The below-ground carbon and nitrogen cycling patterns of different mycorrhizal forests on the eastern Qinghai-Tibetan Plateau

Miaomiao Zhang[1,2], Shun Liu[1,2], Miao Chen[1,2], Jian Chen[1,2], Xiangwen Cao[1,2], Gexi Xu[1,2], Hongshuang Xing[1,2], Feifan Li[1,2] and Zuomin Shi[1,2,3,4]

[1] Key Laboratory of Forest Ecology and Environment of National Forestry and Grassland Administration, Ecology and Nature Conservation Institute, Chinese Academy of Forestry, Beijing, China
[2] Miyaluo Research Station of Alpine Forest Ecosystem, Lixian County, Sichuan, China
[3] Co-Innovation Center for Sustainable Forestry in Southern China, Nanjing Forestry University, Nanjing, China
[4] Institute for Sustainable Plant Protection, National Research Council of Italy, Torino, Italy

ABSTRACT

Mycorrhizal fungi can form symbiotic associations with tree species, which not only play an important role in plant survival and growth, but also in soil carbon (C) and nitrogen (N) cycling. However, the understanding of differences in soil C and N cycling patterns among forests with different mycorrhizal types is still incomplete. In order to determine the similarities and differences of soil C and N cycling patterns in different mycorrhizal forest types, three primary forests dominated by ectomycorrhizal (EcM), arbuscular mycorrhizal (AM) and ericoid mycorrhizal (ErM) trees respectively were studied on the eastern Qinghai-Tibetan Plateau. Indicators associated with soil C and N cycling, including leaf litter quality, soil C and N contents, soil C and N fluxes, and soil microbial biomass C and N contents were measured in each mycorrhizal type forest. The results showed that leaf litter quality was significantly lower with high C:N ratio and lignin: N ratio in ErM forest than that in AM and EcM forests. Soil CO_2 flux (508.25 ± 65.51 mg m^{-2} h^{-1}) in AM forest was significantly higher than that in EcM forest (387.18 ± 56.19 mg m^{-2} h^{-1}) and ErM forest (177.87 ± 58.40 mg m^{-2} h^{-1}). Furthermore, soil inorganic N content was higher in the AM forest than that in EcM and ErM forests. Soil net N mineralization rate (-0.02 ± 0.03 mg kg^{-1} d^{-1}) was lower in ErM forest than that in EcM and AM forests. We speculated that AM and EcM forests were relatively characterized by rapid soil C cycling comparing to ErM forest. The soil N cycling in EcM and ErM forests were lower, implying they were 'organic' N nutrition patterns, and the pattern in ErM forest was more obvious.

Subjects Ecology, Soil Science, Biogeochemistry, Forestry
Keywords Soil C and N cycling, EcM forest, AM forest, ErM forest, Mycorrhizal association

Submitted 21 April 2022
Accepted 16 August 2022
Published 14 September 2022

Corresponding author
Zuomin Shi, shizm@caf.ac.cn

Academic editor
Leonardo Montagnani

Additional Information and Declarations can be found on page 11

DOI **10.7717/peerj.14028**

INTRODUCTION

Most angiosperms are associated with mycorrhizal fungi, which can increase their access to nutrients and water or enhance the stress resistant ability (*Cheeke et al., 2017*; *Tedersoo & Bahram, 2019*). Mycorrhizal fungi are generally divided into three major types according to

their anatomy and function, such as ectomycorrhizal (EcM), arbuscular mycorrhizal (AM) and ericoid mycorrhizal (ErM) fungi (*Read & Perez-Moreno, 2003*). Increasing evidence indicates that functional variations of EcM, AM and ErM fungi may drive soil C and nutrient cycling in varying degrees (*Phillips, Brzostek & Midgley, 2013*; *Tedersoo, Bahram & Zobel, 2020*).

The soils in EcM- and ErM-dominated ecosystems have higher C contents than in AM-dominated ecosystems due to the specific N uptake preference of EcM and ErM fungi (*Averill & Finzi, 2011*; *Cavagnaro, Barrios-Masias & Jackson, 2012*; *Adamczyk et al., 2016*). The effect of mycorrhizal types on soil C contents may be regulated by the chemical differences of plant litter associated with EcM, AM and ErM. Compared to AM plants, EcM and ErM plants have relatively lower growth rates as well as lower leaf litter quality, *i.e.*, higher C:N or lignin:N (*Taylor, Lankau & Wurzburger, 2016*; *Read, Leake & Perez-Moreno, 2004*). Therefore, the leaf litter decomposition rates of EcM and ErM plants are relatively slower than that of AM plants, resulting in increase of soil C content in EcM- and ErM-dominated ecosystems (*Lin et al., 2016*; *Wurzburger & Hendrick, 2009*). Furthermore, *Clemmensen et al. (2021)* indicated that ErM plants decomposed slower than EcM plants and tended to accumulate more soil organic matter. On the contrary, some studies declared that AM soil had more C and N in temperate broadleaved forest (*Craig et al., 2018*). That is possibly attributed to enhancing the mineral-associated organic matter (MAOM) by promoting production and stabilization of microbial residues in AM forest (*Rillig, 2004*; *Cotrufo et al., 2013*). For soil C fluxes, the significant differences in different mycorrhizal forests may be caused by the result of the interactions between mycorrhizal associations and microorganisms (*Shi, Wang & Liu, 2012*; *Hughes et al., 2008*; *Heinemeyer et al., 2007*). Mycorrhizal exudates could promote soil organic matter decomposition and thus increase soil CO_2 flux ('Rhizosphere priming hypothesis', *Brzostek et al., 2015*). Study also shows that mycorrhizal fungi could compete directly with free-living decomposers for organic N which decreasing soil CO_2 flux ('Gadgil effect', *Gadgil & Gadgil, 1971*). Furthermore, niche assembly indicates that mycorrhizal fungi have different ecological niches from other microorganisms and there are no obvious interactions between mycorrhizal fungi and other microorganisms, which do not affect soil CO_2 flux (*Lindahl et al., 2007*).

Compared to the complex differences of soil C cycling, the differences of soil N cycling among different mycorrhizal forests are systematic (*Wang, Wang & Zhang, 2017*). In general, AM fungi adapt to ecosystems with high inorganic N content and lack proteolytic ability, while EcM and ErM fungi adapt to ecosystems with low N cycling rate and rich organic matter (*Hobbie & Högberg, 2012*). Similarly, *Phillips, Brzostek & Midgley (2013)* proposed the mycorrhizal-associated nutrient economy (MANE) framework that AM forest associated with the inorganic N economy and EcM forest with organic N economy. The unique effects of different mycorrhizal trees on soil N cycling are attributed mainly to the changes of their related mycorrhizal fungi traits (*Read & Perez-Moreno, 2003*; *Averill, 2016*). Previous studies showed that EcM and ErM fungi could exudate extracellular enzymes to degrade recalcitrant organic nutrients that could not be absorbed by plants (*Orwin et al., 2011*; *Ward et al., 2021*) and ErM fungi had stronger degradation ability than EcM fungi (*Read & Perez-Moreno, 2003*). AM fungi seem to lack the ability to express C and

N degrading enzymes comparing to EcM and ErM fungi (*Tisserant et al., 2013*). However, AM fungi could increase the absorption of dissolved and inorganic nutrients by changing the traits (biomass and growth rate) of host plant root system (*Bennett & Groten, 2022*).

