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ORIGINAL RESEARCH

Fungal diversity in wood of living trees is higher in oak than in beech, maple or linden, and is affected by tree size and climate

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Abstract
Fungi are abundant in wood of living trees, but few studies have compared the diversity of fungi among different tree species and trees of varying age and size, aspects of importance for conservation planning. We investigated if fungal species richness and species composition in wood vary significantly among the temperate broadleaf tree species beech (*Fagus sylvatica*), linden (*Tilia cordata*), Norway maple (*Acer platanoides*) and pedunculate oak (*Quercus robur*). Each tree species was represented by four stem size classes, and the total sample included 240 trees in southern Norway. Wood cores were collected from individual trees and fungal DNA was amplified using ITS2 rRNA as a marker and subjected to high-throughput sequencing. In total, we detected 1156 fungal OTUs. Oaks had significantly higher richness of fungal OTUs than any of the other tree species and harboured unique communities. Further, oak hosted most species-specific Indicator species (39) and was the only species to host Red-Listed fungal species (five). The circumference (proxy for age) did not significantly affect neither OTU richness nor its overall composition. However, several individual Red List and Indicator species were found only in trees of the largest size class. There was a significant effect of bioclimatic section on species composition. Our results emphasize the important roles of oaks and to some extent large trees as repositories of fungal diversity, which should be considered in conservation planning.

Keywords Biodiversity · Metabarcoding · Ascomycota · Basidiomycota · Temperate broadleaf trees

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Introduction

Large trees are crucial for biodiversity and a multitude of species are dependent on them (Lindenmayer et al. 2012). Accordingly, knowledge about diversity patterns of associated species such as species richness, species composition and the occurrence of red-listed species in various trees is essential for conservation strategies (Prevedello et al. 2018). Many tree species are currently declining, and ancient trees of various species have become rare worldwide (Lindenmayer et al. 2012; Jacobsen et al. 2023) leaving the remaining large and old trees with a disproportionally high value for biodiversity (Bowd et al. 2025). In addition, certain forest types have declined, and for example temperate broadleaf forest has historically lost much ground to agriculture, forestry and other human activities in large parts of Europe (Lindbladh et al. 2014). In many countries in northern Europe, temperate broadleaf forest currently constitutes only a small part of the forested area (Granhus et al. 2012) but has the highest share of red-listed species per area (Henriksen and Hilmo 2015). Thus, both large and old trees and temperate broadleaf forest may be important objects for the study of diversity patterns and conservation priorities, especially concerning taxa that have been underrepresented in previous studies.

A prominent part of the species communities associated with trees consist of the functionally important and mega-diverse fungi (Antonelli et al. 2023; Bowd et al. 2025). Rich fungal communities are associated with bark (e.g. epiphytic lichenized fungi; Smith 2009), roots (e.g. mycorrhizal fungi; Knudsen and Vesterholt 2012) and dead wood (saprotrophic fungi, Nordén et al. 2004). Fungi are also abundant within healthy tissues of trees and may have beneficial effects in living trees such as increased stress tolerance, protection against herbivores and pathogens, and other health- and growth-promoting effects (Terhonen et al. 2019; Sodhi & Saxena 2023). However, fungal communities in living tissues have rarely been studied, leaving their diversity poorly understood. One of the least explored microhabitats is sapwood of living trees (Baldrian 2017; Pellietier et al. 2019; Gilmartin et al. 2022).

Fungi in wood may occur as mycelia or spores and can be detected by techniques that rely on morphological identification after pure cultivation (Rayner and Boddy 1988). Since they typically reveal a much higher diversity than isolation techniques, molecular techniques such a metabarcoding are essential for studying these fungal communities (Tedersoo et al. 2022). Metabarcoding is particularly valuable in conservation-oriented biodiversity studies, as it allows direct comparison of diversity patterns among known species while also capturing the vast array of undescribed and often unculturable species that make up majority of fungal communities (Ryberg and Nilsson 2018).

We used metabarcoding to investigate if species richness and species composition of fungal OTUs vary among the four broadleaf tree species beech (*Fagus sylvatica* L.), linden (*Tilia cordata* Mill.), Norway maple (*Acer platanoides* L.), and pedunculate oak (*Quercus robur* L.). We pose the conservation relevant question of whether a tree's age affects fungal species richness, and composition. To inform spatial planning, we also ask whether fungal species richness and species composition differ among bioclimatic sections forming strong gradients in southern Norway, especially based on the degree of oceanity.

Based on patterns concerning other fungal guilds, we hypothesize that tree species differ in fungal species composition but not in species richness, and that oak and beech share the highest similarity since they both belong to the Fagaceae family. Further, we also hypothesize that older trees accumulate a higher fungal species richness and have a unique species

composition. Regarding climatic variation, we based on previous experience from fruitbody surveys, hypothesize that species richness decreases with increasing oceanity, and that the decrease is accompanied by a change in species composition.

2. Materials and methods

2.1. Tree species and site selection

We choose the native tree species beech, linden, Norway maple (maple below), and pedunculate oak (oak below) because they are important constituents of the temperate broadleaf forest in northern Europe and have an approximately similar distribution in the investigated area (Diekmann 1994). The most common of these species in Norway is oak followed by linden and maple, while beech is considerably less common with only 25─30% as many occurrences as the other tree species (NBIC & GBIF Norway 2024). The two other most important temperate broadleaf trees in this area, *Ulmus glabra* Hudson and *Fraxinus excelsior* L., were excluded since they are currently subject to major diebacks due to fungal diseases.

Using a database of important areas for biodiversity from the Norwegian Environment Agency (https://www.miljodirektoratet.no/tjenester/naturbase/), we identified 28 forest sites with relevant size classes of at least one of the tree species. The sites were situated within the boreonemoral zone mainly with monthly temperatures of −4 ─ 0 °C in January and 12 ─16 °C in July (Moen 1998). The sites were distributed along a stronger gradient of decreasing oceanity from W to E in southern Norway (Fig. 1) falling into four bioclimatic sections as defined by Bakkestuen et al. (2009). Precipitation decreases from 2500─3000 mm per year in the westernmost strongly oceanic section to 700─1000 mm per year in the transition zone to continental climate (Moen 1998). The forest stands were mainly natural forest stands or previous (wooded) pastures and the trees were mainly established through natural regeneration. In a few cases, linden and maple trees appeared to have been planted (as roadside trees or in a park).

2.2. Fieldwork

Fieldwork was performed at the sites in 2019 and 2020. In the field, we searched for trees representing the following four classes of circumference at breast height (130 cm above the ground); A: 120–180 cm, B: 180–240 cm, C: 240–300 cm, and D: > 300 cm. A total of 240 healthy-looking trees were selected. Regarding some tree species it was difficult to find representatives of all size classes. The largest size classes were rare for all tree species except oak (Table 1).

For some tree species, it wasn't possible to find samples from some climatic sections (Table 2).

We collected wood core samples (sapwood and to some extent heartwood) using a 5.15 mm diameter increment borer (Haglöf, Långsele, Sweden), which was flame sterilised between each individual sample.

We removed all bark and an extra 1 mm of sapwood just inside the bark with a flame sterilised knife before sampling. We extracted two 10 cm-long wood cores at 1.2 m above the

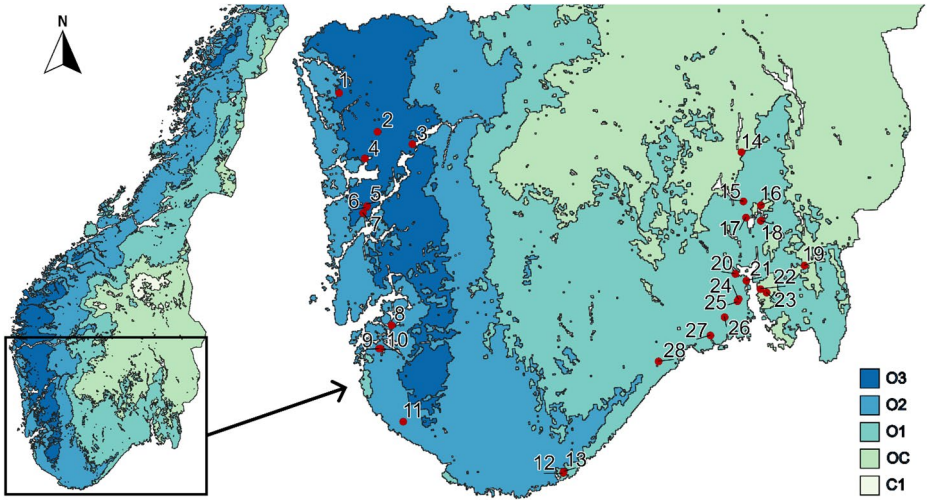

Fig. 1 Map showing areas with varying oceanity in southern Norway. O3 representing strongly oceanic, O2 clearly oceanic, O1 weakly oceanic climate, OC transition zone to weakly continental and C1 Weak continental climate. The placement of the sampling sites is shown as follows; (1) Vollum, (2) Kvennesvatnet, (3) Berge, (4) Floget, (5) Skjelavik, (6) Storhaugen, (7) Agdestein, (8) Hovda, (9) Stavanger 2, (10) Stavanger 1, 11. Nokkåsen, 12. Selåsen, 13. Dolsvann, 14. Øvre Vang, 15. Enli, 16. Dronningberget, 17. Løkenes, 18. Røer, 19. Eidsberg, Floget, 20. Mulåsen, 21. Karljohansvern, 22. Kajalunden, 23. Tasken, 24. Berg Fengsel, 25. Gullkronene, 26. Fokserød, 27. Larvik, 28. Kammerfossåsen

Table 1 The number of samples collected from the different size classes for the four tree species

[a]A: 120-180 cm, B: 180-240 cm, C: 240-300 cm, and D: > 300 cm circumference at 130 cm above the ground.

