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MICROBIAL ECOLOGY AND ENVIRONMENTAL MICROBIOLOGY

Mycorrhizal Fungal Diversity Associated with Six Understudied Ectomycorrhizal Trees in the Republic of Korea

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Abstract

Mycorrhizal fungi are key components of forest ecosystems and play essential roles in host health. The host specificity of mycorrhizal fungi is variable and the mycorrhizal fungi composition for the dominant tree species is largely known but remains unknown for the less common tree species. In this study, we collected soil samples from the roots of six understudied ectomycorrhizal tree species from a preserved natural park in the Republic of Korea over four seasons to investigate the host specificity of mycorrhizal fungi in multiple tree species, considering the abiotic factors. We evaluated the mycorrhizal fungal composition in each tree species using a metabarcoding approach. Our results revealed that each host tree species harbored unique mycorrhizal communities, despite close localization. Most mycorrhizal taxa belonged to ectomycorrhizal fungi, but a small proportion of ericoid mycorrhizal fungi and arbuscular mycorrhizal fungi were also detected. While common mycorrhizal fungi were shared between the plant species at the genus or higher taxonomic level, we found high host specificity at the species/OTU (operational taxonomic unit) level. Moreover, the effects of the seasons and soil properties on the mycorrhizal communities differed by tree species. Our results indicate that mycorrhizal fungi feature host-specificity at lower taxonomic levels.

Keywords Mycorrhizal fungi · Fungal diversity · Microbiome · Metabarcoding · Ectomycorrhizal tree

Introduction

Diverse mycorrhizal fungi are associated underground with their host plants. Extended mycelia from mycorrhizal fungi transport soil nutrients to their host plants, while the fungi receive photosynthetic products in return (Genre et al., 2020). Mycorrhizal fungi also provide protection from phytopathogens, thus enhancing the overall growth and survival of the host plants (Högberg & Högberg, 2002; Smith & Read, 2013). The major types of mycorrhizal fungi are ectomycorrhizal fungi (EcM), arbuscular mycorrhizal fungi (AM), ericoid mycorrhizal fungi (ErM), and orchid mycorrhizal fungi (OM). More than 200,000 plant species form a symbiotic relationship with mycorrhizal fungi, and approximately 6000–7000 species are associated with ectomycorrhizal (EcM) fungi (Brundrett & Tedersoo, 2018). Many symbiont mycorrhizal species exist for a single plant species. Up to 200 EcM species have been detected in an adult *Populus tremula* tree (Bahram et al., 2011), and more than ten EcM species have been reported for each *Pinus* seedling (Obase et al., 2009; Yoo et al., 2022).

Although many mycorrhizal fungi associate with multiple hosts, limited host specificity of mycorrhizal fungi has been reported in various studies (Bruns et al., 2002; Tedersoo et al., 2008; van der Linde et al., 2018). Depending on their host specificity, mycorrhizal species are classified as generalists (i.e., associated with multiple plant species) or specialists (i.e., associated with specific plant species) (Smith et al., 2009). The host specificity of mycorrhizal

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fungi has been reported at the genus and the species levels. The suilloid group, including *Suillus*, *Rhizopogon*, *Truncocolumella*, *Gomphidius*, and *Chroogomphus*, consists of many Pinaceae-specific mycorrhizal fungi (Bruns et al., 2002). At the species level, *Tricholoma matsutake* forms an ectomycorrhizal relationship primarily with *Pinus densiflora* (Vaario et al., 2017), and *Suillus grevillea* with *Larix* (Bruns et al., 2002). The symbiotic relationship between mycorrhizal species and their host plants is dynamic, and fungal species within a single genus may feature differently in host specificities (Lofgren et al., 2021; Nguyen et al., 2016b).

Ectomycorrhizal tree species in Pinales and Fagales are major components of temperate forests in the Republic of Korea (KFRI, 2016; Choung et al., 2020). *Pinus densiflora* and *Quercus mongolica* are the dominant ectomycorrhizal trees in the Republic of Korea and several studies have reported their importance in the forest constitution (Choung et al., 2020) and the composition of specific mycorrhizal fungi associated with *P. densiflora* and *Q. mongoica* (Lee & Eom, 2013; Oh et al., 2018; Park et al., 2021). However, mycorrhizal fungi associated with other tree species in Pinales and Fagales are understudied. As host identity is a principal factor in determining mycorrhizal community (van der Linde et al., 2018; Yang et al., 2021), distinct sets of mycorrhizal fungi are expected from the underexplored trees. The differential mycorrhizal community may be the result of coevolution between fungi and plants (Gao et al., 2013; Rochet et al., 2011), but abiotic conditions, such as phosphorus and nitrogen, also influence the symbiotic relationship between the host plants and mycorrhizal fungi (Yang et al., 2022). Therefore, investigating multiple tree species with the consideration of abiotic factors is required to better understand the plant-mycorrhizal fungal specificity and interaction.

In this study, we analyzed the community composition of mycorrhizal fungi associated with six EcM trees (three conifer species in Pinales and three broadleaf species in Fagales) in a preserved temperate forest of Mt. Jeombong in the Republic of Korea, using high-throughput sequencing of the ribosomal internal transcribed spacer 2 (ITS2) region. The main goal of this study was to find the host-specificity of mycorrhizal fungi in multiple tree species, considering abiotic factors including soil property, season, and sampling year. This study provides an opportunity to compare mycorrhizal fungi in different hosts while controlling possible geographic variations, as the samples were collected in adjacent forests.

Materials and Methods

Sample Collection and DNA Extraction

This study was conducted in a temperate forest of Mt. Jeombong (Seoraksan National Park, Gangwon Province, Republic of Korea) between 2015 and 2017. The following six tree species were selected for analysis (Table 1): three species in Pinales — *Abies holophylla* (Ah), *Larix kaempferi* (Lk), and *Pinus koraiensis* (Pk), and three species in Fagales — *Carpinus laxiflora* (Cl), *Quercus aliena* (Qa), *Quercus serrata* (Qs). Each year, we sampled two tree species from two different monodominant sites covering more than 50 × 50 m. Sampling was conducted per season (four times/year) to address the possible temporal variance of mycorrhizal communities: winter (March), spring (May), summer (July), and autumn (September). From each site, three trees without any visual disease symptoms were selected for the study. The trees were at least 10 m apart from one another. The soil samples of each tree were sampled from surrounding soil from the 0–10 cm soil layer after removing surface debris. The rhizospheric soil samples were collected from

