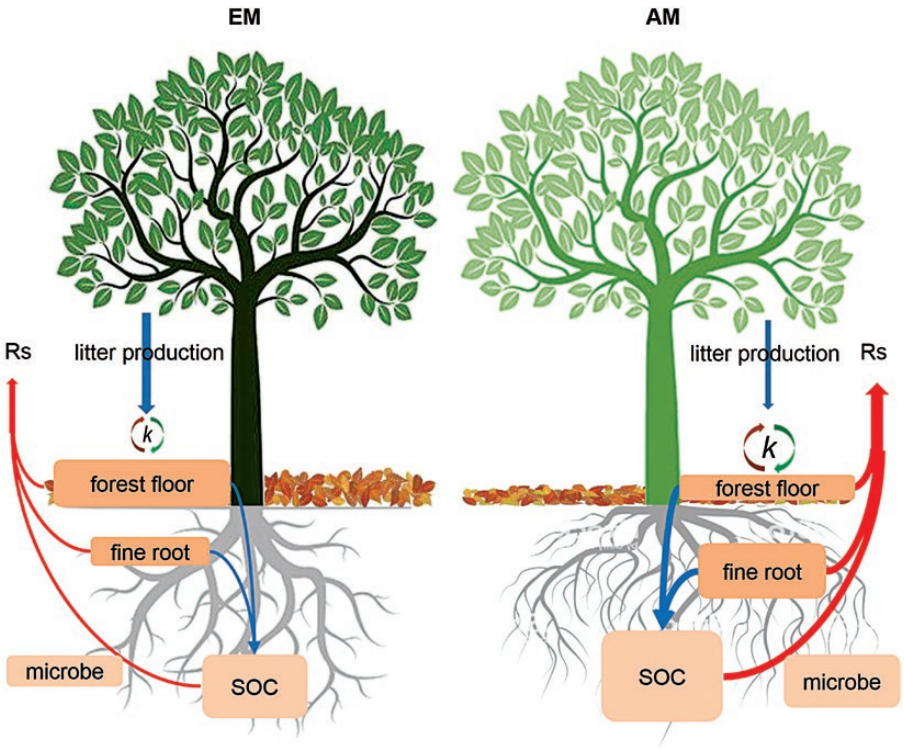

Figure 7: Scheme diagram of tree mycorrhizal type effect on Rs via fine root and litter dynamic in EM (left) and AM (right) stands. Blue and red arrows indicate input and output carbon fluxes, respectively. Boxes indicate carbon pools. Thickness of arrows and areas of boxes represent relative magnitudes of carbon fluxes and pools between EM and AM stands, respectively. Abbreviation: k = litter turnover rate.

Our finding from a long-term common garden experiment in tropical plantations may provide insight as to how tree mycorrhizal type affect soil C processes. Thus, our study will offer some suggestions for modeling and experimental studies on ecosystem C cycle in the future, at least in three aspects. First, mycorrhizal influences on soil C storage and Rs via fine root and litter turnover could be incorporated into Earth system models to improve their prediction of forest C dynamics (Meyer *et al.* 2010). Second, as the distribution of AM trees are expected to expand under climate change (Barceló *et al.* 2019; Kivlin *et al.* 2011), and the growth of some invasive plants was found to benefit from AM (Song *et al.* 2021; Kong *et al.* 2022), the shift of community composition from EM to AM plants might mitigate the influence of CO_2 rising to some extent, possibly resulting in negative feedbacks between climate change and the C cycle, which deserves to be further investigated. Third, fine root turnover is crucial to soil C accumulation and Rs (Luo and Zhou 2006), but it was not measured in this study constrained by the experimental condition. Future studies should measure both the input and output C fluxes to accurately quantify the differences of the C cycle between AM and EM forests.

In summary, with an approximate three-decade common garden experiment, our results highlight the importance of tree mycorrhizal type on Rs and soil C storage in tropical plantations, indicating that planting AM tree species may promote soil C stock in tropical zone. We further found that effects of tree mycorrhizal type on Rs and soil C stock were mainly associated with contrasting fine root biomass and litter dynamics between AM and EM monocultures. This finding supports the linkages among the mycorrhizal association, plant traits and soil C process, suggesting the necessity of integrating the mycorrhizal-associated nutrient economy (Phillips *et al.* 2013) and the microbial efficiency-matrix stabilization processes (Cotrufo *et al.* 2013). These results may also provide some insights to the development of future Earth system models and field experiments, promoting tropical plantation managements.