	Size classes within tree species[a]				
Tree species	A	B	C	D	Total
Beech	14	13	16	7	50
Linden	19	16	21	6	62
Maple	18	17	9	3	47
Oak	17	19	19	26	81
Total	68	65	65	42	240

Table 2 The representation of trees from bioclimatic sections (number of samples)

Bioclimatic section	[a]O3	[a]O2	[a]O1	[a]OC	Total
Beech	8	0	42	0	50
Linden	19	10	18	15	62
Maple	0	0	24	23	47
Oak	30	5	41	5	81
Total	57	15	125	43	240

[a]Bioclimatic sections according to Bakkestuen (2009); O3 strongly oceanic, O2 clearly oceanic, O1 weakly oceanic, OC transition zone to weakly continental climate.

ground; one on the S-facing side, and one on the N-facing side, and pooled the cores from each tree. In the lab, the pieces were surface sterilized for 1 min in 33% hydrogen peroxide and cut in half, the inner 5 cm sections of the cores, corresponding to a depth of 5–10 cm from the outer edge of the wood, were used for DNA sequencing. The sections closer to the surface were reserved for fungal culturing as part of a separate study. To study if the correlation between trunk circumference and age differed among tree species, we collected samples from a total of 28 trees of varying sizes, all growing in forest with semi-open to closed canopies, with a 40 cm long increment borer to count the annual rings. Linden trees proved very difficult to drill, with cores crumbling and falling apart and only four samples were obtained of this tree species.

2.2. DNA sequencing and data analysis

DNA extraction followed the methods used in Menkis et al. (2022). Wood core samples were freeze-dried for 72 h in a VirTis SP Scientific Freeze Dryer (SP Industries Inc., Suffolk, UK) at a temperature of −105 ˚C, and individual samples were milled using Retsch MM400 sample homogeniser (Retsch, Haan, Germany).

For DNA extraction, 2 mL centrifugation screw cap tubes containing 0.4 g of freeze-dried wood powder from the individual trees and a 3% CTAB solution were incubated at 65 ˚C for 1.5 h and later cleaned with chloroform. The upper phase was further purified using the NucleoSpin soil kit (Macherey-Nagel, Düren, Germany), following the recommendations from the producer. DNA concentration in each sample was determined using ND-1000 spectrophotometer (NanoDrop Technologies, Wilmington, DE, USA) and diluted to 10 ng/mL.

Individual DNA samples were amplified by PCR using the primer pair gITS7 (Ihrmark et al. 2012), and ITS4 (White et al. 1990) each containing unique sample identification barcodes. PCR was performed in 15 µl reactions containing an amount of 1.2 µl DNA, 1% Taq polymerase (5 u/µl), (DreamTaq Green, Thermo Scientific, Waltham, USA), 11% of 10 × buffer, 11% dNTPs (10mM), 1% $MgCl_2$ (25 nM), 2% each primer (200 mM) and 72% Milli-Q water. The reactions were put through Applied biosystems 2700 thermal cycler (Applied Biosystems, Foster City, CA, USA) and performed 35 cycles. The process steps for the amplification consisted of initial denaturation at 95 ˚C for 2 min followed by 30 s denaturation at 95 ˚C, 30 s annealing at 56 ˚C, continued with extension for 1 min at 72 ˚C and ended with final extension for 7 min.

The amplified samples were run on a 1.5% agarose gel (Agarose D1, Conda, Madrid, Spain) for 30 min at 300 V and scanned using QuantityOne software. The resulting DNA concentration from the electrophoresis was evaluated using a NanoDrop 3300 flurospectrometer (Thermo Fisher Scientific, Waltham, MA, USA). After evaluation, equimolar mix of all PCR samples were produced as a part of the larger sample and cleaned using EZNA cycle pure kit (Omega Bio-tek, Norcross, GA, USA). The quality was controlled using NanoDrop 3300 flurospectrometer and an Invitrogen QUBIT fluorometer (Fisher Scientific, Loughborough, UK). The sequencing was completed at SciLifeLab (Uppsala Genome Centre, NGI Uppsala, Sweden) using the Pacific Biosciences (PacBio RSII) platform.

The sequences generated were quality-filtered and clustered in the SCATA NGS sequencing pipeline (https://scata.mykopat.slu.se/). Filtering the sequences resulted in removing sequences shorter than 200 bp, primer dimer, homopolymers, and sequences difficult to read

caused by low quality. Also, sequences with missing barcodes or primers were removed. The barcodes and primers were removed but stored as meta-data to keep the information. The sequences were clustered together in different taxa ("Operational Taxonomic Units", OTUs) using single-linkage clustering as a method that was based on 98% similarity. The most commonly occurring sequence was chosen to represent each cluster. The identification of fungal OTUs (species) was done using massBLASTer at the PlutoF biodiversity platform (https://plutof.ut.ee/).

To illustrate and discuss the overall ecological functions of fungi in wood and to comment on the ecology of individual species, we classified functional groups according to the FungalTraits database (Polme et al. 2020). Functional groups were assigned to all species identified at >94% sequence similarity at a genus level and the output was grouped into endophytic, lichenized, parasitic, pathogenic, saprotrophic species, and species with unknown ecology. Fungi with ≤94% sequence similarity at the genus level were classified as unknown. We also recorded secondary ecological functions according to Polme et al. (2020). The distribution of functional groups across the various tree categories will be analysed in a forthcoming paper.

2.3. Statistical analysis

To analyse species richness, we used a generalized linear mixed model (GLMM) with package glmmTMB (Bolker 2015; Brooks et al. 2017) in R Statistical Software. The *number of species* per sample was used as response variable; *tree species* (categoric, four levels, oak as reference level), *bioclimatic section* (categoric, four levels, transition section as reference level), and tree *diameter at breast height* (continuous) were used as predictor variables, with *site* as a random factor. Checking model diagnostics using the DHARMa R package (Hartig 2020) revealed potentially problematic residual distribution and underdispersion in the model run using a Poisson response distribution, but this was rectified by using a Conway-Maxwell-Poisson distribution instead (Conway et al. 2017). Variance inflation factors did not reveal any problems with multicollinearity of predictor variables. P-values were based on the Wald test and confidence intervals were calculated using profile likelihood.

Species accumulation (rarefaction) curves of the four tree species were made using the iNEXT R package (Hsieh et al. 2020), with extrapolation to twice the sample size.

We used PERMANOVA, PERMDISP and Principal coordinates analysis (PCO) plots in Primer 7 (Gotelli and Ellison 2013; Clarke et al. 2014) with the PERMANOVA+addon (Anderson et al. 2008) to examine the differences in species composition and dispersion (beta diversity) between tree species, size classes and bioclimatic sections, as well as the interaction between tree species and size class. Site was used as a random factor in the PERMANOVA model. The analyses were based on the Sørensen similarity index calculated from presence/absence data. PERMDISP analysis performed on presence/absence data can be used as a test of beta diversity (Anderson et al. 2006).

Indicator species analysis (De Cáceres and Legendre 2009) for the four tree species, size classes and bioclimatic sections was performed using the R package indicspecies (https://cran.r-project.org/web/packages/indicspecies/index.html). The analysis estimates (A) the probability that a tree where the species was found is of a specific tree species, i.e. the specificity of each fungal species, and (B) the probability of finding the fungal species in a tree of the tree species in question, i.e. the frequency of each fungal species. The frequency

may here be less relevant since we seek to understand which fungal species is specific to each tree species. On the other hand, very low frequency means that the association is more likely to be due to chance.

The number of species shared between the four tree species was illustrated using a Venn diagram (Oliveros 2015). Fungal species with occurrence in just one tree (singletons) were included in this analysis only to include also the effect of rare, potentially conservation-relevant species.

3. Results

3.1. Taxonomic affiliations and functional groups

High-throughput sequencing of the fungal communities yielded 1,124,002 reads. Among these, 653,419 were of high-quality and were retained, while 470,583 poor-quality reads were excluded. Clustering at 98% similarity of high-quality reads resulted in 1401 non-singleton OTUs and in 2663 singletons, which were excluded. Blast analysis of the 1401 OTUs showed that 1156 OTUs were fungal, while 245 non-fungal species were excluded. Among all fungal taxa, 11.8% could be identified to species, 20.6% to genus, while 67.6% only to higher taxonomic level. The great majority (88.2%) of all taxa thus remained unidentified at the species level and (79.4%) at the genus level. Fungal OTUs were represented by 54,059 high-quality sequences, which were used in downstream analyses. Among fungal OTUs, Ascomycota was represented by 823 (71.2%) OTUs, Basidiomycota by 283 (24.5%) OTUs, Zygomycota by 38 (3.3%) OTUs and Chytridiomycota by 12 (1.0%) OTUs. We identified 1011 OTUs from oak, 160 from beech, 165 from maple and 175 from linden. The 20 most common OTUs are presented in Table 3. Sequences of all fungal non-singletons (1156 taxa) are available from GenBank, accession numbers PQ417958 to PQ419113.