Table 1 GPS coordinates, altitude, and soil properties of sampling sites

Category	Variables	Pinales			Fagales		
		A. holophylla	*P. koraiensis*	*L. kaempferi*	*C. laxiflora*	*Q. serrata*	*Q. aliena*
Geography	GPS	38.0394, 128.4443	38.072, 128.4492	38.0201, 128.4750	38.0327, 128.4598	38.0716, 128.4497	37.9982, 128.4942
	Altitude (m)	885	436	723	809	444	652
	Year	2015	2016	2017	2015	2016	2017
Soil properties	pH	4.4 (0.1)	4.9 (0.1)	4.6 (0.0)	4.6 (0.1)	4.4 (0.0)	4.4 (0.1)
	TOC (%)	5.6 (0.3)	3.8 (0.5)	7.7 (0.4)	5.3 (0.2)	5 (0.3)	13 (2.6)
	TN (%)	0.48 (0.02)	0.27 (0.03)	0.57 (0.02)	0.44 (0.02)	0.38 (0.02)	0.74 (0.08)
	C/N	11.5 (0.1)	13.5 (0.4)	13.5 (0.5)	12.1 (0.3)	13 (0.4)	16 (1.3)
	TP (mg/kg)	1002.3 (183.7)	218.6 (16.0)	689.4 (17.7)	472.5 (21.7)	289.6 (11.3)	1234.9 (78.6)
	Water contents (%)	30.4 (1.6)	19.1 (1.2)	29.2 (1.8)	29.4 (1.7)	24.5 (1.0)	34.9 (7.1)

Values in a parenthesis indicate the standard deviation

root-surrounding soils in three directions around the tree trunk using a sterilized shovel and zipper bag (15 × 20 cm). In total, we collected 72 soil samples (six tree species × three individual trees × four seasons). The soil sampling was permitted by Korea National Arboretum Authority.

All soil samples were stored in an ice box and transported to the laboratory. Triplicate soil samples from each tree were mixed upon arrival and filtered through a 2.0 mm sieve. Each soil sample was divided into two different batches. One batch was sent to the National Instrumentation Center for Environmental Management (Seoul, Republic of Korea) for a soil property analysis within the day of collection. The following soil properties were measured: pH, total organic carbon (TOC), total nitrogen (TN), total phosphorus (TP), and water contents. The carbon-to-nitrogen ratio (C/N) was calculated using the TOC and TN values. The remaining batch was stored at − 80 °C before the next experiment.

DNA Extraction, PCR Amplification, and Sequencing

Within a week after sampling, environmental DNA (eDNA) was extracted from the soil samples using a PowerSoil DNA extraction kit (MoBio) following the manufacturer's instructions. Extraction was performed in triplicate for each sample, using 0.25 g of soil samples per extraction, then pooled into a single sample for subsequent processes. After DNA extraction, the ITS2 region of ribosomal RNA was amplified using ITS3 and ITS4 primers with Illumina sequencing adaptors (White et al., 1990; Yu et al., 2022). To minimize PCR bias, PCR was conducted in triplicate for each sample using an AccuPower PCR Premix Kit (Bioneer). The following PCR conditions were used: 94 °C for 5 min, 35 cycles of 94 °C for 30 s, 55 °C for 30 s, 72 °C for 40 s, and final extension at 72 °C for 10 min. The final PCR reaction volume was 20 µl, containing 10 pmol of each primer and 1 µl of genomic DNA. We verified the PCR amplicons through gel electrophoresis using 1% agarose gel (BIOFACT), then pooled the triplicate samples and purified them using an Expin™ PCR Purification Kit (GeneAll Biotechnology). DNA sequencing was conducted using the Illumina Miseq platform (Macrogen). Raw sequence data were deposited in the NCBI Sequence Read Archive (SRA) under Project ID PRJNA937676.

Sequence Processing

Raw sequencing data were processed using QIIME v. 1.8.0 (Caporaso et al., 2010). Paired sequences were merged using PEAR (Zhang et al., 2014) with quality filtering (length < 200 bps; quality score < 20). Acquired sequences were clustered to operational taxonomic unit (OTU) with 97% similarity, and chimera sequences were filtered using Vsearch version 2.6.2 (Rognes et al., 2016). We assigned the most abundant sequence of each OTU as a representative. Identification of OTUs was conducted by nucleotide BLAST search against the UNITE v7.2 (Kõljalg et al., 2013) and the Seoul National University Fungus Collection (SFC) database. After removing global singletons, co-occurring OTUs were merged with similar but more abundant OTUs with LULU and post-clustering curation method in the default settings (Frøslev et al., 2017). Rare OTUs (< 100 reads) were removed from the analysis for quality control. To filter the mycorrhizal fungi, FUNGuild was used as a primary database (Nguyen et al., 2016a), and manual examination was conducted using the FungalTraits database (Põlme et al., 2020). The filtered mycorrhizal OTUs were normalized to the minimum sequence reads (6450) by single rarefaction for further analysis.

Statistical Analysis

Alpha diversity indices (Chao1 richness and Shannon diversity) were calculated in QIIME, and further analyses were conducted in R v.4.1.2 (R Core Team, 2021). The relative abundance of mycorrhizal fungi (sequence reads of mycorrhizal OTUs/ sequence reads of all OTUs in the sample) was compared for each eDNA sample. Mycorrhizal communities from each tree species were compared through the principal coordinate analysis (PCoA) using the phyloseq R package (McMurdie & Holmes, 2013) based on Jaccard dissimilarity. Permutational multivariate analysis of variance (PERMANOVA) statistical test was performed to test the significance of the tree species, seasons, tree order, and soil properties using adonis in the vegan R package (Oksanen et al., 2013). The influence of soil properties was visualized on a PCoA ordination plot using envfit in the vegan R package. Then, a pairwise PERMANOVA test was performed using the pairwiseAdonis R package with Bonferroni adjustment to test the differences between each tree species (Arbizu, 2017). UPGMA (average linkage clustering) dendrogram was constructed with a Jaccard dissimilarity index to visualize the relationship between the mycorrhizal community composition and host trees. Canonical analysis of principal coordinates (CAP) was performed using capsacle in vegan R package to test the differentiation of mycorrhizal communities between host species after controlling the sampling year. To separate the effect of environmental factors and tree species on community composition, we constructed a variation partitioning using varpart in the vegan R package. UpSet plots were generated using the UpSetR package to describe the co-occurrence patterns of mycorrhizal taxa (genus/OTU) between tree species (Conway et al., 2017). Host-specific OTUs were identified through the indicator species analysis using multipatt function in the indicspecies package (De Cáceres & Legendre, 2009).

Results

Sequencing Results

A total of 17,126,495 sequences and 12,085 OTUs were obtained from Illumina Miseq sequencing after filtering low-quality and non-fungal sequences. The average length of obtained OTUs were 373.56 ± 44.69 bp. Post-clustering and removal of rare OTUs (< 100 reads) resulted in 16,928,597 sequence reads and 3425 OTUs. For mycorrhizal fungi, 7,880,338 reads and 566 OTUs were acquired, and 565 OTUs remained after normalization to 6450 reads per sample. On average, 235.83 ± 29.38 mycorrhizal OTUs were found per tree species, and 70.28 ± 27.38 mycorrhizal OTUs were found per individual tree.

The seasonal pattern of alpha diversity in the mycorrhizal community could not be generalized by tree species (Fig. S1). Although no index showed statistical significance, the season with the highest richness was either spring (Lk, Qa, Qs) or winter (Ah, Cl, Pk), and the season with the highest diversity was winter (Ah, Cl), autumn (Qs, Pk, Lk), or spring (Qa). The relative abundance of mycorrhizal fungi was the highest in summer for three tree species (Ah, Lk, Qs), but the others had different patterns.