Different mycorrhizal fungi have great differences in species diversity, C demands (*Orwin et al., 2011*), and the ability of enzymes to access different forms of N (*Hobbie & Högberg, 2012*). Previous studies on the differences of soil C and N cycling patterns in different mycorrhizal ecosystems focused predominantly on the global scale (*Tedersoo & Bahram, 2019*; *Lin et al., 2016*), and a few studies were carried out on a small scale (*Cheeke et al., 2017*; *Taylor, Lankau & Wurzburger, 2016*). Additionally, most of these investigations focused mainly on EcM- and AM-dominated ecosystems (*Keller & Phillips, 2018*; *Fang et al., 2020*). Comparative study on soil C and N cycling patterns among EcM-, AM- and ErM-dominanted ecosystems is still incomplete.

The eastern Qinghai-Tibetan Plateau presents a unique natural environment which plays an important role in soil and water conservation, climate regulation and biodiversity protection (*Wang et al., 2007*). It is an important ecological barrier in the middle and upper reaches of the Yangtze River (*Chen, 2019*). In-depth explorations of the biogeochemical cycle of the forest ecosystems with different mycorrhizal types are essential to understand their ecological functions. To understand more the below-ground C and N cycling patterns of different mycorrhizal forests on the eastern Qinghai-Tibetan Plateau, key parameters related to soil C and N cycling (*Lin et al., 2016*) of three primary forests (including EcM, AM and ErM) were studied. Our hypotheses were: (1) AM forest had faster C cycling than EcM and ErM forests ; (2) AM forest had faster N cycling (soil C:N, soil net nitrification rate and soil net N mineralization rate) dominated by soil inorganic N (NH_4^+-N and NO_3^--N) forms, EcM and ErM forests relied more on soil organic N.

MATERIALS & METHODS

Study site

The study was conducted in the upper reaches of the Minjiang River, western Sichuan Province. This area is located in the outermost part of the fold belt on the eastern Qinghai-Tibetan Plateau (*Xu et al., 2021*). The average annual temperature is 2∼4 °C, the highest temperature in summer is 23.7 °C, and the lowest temperature in winter is −18.1 °C. Annual precipitation is 700∼1,000 mm and concentrated mainly in the growing season (*Chen, 2019*). The soil in this area is defined as mountain brown soil, mountain brown cinnamon soil and subalpine meadow soil according to the Chinese soil taxonomic classification (*Liu, 2010*; *Feng et al., 2017*; *Chen, 2019*).

Experimental design

Three primary forests with different mycorrhizal types under similar soil and climate conditions, including *Abies faxoniana* primary forest (EcM), *Cupressus chengiana* primary forest (AM) and *Rhododendron phaeochrysum* primary forest (ErM) were selected in June 2018. Eight 15 m × 15 m sample plots (≥90% the dominant tree species by basal area in each sample plot) for each forest type were randomly set. In each forest type, the distance

Table 1 The basic stand information of the three forests. Data in the table were mean ± standard error ($n = 8$ in each case).

Forest type	*Abies faxoniana* primary forest	*Cupressus chengiana* primary forest	*Rhododendron phaeochrysum* primary forest
Mycorrhizal type	EcM	AM	ErM
Geographical coordinate	31°35′N, 102°48′E	31°54′N, 102°2′E	31°53′N, 102°46′E
Elevation (m)	3,295	2,802	3,805
Stand DBH (cm)	34.87 ± 13.56	21.63 ± 1.88	4.36 ± 0.50
Stand height (m)	18.52 ± 1.14	8.16 ± 0.79	2.64 ± 0.16
Canopy density	0.90	0.90	0.95
Soil pH	5.48	6.70	5.01

between any two sample plots was more than 50 m. The basic stand information of each forest type was given in Table 1.

Soil and leaf litter sampling

Soil (0–10 cm) samples were collected from four corners and central position of each plot with a soil drill in July 2019. The five soil samples were mixed into a zipper storage bag and transported to laboratory in ice box within 3 h. Moreover, Five to six trees without diseases and pests in each plot were selected randomly. Leaf litter without decay or degradation under each tree was sampled (over 100 g) and mixed together to form one sample per plot (*Barajas-Guzmán & Alvarez-Sánchez, 2003*).

Soil N mineralization

Five polyvinyl chloride cores (PVC cores, 15 cm height and 5 cm diameter) were buried into depth of 10 cm in the vicinity of the soil sampling location in July 2019 (*Becker et al., 2015*). Top of the PVC core was covered with permeable plastic film and bottom was with gauze to segregate water and allowed gas flow (*Kong et al., 2019*). Soil samples in the PVC cores were taken out after *in situ* for one month and sent to the laboratory for measurements. Soil net N mineralization rate and soil net nitrification rate were quantified by the difference between NH_4^+-N and NO_3^--N per month (*Wang et al., 2017*).

Soil respiration

Soil respiration was measured by static chambers and the gas chromatography technique. In September 2018, three static chamber bottoms were installed in each sample plot. The polyvinyl chloride cores (PVC cores, 10 cm height and 25 cm diameter) were buried 5 cm underground and reduced soil compaction in this process. The PVC cores were hollow and their tops and bottoms were unwrapped (*Heinemeyer et al., 2007*; *Tomè et al., 2016*). The living plants in the polyvinyl chloride cores were removed to reduce the impact of surface vegetation on soil CO_2 flux (*Wang et al., 2010*). In July 2019, a PVC portable opaque chamber (cylindrical, 30 cm height and 25 cm diameter) fitted with a fan to mix the air was attached to bottom of static chamber (*Wu et al., 2018*). For gas collection, a rubber tube (20 cm length and 5 mm diameter) was attached to top of the portable opaque chamber, which was kept closed throughout sampling to ensure airtightness (*Wanyama et al., 2019*). Gas samples were taken on sunny days between 9 am and 11 am (*Cheng et al., 2010*). A

gas sample was extracted from the static chamber using a gas-tight syringe (100 ml) at 0 min, 15 min, 30 min, and 45 min, respectively and injected into a gas preservation bag. The gas samples were sealed and transported to the laboratory within 48 h (*Wang et al., 2010*). Carbon dioxide concentration was analyzed by a gas chromatograph equipped with a flame ionization detector (Agilent4890D; Agilent Technologies, Santa Clara, CA, USA). The calculation method of CO_2 fluxes was referred to *Chen et al. (2021)*.

Measurements of soil and leaf litter

Soil samples were divided into two parts after passing through a 2-mm sieve; one was stored at −20 °C to measure the dissolved organic C (DOC), NH_4^+-N, NO_3^--N, microbial biomass C (MBC) and microbial biomass N (MBN). Microbial biomass C:N ratio could be used to characterize the ratio of soil fungi to bacteria (*Bardgett et al., 2005*). The other part of soil was dried naturally at room temperature to measure soil organic C (SOC), soil total N (TN) and soil pH. In addition, leaf litter samples were dried at 70 °C to obtain constant weight, then ground and stored for analyzing organic C, TN and lignin contents.

Soil DOC content was measured by TOC analyzer (TOC-5000 Analyzer, Shimadzu and Kyoto, Japan) setting the 1:5 ratio of soil material to deionized water. The NO_3^--N and NH_4^+-N were determined by automatic flow injection analyzer (FIAstar 5000 Analyzer, FOSS, Hillerød, Denmark). The MBC and MBN were determined by chloroform fumigation -K_2SO_4 extraction. SOC and leaf litter organic C contents were measured using the wet oxidation method with $K_2Cr_2O_7$ and H_2SO_4, and $FeSO_4$ titration. TN content was determined using the Kjeldahl method (*Liu et al., 2021*). Soil pH was determined by the glass electrode meter method setting the 1:2.5 (w/v) ratio of soil material to deionized water. Leaf litter lignin content was analyzed using concentrated sulfuric acid method (*Singh et al., 2021*). The C:N ratio was the ratio of organic C to TN.