Among fungal functional groups, the majority of OTUs were saprotrophic, followed by pathogenic and parasitic OTUs. Only a smaller part was lichenized or endophytic OTUs according to the definition by Rodriguez et al. (2009; Figs. 2 and 3). When also secondary ecological functions were taken into account, 12.7% of the classified genera contained endophytic species (data not shown). The proportion of saprotrophs was higher when considering the frequency of sequence reads (Fig. 3) as compared to the number of species (Figs. 2 and 3).

3.2. Species richness

The GLMM analysis showed that Norway maple, beech, and linden harbour on average significantly fewer fungal OTUs per sampled tree (species richness) than oak ($p < 5 \times 10^{-17}$ for all three, Table 4). In percent, beech had 17%, linden 13% and Norway maple 10% of the species richness of oak. The three bioclimatic sections other than the transition section all had estimated on average lower fungal species richness per tree, but with wide and overlapping confidence intervals (Table 4). Trunk circumference did not have a significant effect on species richness.

Table 3 Occurrence and relative abundance of the 20 most common fungal OTUs associated with wood of beech, maple, linden, and oak in norway. ASV=Amplicon sequence variant

Fungal species (OTU)	Ref. seq.[a]	Sp. hypothesis[b]	Seq. sim.[c] (%)	Mismatch (basepairs)	ASVs[d]	Maple (%)	Beech (%)	Oak (%)	Linden (%)	All (%)
Tremellomycetes sp.	UDB028113	SH1556067.08FU	85.4	30	3732	21.4	1.7	6.5	0.2	6.9
Armillaria lutea	UDB037624	SH1577896.08FU	100	0	3655	8.3	10.4	2.5	33.7	6.7
Mycosphaerella tassiana	UDB0780758	SH1572816.08FU	100	0	2420	10.8	16.0	2.7	0.7	3.9
Mollisia sp.	UDB0780679	SH1647282.08FU	96.6	8	7256	-	-	4.2	5.1	3.7
Penicillium citreonigrum	UDB023807	SH1529989.08FU	100	0	1756	1.7	4.8	3.6	0.1	3.2
Laetiporus sulphureus	UDB031946	SH1562644.08FU	99.7	1	1524	-	-	3.7	-	2.8
Helotiales sp.	UDB025172	SH1509542.08FU	95.9	9	1428	-	-	3.5	-	2.6
Calycellina sp.	UDB0778858	SH1564446.08FU	99.6	1	1324	0.0	0.0	2.7	4.1	2.4
Cadophora luteo-olivacea	UDB028476	SH1545862.08FU	93.8	15	1147	8.8	5.8	0.1	6.5	1.9
Aureobasidium pullulans	UDB035654	SH1515060.08FU	100	0	1173	3.0	11.6	1.1	0.7	1.8
Ascocoryne solitaria	UDB0778476	SH1551049.08FU	99.6	1	883	0.0	0.0	1.3	6.2	1.6
Mycena abramsii	UDB07672647	SH0748989.10FU	98.36	3	723	-	0.0	1.4	3.1	1.3
Baeospora myosura	UDB037618	SH1600467.08FU	100	0	690	-	-	1.7	-	1.3
Chaetothyriales sp.	UDB027816	SH1648681.08FU	77.3	61	685	-	-	1.7	-	1.3
Hymenochaete rubiginosa	UDB031456	SH1600724.08FU	100	0	659	-	-	1.6	-	1.2
Meliniomyces sp.	UDB026396	SH1523753.08FU	93.3	16	642	-	-	1.6	-	1.2
Mollisia sp.	UDB0780685	SH1647286.08FU	99.2	2	639	-	-	1.1	3.0	1.2
Ascomycota sp	UDB035461	SH1564481.08FU	88.16	29	633	-	-	1.6	-	1.2
Mortierella sp.	UDB028005	SH1557019.08FU	95.13	17	607	1.2	0.1	1.1	1.5	1.1
Debaryomyces sp.	UDB033935	SH1516571.08FU	99.65	1	583	-	-	1.3	0.02	1.0
Total of 20 species						55.2	50.6	44.9	65.0	48.2

[a]Reference sequence, bSpecies hypothesis, cPercent sequence similarity, d ASV = Amplicon

3.2. Species accumulation curves

The species accumulation curves showed that oaks had a much steeper species accumulation than Norway maple, beech and linden (Fig. 4). The curves of the latter three tree species were approaching a plateau (somewhere around 250 species), while the accumulation curve for oak was still clearly rising (well above 1000 species).

3.3. Species composition

The total number of singleton species (species occurring in only one sample) was 479, and oak had a substantially higher proportion of these than the other tree species (39%, see Table 5).

The principal coordinates analysis (PCO) showed that oak was clearly separated in terms of fungal species composition, while the three other tree species were largely overlapping (Fig. 5; see also Fig. 6 for the number of fungal species shared between the four tree species). A relatively low amount of the sample variation is explained in PCO axes 1 and 2; 6.7% and 5.1%, respectively. While this means interpretation should be done with care, the PERMANOVA supported the PCO visualisation, demonstrating a significant effect of the tree species on fungal species composition (Sums of squares = 32 932, pseudo-F (3) = 3.02, $p = 0.001$).

The overall effect of size class was not significant, but there was a significant interaction between tree species and size class (SS = 37 753, pseudo-F (9) = 1.15, $p = 0.008$), indicating that the effect of tree species on fungal species composition is dependent on trunk size and/or vice versa. Pairwise tests (see Table 6) revealed that while oak was significantly different from linden in all size classes, it was different from beech only in classes C and D, and from maple in none of the size classes (note that the sample did not allow for a pairwise test between maple and oak in size class D). The PERMDISP analysis indicated highest multivariate dispersion (i.e. beta diversity) in linden (mean: 66.8 SE: ± 0.53), followed by beech (65.6 ± 0.98), maple (62.4 ± 1.41) and oak (62.1 ± 0.38), with significant differences between linden and maple ($p = 0.006$), oak and linden ($p = 0.001$) and oak and beech ($p = 0.001$). This was also reflected in the PCO plot, where the oak samples were more tightly clustered than of the other tree species (Fig. 5). Size class A had significantly higher multivariate dispersion (67.53 ± 0.46) than classes B (65.05 ± 0.68, $p = 0.004$), C (65.87 ± 0.56, $p = 0.023$) and D (64.55 ± 0.73, $p = 0.001$), indicating decreasing beta diversity with size/age.

There was a significant effect of bioclimatic section on species composition (SS = 27 626, pseudo-F (3) = 1.73, $p = 0.001$). For the bioclimatic sections, the transition section had significantly lower multivariate dispersion (60.5 ± 1.44) than both the weakly (66.27 ± 0.43, $p = 0.001$) and the strongly (66.23 ± 0.61, $p = 0.001$) oceanic sections.

The result of the tree ring analysis showed that maple had 4.9 ± 2.1 SD ($n = 5$), beech 4.5 ± 0.6 ($n = 10$), oak 4.7 ± 1.0 ($n = 8$), and linden 4.1 ± 0.5 ($n = 4$) annual rings per cm (Table 9). The result of the reverse pairwise PERMANOVA test (size class within tree species), only indicated an effect on species composition with increasing age in a single case (maple size class A vs. B; Table 10).

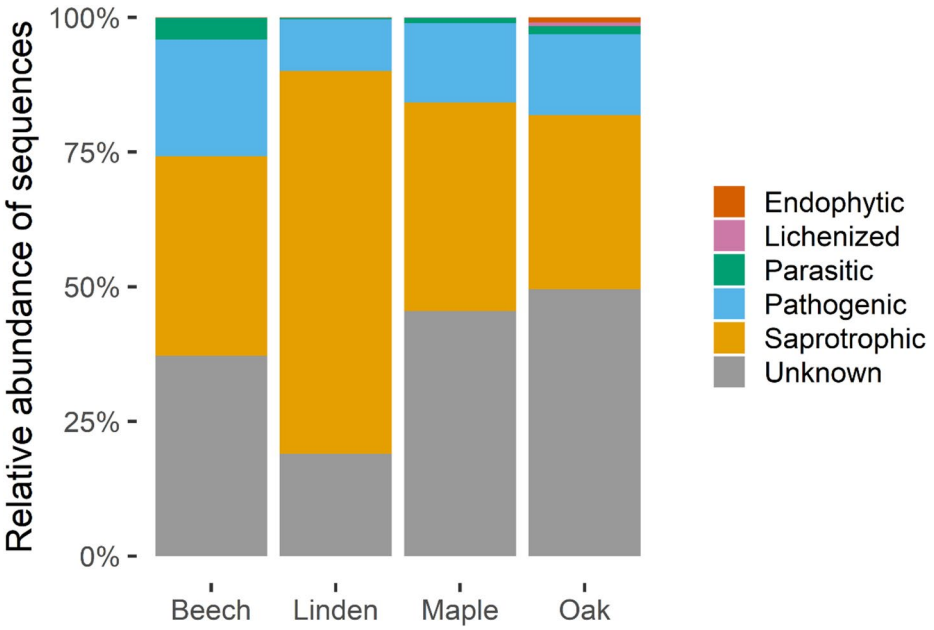

Fig. 2 Relative proportion of sequence reads representing each of the fungal functional groups in wood of the different tree species