Taxonomy of the Mycorrhizal Community

Overall, mycorrhizal species in 4 phyla, 9 classes, 18 orders, 48 families, and 77 genera were found from the six tree species analyzed. Basidiomycota was the most dominant phylum (> 75%) in all tree species, followed by Ascomycota, Glomeromycota, and Mucoromycota (Supplementary File 1). At the genus level, the most dominant mycorrhizal genus was *Russula*, followed by *Sebacina, Amanita, Lactifluus, Oidiodendron, Inocybe, Clavulina, Piloderma, Suillus, Amphinema, Hygrophorus, Tomentella, Cenococcum, Tylospora, Cortinarius, Laccaria*, and *Tuber* (relative abundance > 1%). These genera accounted for 82.94% of the total sequences (Fig. 1A, Fig. S2). However, the dominant mycorrhizal type differed by tree species. Overall, EcM was the most dominant guild in the mycorrhizal community (93.46%), followed by ErM (6.13%), AM (0.37%), and OM (0.04%). EcM accounted for the highest proportion for all tree species, and ErM also accounted for a high proportion in Lk, Pk, and Qs.

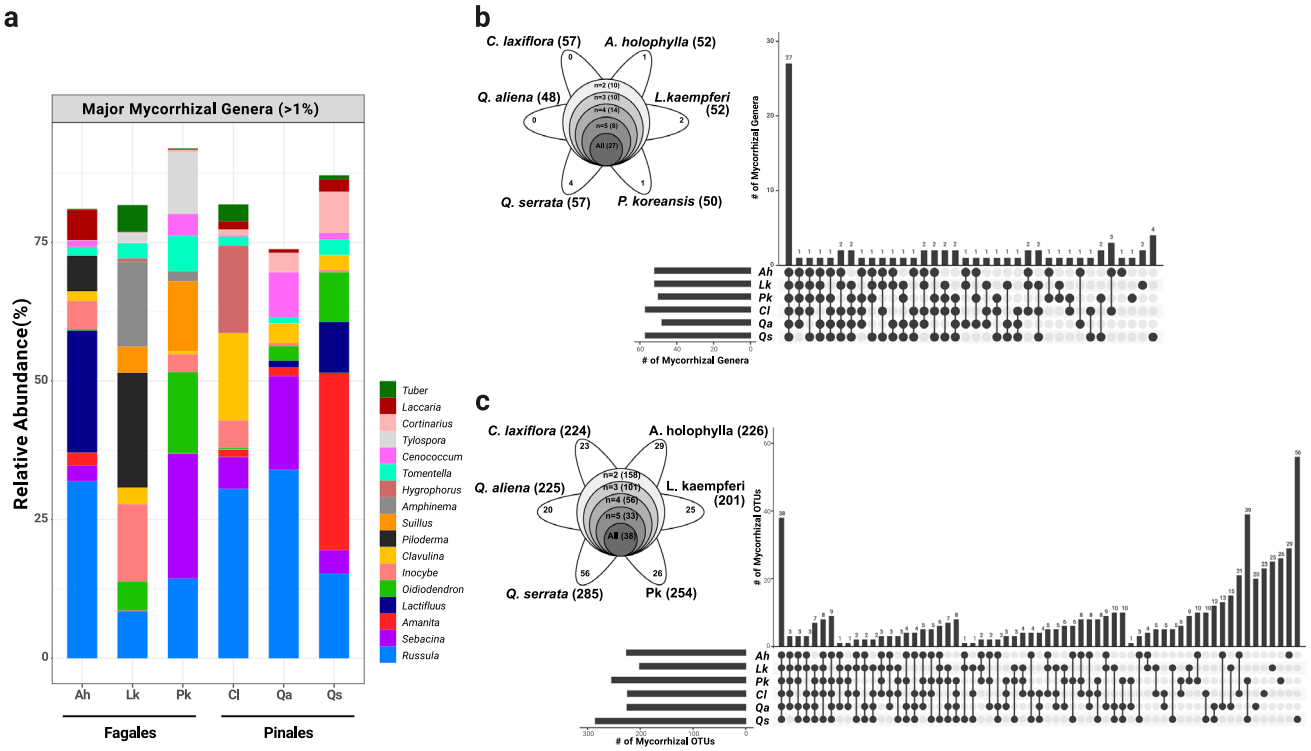

Fig. 1 **A** Genus-level composition of major mycorrhizal fungi (> 1%) associated with six host trees analyzed in this study. The average relative abundance of sequence reads is shown for each tree species. Venn-diagram and Upset plot of generalist and host-specific mycorrhizal **B** genera and **C** OTUs. The total number of mycorrhizal genera for each host tree is described on the bottom left, and overlapping genera are marked with dark circles and a bar on the bottom. *Ah Abies holophylla*; *Lk Larix kaempferi*; *Pk Pinus koreansis*; *Cl Carpinus laxiflora*; *Qa Quercus aliena*; *Qs Quercus serrata*

Host Specificty of Mycorrhizal Communities Between Tree Species

At the genus level, 27 mycorrhizal genera were detected from all six trees, including the most dominant mycorrhizal genera: *Russula*, *Sebacina*, *Amanita*, *Lactifluus*, and *Oidiodendron*. More than a third of mycorrhizal fungal genera overlapped between the six tree species (Fig. 1B), and a fourth or fifth of the genera were found from all tree species when the data was separated by season. In contrast, the number of tree-specific genera was generally low (Fig. S3). Only eight genera were detected explicitly from a single tree species: Qs–*Ramaria, Sarcodon, Asterophora*, and *Coltricia*, Lk–*Wilcoxina* and *Odontia*, Pk–*Membranomyces*, and Ah–*Claroideoglomus* (Fig. 1B). For each season, the number of Qs-specific mycorrhizal genera was higher than that of the other hosts, except for winter (Fig. S3).

There were more tree-specific OTUs than the tree-specific genera (Fig. 1C, Fig. S4). Regarding frequency, only 38 OTUs were found commonly in all six trees. They were EcM and ErM OTUs, including *Cenococcum geophilum* (OTU 41), *Acephala applanate* (OTU 3399), *Pezoloma ericae* (OTU 1361), *Laccaria laccata* (OTU 469), *Astraeus hygrometricus* (OTU 94), and *Oidiodendron chlamydosporicum* (OTU 10). Several AM OTUs (identified as *Archaeospora* sp. and Glomeraceae sp.) were also found in all host species, but they were of lower abundance than that of the EcM or ErM. A total of 179 tree-specific OTUs were found, and the number of Qs-specific OTUs was higher than that of the other trees: 29 (Ah), 23 (Cl), 25 (Lk), 26 (Pk), 20 (Qa), and 56 (Qs). Some tree-specific OTUs included *Cortinarius rigidipes* (Ah), *Lactarius* aff. *decipiens* (Cl), *Inocybe ericetorum* (Lk), *Inocybe straminipes* (Pk), *Tricholoma atroviolaceum* (Qa), and *Lactarius* cf. *chrysorrheus* (Qs).