Statistical analysis

A one-way analysis of variance (ANOVA) was performed to examine the differences of litter quality, contents of soil C and N, soil C and N fluxes and microbial biomass C and N among the three forests. Tukey's HSD method was used for multiple comparisons. SPSS was used for data processing and analysis, and Origin 8.0 was used to create figure.

RESULTS

Leaf litter quality

Leaf litter in ErM forest had significantly higher level in lignin: N ratio (45.02 $\pm$ 5.59) than that in AM forest (33.08 $\pm$ 2.13) and EcM forest (26.63 $\pm$ 5.29) ($P < 0.05$) (Fig. 1A). Similarly, leaf litter C:N ratio in ErM forest was the highest (83.05 $\pm$ 10.98), followed by AM forest (60.19 $\pm$ 5.81) and EcM forest (44.97 $\pm$ 9.64) ($P < 0.05$) (Fig. 1B).

Contents of soil C and N

There were no significant differences in SOC contents among the three forests (Fig. 2A). Soil DOC content in EcM forest (106.17 $\pm$ 16.02 mg kg^{-1}) was significantly higher than that in AM forest (79.89 $\pm$ 20.98 mg kg^{-1}) and ErM forest (70.15 $\pm$ 5.61 mg kg^{-1}) ($P <$

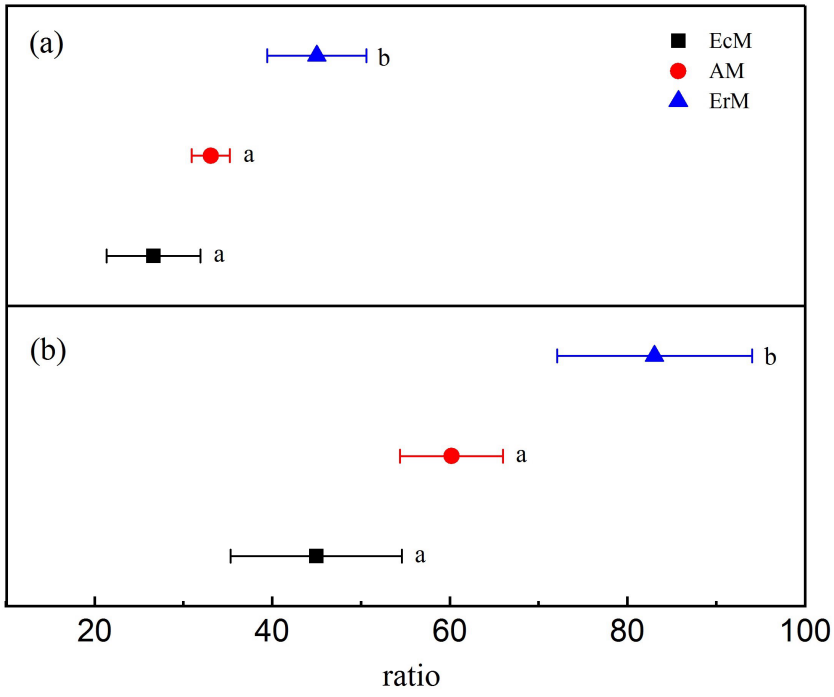

Figure 1 Differences in leaf litter quality among the three forests. Leaf litter lignin: N ratios (A) and leaf litter C:N ratios (B) in *Abies faxoniana* primary forest (EcM), *Cupressus chengiana* primary forest (AM) and *Rhododendron phaeochrysum* primary forest (ErM). Data in the figure were mean ± standard errors (parallel bars; $n = 8$ in each case). a, b and c indicated significant differences among the three forests ($P < 0.05$) according to a one-way analysis of variance.

Full-size DOI: 10.7717/peerj.14028/fig-1

0.05) (Fig. 2D). Moreover, the soil C:N ratio was also significantly higher in EcM forest than that in AM and ErM forests ($P < 0.05$) (Fig. 2C).

Soil TN content in EcM forest was significantly lower than in other forests ($P < 0.05$) (Fig. 2B). Furthermore, soil NH_4^+-N content in EcM forest (4.74 ± 2.40 mg kg^{-1}) was significantly lower than that in AM forest (10.26 ± 2.79 mg kg^{-1}) and ErM forest (12.61 ± 2.57 mg kg^{-1}) ($P < 0.05$) (Fig. 2E). AM forest had the highest soil NO_3^--N content and there was no significant difference between EcM and ErM forests (Fig. 2F).

Soil C and N fluxes

There were significant differences of soil CO_2 flux among AM forest (508.25 ± 65.51 mg m^{-2} h^{-1}), EcM forest (387.18 ± 56.19 mg m^{-2} h^{-1}) and ErM forest (177.87 ± 58.40 mg m^{-2} h^{-1}) ($P < 0.05$) (Fig. 3A). Moreover, there were no significant differences in soil net nitrification rates (NN) among the three forests (Fig. 3B). Soil net N mineralization rate (NNM) in ErM forest was negative (−0.02 ± 0.03 mg kg^{-1} d^{-1}) and had significant differences with the other two forests ($P < 0.05$) (Fig. 3C).

Soil microbial biomass C and N

The MBC and MBN contents in ErM forest were significantly higher than those in AM forest ($P < 0.05$) but there were no significant differences with EcM forest (Figs. 4A and