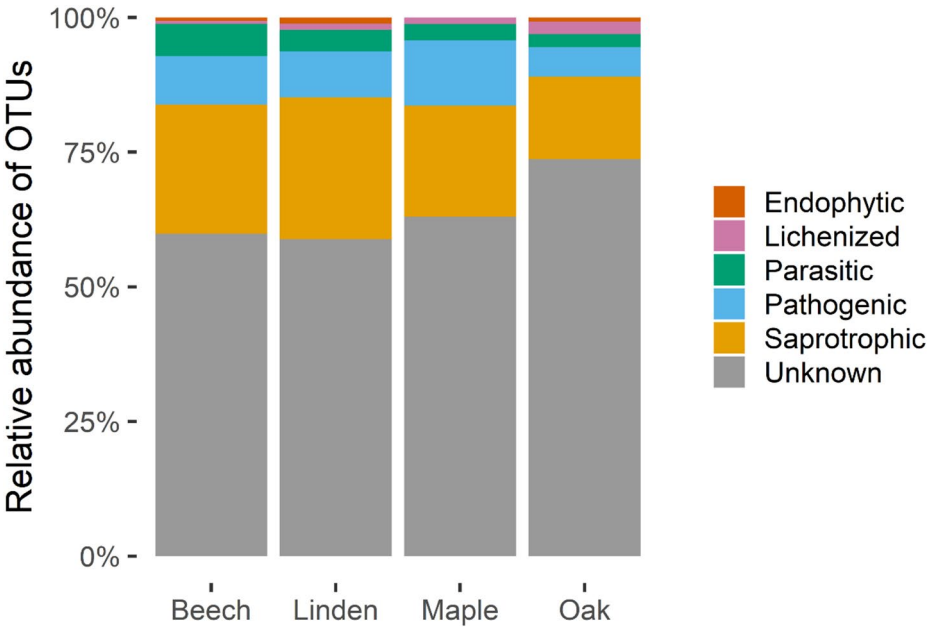

Fig. 3 Relative proportion of fungal species representing each of the fungal functional groups in wood of the different tree species

Table 4 Results of the GLMM analysis. "Oak" and "OC" (Transition section) were used as reference levels

Fixed effects					
Factor		Estimate	CI[a] 2.5%	CI[a] 97.5%	p
Intercept		74.33	23.36	93.81	0.000
Tree species (Oak)		*1.00*	*(Reference level)*		
Tree species (Beech)		0.17	0.11	0.26	0.000
Tree species (Linden)		0.13	0.10	0.18	0.000
Tree species (Maple)		0.10	0.06	0.15	0.000
Bioclimatic section (OC[b] Transition)		*1.00*	*(Reference level)*		
Bioclimatic section (Weakly oceanic)		0.53	0.39	1.82	0.058
Bioclimatic section (Intermediately oceanic)		0.63	0.26	1.51	0.289
Bioclimatic section (Strongly oceanic)		0.41	0.29	1.46	0.013
Circumference at breast height (cm)		1.00	0.95	1.14	0.400
Random effects					
Factor	SD				
Site	1.69				

[a]CI Confidence interval, [b]OC Transition section.

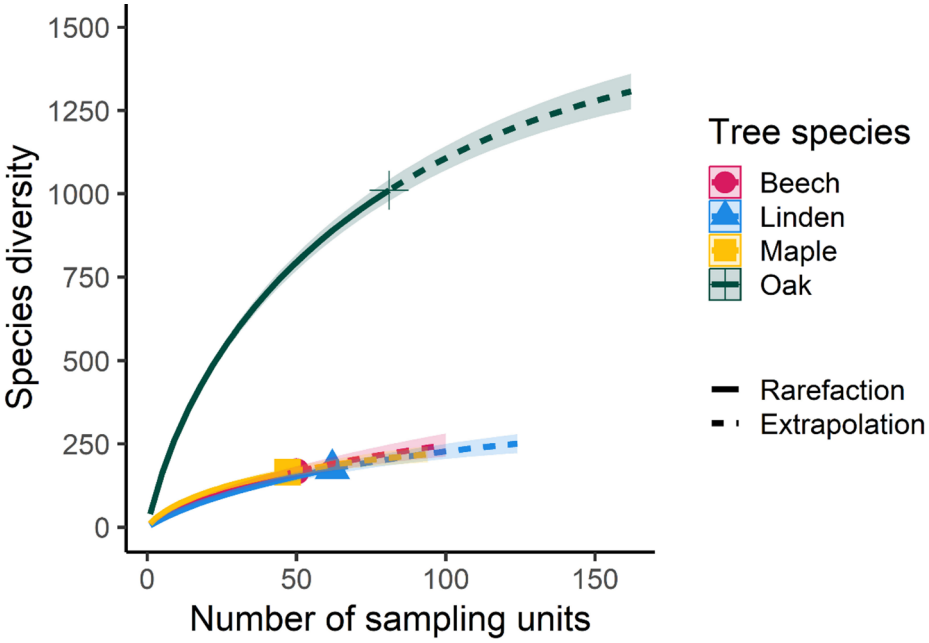

Fig. 4 Species accumulation curves for the four tree species showing the total number of fungal OTUs with rarefaction (interpolation) and extrapolation to twice the sample size. Envelopes indicate 95% confidence intervals

Table 5 The number and percentage of Singleton fungal species identified in each of the four tree species

Singleton fungal species among the tree species			
Tree species	Total no. of species	No. of singleton species	Percentage of singletons
Beech	167	32	19%
Linden	175	21	12%
Maple	165	33	20%
Oak	1011	393	39%
Total	1156	479	41%

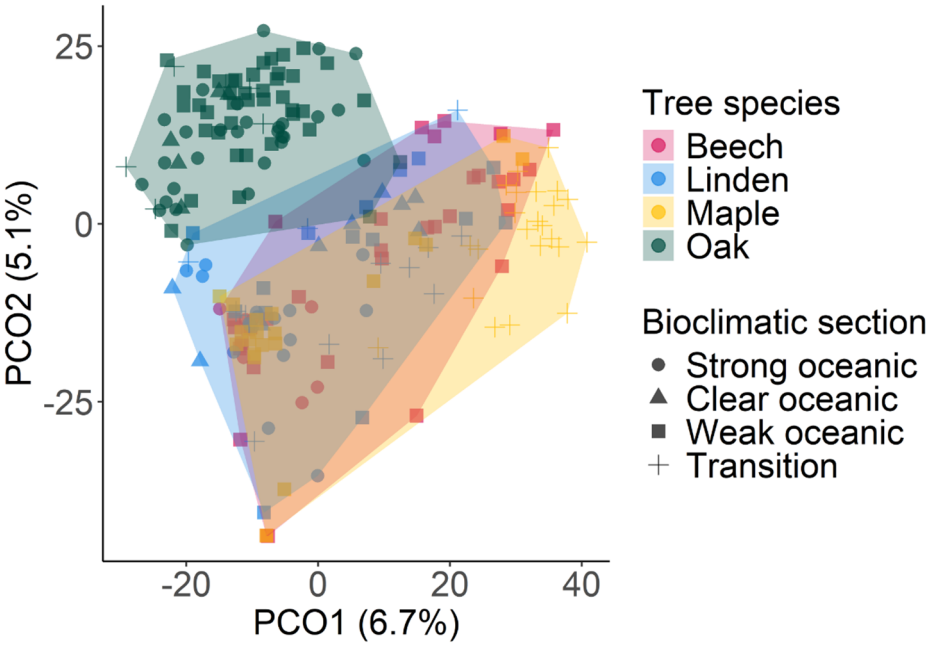

Fig. 5 PCO of the relationship among beech, linden, maple, oak in terms of fungal species composition. PCO1 represents 6.7% of the total variation, and axis PCO2 represents 5.1% of the total variation of the total samples

3.3.1. Red-listed species

Five of the encountered species were listed in the Norwegian Red List (Artsdatabanken 2021). Two of the species were lichenized, two wood-decaying species and one soil-living saprophytic. They were all found in oak of larger dimensions (eight oaks in total; Table 7).