Indicator species analysis showed that the number of Pk-specific OTUs (20 OTUs) was greater than that of the other host trees (3–7 OTUs). *Suillus pictus* (OTU 40), *Tuber pseudosphaerosporum* (OTU 43), *Inocybe ericetorum* (OTU 251), *Sebacina* sp. (OTU 6), and *Oidiodendron echinulatum* (OTU 181) were identified as Pk-specific OTUs (Table 2). For the other host tree species, *Amanita pantherina* (OTU 100) and *Russula densifolia* (OTU 26) preferred Qs, while *Cenococcum geophilum* (OTU 456), *Russula risigallina* (OTU 12,127), and *Lactarius quietus* (OTU 531) preferred Qa. The mycorrhizal OTUs in the same genera, such as those of *Sebacina*, *Cenococcum*, and *Oidiodendron*, were often found as host-specific OTUs of different tree species in indicator species analysis (Table 2).

Effect of Host Tree Species and Environmental Factors on Mycorrhizal Community

The influence of tree species on the mycorrhizal community was affirmed in PERMANOVA and pairwise PERMANOVA tests, which showed a significant impact on the overall community ($R^2 = 0.24$, $p = 0.001$), and between individual tree species ($R^2 = 0.16$–0.33, $p = 0.001$). A similar result was observed in UPGMA tree (Fig. 2A and B). The influence of tree order was also significant, but slightly lower than that of between tree species ($R^2 = 0.05$, $p = 0.001$). Mycorrhizal communities from the same host were more likely to be clustered. Although mycorrhizal communities from several hosts were clearly grouped by the host identity (Cl, Lk, Pk, Qs), those from the other hosts did not form a definite clustering (Ah, Qa). The mycorrhizal communities were significantly affected by the host trees when the sampling year was in control ($p = 0.001$, Fig. S5).

The effects of the seasons and soil properties on the mycorrhizal communities differed by tree species (Table 3, Fig. S6). The season was the most substantial factor in the mycorrhizal communities, except for those in Lk. For the other environmental factors, pH, TP, and C/N ratio were the dominant factors in more than half of host species, but their influence differed by tree species. Interestingly, mycorrhizal fungi found in the Lk site soil were not influenced by any environmental factors. The tree species, seasons, and soil properties each explained 31.5%, 4.0%, and 14.7% of the variance in the mycorrhizal communities, respectively (Fig. 2C). Overall, the tree identity had the highest contribution to variance (21.0%), followed by the soil property (3.9%) and seasons (3.7%).

Discussion

In this study, we found that host specificity was a significant factor in the determination of mycorrhizal community composition. The pronounced effect of the host tree was predominantly found at the OTU level rather than the genus level. Only a few OTUs were detected across all six tree species, and more generalists were found at the genus level. The evident influence of host tree identity on mycorrhizal fungi is consistent with previous studies that reported on the effect of host plants on the differential mycorrhizal communities (Rincón et al., 2015; Tedersoo et al., 2014; Wu et al., 2018).

Mycorrhizal communities featured different co-occurrence patterns in the genus and OTU levels. Most mycorrhizal genera were shared between host trees, and the complete list of mycorrhizal genera was consistent with the results from previous studies conducted in various host plant groups (Gong et al., 2022; Deng et al., 2019; Wang et al., 2019), as well as our previous studies conducted from the same

Table 2 List of host-specific OTUs obtained by indicator species analysis

Type	Host	OTU (Identity)	Indicator value	*p* value	Relative abundance (%)
Pinales	*Abies holophylla*	OTU 123 (*Piloderma* sp)	0.550	0.029	0.831
		OTU 159 (*Trichophaea* sp)	0.520	0.044	0.161
		OTU 141 (*Amanita longistriata*)	0.371	0.029	0.134
	Larix kaempferi	OTU 5 (*Piloderma* sp)	0.876	0.001	3.449
		OTU 60 (*Trichophaea* sp)	0.826	0.001	0.778
		OTU 297 (*Tuber* sp)	0.671	0.017	0.115
		OTU 112 (*Oidiodendron griseum*)	0.732	0.003	0.304
		OTU 21 (*Amphinema* sp)	0.713	0.004	1.518
		OTU 338 (*Inocybe* sp)	0.518	0.029	0.131
		OTU 543 (*Tomentella sublilacina*)	0.460	0.027	0.104
	Pinus koreansis	OTU 6 (*Sebacina* sp)	0.725	0.002	1.577
		OTU 66 (*Oidiodendron griseum*)	0.750	0.003	0.423
		OTU 69 (*Meliniomyces* sp)	0.718	0.003	0.311
		OTU 17 (*Suillus placidus*)	0.604	0.005	0.858
		OTU 181 (*Oidiodendron echinulatum*)	0.627	0.006	0.248
		OTU 87 (*Tomentella* sp)	0.624	0.007	0.225
		OTU 103 (*Oidiodendron* sp)	0.517	0.028	0.169
		OTU 40 (*Suillus pictus*)	0.594	0.031	0.916
		OTU 72 (*Sebacina incrustans*)	0.416	0.032	0.646
		OTU 7 (*Tylospora* sp)	0.560	0.032	2.153
		OTU 16,517 (*Sebacina incrustans*)	0.497	0.035	0.170
		OTU 51 (*Inocybe* sp)	0.398	0.041	0.491
		OTU 162 (*Inocybe* sp)	0.608	0.003	0.164
		OTU 43 (*Tuber pseudosphaerosporum*)	0.576	0.030	0.711
		OTU 224 (*Amphinema* sp)	0.487	0.027	0.110
		OTU 83 (*Amphinema* sp)	0.482	0.031	1.094
		OTU 3411 (*Pezoloma ericae*)	0.472	0.030	0.522
		OTU 2 (Thelephoraceae sp)	0.436	0.032	0.158
		OTU 114 (*Pseudotomentella tristis*)	0.324	0.031	0.177
		OTU 251 (*Inocybe ericetorum*)	0.304	0.044	0.107
Fagales	*Carpinus laxiflora*	OTU 82 (*Tuber* sp)	0.627	0.007	0.616
		OTU 25 (*Hygrophorus* sp)	0.586	0.032	2.622
		OTU 143 (*Sebacina* sp)	0.564	0.032	0.168
		OTU 235 (*Helvellosebacina* sp)	0.523	0.036	0.255
		OTU 149 (*Sebacina* sp)	0.482	0.030	0.130
	Quercus aliena	OTU 456 (*Cenococcum* sp)	0.505	0.030	0.387
		OTU 322 (Pezizaceae sp)	0.498	0.030	0.412
		OTU 12,127 (*Russula risigallina*)	0.448	0.039	0.154
		OTU 531 (*Lactarius quietus*)	0.316	0.046	0.146
	Quercus serrata	OTU 42 (*Russula* sp)	0.700	0.005	0.862
		OTU 46 (*Sebacina* sp)	0.666	0.004	0.507
		OTU 1 (*Amanita* sp)	0.636	0.005	3.277
		OTU 100 (*Amanita pantherina*)	0.479	0.026	0.329
		OTU 247 (*Cortinarius* sp)	0.474	0.029	0.140
		OTU 26 (*Russula densifolia*)	0.427	0.034	0.776
Conifer + Conifer	Ah + Pk	OTU 19 (*Russula violeipes*)	0.608	0.012	2.617
	Lk + Pk	OTU 3399 (*Acephala applanata*)	0.629	0.012	0.119
		OTU 11,827 (*Oidiodendron maius*)	0.470	0.041	0.163
Conifer + Broadleaf	Pk + Qa	OTU 41 (*Cenococcum geophilum*)	0.541	0.018	0.917
	Pk + Qs	OTU 10 (*Oidiodendron chlamydosporicum*)	0.839	0.000	2.519
	Lk + Qs	OTU 45 (*Oidiodendron maius*)	0.598	0.013	0.766
	Lk + Pk + Qs	OTU 68 (*Oidiodendron echinulatum*)	0.511	0.038	0.303