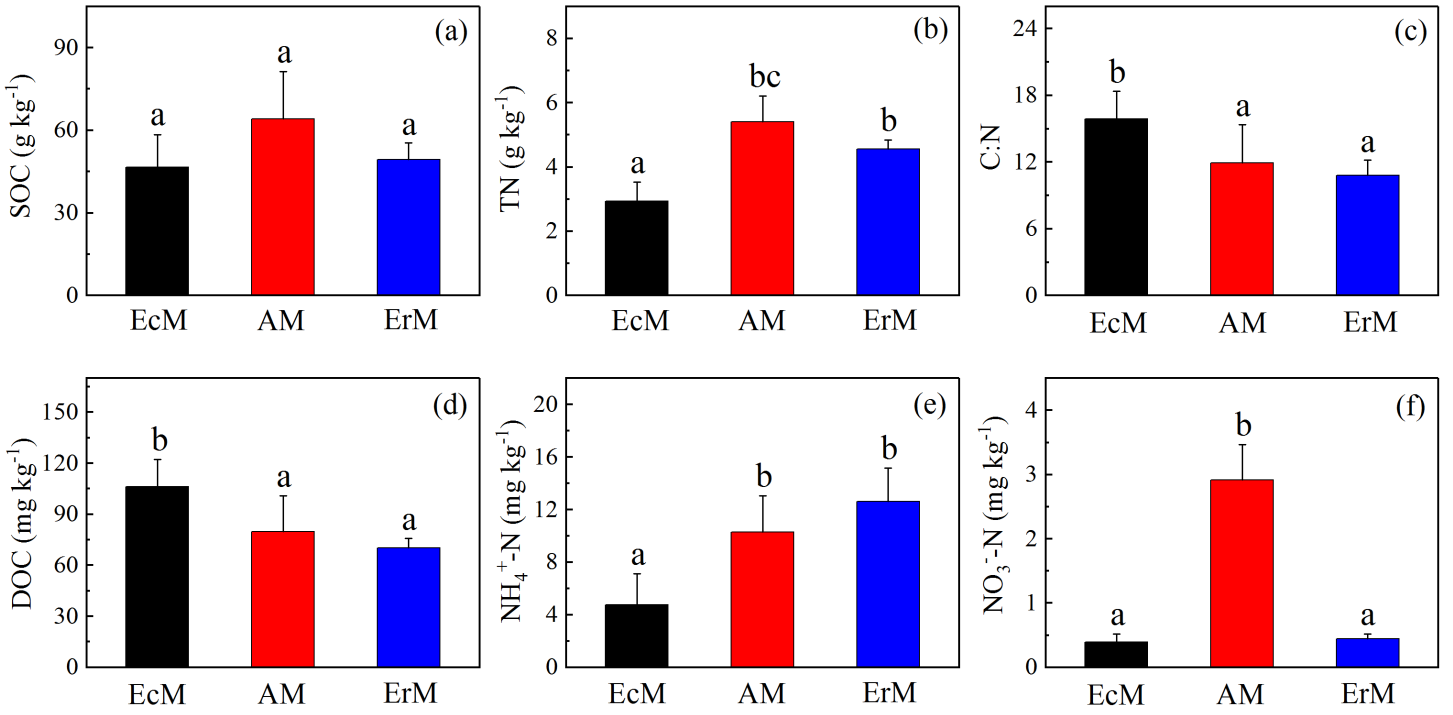

Figure 2 **Differences in contents of soil C and N among the three forests.** SOC contents (A), soil TN contents (B), soil C:N ratios (C), mean soil DOC contents (D), soil NH_4^+-N contents (E) and soil NO_3^--N contents (F) in *Abies faxoniana* primary forest (EcM), *Cupressus chengiana* primary forest (AM) and *Rhododendron phaeochrysum* primary forest (ErM). Data in the figure were mean ± standard errors (vertical bars; $n = 8$ in each case). a, b and c indicated significant differences among the three forests ($P < 0.05$) according to a one-way analysis of variance.

Full-size DOI: 10.7717/peerj.14028/fig-2

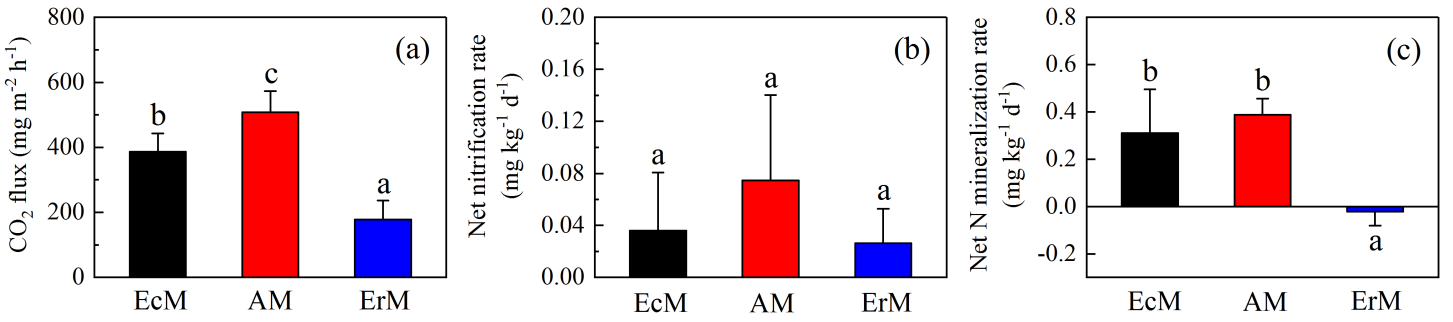

Figure 3 **Differences in soil C and N fluxes among the three forests.** Soil CO_2 fluxes (A), soil net nitrification rates (B) and soil net N mineralization rates (C) in *Abies faxoniana* primary forest (EcM), *Cupressus chengiana* primary forest (AM) and *Rhododendron phaeochrysum* primary forest (ErM). Data in the figure were mean ± standard errors (vertical bars; $n = 8$ in each case). a, b and c indicated significant differences among the three forests ($P < 0.05$) according to a one-way analysis of variance.

Full-size DOI: 10.7717/peerj.14028/fig-3

4B). There was significant difference in MBC:MBN ratio between ErM forest (3.36 ± 1.03) and AM forest (2.73 ± 0.75) ($P < 0.05$), while the ratio in EcM forest (3.08 ± 0.91) had no significant difference with the other two forests (Fig. 4C).

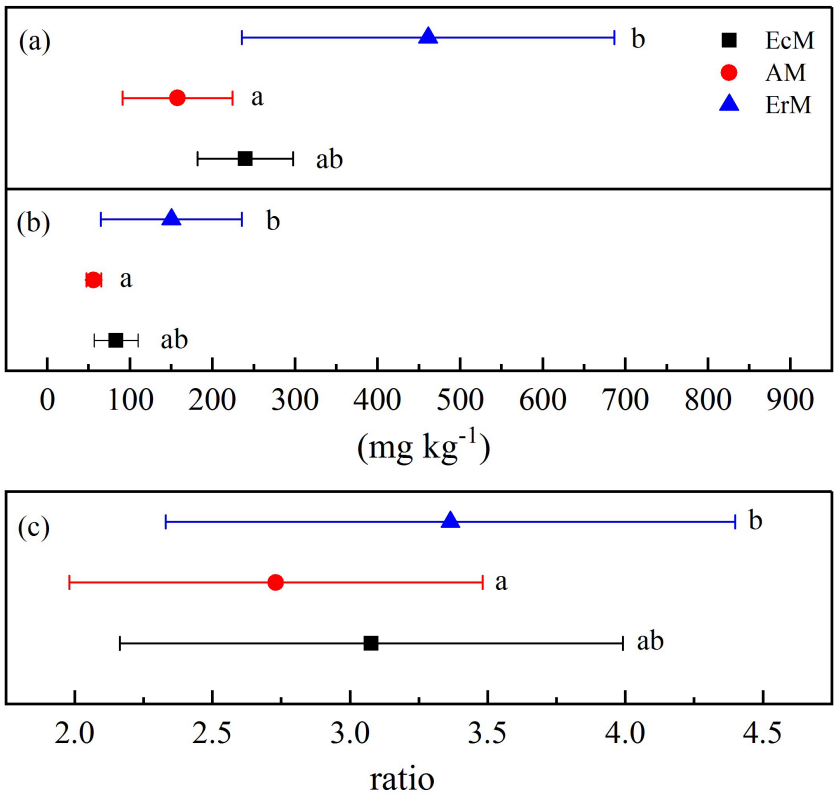

Figure 4 Differences in soil microbial biomass C and N contents among the three forests. Soil microbial biomass C contents (A), soil microbial biomass N contents (B) and soil microbial biomass C:N ratios (C) in *Abies faxoniana* primary forest (EcM), *Cupressus chengiana* primary forest (AM) and *Rhododendron phaeochrysum* primary forest (ErM). Data in the figure were mean ± standard errors (parallel bars; $n = 8$ in each case). a, b and c indicated significant differences among the three forests ($P < 0.05$) according to a one-way analysis of variance.