3.3.2. Indicator species

Oak had the highest number of species (104 or 10.3%) showing 100% specificity within the dataset, maple had 20 (12.1%), beech had three species (1.9%) and linden had three species (1.7%). Regarding size classes, size class A had zero species showing 100% specificity, size class B had three, size class C had zero, and size class D had 39. For the bioclimatic sections,

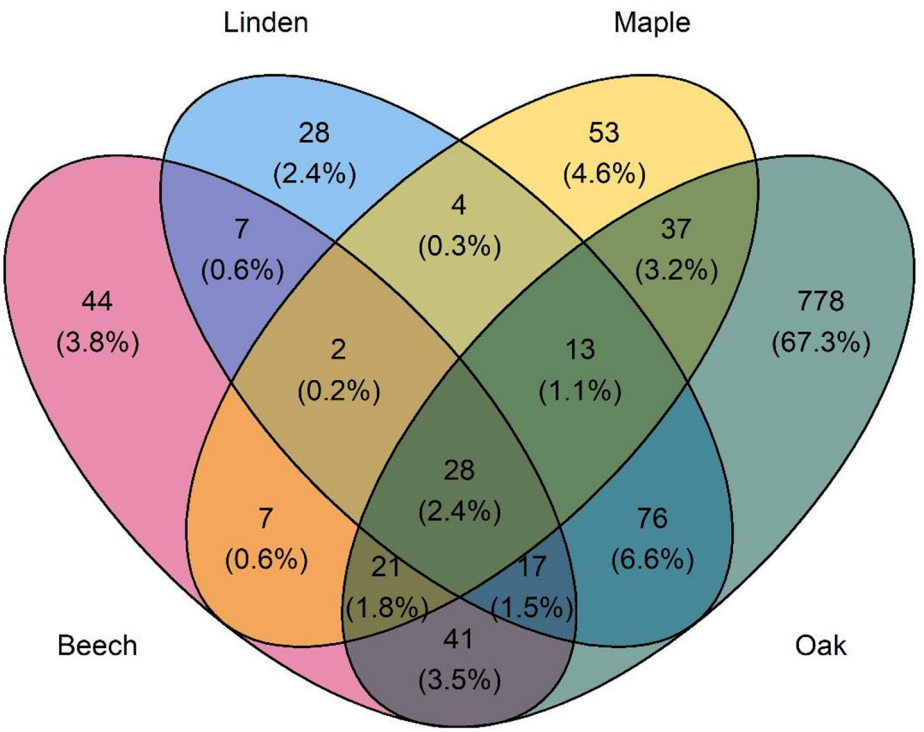

Fig. 6 Venn diagram showing the number of species shared between the four tree species including singletons

Table 6 Results of pairwise PERMANOVA tests of difference in species composition among tree species within size classes. Numbers given are *p*-values based on permutation test

Size A	Oak		
Beech	$t(13)=1.11$, $p=0.365$	Beech	
Linden	$t(18)=1.29$, $p=0.016$	No test	Linden
Maple	$t(17)=1.12$, $p=0.398$	$t(20)=1.23$, $p=0.068$	$t(22)=0.96$, $p=0.609$
Size B	Oak		
Beech	$t(13)=1.23$, $p=0.145$	Beech	
Linden	$t(18)=1.29$, $p=0.026$	No test	Linden
Maple	$t(18)=1.26$, $p=0.121$	No test	$t(18)=1.15$, $p=0.200$
Size C	Oak		
Beech	$t(17)=1.36$, $p=0.021$	Beech	
Linden	$t(19)=1.41$, $p=0.004$	No test	Linden
Maple	$t(14)=1.29$, $p=0.059$	No test	No test
Size D	Oak		
Beech	$t(16)=1.32$, $p=0.025$	Beech	
Linden	$t(15)=1.30$, $p=0.034$	$t(3)=1.03$, $p=0.433$	Linden
Maple	No test	No test	No test

Table 7 Red-listed species identified, red-list category, functional role, the number of finds, tree species, size class of tree and bioclimatic section

Species	Ref. seq.[a]	Sp. hypothesis[b]	Seq. sim. (%)[c]	ASV[d]	Red List cat.[e]	Fr[f]	Trees	Size[g]	Oc.[h]
Caloplaca lucifuga	OQ717772	SH1021019.10FU	100	40	NT	L	2 oaks	D	O1
Gyalecta carneola	OQ717850	SH1040988.10FU	100	11	VU	L	oak	C	O1
Lentaria byssiseda	UDB019556	SH0988816.10FU	100	95	NT	S	3 oaks	B, C,D	O1
Mutinus caninus	GQ981513	SH0741746.10FU	99.65	2	VU	S	1 oak	D	O2
Phlebia centrifuga	UDB0799107	SH0946793.10FU	100	3	NT	S	1 oak	D	O3

[a]Reference sequence, [b]Species hypothesis, [c]Percent sequence similarity, [d]ASV = Amplicon sequence variant, [e]Red List category, [f]Functional role; L = Lichenized, S = Saprotrophic, [g]Tree size class, [h]Oceanity; O3 strongly oceanic, O2 clearly oceanic, O1 weakly oceanic climate.

strongly oceanic had 12, Intermediately oceanic six, Weakly oceanic one, and Transition 12 species showing 100% specificity. Most species had low stat values due to low frequency. The (up to) three Indicator species with the highest stat value and 100% specificity for each tree species, size class and bioclimatic section are presented in Table 8.

4. Discussion

We compared fungal communities and diversity in wood among several tree species and age groups, using standardized methods for accurate assessment. Fungal communities in living trees are known to be very diverse (Agostinelli et al. 2018; Menkis et al. 2022) but diversity patterns in wood were not previously directly compared among tree species using molecular methods. Our results shed new light on the diversity of fungal communities in middle-aged to ancient broadleaf trees. That the great majority of all taxa remained unidentified at the species level was expected given that the number of described fungal species corresponds to only a few percent of the estimated true number of fungal species (Blackwell 2011; Antonelli et al. 2023). After commenting on our major results, we discuss the conservation relevance of our findings.

4.1. Tree species vs. fungal diversity

Our hypothesis that tree species differ only in fungal species composition but not in species richness was not supported. We found that oak host unique and significantly richer fungal communities compared to the other studied broadleaf tree species. Further, the species accumulation curve indicated that there is still a substantial number of unsampled fungal species present in the species pool. Oak is known to host distinct and diverse communities of e.g. lichenized fungi, wood-decaying fungi, and insects (Mitchell et al. 2019; Sundberg et al. 2019). The reasons for this comparatively high fungal species richness associated with oak are unknown but we suggest they may involve the long evolutionary history of the large genus *Quercus* and its wide distribution range, both factors potentially contributing to

Table 8 Indicator species analyses for tree species, size classes and bioclimatic sections based on de Cáceres and legendre (2009)

Beech						
Species	Ref. seq.[a]	Sp. hypothesis[b]	Seq. sim. (%)[c]	B[d]	Stat	p
Mucidula mucida	UDB034175	SH0818254.10FU	99.16	0.060	0.245	0.021
Neocucurbitaria juglandicola	OQ306533	SH0812192.10FU	99.2	0.060	0.245	0.014
Aposphaeria sp.	PP097810	SH0878671.10FU	100	0.060	0.245	0.019
Linden						
Merismodes sp.	UDB016262	SH0895836.10FU	99.4	0.048	0.220	0.029
Helotiales	MK627123	-	100	0.048	0.220	0.024
Mortierellaceae	MW237938	-	100	0.048	0.220	0.034
Maple						
Phomopsis sp.	MW765220	SH1011994.10FU	99.2	0.192	0.438	0.001
Fungi	KP891071	-	91.4	0.170	0.413	0.001
Mortierella sp.	MF423509	SH0960692.10FU	95.9	0.106	0.326	0.002
Oak						
Fungi	MW216203	SH0983948.10FU	95.9	0.395	0.629	0.001
Fungi	MT266073	-	100	0.259	0.509	0.001
Pleurocybella porrigens	UDB0781063	SH0890536.10FU	100	0.247	0.497	0.001
Size class B						
Merismodes sp.	UDB016262	SH0895836.10FU	99.4	0.046	0.215	0.049
Phaeophyscia orbicularis	OQ718018	SH0867317.10FU	99.6	0.046	0.215	0.039
Ascomycota	KJ827805	-	87.8	0.046	0.215	0.049
Size class D						
Agaricomycetes	OX033148	-	98.6	0.071	0.267	0.007
Chaetothyriales	MN902451	-	97.6	0.071	0.267	0.003
Cantharellaceae	MW238077	-	98.8	0.071	0.267	0.005
Strongly oceanic						
Pseudocercospora sp.	KP889581	SH0851812.10FU	99.2	0.140	0.375	0.012
Lophiostoma sp.	OR481148	-	99.2	0.123	0.350	0.005
Kuraishia sp.	OR481183	-	100	0.123	0.350	0.003
Intermediately oceanic						
Ricasolia amplissima	MK811832	SH0860360.10FU	100	0.200	0.447	0.001
Candelaria concolor	OQ717774	SH0868355.10FU	99.6	0.133	0.365	0.001
Fungi	MN660627	-	99.7	0.133	0.365	0.003
Weakly oceanic						
Fungi	5064_166	MW757649	100	0.088	0.297	0.04
Transition						
Phomopsis sp.	MW765220	SH1011994.10FU	99.2	0.209	0.457	0.001
Mortierella sp.	MF423509	SH0960692.10FU	95.9	0.116	0.341	0.004
Fungi	MG828225	-	97.9	0.070	0.264	0.010

[a]Reference sequence, [b]Species hypothesis, [c]Percent sequence similarity, [d]The value B is an estimate of the probability of finding the fungal species in a tree of the tree species in question.

a large species pool of oak-associated organisms. Furthermore, oak is a more common tree species than the other three in the studied area, potentially further enhancing this tendency. It is also possible that factors related to microhabitat within the wood may play a role. Oak differs from the other species studied by having ring-porous wood, potentially adding to the heterogeneity of niches within the wood. Additional tree species with ring-porous wood

such as *Fraxinus* and *Ulmus* species should be included in future studies to study if this speculation is supported.