Table 2 (continued)

Only major OTUs (> 0.1%) were described

Ah Abies holophylla; *Lk Larix kaempferi*; *Pk Pinus koreansis*; *Cl Carpinus laxiflora*; *Qa Quercus aliena*; *Qs Quercus serrata*

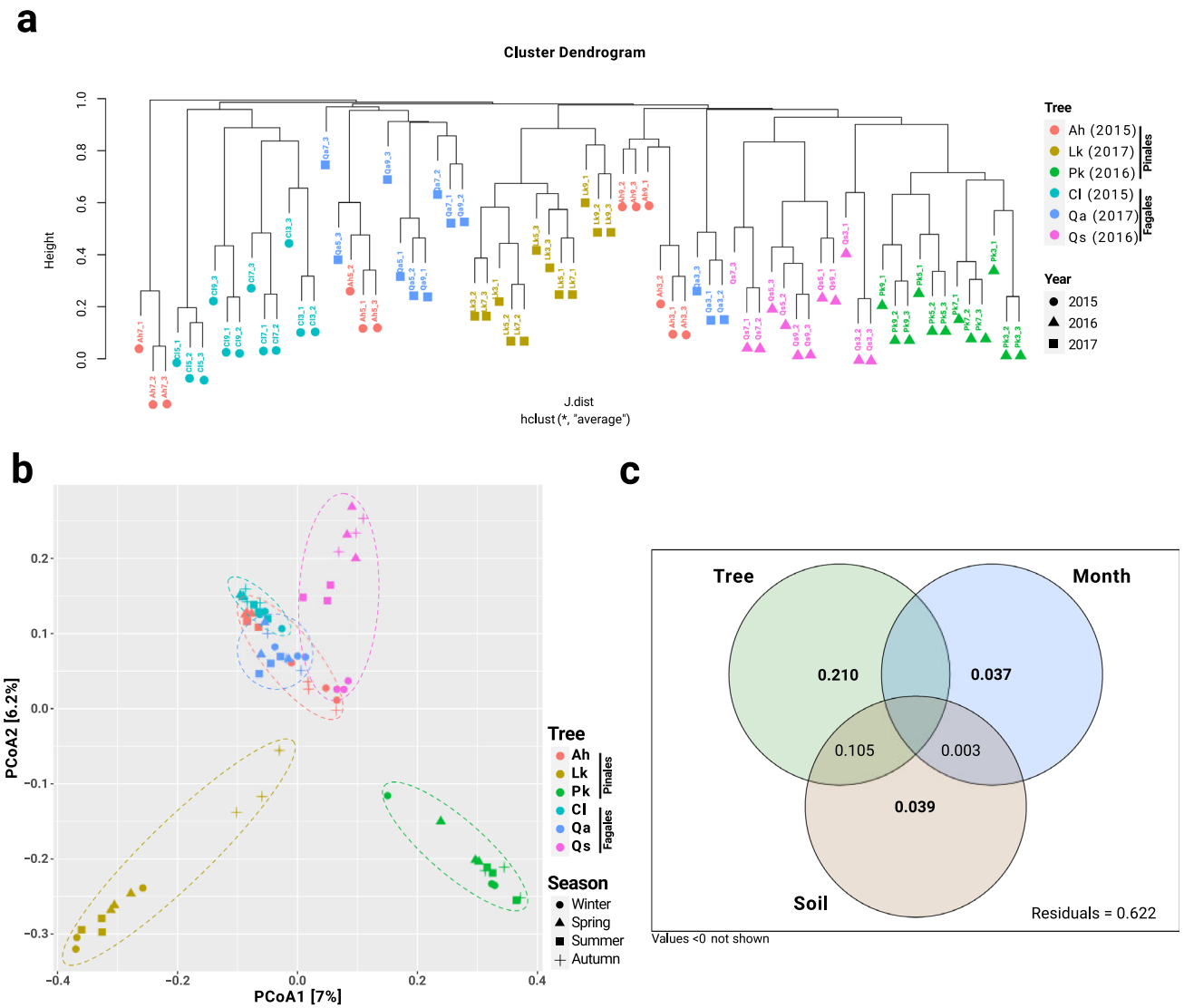

Fig. 2 **A** UPGMA dendrogram of mycorrhizal communities. **B** Principal coordinate analysis (PCoA) ordination of the mycorrhizal community structure associated with six host trees in this study. **C** Result of variation partitioning analysis showing the effect of host tree species, soil property, and season. Proportions of the explained variations are indicated with numbers (adjusted R^2 values). *Ah Abies holophylla*; *Lk Larix kaempferi*; *Pk Pinus koreansis*; *Cl Carpinus laxiflora*; *Qa Quercus aliena*; *Qs Quercus serrata*

area (Oh et al., 2018; Park et al., 2020). *Russula* was identified as the most dominant host-generalist genus, followed by *Sebacina* and *Oidiodendron*. The most mycorrhizal genera were detected from more than half of the tree species assessed in our study. For instance, *Lactifluus*, *Piloderma*, and *Russula* were found in multiple hosts. Though, indicator species analysis revealed possible preferences for a tree species from EcM genera in terms of relative abundance (*Amanita* for Qs, *Lactifluus* for Ah and Qs, *Piloderma* for Ah and Lk, and *Clavulina* for Cl).