Full-size DOI: 10.7717/peerj.14028/fig-4

DISCUSSION

Soil C cycling in different mycorrhizal forests

Compared to EcM and ErM forests, the AM forest had the highest soil CO_2 flux (Fig. 3A). Previous studies showed that EcM and ErM fungi preferred and decomposed selectively soil organic N (*Lin et al., 2016*; *Ward et al., 2021*). This nutrient strategy would drive N limitation of soil free-living decomposers, thereby slowing down the soil respiration (*Averill, Turner & Finzi, 2014*). Meanwhile, AM forest had lower microbial biomass C:N ratio than ErM forest in our study (Fig. 4C). It implied that the ratio of soil fungi to bacteria in AM forest was lower than that in ErM forest. AM forest tend to have bacteria-dominated food webs, which may result in AM forest having greater heterotrophic respiration (*Taylor, Lankau & Wurzburger, 2016*).

Simultaneously, the ErM forest had the slowest soil CO_2 flux in our study, which might be related to the quality of its leaf litter (Figs. 1 and 3A). Comparing to AM and EcM forests, leaf litter in ErM forest had higher C:N ratio and lignin: N ratio (Fig. 1) which could represent lower quality of leaf litter (*Midgley, Brzostek & Phillips, 2015*). The lower

quality may lead to accumulation of plant-derived compounds in the soil, which decreases soil CO_2 flux (*Taylor, Lankau & Wurzburger, 2016*; *Keller & Phillips, 2018*). In addition, as one of the main pathways of soil C output, soil DOC is mainly related to soil characteristic, decomposition of litter and humus (*Kindler et al., 2011*). Our results showed that soil DOC content in EcM forest was higher (Fig. 2D). We speculated that higher quality of leaf litter might be one of the reasons for the higher soil DOC content in EcM forest (Figs. 1 and 2D). Overall, our results showed that soil C cycling in AM and EcM forests were relatively faster than that in ErM forest (Fig. 5).

Soil N cycling in different mycorrhizal forests

Our results showed that soil N cycling was more open in the AM forest than that in EcM and ErM forests (Fig. 5). These were reflected mainly in soil C:N, soil inorganic N content, soil net nitrification rate and soil net N mineralization rate (Figs. 2C, 2E, 2F, 3B and 3C). The more open N cycle pattern of AM forest confirms the long-held view that EcM, AM and ErM plants and related fungi differ in their acquisition, utilization and impact on soil N (*Lin et al., 2016*; *Craig et al., 2018*). In our study, soil C:N was lower in AM forest than that in EcM forest (Fig. 2C). Previous researches indicated that EcM fungi could obtain stable C sources from hosts (*Smith & Read, 2008*; *Lindahl & Tunlid, 2015*). As a result, EcM fungi could use selectively organic substrates to obtain N which might lead to higher soil C:N and slower N cycling (*Lin et al., 2016*). Our results showed that the AM forest had higher soil inorganic N content (Figs. 2E and 2F) which was consistent with previous study (*Tedersoo & Bahram, 2019*). Additionally, there were no significant differences in soil net nitrification rates among AM, EcM and ErM forests (Fig. 3B). Soil net N mineralization rate was lower in the ErM forest and there was no significant difference between AM and EcM forests (Fig. 3C). These results were not entirely consistent with previous studies, because the examinations pointed that soil net nitrification rate and soil net N mineralization rate in AM forests were higher than those in EcM forests (*Saifuddin et al., 2021*; *Lin et al., 2016*). However, there were other studies showed that EcM trees had stronger positive effect on soil net nitrification rate and soil net N mineralization rate than AM trees (*Phillips & Fahey, 2006*). *Chen et al. (2018)* indicated that there might be variations in soil net nitrification rate and soil net N mineralization rate in AM and EcM forests with different dominant tree species.

Previous studies showed that AM fungi absorbed primarily soil inorganic N (*Read & Perez-Moreno, 2003*; *Tedersoo, Bahram & Zobel, 2020*). Meanwhile, some studies suggested that EcM and ErM fungi had genetic potential to produce oxidase and glycoside hydrolase which could decompose recalcitrant organic matter (*Op De Beeck et al., 2018*; *Ward et al., 2021*) and mobilized N from organic matter (*Orwin et al., 2011*; *Shah et al., 2016*). *Phillips, Brzostek & Midgley (2013)* proposed that there was inorganic N economy in AM-dominated ecosystem but organic N economy in EcM-dominated ecosystem. In our study, more open soil N cycling implied that there was inorganic N pattern in AM forest (Fig. 5). In addition, our results showed that soil net N nitrification rate in ErM forest was positive and soil net N mineralization rate was negative (Figs. 3B and 3C), indicating that soil net ammonification rate in ErM forest was negative. Consumption of soil NH_4^+-N