Supplementary Material

Supplementary material is available at *Journal of Plant Ecology* online.

Table S1: Tree species, mycorrhizal associations (Myc), stand age, diameter at breast height (DBH) of

arbuscular mycorrhizal- (AM) and ectomycorrhizal-associated (EM) monocultures.
Table S2: Effects of tree mycorrhizal type (Myc), stand ages (age) and their interactions (Myc × age) on soil respiration (Rs) and soil organic carbon content (SOC).
Table S3: Models of soil respiration (ln Rs) against soil temperature (Ts) and water volumetric content (Ws) at 5 cm depth of six monocultures.
Table S4: Effects of tree mycorrhizal type (Myc), plot basal area (BA) and their interaction (Myc × BA) on annual soil respiration.
Table S5: Spearman's rank correlation coefficients among factors on plot level.
Figure S1: Daily soil temperature (Ts), daily water volumetric content (Ws) at 5 cm depth and daily precipitation recorded at the local weather station in 2014.
Figure S2: Variation partitioning for the soil respiration.
Figure S3: The piecewise structural equation model of mechanisms of tree mycorrhizal type effect (Myc) on soil respiration (Rs) considering soil organic carbon content (SOC).

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Conflict of interest statement. The authors declare that they have no conflict of interest.

Authors' Contributions

G. Zhang and X. Zhou designed, and oversaw the research in consultation with other coauthors. G. Zhang and G. Zhou synthesized data, conducted experiment and wrote manuscript. M. Delgado-Baquerizo discussed and edited the manuscript. L. Zhou, J. Shao, R. Liu, J. Gao, Y. He, H. Liu, Z. Du and J. Tang conducted experiment together.

REFERENCES

Asmelash F, Bekele T, Birhane E (2016) The potential role of arbuscular mycorrhizal fungi in the restoration of degraded lands. *Front Microbiol* **7**:1095.

Averill C, Hawkes CV (2016) Ectomycorrhizal fungi slow soil carbon cycling. *Ecol Lett* **19**:937–947.

Averill C, Turner BL, Finzi AC (2014) Mycorrhiza-mediated competition between plants and decomposers drives soil carbon storage. *Nature* **505**:543–545.

Barceló M, van Bodegom PM, Soudzilovskaia NA (2019) Climate drives the spatial distribution of mycorrhizal host plants in terrestrial ecosystems. *J Ecol* **107**:2564–2573.

Brundrett MC, Tedersoo L (2018) Evolutionary history of mycorrhizal symbioses and global host plant diversity. *New Phytol* **220**:1108–1115.

Cao JX, Liu WJ, Sha LQ (2009) Spatial heterogeneity of soil physical and chemical properties in Xishuangbanna tropical botanical garden. *J Yunnan Univ Nat Sci Ed* **31**:325–330.

Chong IG, Jun CH (2005) Performance of some variable selection methods when multicollinearity is present. *Chemometr Intell Lab Syst* **78**:103–112.

Cornelissen JHC, Aerts R, Cerabolini B, *et al*. (2001) Carbon cycling traits of plant species are linked with mycorrhizal strategy. *Oecologia* **129**:611–619.

Cotrufo MF, Wallenstein MD, Boot CM, *et al*. (2013) The microbial efficiency-matrix stabilization (MEMS) framework integrates plant litter decomposition with soil organic matter stabilization: do labile plant inputs form stable soil organic matter? *Glob Change Biol* **19**:988–995.

Craig ME, Turner BL, Liang C, *et al*. (2018) Tree mycorrhizal type predicts within-site variability in the storage and distribution of soil organic matter. *Glob Change Biol* **24**:3317–3330.

Dariah A, Marwanto S, Agus F (2014) Root- and peat-based CO_2 emissions from oil palm plantations. *Mitig Adapt Strateg Glob Change* **19**:831–843.

Ekblad A, Bostrom B, Holm A, *et al*. (2005) Forest soil respiration rate and delta C-13 is regulated by recent above ground weather conditions. *Oecologia* **143**:136–142.