At the OTU level, however, high host-specificity was found in indicator species analysis. *Lactifluus bertillonii* (OTU 12) was dominant in Ah, while *Lf. acicularis* (OTU 52) constituted a large proportion of the mycorrhizal community for Qs. This pattern aligned with previous observations from the field study — the fruiting bodies of *Lf.*

Table 3 The effect of seasons and soil properties on mycorrhizal communities detected by PERMANOVA tests

Variables	Pinales			Fagales		
	A. holophylla	*P. koraiensis*	*L. kaempferi*	*C. laxiflora*	*Q. aliena*	*Q. serrata*
pH	0.241***	0.188***	0.083	0.154*	0.103	0.093
TOC (%)	0.03	0.141	0.069	0.116	0.087	0.170***
TN (%)	0.088	0.14	0.068	0.111	0.097	0.17
C/N	0.176*	0.147*	0.063	0.125	0.193**	0.227
TP (mg/kg)	0.186*	0.190*	0.071	0.196***	0.196**	0.113
Water (%)	0.183**	0.101	0.066	0.109	0.092	0.259***
Season	0.657***	0.724***	0.305	0.598***	0.397**	0.582***

*, **, and *** indicates significance (p value < 0.05, 0.01, or 0.001, respectively)

pilosus and *Lf. acicularis* were frequently found in the Ah and Qs sites, respectively, during summer (Lee et al., 2018). Similarly, different host-specificity of congeneric fungal species was reported in a bioassay test for *S. glandulosus* and *S. punctatipes* against multiple host species (Pérez-Pazos et al., 2021). This results indicate possible species level host-specificity of mycorrhizal fungi.

While all hosts were ectomycorrhizal trees, other types of mycorrhizal fungi (AM, OM, and ErM) were also found in small proportions. Similar results were reported in previous studies, where AM or ErM was detected from the root of ectomycorrhizal trees (Park et al., 2020; Toju & Sato, 2018). While we could not detect AM fungi in *P. densiflora* (Lee & Eom, 2013; Park et al., 2021), a small proportion of AM fungi was detected from a congeneric *P. koreansis*, highlighting the importance of host-specificity in host-symbiont associations. The only ErM fungi, *Oidiodendron* was found abundantly in our study and is known to be frequently detected in non-ericaceous hosts (Park et al., 2020; Zhao et al., 2020). They could be associated with other tree species as soil samples were used in this study, but they might act as root endophytes in the absence of other ErM species (Abuzinadah & Read, 1989; Martino et al., 2018). The host preference of these species aligned with those reported in previous studies (Huang et al., 2018; Rasmussen et al., 2018). For a deeper understanding of host-symbiont fungi, more intensive species-level analysis and further improvements in the current database are necessary (Kõljalg et al., 2013; Nguyen et al., 2016a; Peay, 2014; Põlme et al., 2020).

The season and the soil properties were significant factors in determining mycorrhizal community composition and diversity in most previous studies. We observed a significant influence of both variables on the mycorrhizal communities in this study, which is consistent with that of most other EcM trees (Santalahti et al., 2016; Voříškova et al., 2014). As EcM species often have different type of nutrition acquisition strategy following their exploration type (Defrenne et al., 2019) and genome (Ryberg et al., 2022), differences in major EcM species between trees could affect their response to environmental factors. pH, TP, and C/N ratio affected the mycorrhizal community composition of most tree species, while TOC and water contents significantly influenced the mycorrhizal community in only a few tree species, and TN was not a significant factor in all of tree species in our study. There have been reports indicating its significant influence of pH on the forest soil microbial communities, including EcM fungi (Lladó et al., 2018). In the case of phosphorus, it is known that EcM diversity plays an important role in phosphorus absorption in soil (Köhler et al., 2018). C/N ratio, on the other hand, can be influenced by nitrogen uptake by mycorrhizal fungi (Kuyper, 2017). However, depending on the tree species or niche, the influence of soil factors on fungal communities can vary. For instance, nitrogen is a crucial factor in mycorrhizal fungal communities (Cox et al., 2010). In contrast, the non-significant relationship between TN and mycorrhizal fungal communities was found in our study. Meanwhile, in Lk, the mycorrhizal community composition was not significantly affected by the seasons or the soil properties. A similar result was found in previous studies for Lk in a subtropical forest (Matsuoka et al., 2016) and *Carpinus cordata* in a temperate forest (Park et al., 2020).

In conclusion, our results revealed the mycorrhizal community composition associated with less studied ectomycorrhizal tree species in a preserved forest of Mt. Jeombong. We detected a high diversity of mycorrhizal fungi composed mainly of EcM fungi and a small portion of ErM, AM, and OM fungi. Most mycorrhizal genera were shared among the six studied tree species. At the OTU level, however, only a small number of OTUs were shared among the tree species, and most were host species-specific. Our study provides valuable data on mycorrhizal fungal diversity associated with native trees in the Republic of Korea. Our investigation of mycorrhizal diversity would enable better forest management capabilities in the future.

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Data Availability Raw sequence data is available in NCBI Sequence Read Archive (SRA) under Project ID PRJNA937676.

Declarations

Conflict of Interest The authors declare no competing interests.

Ethical Approval This study does not contain any experiments with animal or human participants.

References

Abuzinadah, R. A., & Read, D. J. (1989). The role of proteins in the nitrogen nutrition of ectomycorrhizal plants. *New Phytologist*, *112*, 61–68.

Arbizu, P. M. (2017). pairwiseAdonis: Pairwise multilevel comparison using adonis. *R package version 0.4*. https://github.com/pmartinezarbizu/pairwiseAdonis.

Bahram, M., Põlme, S., Kõljalg, U., & Tedersoo, L. (2011). A single european aspen (*Populus tremula*) tree individual may potentially harbour dozens of *Cenococcum geophilum* ITS genotypes and hundreds of species of ectomycorrhizal fungi. *FEMS Microbiology Ecology*, *75*, 313–320.

Brundrett, M. C., & Tedersoo, L. (2018). Evolutionary history of mycorrhizal symbioses and global host plant diversity. *New Phytologist*, *220*, 1108–1115.

Bruns, T. D., Bidartondo, M. I., & Taylor, D. L. (2002). Host specificity in ectomycorrhizal communities: What do the exceptions tell us? *Integrative and Comparative Biology*, *42*, 352–359.

Caporaso, J. G., Kuczynski, J., Stombaugh, J., Bittinger, K., Bushman, F. D., Costello, E. K., Fierer, N., Peña, A. G., Goodrich, J. K., Gordon, J. I., et al. (2010). QIIME allows analysis of high-throughput community sequencing data. *Nature Methods*, *7*, 335–336.

Choung, Y., Lee, J., Cho, S., & Noh, J. (2020). Review on the succession process of *Pinus densiflora* forests in South Korea: Progressive and disturbance-driven succession. *Journal of Ecology and Environment*, *44*, 16.

Conway, J. R., Lex, A., & Gehlenborg, N. (2017). UpSetR: An R package for the visualization of intersecting sets and their properties. *Bioinformatics*, *33*, 2938–2940.

Cox, F., Barsoum, N., Lilleskov, E. A., & Bidartondo, M. I. (2010). Nitrogen availability is a primary determinant of conifer mycorrhizas across complex environmental gradients. *Ecology Letters*, *13*, 1103–1113.

De Cáceres, M., & Legendre, P. (2009). Associations between species and groups of sites: Indices and statistical inference. *Ecology*, *90*, 3566–3574.

Defrenne, C. E., Philpott, T. J., Guichon, S. H. A., Roach, W. J., Pickles, B. J., & Simard, S. W. (2019). Shifts in ectomycorrhizal fungal communities and exploration types relate to the environment and fine-root traits across interior Douglas-fir forests of western Canada. *Frontiers in Plant Science*, *10*, 643.