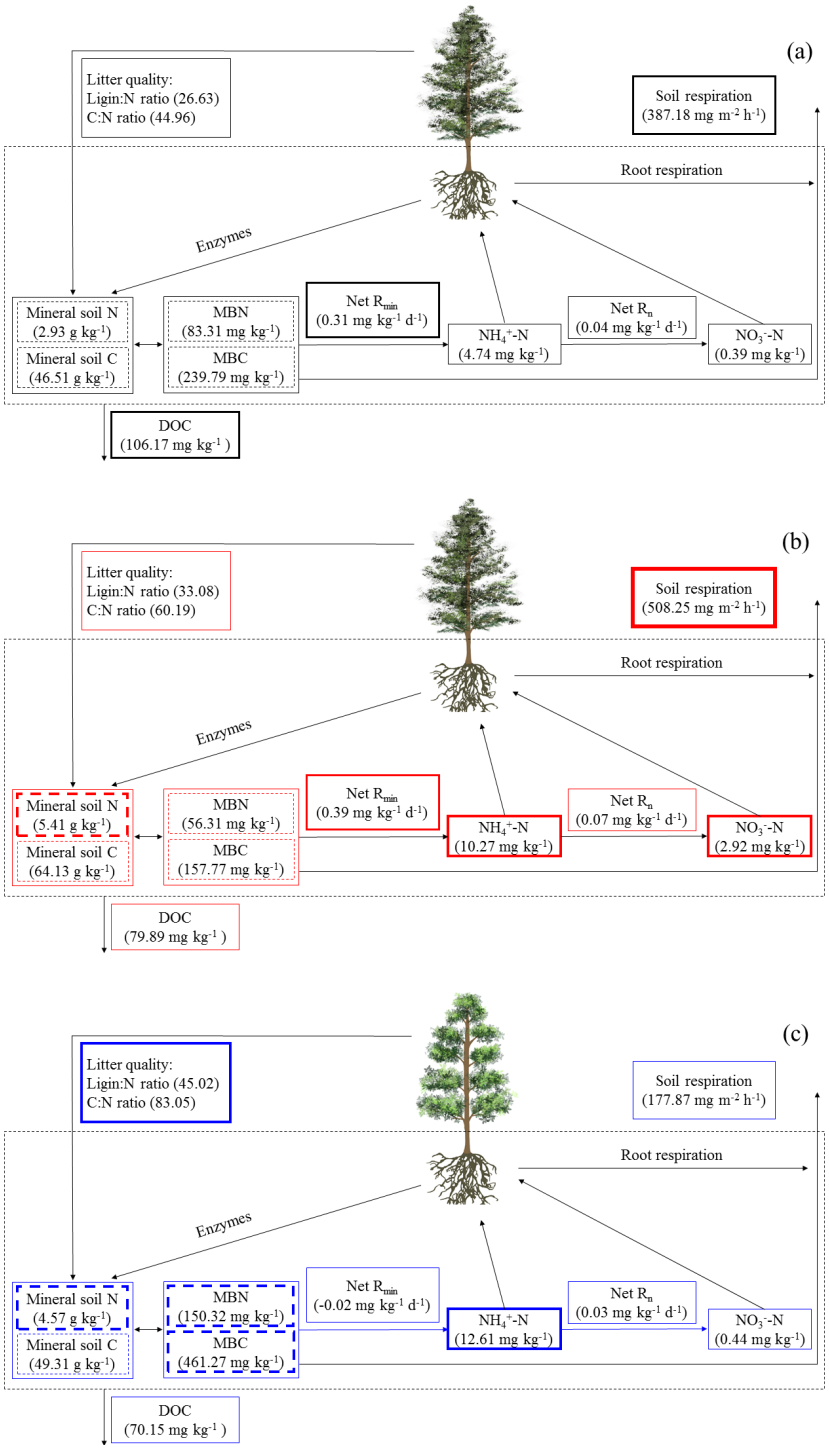

Figure 5 The soil C and N cycling in *Abies faxoniana* primary forest (EcM) (A), *Cupressus chengiana* primary forest (AM) (B) and *Rhododendron phaeochrysum* primary forest (ErM) (C). Data in the figure were mean values ($n = 8$ in each case). DOC, dissolved organic C; MBC, microbial biomass C; MBN, microbial biomass N; Net R_{min}, net N mineralization rate; Net R_n, net nitrification rate. The bolder frame presented the larger value.

Full-size DOI: 10.7717/peerj.14028/fig-5

is higher than its production might mean that microorganisms absorb a lot of nutrients to maintain growth and reproduction of their populations (*Miller et al., 2009*; *Liu et al., 2021*), which corresponds to our results of higher soil microbial biomass C and N contents in ErM forest (Figs. 4A and 4B). Moreover, some studies showed that ErM fungi had wider and stronger ability to decompose organic matter than EcM fungi (*Read, 1991*; *Read & Perez-Moreno, 2003*).Therefore, we speculated that ErM forest had higher demand of organic N and soil net N mineralization was dominated by net ammonification.

CONCLUSIONS

Soil C and N cycling patterns in three primary forests with different mycorrhizal types were compared in our study. AM and EcM forests had relatively faster soil C cycling than that in the ErM forest. Further, the AM forest had lower soil C:N, higher soil inorganic N content and soil net N mineralization rate, indicating AM forest might absorb more soil inorganic N. EcM and ErM forests might demand more organic N sources. These findings provide an in-depth understanding on differences in the way of nutrient acquisition of forests with different mycorrhizal types, which plays an important role in evaluating soil C and N cycling of the forest ecosystems on the eastern Qinghai-Tibetan Plateau.

ACKNOWLEDGEMENTS

We thank Qiuhong Feng from Sichuan Academy of Forestry for her help during sampling. We are also grateful to the reviewers and academic editor for their professional comments and suggestions.

ADDITIONAL INFORMATION AND DECLARATIONS

Funding

This work was supported by the Scientific Research and Development Project of the Ecology and Nature Conservation Institute, the Chinese Academy of Forestry (CAF) (99805-2020) and the Fundamental Research Funds of CAF (CAFYBB2018ZA003). The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

Grant Disclosures

The following grant information was disclosed by the authors:
Scientific Research and Development Project of the Ecology and Nature Conservation Institute, Chinese Academy of Forestry (CAF): 99805-2020.
Fundamental Research Funds of CAF: CAFYBB2018ZA003.

Competing Interests

The authors declare there are no competing interests.

Author Contributions

- Miaomiao Zhang conceived and designed the experiments, performed the experiments, analyzed the data, prepared figures and/or tables, authored or reviewed drafts of the article, and approved the final draft.
- Shun Liu conceived and designed the experiments, performed the experiments, authored or reviewed drafts of the article, and approved the final draft.
- Miao Chen performed the experiments, prepared figures and/or tables, and approved the final draft.
- Jian Chen performed the experiments, prepared figures and/or tables, and approved the final draft.
- Xiangwen Cao performed the experiments, prepared figures and/or tables, and approved the final draft.
- Gexi Xu conceived and designed the experiments, prepared figures and/or tables, and approved the final draft.
- Hongshuang Xing analyzed the data, prepared figures and/or tables, and approved the final draft.
- Feifan Li analyzed the data, prepared figures and/or tables, and approved the final draft.
- Zuomin Shi conceived and designed the experiments, authored or reviewed drafts of the article, and approved the final draft.

Field Study Permissions

The following information was supplied relating to field study approvals (*i.e.*, approving body and any reference numbers):

Field experiments were approved by Ecology and Nature Conservation Institute, Chinese Academy of Forestry (project number: 99805-2020).

Data Availability

The following information was supplied regarding data availability:

The raw data is available in the Supplemental Files.

Supplemental Information

Supplemental information for this article can be found online at http://dx.doi.org/10.7717/peerj.14028#supplemental-information.

REFERENCES

Adamczyk B, Ahvenainen A, Sietiö OM, Kanerva S, Kieloaho AJ, Smolander A, Kitunen V, Saranpää P, Laakso T, Straková P, Heinonsalo J. 2016. The contribution of ericoid plants to soil nitrogen chemistry and organic matter decomposition in boreal forest soil. *Soil Biology and Biochemistry* **103**:394–404 DOI 10.1016/j.soilbio.2016.09.016.

Averill C. 2016. Slowed decomposition in ectomycorrhizal ecosystems is independent of plant chemistry. *Soil Biology and Biochemistry* **102**:52–54 DOI 10.1016/j.soilbio.2016.08.003.

Averill C, Finzi A. 2011. Increasing plant use of organic nitrogen with elevation is reflected in nitrogen uptake rates and ecosystem δ^{15}N. *Ecology* **92(4)**:883–891 DOI 10.1890/10-0746.1.

Averill C, Turner BL, Finzi AC. 2014. Mycorrhiza-mediated competition between plants and decomposers drives soil carbon storage. *Nature* **505(7484)**:543–545 DOI 10.1038/nature12901.

Barajas-Guzmán G, Alvarez-Sánchez J. 2003. The relationships between litter fauna and rates of litter decomposition in a tropical rain forest. *Applied Soil Ecology* **24(1)**:91–100 DOI 10.1016/S0929-1393(03)00069-6.

Bardgett RD, Bowman WD, Kaufmann R, Schmidt S. 2005. A temporal approach to linking aboveground and belowground ecology. *Trends in Ecology & Evolution* **20(11)**:634–641 DOI 10.1016/j.tree.2005.08.005.

Becker H, Uri V, Aosaar J, Varik M, Ü Mander, Soosaar K, Hansen R, Teemusk A, Morozov G, Kutti S, Lõhmus K. 2015. The effects of clear-cut on net nitrogen mineralization and nitrogen losses in a grey alder stand. *Ecological Engineering* **85**:237–246 DOI 10.1016/j.ecoleng.2015.10.006.

Bennett AE, Groten K. 2022. The costs and benefits of plant-arbuscular mycorrhizal fungal interactions. *Annual Review of Plant Biology* **73**:18.1–18.24 DOI 10.1146/annurev-arplant-102820-124504.

Brzostek ER, Dragoni D, Brown ZA, Phillips RP. 2015. Mycorrhizal type determines the magnitude and direction of root-induced changes in decomposition in a temperate forest. *New Phytologist* **206(4)**:1274–1282 DOI 10.1111/nph.13303.