Gadgil RL, Gadgil PD (1971) Mycorrhiza and litter decomposition. *Nature* **233**:133.

Huang XM, Liu SR, Wang H, *et al*. (2014) Changes of soil microbial biomass carbon and community composition through mixing nitrogen-fixing species with *Eucalyptus urophylla* in subtropical China. *Soil Biol Biochem* **73**:42–48.

Kaiser C, Frank A, Wild B, *et al*. (2010) Negligible contribution from roots to soil-borne phospholipid fatty acid fungal biomarkers 18:2ω6,9 and 18:1ω9. *Soil Biol Biochem* **42**:1650–1652.

Keller AB, Phillips RP (2019) Leaf litter decay rates differ between mycorrhizal groups in temperate, but not tropical, forests. *New Phytol* **222**:556–564.

Kivlin SN, Hawkes CV, Treseder KK (2011) Global diversity and distribution of arbuscular mycorrhizal fungi. *Soil Biol Biochem* **43**:2294–2303.

Kong LJ, Chen X, Yerger EH, *et al*. (2022) Arbuscular mycorrhizal fungi enhance the growth of the exotic species *Ambrosia artemisiifolia*. *J Plant Ecol* **15**:581–595.

Kuzyakov Y (2006) Sources of CO_2 efflux from soil and review of partitioning methods. *Soil Biol Biochem* **38**:425–448.

Lang AK, Jevon FV, Ayres MP, *et al*. (2020) Higher soil respiration rate beneath arbuscular mycorrhizal trees in

a northern hardwood forest is driven by associated soil properties. *Ecosystems* **23**:1243–1253.

Lefcheck JS (2016) Piecewisesem: piecewise structural equation modelling in R for ecology, evolution, and systematics. *Methods Ecol Evol* **7**:573–579.

Leifheit EF, Veresoglou SD, Lehmann A, *et al*. (2014) Multiple factors influence the role of arbuscular mycorrhizal fungi in soil aggregation—a meta-analysis. *Plant Soil* **374**:523–537.

Lin GG, Guo DL, Li L, *et al*. (2018) Contrasting effects of ectomycorrhizal and arbuscular mycorrhizal tropical tree species on soil nitrogen cycling: the potential mechanisms and corresponding adaptive strategies. *Oikos* **127**:518–530.

Lin GG, Mccormack ML, Ma CE, *et al*. (2017) Similar below-ground carbon cycling dynamics but contrasting modes of nitrogen cycling between arbuscular mycorrhizal and ectomycorrhizal forests. *New Phytol* **213**:1440–1451.

Lindahl B, Tunlid A (2014) Ectomycorrhizal fungi—potential organic matter decomposers, yet not saprotrophs. *New Phytol* **205**:1443–1447.

Lorenz M, Thiele-Bruhn S (2019) Tree species affect soil organic matter stocks and stoichiometry in interaction with soil microbiota. *Geoderma* **353**:35–46.

Luke McCormack M, Adams TS, Smithwick EAH, *et al*. (2012) Predicting fine root lifespan from plant functional traits in temperate trees. *New Phytol* **195**:823–831.

Luo YQ, Zhou XH (2006) *Soil Respiration and the Environment.* San Diego, CA: Academic Press.

Metcalfe DB, Fisher RA, Wardle DA (2011) Plant communities as drivers of soil respiration: pathways, mechanisms, and significance for global change. *Biogeosciences* **8**:2047–2061.

Meyer A, Grote R, Polle A, *et al*. (2010) Simulating mycorrhiza contribution to forest C- and N cycling—the MYCOFON model. *Plant Soil* **327**:493–517.

Phillips RP, Brzostek E, Midgley MG (2013) The mycorrhizal-associated nutrient economy: a new framework for predicting carbon-nutrient couplings in temperate forests. *New Phytol* **199**:41–51.

Poorter L, van der Sande MT, Thompson J, *et al*. (2015) Diversity enhances carbon storage in tropical forests. *Glob Ecol Biogeogr* **24**:1314–1328.

R Core Team (2020) *R: A Language and Environment for Statistical Computing*. Vienna, Austria: R Foundation for Statistical Computing. https://www.r-project.org/ (26 April 2022, date last accessed).

Rillig M (2004) Arbuscular mycorrhizae, glomalin, and soil aggregation. *Can J Soil Sci* **84**:355–363.