The unique species composition of fungi in oak wood was rather expected given prior knowledge of communities of wood-decaying fungi (Nordén et al. 2004) and lichenized fungi (Paltto et al. 2011) associated with oak even though we did expect a certain overlap with fungal communities in beech. We hypothesized that oak and beech share the highest similarity since they both belong to the *Fagaceae* family because some studies have shown phylogenetic congruence between trees and their associated fungi (see e.g. Liu et al. 2016), but our hypothesis was not supported. Instead, the beech fungal community showed marked overlap with that in maple belonging to *Sapindaceae* and linden belonging to *Malvaceae* (Byng 2014). That maple (12.1%), and oak (10.3%) both had a relatively high share of species-specific fungi while beech (1.9%) and linden (1.7%) had a much lower share was also surprising and represent another pattern for which we currently have no explanation. The top Indicator species for beech, *Mucidula mucida*, is a common toadstool on beech trunks while the second, *Neocucurbitaria juglandicola*, is a dothideomycete described from walnut trees. The true identity of this species might be an undescribed species of *Neocucurbitaria* or a species of the genus lacking a reference sequence, and this is probably true also for *Aposphaeria* sp. since most dothideomycete genera have not been sufficiently studied taxonomically and sequenced. On linden, the most common species, *Merismodes* sp. is a small Agaricales species often found on dead wood, and the other Indicator species is an unknown Helotiales discomycete and a representative of Mortierellaceae (Mucoromyceta). Maple and oak showed similar situations with few hits for specific species but instead higher taxa. The results emphasize our poor taxonomic knowledge of many fungal taxa even in northern Europe. *Pleurocybella porrigens* on oak was an interesting exception, being a common agaricoid fungus often found on spruce stumps it also seems to occur as an endophytic species in living oaks.

The lower beta diversity of oak indicated that many species specialized on oak are well dispersed in the landscape, perhaps as a legacy of the former larger and more coherent distribution of oak forest in S Norway before it was extensively cut back, mainly for ship building (Moore 2010). Regarding the remaining three tree species, it is known that beech have had a small distribution in the area also historically (Bjune et al. 2012) and this is probably also true for maple. Linden has a long history in the whole area with peaks during warmer historic periods (de Benedetti et al. 2022).

4.2. Effects of tree size (age)

Even though limitations of the fieldwork only allowed us to core a subsample of trees, we consider the tree ring analysis to indicate that the increment in girth per year did not differ substantially among the tree species. However, to draw more strongly founded conclusions on this matter, future studies should implement tree ring analysis for each sampled tree and use age as a variable in statistical models.

Our hypothesis that older trees have accumulated a higher fungal species richness and have a unique species composition was not supported. It should be noted however, that size class D trees had by far the highest number of species-specific Indicator species (including e.g. the heart-rotting polyporoid basidiomycete *Laetiporus sulphureus*), and a majority of finds of Red List species, indicating an effect of size class (age) on individual species. The

limited effect of tree age on fungal species richness and species composition may be due to stability of the chemistry of wood with ageing of the tree. It is known that the bark of trees changes structurally and chemically with age leading to species turnover of epiphytic lichenized fungi (Fritz et al. 2009), but few studies have reported similar changes of the environment inside ageing but healthy trees (but see Thurner et al. 2024 who showed that nitrogen levels in various tissues decrease with tree size).

The decreasing beta diversity with size/age may indicate more similar microhabitat conditions with age. It may alternatively be an effect of older trees being less isolated than younger trees since sites with older trees usually also had younger trees, while some of the sites sampled for younger trees lacked the older tree generation. Further, older trees may continuously accumulate species from the species pool during their lifetime which may tend to harmonize species composition.

4.3. Regional variation

We interpret the lower species richness in the western-most climatic section mainly as an effect of increasing oceanity along the W-E gradient. Some fungi such as polyporoid basidiomycetes in dead wood seem to be less common in oceanic than in continental areas (Heilmann-Clausen and Boddy 2008; own observations), but the reasons for the overall lower species richness of wood fungi in oceanic areas are not known. One possible explanation is that the higher yearly variation in temperatures in more continental areas enable fungal species with a wider range of temperature optima to co-exist. The significant effect of bioclimatic section on fungal species composition was expected considering the analogy with lichenized fungi, which show a very pronounced species turnover with oceanity (Smith 2009). In a few cases, lichenized fungi were also part of the wood communities. Among the Indicator species for the bioclimatic sections with 100% specificity, *Ricasolia amplissima*, the giant candlewax lichen, is an iconic Indicator of old broadleaf trees in oceanic climate.

4.5. Ecological function

Based on assignment of main ecological functions at the genus-level, our results indicated that the share of endophytic species (in the narrow sense of Rodriguez et al. 2009 and Polme et al. 2020) was low, and that the fungal communities were dominated by saprotrophic, pathogenic, and parasitic species. However, many genera include endophytic species, and there are multiple occasions where the same species has diverse functions or members of the same genus display different trophic strategies (Cline et al. 2017; Selosse et al. 2018). The endophytic lifestyle is asymptomatic, commensal or weakly mutualistic (Rodriguez et al. 2009), but the demarcation to the pathogenic lifestyle may not always be entirely clear-cut. According to Sieber (2007) it is likely that majority of fungal species in living trees classified as pathogens have co-evolved with their hosts and are not highly virulent. Certain species may therefore be endophytic in living wood despite commonly being seen as belonging to other guilds. On the other hand, many fungi in living wood tissues may also be latent invaders causing little harm in the living tree but that decay wood when it becomes non-functional or may become pathogenic in stressed or harmed trees (Parfitt et al. 2010).

The 20 most common species appear to represent a diverse array of ecological functions and specificities, some being known as ubiquitous molds, while *Mycosphaerella (Davi-*

diella) tassiana is an ascomycete species which sexual stage is considered common on various plant material, especially in alpine areas (Eriksson 2014), and *Calycellina* sp. and *Ascocoryne solitaria* are discomycetes on dead wood or plant material. Regarding the high relative abundances of the two toadstools *Baeospora myosura* and *Mycena abramsii*, the latter is interesting in that it occurs not only on dead wood but also on the basis of living trees (Ryman and Holmåsen 1992), indicating that it might be present in the bark or possibly also inside the functional wood. However, with 98.4% sequence similarity (based on a 297 basepair sequence) it is also possible that our sequences may represent an unrecognized *Mycena* species. The presence of *B. myosura* (identified by 100% sequence similarity based on a 303 basepair sequence) is even more intriguing as it its fruitbodies are typically found on conifer cones. We refrain from speculating and leave for future studies to study the possible endophytic life stages of this species. An example of a wood-decaying species typical of its host tree is *Laetiporus sulphureus*, the "chicken of the woods" polypore especially common on old oak trees. It is possible that the occurrence of this species is due to the inclusion of heart wood since it is well-known as a heart-rotting species (Ryvarden and Gilbertson 1993).

Metabarcoding has the capacity to reveal important information regarding e.g. Red List species not available through fruitbody surveys. The occurrence of *Phlebia centrifuga* (100% similarity; 296 basepair sequence) in an ancient oak in a strongly oceanic area was such a revelation. This species is otherwise typically known from spruce logs in old boreal forest in rather continental areas. The occurrence of *Mutinus caninus* (99.7% similarity; 285 basepair sequence) in a similar oak tree is also interesting, given that this species is saprophytic and forms basidiomata on the forest floor. These species may appear to have previously unknown ecological functions in living trees. However, we recognize that three of the five species listed in Table 7 are singletons, and the others have low occurrences. It is possible that some of the occurrences represent propagules rather than active colonizers of functional wood given the sensitivity of microbial community metabarcoding approaches to environmental contamination. Further studies using targeted sampling or independent verification methods are needed to confirm the ecological roles of these species in wood. Further, metabarcoding studies of fungi reveal only the presence of DNA, not if the fungi are active and or only present as dormant spores. It is therefore uncertain if detection of a certain species means it can complete its life cycle and contribute to the long-term survival of the population. This should be examined in future studies.

The apparent presence of DNA from epiphytic lichenized fungi relatively deep inside living wood may indicate that some lichenized fungi have additional unknown ecological functions within their substrates. Although we are cautious to draw this conclusion from our study, it is worth noting that a similar pattern concerning certain lichenized fungi has been observed in dead wood (Kubartová et al. 2012; Tuovinen et al. 2015). Further, Favero-Longo and Piervittori (2010) noted that hyphae of some epiphytic lichens have been reported to penetrate bark and wood, and to have reached the xylem vessels of their host trees. Tuovinen (2015) concluded that the discovery of various mycobiont taxa in dead wood merits further investigation to determine whether some of these mycobionts have the potential for a free-living saprotrophic phase.