Cavagnaro TR, Barrios-Masias FH, Jackson LE. 2012. Arbuscular mycorrhizas and their role in plant growth, nitrogen interception and soil gas efflux in an organic production system. *Plant and Soil* **353(1–2)**:181–194 DOI 10.1007/s11104-011-1021-6.

Cheeke TE, Phillips RP, Brzostek ER, Rosling A, Bever JD, Fransson P. 2017. Dominant mycorrhizal association of trees alters carbon and nutrient cycling by selecting for microbial groups with distinct enzyme function. *New Phytologist* **214(1)**:432–442 DOI 10.1111/nph.14343.

Chen HH. 2019. Forest community diversity, carbon and nitrogen cycling modes and their relationships with mycorrhizal fungi in the subalpine area of western Sichuan. Dissertation, Chinese Academy of Forestry DOI 10.27625/d.cnki.gzlky.2019.000062.

Chen X, Ding ZJ, Tang M, Zhu B. 2018. Greater variations of rhizosphere effects within mycorrhizal group than between mycorrhizal group in a temperate forest. *Soil Biology and Biochemistry* **126**:237–246 DOI 10.1016/j.soilbio.2018.08.026.

Chen Q, Long C, Chen JW, Cheng XL. 2021. Differential response of soil CO_2, CH_4, and N_2O emissions to edaphic properties and microbial attributes following afforestation in central China. *Global Change Biology* **00**:1–13 DOI 10.1111/gcb.15826.

Cheng XL, Luo YQ, Xu Q, Lin GH, Zhang QF, Chen JK, Li B. 2010. Seasonal variation in CH_4 emission and its ^{13}C-isotopic signature from Spartina alterniflora and Scirpus mariqueter soils in an estuarine wetland. *Plant and Soil* **327(1–2)**:85–94 DOI 10.1007/s11104-009-0033-y.

Clemmensen KE, Durling MB, Michelsen A, Hallin S, Finlay RD, Lindahl BD. 2021. A tipping point in carbon storage when forest expands into tundra is related to mycorrhizal recycling of nitrogen. *Ecology Letters* **24**:1193–1204 DOI 10.1111/ele.13735.

Cotrufo MF, Wallenstein MD, Boot CM, Denef K, Paul E. 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? *Global Change Biology* **19(4)**:988–995 DOI 10.1111/gcb.12113.

Craig ME, Turner BL, Liang C, Clay K, Johnson DJ, Phillips RP. 2018. Tree mycorrhizal type predicts within-site variability in the storage and distribution of soil organic matter. *Global Change Biology* **24(8)**:3317–3330 DOI 10.1111/gcb.14132.

Fang M, Liang MX, Liu XB, Li WB, Huang E, Yu SX. 2020. Abundance of saprotrophic fungi determines decomposition rates of leaf litter from arbuscular mycorrhizal and ectomycorrhizal trees in a subtropical forest. *Soil Biology and Biochemistry* **149**:107966 DOI 10.1016/j.soilbio.2020.107966.

Feng QH, Shi ZM, Xu ZJR, Miao N, Tang JC, Liu XL, Zhang L. 2017. Phenotypic variations in cones and seeds of natural *Cupressus chengiana* populations in China. *Chinese Journal of Ecology* **28**:748–756 DOI 10.13287/j.1001-9332.201703.001.

Gadgil RL, Gadgil PD. 1971. Mycorrhiza and litter decomposition. *Nature* **233**:133 DOI 10.1038/233133a0.

Heinemeyer A, Hartley IP, Evans SP, Fuente JACDL, Ineson P. 2007. Forest soil CO_2 flux: uncovering the contribution and environmental responses of ectomycorrhizas. *Global Change Biology* **13(8)**:1786–1797 DOI 10.1111/j.1365-2486.2007.01383.x.

Hobbie EA, Högberg P. 2012. Nitrogen isotopes link mycorrhizal fungi and plants to nitrogen dynamics. *New Phytologist* **196(2)**:367–382 DOI 10.1111/j.1469-8137.2012.04300.x.

Hughes JK, Hodge A, Fitter AH, Atkin OK. 2008. Mycorrhizal respiration: implications for global scaling relationships. *Trends in Plant Science* **13(11)**:583–588 DOI 10.1016/j.tplants.2008.08.010.

Keller AB, Phillips RP. 2018. Leaf litter decay rates differ between mycorrhizal groups in temperate, but not tropical, forests. *New Phytologist* **222(1)**:556–564 DOI 10.1111/nph.15524.

Kindler R, Siemens J, Kaiser K, Walmsley DC, Bernhofer C, Buchmann N, Cellier P, Eugster W, Gleixner G, Grûnwald T. 2011. Dissolved carbon leaching from soil is a crucial component of the net ecosystem carbon balance. *Global Change Biology* **17(2)**:1167–1185 DOI 10.1111/j.1365-2486.2010.02282.x.

Kong WB, Yao YF, Zhao ZN, Qin X, Zhu HS, Wei XR, Shao MA, Wang Z, Bao KQ, Su M. 2019. Effects of vegetation and slope aspect on soil nitrogen mineralization during the growing season in sloping lands of the Loess Plateau. *Catena* **172**:753–763 DOI 10.1016/j.catena.2018.09.037.

Lin GG, Mccormack ML, Ma C, Guo DL. 2016. Similar below-ground carbon cycling dynamics but contrasting modes of nitrogen cycling between arbuscular mycorrhizal and ectomycorrhizal forests. *New Phytologist* **213**:1440–1451 DOI 10.1111/nph.14206.

Lindahl BD, Ihrmark K, Boberg J, Trumbore SE, Högberg P, Stenlid J, Finlay RD. 2007. Spatial separation of litter decomposition and mycorrhizal nitrogen uptake in a boreal forest. *New Phytologist* **173(3)**:611–620 DOI 10.1111/j.1469-8137.2006.01936.x.

Lindahl BD, Tunlid A. 2015. Ectomycorrhizal fungi-potential organic matter decomposers, yet not saprotrophs. *New Phytologist* **205(4)**:1443–1447 DOI 10.1111/nph.13201.

Liu Y. 2010. Patterns of above-and belowground biodiversity of alpine timberline ecotone and forest-meadow ecotone in Western Sichuan. Dissertation, Sichuan Agricultural University.

Liu S, Luo D, Cheng RM, Wu JM, Yang HG, Shi ZM. 2021. Temporal variability in soil net nitrogen mineralization among forest regeneration patterns in eastern Tibetan Plateau. *Ecological Indicators* **128**:107811 DOI 10.1016/j.ecolind.2021.107811.

Midgley MG, Brzostek E, Phillips RP. 2015. Decay rates of leaf litters from arbuscular mycorrhizal trees are more sensitive to soil effects than litters from ectomycorrhizal trees. *Journal of Ecology* **103(6)**:1454–1463 DOI 10.1111/1365-2745.12467.

Miller AE, Schimel JP, Sickman JO, Skeen K, Meixner T, Melack JM. 2009. Seasonal variation in nitrogen uptake and turnover in two high-elevation soils: mineralization responses are site-dependent. *Biogeochemistry* **93(3)**:253–270 DOI 10.1007/s10533-009-9301-4.

Op De Beeck M, Troein C, Peterson C, Persson P, Tunlid A. 2018. Fenton reaction facilitates organic nitrogen acquisition by an ectomycorrhizal fungus. *New Phytologist* **218**:335–343 DOI 10.1111/nph.14971.