Deng, J., Yin, Y., Luo, J., Zhu, W., & Zhou, Y. (2019). Different revegetation types alter soil physical-chemical characteristics and fungal community in the Baishilazi Nature Reserve. *PeerJ*, *6*, e6251.

Frøslev, T. G., Kjøller, R., Bruun, H. H., Ejrnæs, R., Brunbjerg, A. K., Pietroni, C., & Hansen, A. J. (2017). Algorithm for post-clustering curation of DNA amplicon data yields reliable biodiversity estimates. *Nature Communications*, *8*, 1188.

Gao, C., Shi, N. N., Liu, Y. X., Peay, K. G., Zheng, Y., Ding, Q., Mi, X. C., Ma, K. P., Wubet, T., Buscot, F., et al. (2013). Host plant genus-level diversity is the best predictor of ectomycorrhizal fungal diversity in a chinese subtropical forest. *Molecular Ecology*, *22*, 3403–3414.

Genre, A., Lanfranco, L., Perotto, S., & Bonfante, P. (2020). Unique and common traits in mycorrhizal symbioses. *Nature Reviews Microbiology*, *18*, 649–660.

Gong, S., Feng, B., Jian, S. P., Wang, G. S., Ge, Z. W., & Yang, Z. L. (2022). Elevation matters more than season in shaping the heterogeneity of soil and root associated ectomycorrhizal fungal community. *Microbiology Spectrum*, *10*, e01950–e01921.

Högberg, M. N., & Högberg, P. (2002). Extramatrical ectomycorrhizal mycelium contributes one-third of microbial biomass and produces, together with associated roots, half the dissolved organic carbon in a forest soil. *New Phytologist*, *154*, 791–795.

Huang, H., Zi, X. M., Lin, H., & Gao, J. Y. (2018). Host-specificity of symbiotic mycorrhizal fungi for enhancing seed germination, protocorm formation and seedling development of over-collected medicinal orchid, *Dendrobium devonianum*. *Journal of Microbiology*, *56*, 42–48.

Köhler, J., Yang, N., Pena, R., Raghavan, V., Polle, A., & Meier, I. C. (2018). Ectomycorrhizal fungal diversity increases phosphorus uptake efficiency of european beech. *New Phytologist*, *220*, 1200–1210.

Kõljalg, U., Nilsson, R. H., Abarenkov, K., Tedersoo, L., Taylor, A. F., Bahram, M., Bates, S. T., Bruns, T. D., Bengtsson-Palme, J., Callaghan, T. M., et al. (2013). Towards a unified paradigm for sequence-based identification of fungi. *Molecular Ecology*, *22*, 5271–5277.

Korea Forest Research Institute, KFRI. (2016). *Annual Report of Monitoring for Forest Insect Pests and Diseases in Korea*. Seoul, Republic of Kore: Korea Forest Research Institute.

Kuyper, T. W. (2017). Carbon and Energy Sources of Mycorrhizal Fungi: Obligate Symbionts or Latent Saprotrophs? In N. C. Johnson, C. Gehring, & J. Jansa (Eds.), *Mycorrhizal Mediation of Soil: Fertility Structure and Carbon Storage* (pp. 357–374). Cambridge, Massachusetts, USA: Elsevier.

Lee, E. H., & Eom, A. H. (2013). Ectomycorrhizal fungal communities of red pine (*Pinus densiflora*) seedlings in disturbed sites and undisturbed old forest sites. *Mycobiology*, *41*, 77–81.

Lee, H., Park, J. Y., Wisitrassameewong, K., Kim, M. J., Park, M. S., Kim, N. K., Lee, J. K., & Lim, Y. W. (2018). First report of eight milkcap species belonging to Lactarius and Lactifluus in Korea. *Mycobiology, 46*, 1–12.

Lladó, S., López-Mondéjar, R., & Baldrian, P. (2018). Drivers of microbial community structure in forest soils. *Applied Microbiology and Biotechnology*, *102*, 4331–4338.

Lofgren, L. A., Nguyen, N. H., Vilgalys, R., Ruytinx, J., Liao, H. L., Branco, S., Kuo, A., LaButti, K., Lipzen, A., Andreopoulos, W., et al. (2021). Comparative genomics reveals dynamic genome evolution in host specialist ectomycorrhizal fungi. *New Phytologist*, *230*, 774–792.

Martino, E., Morin, E., Grelet, G. A., Kuo, A., Kohler, A., Daghino, S., Barry, K. W., Cichocki, N., Clum, A., Dockter, R. B., et al. (2018). Comparative genomics and transcriptomics depict ericoid mycorrhizal fungi as versatile saprotrophs and plant mutualists. *New Phytologist*, *217*, 1213–1229.

Matsuoka, S., Kawaguchi, E., & Osono, T. (2016). Temporal distance decay of similarity of ectomycorrhizal fungal community composition in a subtropical evergreen forest in Japan. *FEMS Microbiology Ecology*, *92*, fiw061.

McMurdie, P. J., & Holmes, S. (2013). Phyloseq: An R package for reproducible interactive analysis and graphics of microbiome census data. *Plos One*, *8*, e61217.

Nguyen, N. H., Song, Z., Bates, S. T., Branco, S., Tedersoo, L., Menke, J., Schilling, J. S., & Kennedy, P. G. (2016a). FUNGuild: An open annotation tool for parsing fungal community datasets by ecological guild. *Fungal Ecology*, *20*, 241–248.

Nguyen, N. H., Vellinga, E. C., Bruns, T. D., & Kennedy, P. G. (2016b). Phylogenetic assessment of global *suillus* ITS sequences supports morphologically defined species and reveals synonymous and undescribed taxa. *Mycologia*, *108*, 1216–1228.

Obase, K., Cha, J. Y., Lee, J. K., Lee, S. Y., Lee, J. H., & Chun, K. W. (2009). Ectomycorrhizal fungal communities associated with *Pinus thunbergii* in the eastern coastal pine forests of Korea. *Mycorrhiza*, *20*, 39–49.

Oh, S. Y., Cho, H. J., Eimes, J. A., Han, S. K., Kim, C. S., & Lim, Y. W. (2018). Guild patterns of basidiomycetes community associated with *Quercus mongolica* in Mt. Jeombong, Republic of Korea. *Mycobiology*, *46*, 13–23.

Oksanen, J., Blanchet, F. G., Kindt, R., Legendre, P., Minchin, P. R., O'Hara, B., Simpson, G. L., Leslie, G., Solymos, P., Stevens, H. (2013). The vegan R package version 2.0–8: community ecology. Available at: http://CRAN.R-project.org/package=vegan.

Park, K. H., Oh, S. Y., Yoo, S., Fong, J. J., Kim, C. S., Jo, J. W., & Lim, Y. W. (2020). Influence of season and soil properties on fungal communities of neighboring climax forests (*Carpinus cordata* and *Fraxinus rhynchophylla*). *Frontiers in Microbiology*, *11*, 572706.

Park, K. H., Yoo, S., Park, M. S., Kim, C. S., & Lim, Y. W. (2021). Different patterns of belowground fungal diversity along altitudinal gradients with respect to microhabitat and guild types. *Environmental Microbiology Reports*, *13*, 649–658.