5. Conservation relevance/conclusions

Our results may be of importance for conservation planning because they provide insights into the diversity patterns of fungal communities, their potential ecological roles, and the identification of species or habitats that may require targeted conservation efforts.

Fungi in wood of broadleaf trees form very rich communities, which should be considered in conservation. Especially, our findings illustrate the important role of oaks as repositories of fungal diversity, including Red List species, adding to the many arguments for conservation and restoration of oak forests and oaks in wooded pastures.

The lack of an overall effect of tree size/age indicates that all broadleaf trees with circumference > 120 cm may be equally important for the diversity of sapwood fungi. However, certain individual species did occur only in the largest trees. Species only occurring in ancient trees may be of conservation concern and the occurrence of such species should be investigated further.

The lower species richness in oceanic climatic sections should be researched further in studies controlling for landscape effects.

Appendices

See Tables (9, 10).

Table 9 Trees drilled with increment borer for ageing

Site	Tree species	Circumference (cm)	Length of core (cm)	Number of rings per cm	Estimated age[a] (years)
Løkenes	maple	159	23.00	5.52	123
Eidsberg	maple	167	33.70	2.64	81
Løkenes	maple	178	22.60	5.44	119
Berg fengsel	maple	212	24.50	3.31	127
Løkenes	maple	344	30.70	3.88	145
Løkenes	maple	125	11.1	4.23	84
Kajalunden	beech	119	26.40	3.79	73
Kajalunden	beech	134	24.80	5.04	85
Kajalunden	beech	150	34.80	3.76	125
Gullkronene	beech	220	27.80	5.25	155
Gullkronene	beech	235	18.50	3.95	165
Kajalunden	beech	240	29.00	5.45	145
Kajalunden	beech	255	28.20	4.43	221
Gullkronene	beech	270	29.80	4.70	161
Gullkronene	beech	285	24.10	3.98	238
Gullkronene	beech	292	33.40	4.40	218
Løkenes	oak	106	33.30	5.11	118
Løkenes	oak	162	21.40	3.55	76
Løkenes	oak	210	32.20	4.01	115
Løkenes	oak	227	22.60	4.03	146
Løkenes	oak	231	27.60	3.80	188
Gullkronene	oak	300	35.50	5.24	191
Gullkronene	oak	303	35.30	4.53	218
Løkenes	oak	319	17.60	6.70	266
Løkenes	linden	144	28.60	3.78	94
Løkenes	linden	169	21.40	4.11	108
Bygdøy	linden	250	34.80	3.74	197
Bygdøy	linden	265	22.20	4.95	158

[a]In cases with partial core length, age was estimated by calculating the average number of rings per cm on the partial core and multiplying it with the trees stem radius

Table 10 Results of the reverse pairwise PERMANOVA test (size class within tree species), indicating an effect on species composition with increasing age in a single case (maple size class A vs. B)

Oak	Size A		
Size B	$t(18) = 1.04$, $p = 0.365$	Size B	
Size C	$t(19) = 0.98$, $p = 0.538$	$t(22) = 1.05$, $p = 0.326$	Size C
Size D	$t(24) = 1.10$, $p = 0.192$	$t(28) = 1.12$, $p = 0.147$	$t(27) = 1.03$, $p = 0.391$
Beech	Size A		
Size B	$t(16) = 1.01$, $p = 0.418$	Size B	
Size C	$t(19) = 1.00$, $p = 0.470$	$t(17) = 1.06$, $p = 0.335$	Size C
Size D	$t(11) = 1.05$, $p = 0.391$	$t(9) = 1.00$, $p = 0.458$	$t(13) = 1.07$, $p = 0.323$
Linden	Size A		
Size B	$t(19) = 0.88$, $p = 0.767$	Size B	
Size C	$t(21) = 1.15$, $p = 0.132$	$t(18) = 1.12$, $p = 0.293$	Size C
Size D	$t(11) = 1.10$, $p = 0.269$	$t(8) = 1.02$, $p = 0.551$	$t(13) = 0.95$, $p = 0.566$
Maple	Size A		
Size B	$t(23) = 1.31$, $p = \mathbf{0.019}$	Size B	
Size C	$t(18) = 1.17$, $p = 0.169$	$t(18) = 1.03$, $p = 0.352$	Size C
Size D	No test	$t(12) = 0.90$, $p = 0.661$	$t(8) = 1.08$, $p = 0.330$

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Author contributions B.N. designed the study and wrote most of the manuscript, M.A. took part in the field work and performed most of the lab work, O.G. had the main responsibility for the statistical analysis, A.M was responsible for the bioinformatics. All authors contributed to earlier versions of the manuscript and read and approved the final manuscript.

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Data availability Sequences of all fungal non-singletons (1156 taxa) are available from GenBank, accession numbers PQ417958 to PQ419113.

Declarations

Competing interests The authors declare no competing interests.

References

Agostinelli M, Cleary M, Martín JA, Albrectsen BR, Witzell J (2018) Pedunculate Oaks (Quercus robur L.) differing in vitality as reservoirs for fungal biodiversity. Front Microbiol 9. https://doi.org/10.3389/fmicb.2018.01758

Anderson MJ, Ellingsen KE, McArdle BH (2006) Multivariate dispersion as a measure of beta diversity. Ecol Lett 9:683–693. https://doi.org/10.1111/j.1461-0248.2006.00926.x

Anderson MJ, Gorley RN, Clarke KR (2008) PERMANOVA+ for PRIMER: guide to software and statistical methods. PRIMER-e, Plymouth, UK

Antonelli A, Fry C, Smith RJ et al (2023) State of the World's Plants and Fungi 2023. https://doi.org/10.34885/wnwn-6s63

Artsdatabanken (2021) Norsk rødliste for arter 2021

Baldrian P (2017) Forest microbiome: diversity, complexity and dynamics. FEMS Microbiol Rev 41:109–130

Bakkestuen V, Erikstad L, Halvorsen R (2009) Klimaendringer og Norges vegetasjon. Hvordan påvirkesvegetasjonsmodeller av ulike klimascenarier?– NINA Rapport 524:1–24

Bjune A, Helvik I, Birks H (2012) The Fagus sylvatica forests in the Larvik region, south-eastern norway: their origin and history. Veg History Archaeobotany 22. https://doi.org/10.1007/s00334-012-0371-1

Blackwell M (2011) The fungi: 1, 2, 3. 5.1 million species? Am J Bot 98:426–438. https://doi.org/:DOIhttps://doi.org/10.3732/ajb.1000298

Bolker B (2015) Linear and generalized linear mixed models. In: Fox GA, Negrete-Yankelevich S, Sosa VJ (eds) Ecological statistics: contemporary theory and application, first edition. Oxford University Press, pp 309–333

Bowd E, Boddy L, Greenwood L, Hopkins A, May T, Nordén B (2025) The importance of old and large trees for fungi and fungi for old and large trees. In: Blicharska M, Lindenmayer D, Mikusinski G (eds) Large and old trees - ecological and cultural significance. Springer, pp 200–234

Brooks ME, Kristensen K, van Benthem KJ, Magnusson A, Berg CW, Nielsen A, Skaug HJ, Maechler M, Bolker BM (2017) GlmmTMB balances speed and flexibility among packages for Zero-inflated generalized linear mixed modeling. R J 9:378–400

Byng JW (2014) The Flowering Plants Handbook, Plant Gateway. https://doi.org/10.13140/2.1.1849.8566

Clarke KR, Gorley RN, Somerfield PJ, Warwick RM (2014) Change in marine communities: an approach to statistical analysis and interpretation, 3rd edn. PRIMER-e, Plymouth, UK

Cline CL, Schilling SJ, Menke J, Groenhof E, Kennedy GP (2017) Ecological and functional effects of fungal endophytes on wood decomposition. Funct Ecol 32:181–191. https://doi.org/10.1111/1365-2435.12949

Conway JR, Lex A, Gehlenborg N (2017) UpSetR: an R package for the visualization of intersecting sets and their properties. Bioinformatics 33:2938–2940. https://doi.org/10.1093/bioinformatics/btx364

De Benedetti C, Gerasimenko N, Ravazzi C, Magri D (2022) History of Tilia in Europe since the eemian: past distribution patterns. Rev Palaeobot Palynol 307:104778. https://doi.org/10.1016/j.revpalbo.2022.104778

De Cáceres M, Legendre P (2009) Associations between species and groups of sites: indices and statistical inference. Ecology 90:3566–3574. https://doi.org/10.1890/08-1823.1

Diekmann M (1994) Deciduous forest vegetation in Boreo-nemoral Scandinavia / Martin Diekmann. Opulus Press AB

Eriksson OE (2014) Checklist of the nonlichenized Ascomycota of Sweden. Acta Universitatis Upsaliensis Symbolae Botanicae Upsalienses 36:1–499

Favero-Longo SE, Piervittori R (2010) Lichen-plant interactions. J Plant Interact 5(3):163–177. https://doi.org/10.1080/17429145.2010.492917

Fritz Ö, Brunet J, Caldiz M (2009) Interacting effects of tree characteristics on the occurrence of rare epiphytes in a Swedish Beech forest area. Bryologist 112:488–505. https://doi.org/10.1639/0007-2745-112.3.488