Orwin KH, Kirschbaum MUF, John MGSSt, Dickie LA. 2011. Organic nutrient uptake by mycorrhizal fungi enhances ecosystem carbon storage: a model-based assessment. *Ecology Letters* **14(5)**:493–502 DOI 10.1111/j.1461-0248.2011.01611.x.

Phillips RP, Brzostek E, Midgley MG. 2013. The mycorrhizal-associated nutrient economy: a new framework for predicting carbon-nutrient couplings in temperate forests. *New Phytologist* **199(1)**:41–51 DOI 10.1111/nph.12221.

Phillips RP, Fahey TJ. 2006. Tree species and mycorrhizal associations influence the magnitude of rhizosphere effects. *Ecology* **87**:1302–1313 DOI 10.1890/0012-9658(2006)87[1302:TSAMAI]2.0.CO;2.

Read DJ. 1991. Mycorrhizas in ecosystems. *Experientia* **47**:376–391 DOI 10.1007/BF01972080.

Read D, Leake JR, Perez-Moreno J. 2004. Mycorrhizal fungi as drivers of ecosystem processes in heathland and boreal forest biomes. *Canadian Journal of Botany* **82(8)**:1243–1263 DOI 10.1139/b04-123.

Read DJ, Perez-Moreno J. 2003. Mycorrhizas and nutrient cycling in ecosystems-a journey towards relevance? *New Phytologist* **157(3)**:475–492 DOI 10.1046/j.1469-8137.2003.00704.x.

Rillig MC. 2004. Arbuscular mycorrhizae, glomalin, and soil aggregation. *Canadian Journal of Soil Science* **84(84)**:355–363 DOI 10.4141/S04-003.

Saifuddin M, Bhatnagar JM, Phillips RP, Finzi AC. 2021. Ectomycorrhizal fungi are associated with reduced nitrogen cycling rates in temperate forest soils without corresponding trends in bacterial functional groups. *Oecologia* **196**:863–875 DOI 10.1007/s00442-021-04966-z.

Shah F, Nicolás C, Bentzer J, Ellström M, Smits M, Rineau F, Canbäck B, Floudas D, Carleer R, Lackner G, Braesel J, Hoffmeister D, Henrissat B, Ahrén D, Johansson T, Hibbett DS, Martin F, Persson P, Tunlid A. 2016. Ectomycorrhizal fungi decompose soil organic matter using oxidative mechanisms adapted from saprotrophic ancestors. *New Phytologist* **209**:1705–1719 DOI 10.1111/nph.13722.

Shi Z, Wang F, Liu Y. 2012. Response of soil respiration under different mycorrhizal strategies to precipitation and temperature. *Journal of Soil Science and Plant Nutrition* **12(3)**:411–420 DOI 10.4067/S0718-95162012005000003.

Singh L, Thakur D, Sharma MK, Chawla A. 2021. Dynamics of leaf litter decomposition in the timberline zone of western Himalaya. *Acta Oecologica* **111**:103715 DOI 10.1016/j.actao.2021.103715.

Smith SE, Read DJ. 2008. Mycorrhizal symbiosis. *The Quarterly Review of Biology* **3(3)**:273–281 DOI 10.1097/00010694-198403000-00011.

Taylor MK, Lankau RA, Wurzburger N. 2016. Mycorrhizal associations of trees have different indirect effects on organic matter decomposition. *Journal of Ecology* **104**:1576–1584 DOI 10.1111/1365-2745.12629.

Tedersoo L, Bahram M. 2019. Mycorrhizal types differ in ecophysiology and alter plant nutrition and soil processes. *Biological Reviews* **94(5)**:1857–1880 DOI 10.1111/brv.12538.

Tedersoo L, Bahram M, Zobel M. 2020. How mycorrhizal associations drive plant population and community biology. *Science* **367(6480)**:eaba1223 DOI 10.1126/science.aba1223.

Tisserant E, Malbreil M, Kuo A, Kohler A, Symeonidi A, Balestrini R, Charron P, Duensing N, Frey NFD, Gianinazzi-Pearson V, Gilbert LB, Handa Y, Herr JR, Hijri M, Koul R, Kawaguchi M, Krajinski F. 2013. Genome of an arbuscular mycorrhizal fungus provides insight into the oldest plant symbiosis. *Proceedings of the National Academy of Sciences of the United States of America* **110(1)**:20117–20122 DOI 10.1073/pnas.1313452110.

Tomè E, Ventura M, Folegot S, Zanotelli D, Montagnani L, Mimmo T, Tonon G, Tagliavini M, Scandellari F. 2016. Mycorrhizal contribution to soil respiration in an apple orchard. *Applied Soil Ecology* **101**:165–173 DOI 10.1016/j.apsoil.2016.01.016.

Wang H, Liu SR, Mo JM, Zhang T. 2010. Soil-atmosphere exchange of greenhouse gases in subtropical plantations of indigenous tree species. *Plant and Soil* **335(1–2)**:213–227 DOI 10.1007/s11104-010-0408-0.

Wang HL, Na D, Wu DY, Hu S, Kou M. 2017. Long-term net transformation and quantitative molecular mechanisms of soil nitrogen during natural vegetation recovery of abandoned farmland on the Loess Plateau of China. *Science of the Total Environment* **607–608**:152–159 DOI 10.1016/j.scitotenv.2017.07.014.

Wang GX, Wang TB, Li YS, Cheng HY. 2007. Influences of alpine ecosystem responses to climatic change on soil properties on the Qinghai-Tibet Plateau, China. *Catena* **70**(**3**):506–514 DOI 10.1016/j.catena.2007.01.001.

Wang XQ, Wang CK, Zhang TD. 2017. New perspectives on forest soil carbon and nitrogen cycling processes: roles of arbuscular mycorrhizal versus ecto-mycorrhizal tree species. *Chinese Journal of Plant Ecology* **41**(**10**):1113–1125 DOI 10.17521/cjpe.2017.0116.

Wanyama I, Pelster DE, Butterbach-Bahl K, Verchot V, Martius C, Rufino MC. 2019. Soil carbon dioxide and methane fluxes from forests and other land use types in an African tropical montane region. *Biogeochemistry* **143**:171–190 DOI 10.1007/s10533-019-00555-8.

Ward EB, Duguid MC, Kuebbing SE, Lendemer JC, WarrenI IRJ, Bradford MA. 2021. Ericoid mycorrhizal shrubs alter the relationship between tree mycorrhizal dominance and soil carbon and nitrogen. *Journal of Ecology* **109**(**10**):3524–3540 DOI 10.1111/1365-2745.13734.

Wu JJ, Li QX, Chen JW, Lei Y, Zhang Q, Yang F, Zhang DD, Zhang QF, Cheng XL. 2018. Afforestation enhanced soil CH_4 uptake rate in subtropical China: evidence from carbon stable isotope experiments. *Soil Biology and Biochemistry* **118**:199–206 DOI 10.1016/j.soilbio.2017.12.017.

Wurzburger N, Hendrick RL. 2009. Plant litter chemistry and mycorrhizal roots promote a nitrogen feedback in a temperate forest. *Journal of Ecology* **97**(**3**):528–536 DOI 10.1111/j.1365-2745.2009.01487.x.

Xu GX, Chen HH, Shi ZM, Liu S, Cao XW, Zhang MM, Chen M, Chen J, Xiong K, Yang HG, Zhao GD. 2021. Mycorrhizal and rhizospheric fungal community assembly differs during subalpine forest restoration on the eastern Qinghai-Tibetan Plateau. *Plant and Soil* **458**(**1–2**):245–259 DOI 10.1007/s11104-019-04400-7.