Shipley B (2000) A new inferential test for path models based on directed acyclic graphs. *Struct Equ Model* **7**:206–218.

Smith SE, Smith FA (2011) Roles of arbuscular mycorrhizas in plant nutrition and growth: new paradigms from cellular to ecosystem scales. *Annu Rev Plant Biol* **62**:227–250.

Song DL, Zhao YF, Tang FP, *et al*. (2021) Effects of arbuscular mycorrhizal fungi on *Solidago canadensis* growth are independent of nitrogen form. *J Plant Ecol* **14**:648–661.

Talbot J, Allison SD, Treseder K (2008) Belowground responses to climate change decomposers in disguise: mycorrhizal fungi as regulators of soil C dynamics in ecosystems under global change. *Funct Ecol* **22**:955–963.

Tang JW, Cao M, Zhang JH, *et al*. (2010) Litterfall production, decomposition and nutrient use efficiency varies with tropical forest types in Xishuangbanna, SW China: a 10-year study. *Plant Soil* **335**:271–288.

Taylor MK, Lankau RA, Wurzburger N (2016) Mycorrhizal associations of trees have different indirect effects on organic matter decomposition. *J Ecol* **104**:1576–1584.

Terrer C, Vicca S, Hungate BA, *et al*. (2016) Mycorrhizal association as a primary control of the CO_2 fertilization effect. *Science* **353**:72–74.

Townsend AR, Cleveland CC, Houlton BZ, *et al*. (2011) Multi-element regulation of the tropical forest carbon cycle. *Front Ecol Environ* **9**:9–17.

Trogisch S, He JS, Hector A, *et al*. (2016) Impact of species diversity, stand age and environmental factors on leaf litter decomposition in subtropical forests in China. *Plant Soil* **400**:337–350.

Vesterdal L, Elberling B, Christiansen JR, *et al*. (2012) Soil respiration and rates of soil carbon turnover differ among six common European tree species. *For Ecol Manag* **264**:185–196.

Wang XQ, Wang CK (2018) Mycorrhizal associations differentiate soil respiration in five temperate monocultures in northeast China. *For Ecol Manag* **430**:78–85.

Wang XL, Wang XL, Zhang WX, *et al*. (2016) Invariant community structure of soil bacteria in subtropical coniferous and broadleaved forests. *Sci Rep* **6**:11.

Wang CK, Yang JY, Zhang QZ (2006) Soil respiration in six temperate forests in China. *Glob Change Biol* **12**:2103–2114.

Wang YH, Shao CL, Qiu YJ, *et al.* (2022) Arbuscular mycorrhizal fungi protect a subtropical tree species exposed to simulated acid rain by accelerating photosynthetic ability, antioxidant enzymes and osmolyte accumulation. *J Plant Ecol* **15**:1036–1048.

Wardle DA, Bardgett RD, Klironomos JN, *et al*. (2004) Ecological linkages between aboveground and belowground biota. *Science* **304**:1629–1633.

Withington JM, Reich PB, Oleksyn J, *et al*. (2006) Comparisons of structure and life span in roots and leaves among temperate trees. *Ecol Monogr* **76**:381–397.

Wurzburger N, Brookshire ENJ, McCormack ML, *et al*. (2017) Mycorrhizal fungi as drivers and modulators of terrestrial ecosystem processes. *New Phytol* **213**:996–999.

Zhang JL, Zhang SB, Chen YJ, *et al*. (2015) Nutrient resorption is associated with leaf vein density and growth performance of dipterocarp tree species. *J Ecol* **103**:541–549.

Zhou G, Zhou X, Liu R, *et al*. (2020) Soil fungi and fine root biomass mediate drought-induced reductions in soil respiration. *Funct Ecol* **34**:2634–2643.

Zhou G, Zhou X, Zhang T, *et al*. (2017) Biochar increased soil respiration in temperate forests but had no effects in subtropical forests. *For Ecol Manag* **405**:339–349.

Zhu K, McCormack ML, Lankau RA, *et al*. (2018) Association of ectomycorrhizal trees with high carbon-to-nitrogen ratio soils across temperate forests is driven by smaller nitrogen not larger carbon stocks. *J Ecol* **106**:524–535.