Gilmartin CE, Jusino AM, Pyne JE, Banik TM, Lindner LD, Boddy L (2022) Fungal endophytes and origins of decay in Beech (F. silvatica) sapwood. Fungal Ecol 59:101161. https://doi.org/10.1016/j.funeco.2022.101161

Gotelli NJ, Ellison AM (2013) A Primer of Ecological Statistics, Second Edition. Sinauer Associates

Granhus A, Hylen G, Ørnelund-Nilsen J-E (2012) Statistics of forest conditions and resources in Norway 2005–2009. SoL, Ås

Hartig F (2020) DHARMa: Residual Diagnostics for Hierarchical (Multi-Level / Mixed) Regression Models. https://CRAN.R-project.org/package=DHARMa

Heilmann-Clausen J, Boddy L (2008) Distribution patterns of wood-decay basidiomycetes. In: Boddy L, Frankland JC, van West P (eds) Ecology of saprotrophic Basidiomycetes. Elsevier, Amsterdam, pp 263–275

Henriksen S, Hilmo O (2015) The Norwegian red list of species. Artsdatabanken, Trondheim

Hsieh TC, Ma KH, Chao A (2020) iNEXT: iNterpolation and EXTrapolation for species diversity. http://chao.stat.nthu.edu.tw/wordpress/software-download/
https://www.artsdatabanken.no/lister/rodlisteforarter/2021
Ihrmark K, Bodeker ITM, Cruz-Martinez K, Friberg H, Kubartova A, Schenck J, Strid Y, Stenlid J, Brandstrom-Durling M, Clemmensen KE, Lindahl BD (2012) New primers to amplify the fungal ITS2 region - evaluation by 454-sequencing of artificial and natural communities. FEMS Microbiol Ecol 82:666–677. https://doi.org/10.1111/j.1574-6941.2012.01437.x
Jacobsen R, Birkemoe T, Evju M, Skarpaas O, Sverdrup-Thygeson A (2023) Veteran trees in decline: stratified National monitoring of Oaks in Norway. For Ecol Manag 527:120624. https://doi.org/10.1016/j.foreco.2022.120624
Knudsen H, Vesterholt J (eds) (2012) Funga nordica: agaricoid, boletoid, clavarioid, cyphelloid and gastroid genera. Nordsvamp, Copenhagen
Kubartová A, Ottosson E, Dahlberg A, Stenlid J (2012) Patterns of fungal communities among and within decaying logs, revealed by 454 sequencing. Mol Ecol 21:4514–4532. https://doi.org/10.1111/j.1365-294X.2012.05723.x
Lindbladh M, Axelsson AL, Hultberg T, Brunet J, Felton A (2014) From broadleaves to spruce– the borealization of Southern Sweden. Scand J for Res 29:686–696. https://doi.org/10.1080/02827581.2014.960893
Lindenmayer DB, Laurance WF, Franklin JF (2012) Global decline in large old trees. Science Ecology Dec 7;338(6112):1305-6. https://doi.org/10.1126/science.1231070
Liu X, Liang M, Etienne RS, Gilbert GS, Yu S (2016) Phylogenetic congruence between subtropical trees and their associated fungi. Ecol Evol 6:8412–8422. https://doi.org/10.1002/ece3.2503
Menkis A, Redr D, Bengtsson V, Hedin J, Niklasson M, Nordén B, Dahlberg A (2022) Endophytes dominate fungal communities in six-year-old veteranisation wounds in living oak trunks. Fungal Ecol 59:101020. https://doi.org/10.1016/j.funeco.2020.101020
Mitchell RJ, Bellamy PE, Ellis CJ, Hewison RL, Hodgetts NG, Iason GR, Littlewood NA, Newey S, Stockan JA, Taylor AFS (2019) Collapsing foundations: the ecology of the British oak, implications of its decline and mitigation options. Biol Conserv 233:16–327. https://doi.org/10.1016/j.biocon.2019.03.040)
Moen A (1998) Nasjonalatlas for norge: Vegetasjon. Statens kartverk, Hønefoss
Moore JW (2010) Amsterdam is standing on norway' part II: the global North Atlantic in the ecological revolution of the long seventeenth century. J Agrarian Change 10:188–227. https://doi.org/10.1111/j.1471-0366.2009.00262.x
NBIC and GBIF Norway (2024) Species Map. Vis utvalg i kart| Artskart 2 (artsdatabanken.no). Accessed June 8, 2024
Nordén B, Ryberg M, Götmark F, Olausson B (2004) Relative importance of coarse and fine Woody debris for the diversity of wood-inhabiting fungi in temperate broadleaved forest. Biol Conserv 117:1–10. https://doi.org/10.1016/S0006-3207(03)00235-0
Oliveros JC (2015) Venny. An interactive tool for comparing lists with Venn's diagrams. https://bioinfogp.cnb.csic.es/tools/venny/index.html
Paltto H, Nordberg A, Nordén B, Snäll T (2011) Development of secondary woodland in oak wood pastures reduces the richness of rare epiphytic lichens. PLoS ONE 6:e24675. https://doi.org/10.1371/journal.pone.0024675
Parfitt D, Hunt J, Dockrell D, Rogers HJ, Boddy L (2010) Do all trees carry the seeds of their own destruction? PCR reveals numerous wood decay fungi latently present in sapwood of a wide range of angiosperm trees. Fungal Ecol 3:338–346. https://doi.org/10.1016/j.funeco.2010.02.001
Pellietier TP, Zak RD, Salley OS (2019) Environmental filtering structures fungal endophyte communities in tree bark. Mol Ecol 28:5188–5198. https://doi.org/10.1111/mec.15237
Polme S, Abarenkov K, Nilsson et al (2020) FungalTraits: a user-friendly traits database of fungi and fungus-like stramenopiles. Fungal Divers 105:1–16. https://doi.org/10.1007/s13225-020-00466-2
Prevedello JA, Almeida-Gomes M, Lindenmayer DB (2018) The importance of scattered trees for biodiversity conservation: A global meta-analysis. J Appl Ecol 55:205–214. https://doi.org/10.1111/1365-2664.12943
Rayner ADM, Boddy L (1988) Fungal decomposition of wood: its biology and ecology. Wiley, Chichester
Rodriguez RJ, White J Jr, Arnold AE, Redman RS (2009) Fungal endophytes: diversity and functional roles. New Phytol 182:314–330. https://doi.org/10.1111/j.1469-8137.2009.02773.x
Ryberg M, Nilsson RH (2018) New light on names and naming of dark taxa. MycoKeys 30:31–39. https://doi.org/10.3897/mycokeys.30.24376
Ryman SG, Holmåsen I (1992) Svampar: En Fälthandbok. Interpublishing, Stockholm
Ryvarden L, Gilbertson RL (1993) European polypores. Part 1. Synop Fungorum 6:1–387
Selosse MA, Schneider-Maunoury L, Martos F (2018) Time to re-think fungal ecology? fungal ecological niches are often prejudged. New Phytol 217:968–972. https://doi.org/10.1111/nph.14983
Sieber NT (2007) Endophytic fungi in forest trees: are they mutualists? Fungal biology reviews. 2175–89. https://doi.org/10.1016/j.fbr.2007.05.004

Smith CW (ed) (2009) The lichens of Great Britain and Ireland British Lichen Society, 1046 S
Sodhi GK, Saxena S (2023) Role of endophytic Fungi in promoting plant health. In: Singh I, Rajpal VR, Navi SS (eds) Fungal resources for sustainable economy. Springer, Singapore. https://doi.org/10.1007/978-981-19-9103-5
Sundberg S, Carlberg T, Sandström J, Thor G (red.) (eds) (2019) Värdväxters Betydelse för Andra organismer– med Fokus På Vedartade värdväxter. ArtDatabanken rapporterar 22. ArtDatabanken SLU, Uppsala
Tedersoo L, Bahram M, Zinger L, Nilsson RH, Kennedy PG, Yang T, Anslan S, Mikryukov V (2022) Best practices in metabarcoding of fungi: from experimental design to results. Mol Ecol 31:2769–2795. https://doi.org/10.1111/mec.16460
Terhonen E, Blumenstein K, Kovalchuk A, Asiegbu FO (2019) Forest tree microbiomes and associated fungal endophytes: functional roles and impact on forest health. Forests 10:42. https://doi.org/10.3390/f10010042
Thurner M, Yu K, Manzoni S, Prokushkin A, Thurner MA, Wang Z, Hickler T (2024) Nitrogen concentrations in boreal and temperate tree tissues vary with tree age/size, growth rate and climate. EGUsphere. https://doi.org/10.5194/egusphere-2024-1794
Tuovinen V, Svensson M, Kubartová A, Ottosson E, Stenlid J, Thor G, Dahlberg A (2015) No support for occurrence of free-living Cladonia mycobionts in dead wood. Fungal Ecol 14:130–132. https://doi.org/10.1016/j.funeco.2014.12.003
White TJ, Bruns T, Lee S, Taylor J (1990) Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In: Innis MA, Gelfand DH, Sninsky JJ, White TJ: PCR protocols: A guide to methods and applications. San Diego, USA, Academic Press, Inc.: pp 315–322