Peay, K. G. (2014). Back to the future: Natural history and the way forward in modern fungal ecology. *Fungal Ecology*, *12*, 4–9.

Pérez-Pazos, E., Certano, A., Gagne, J., Lebeuf, R., Siegel, N., Nguyen, N., & Kennedy, P. G. (2021). The slippery nature of ectomycorrhizal host specificity: *Suillus* fungi associated with novel pinoid (*Picea*) and abietoid (*Abies*) hosts. *Mycologia*, *113*, 891–901.

Põlme, S., Abarenkov, K., Henrik Nilsson, R., Lindahl, B. D., Clemmensen, K. E., Kauserud, H., Nguyen, N., Kjøller, R., Bates, S. T., Baldrian, P., et al. (2020). FungalTraits: A user-friendly traits database of fungi and fungus-like stramenopiles. *Fungal Diversity*, *105*, 1–16.

R Core Team. (2021). *R: A Language and Environment for Statistical Computing*. Vienna, Austria: R Foundation for Statistical Computing.

Rasmussen, A. L., Busby, R. R., & Hoeksema, J. D. (2018). Host preference of ectomycorrhizal fungi in mixed pine-oak woodlands. *Canadian Journal of Forest Research*, *48*, 153–159.

Rincón, A., Santamaría-Pérez, B., Rabasa, S. G., Coince, A., Marçais, B., & Buée, M. (2015). Compartmentalized and contrasted response of ectomycorrhizal and soil fungal communities of Scots pine forests along elevation gradients in France and Spain. *Environmental Microbiology*, *17*, 3009–3024.

Rochet, J., Moreau, P. A., Manzi, S., & Gardes, M. (2011). Comparative phylogenies and host specialization in the alder ectomycorrhizal fungi Alnicola, Alpova and Lactarius (Basidiomycota) in Europe. *BMC Evolutionary Biology, 11*, 40.

Rognes, T., Flouri, T., Nichols, B., Quince, C., & Mahé, F. (2016). VSEARCH: A versatile open source tool for metagenomics. *PeerJ*, *4*, e2584.

Ryberg, M., Kalsoom Khan, F., & Sánchez-García, M. (2022). On the evolution of ectomycorrhizal fungi. *Mycosphere*, *13*, 1–12.

Santalahti, M., Sun, H., Jumpponen, A., Pennanen, T., & Heinonsalo, J. (2016). Vertical and seasonal dynamics of fungal communities in boreal Scots pine forest soil. *FEMS Microbiology Ecology*, *92*, fiw170.

Smith, S., & Read, D. (2013).*Mycorrhizal symbiosis*. Cambridge: Cambridge University Press.

Smith, M. E., Douhan, G. W., Fremier, A. K., & Rizzo, D. M. (2009). Are true multihost fungi the exception or the rule? Dominant ectomycorrhizal fungi on *Pinus sabiniana* differ from those on co-occurring *Quercus* species. *New Phytologist*, *182*, 295–299.

Tedersoo, L., Jairus, T., Horton, B. M., 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.

Tedersoo, L., Bahram, M., Põlme, S., Kõljalg, U., Yorou, N. S., Wijesundera, R., Villarreal Ruiz, L., Vasco-Palacios, A. M., Thu, P. Q., et al. (2014). Fungal biogeography. Global diversity and geography of soil fungi. *Science*, *346*, 1256688.

Toju, H., & Sato, H. (2018). Root-associated fungi shared between arbuscular mycorrhizal and ectomycorrhizal conifers in a temperate forest. *Frontiers in Microbiology*, *9*, 433.

Vaario, L. M., Yang, X., & Yamada, A. (2017). Biogeography of the japanese gourmet fungus, Tricholoma matsutake: A review of the distribution and functional ecology of matsutake. In L. Tedersoo (Ed.), *Biogeography of Mycorrhizal Symbiosis* (pp. 319–344). Berlin: Springer.

van der Linde, S., Suz, L. M., Orme, C. D. L., Cox, F., Andreae, H., Asi, E., Atkinson, B., Benham, S., Carroll, C., Cools, N., et al. (2018). Environment and host as large-scale controls of ectomycorrhizal fungi. *Nature*, *558*, 243–248.

Voříšková, J., Brabcová, V., Cajthaml, T., & Baldrian, P. (2014). Seasonal dynamics of fungal communities in a temperate oak forest soil. *New Phytologist*, *201*, 269–278.

Wang, Y. L., Gao, C., Chen, L., Ji, N. N., Wu, B. W., Li, X. C., Lü, P. P., Zheng, Y., & Guo, L. D. (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, T. J., Bruns, T., Lee, S., & Taylor, J. (1990). Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In M. A. Innis, D. H. Gelfand, J. J. Shinsky, & T. J. White (Eds.), *PCR protocols: A guide to methods applications* (pp. 315–322). Cambridge: Academic Press.

Wu, B. W., Gao, C., Chen, L., Buscot, F., Goldmann, K., Purahong, W., Ji, N. N., Wang, Y. L., Lü, P. P., Li, X. C., et al. (2018). Host phylogeny is a major determinant of Fagaceae-associated ectomycorrhizal fungal community assembly at a regional scale. *Frontiers in Microbiology*, *9*, 2409.

Yang, T., Tedersoo, L., Fu, X., Zhao, C., Liu, X., Gao, G., Cheng, L., Adams, J. M., & Chu, H. (2021). Saprotrophic fungal diversity predicts ectomycorrhizal fungal diversity along the timberline in the framework of island biogeography theory. *ISME Communications*, *1*, 15.

Yang, N., Hua, J., Zhang, J., Liu, D., Bhople, P., Li, X., Zhang, Y., Ruan, H., Xing, W., & Mao, L. (2022). Soil nutrients and plant diversity affect ectomycorrhizal fungal community structure and functional traits across three subalpine coniferous forests. *Frontiers in Microbiology*, *13*, 1016610.

Yoo, S., Cho, Y., Park, K. H., & Lim, Y. W. (2022). Exploring fine-scale assembly of ectomycorrhizal fungal communities through phylogenetic and spatial distribution analyses. *Mycorrhiza*, *32*, 439–449.

Yu, Y., Yang, Q., Petropoulos, E., & Zhu, T. (2022). ITS3/ITS4 outperforms other ITS region and 18S rRNA gene primer sets for amplicon sequencing of soil fungi. *European Journal of Soil Science*, *73*, e13329.

Zhang, J., Kobert, K., Flouri, T., & Stamatakis, A. (2014). PEAR: A fast and accurate Illumina paired-end reAd mergeR. *Bioinformatics*, *30*, 614–620.

Zhao, P. S., Guo, M. S., Gao, G. L., Zhang, Y., Ding, G. D., Ren, Y., & Akhtar, M. (2020). Community structure and functional group of root-associated Fungi of *Pinus sylvestris* var.

Mongolica across stand ages in the Mu us Desert. *Ecology and Evolution*, *10*, 3032–3042.