

## RESEARCH ARTICLE

# Vineyard maturity increases arbuscular mycorrhizal and decreases plant pathogen fungal relative abundance in bulk soil across a 1,000 km Chilean gradient

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## Funding information

Fondo Nacional de Desarrollo Científico y Tecnológico, Grant/Award Number: 1211655; Fondecyt Regular, Grant/Award Numbers: 1211655, 1240186 (2024); Estonian Research Council, Grant/Award Number: PRG1065;

## Societal Impact Statement

Wine production has a significant impact on the global economy. Despite this, the microbial composition of the *terroir* has not been studied sufficiently, particularly in the southern hemisphere. Here, we investigated how bulk soil fungal communities are affected by several abiotic and biotic factors across 1,000 km of Chilean vineyards. We found that geographical distance was the main driver of vineyard soil fungal communities. Irrespective of variety, older vineyards host a higher relative abundance of arbuscular mycorrhizal fungi (AMF) and a lower relative abundance of plant pathogenic fungi. This result could lead to significant recommendations on when to apply AMF-based inoculants.

## Summary

- The microbial dimension of the *terroir* has been increasingly recognized as an essential factor determining vineyard productivity and quality. Despite this, few studies have analyzed the factors affecting soil fungal communities of vineyards in the southern hemisphere. Across a 1,000 km gradient encompassing 34 Chilean vineyards, we investigated the effects of grapevine variety, geographical distance, maturity, and soil chemical properties on the diversity and composition of soil fungi.
- We implemented standard soil chemical analyses and ITS-based DNA metabarcoding of bulk soil.
- We found that the total soil fungal composition was significantly affected by geographical distance but not by grapevine variety and maturity. Soil chemical dissimilarity also significantly affected soil fungal composition. When analyzing specific fungal guilds, we found a contrasting successional pattern: arbuscular mycorrhizal fungal relative abundance was significantly higher at medium and old-maturity vineyards compared with young vineyards, on which plant pathogenic fungi had a markedly lower abundance.

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ANID – Chile projects SIA, Grant/Award Number: SA77210019 (2021); Corporación de Fomento de la Producción (CORFO), Grant/Award Number: PI-4452 (2020); Freundeskreis Trierer Universität e.V.; Fondecyt Iniciación, Grant/Award Number: 11230870 (2023)

- Our results present several caveats: the molecular marker was not the most commonly used for arbuscular mycorrhizal fungi, bulk soil was sampled (instead of roots), and all abundances were relative. Still, this study constitutes one of the most extensive studies on soil fungi – both for vineyards and Chile – and might have practical implications, as knowing the drivers of fungal and mycorrhizal biodiversity can inform agricultural management decisions.

#### KEYWORDS

arbuscular mycorrhizal fungi, biotic stress, Chilean vineyards, grapevines, terroir

## 1 | INTRODUCTION

Grapevine (*Vitis vinifera* L.) is among the world's oldest cultivated crops, and the wine sector is of outstanding cultural and economic importance (Powell, 2003; OIV, 2023). Investigating the complex interactions determining vineyard productivity and grape quality has received much attention and resulted in the introduction of the *terroir* concept (Van Leeuwen & Seguin, 2006). The *terroir* is “concerned with the relationship between the characteristics of an agricultural product (quality, taste, style) and its geographic origin” (Van Leeuwen & Seguin, 2006), which influences such characteristics. In addition to climate, factors such as grape variety, agricultural management (Van Leeuwen et al., 2004, 2018), and soil physico-chemical properties – such as soil texture (Deloire et al., 2004; Keller, 2015), mineral nutrient content, and pH (Mpelasoka et al., 2003; Verdenal et al., 2021) – are well known to affect grapevines' productivity quality. It is also increasingly understood that the microbial dimension of the *terroir* significantly impacts grapevine growth, health, and quality (Bokulich et al., 2014; Gobbi et al., 2022). For example, the soil microbiome may act as a source for grape- and grape juice-associated fungi (Liu et al., 2020), of plant pathogens causing root infection (Agustí-Brisach et al., 2013), and of mycorrhizal fungi that establish a beneficial symbiosis with plants (Trouvelot et al., 2015). Further, the soil microbiome may indirectly affect grapevines, i.e., via its impact on physico-chemical soil processes, such as nutrient cycling and soil aggregation (Hartmann & Six, 2023). Albeit our understanding of microbial diversity in vineyards is rapidly advancing, there are still uncertainties, mainly those related to its driving factors at different scales.

The recent benchmark study by Gobbi et al. (2022) identified climate and the spatial distance between vineyards – on a global and national scale – as the primary variables explaining beta diversity in fungal communities. They sampled bulk soil from 200 vineyards in four continents and found that the impact of geographical distance at the scale of continent, country, and region was significant in all instances (Gobbi et al., 2022). The general trend of such a study was that “the further the distance, the more diverse the community” (Gobbi et al., 2022). Spatial distance on larger scales (i.e., from landscape to continent or 100 m to >100 km) is typically correlated with marked gradients in geomorphology and soil characteristics (Berg, 2012; Frey, 2015). Similarly to Gobbi et al. (2022), Bokulich et al. (2014) also

found that vineyard climate and geographic location are important drivers of soil fungal communities, but in addition, the latter authors found that grape variety also affected such communities. Unfortunately, in large-scale and global-scale studies, as that by Gobbi et al. (2022), the southern hemisphere is often underrepresented ( $n = 39$ ) when compared to the northern hemisphere ( $n = 201$ ).

At finer scales, the soil microbiome is influenced by agricultural management practices (Likar et al., 2017; Vega-Avila et al., 2015; Zaller et al., 2018), grapevine genotype (Dries et al., 2021; Holland et al., 2014), and soil fertility (Burns et al., 2015; Zarraonaindia et al., 2015). Additionally, few studies have assessed how the vineyard soil microbiome may shift over time (Oyuela Aguilar et al., 2020; Pingel et al., 2019). For example, mycorrhizal communities have been shown to depend on the host development stage, i.e., grapevine plant age (Balestrini et al., 2010; Schreiner & Mihara, 2009). Consequently, it is not unlikely to assume that other soil fungal guilds (i.e., saprotrophic and plant pathogenic fungi) might also shift with increasing vineyard maturity, typically ranging from 30 to 50 years – depending on variety (Riffle et al., 2021).

Chile is the largest wine producer in the southern hemisphere (OIV, 2023). Despite this, few studies have assessed soil and *terroir* microbial biodiversity in Chilean vineyards. The pioneer work of Castañeda and Barbosa (2017) comparatively assessed soil microbial diversity in three Mediterranean vineyards from central Chile with nearby native forests. Despite some reported differences in soil physico-chemical properties (i.e., pH and texture), they found that both habitats shared most (87.1%) of the soil bacterial species, suggesting a high similarity of bacterial communities in these neighboring soil environments. Regarding the fungal community, the study reported lower abundances of Ascomycota and Basidiomycota in the studied vineyards as compared to the adjacent forest soils.

Therefore, this study was conducted to document the diversity and composition of soil fungi in Chilean vineyards. We assessed how grapevine variety, soil chemical properties, vineyard location, and grapevine maturity affect soil fungal diversity and composition. Given previous global-scale studies (Gobbi et al., 2022), we predicted that (i) spatial distance between vineyards affects bulk soil fungal diversity/composition and that (ii) the composition of bulk soil fungi shifts on a temporal scale, i.e., with increasing vineyard maturity. This should provide a better understanding of spatio-temporal patterns of soil fungal ecology in Chilean vineyard agroecosystems.

## 2 | MATERIALS AND METHODS

### 2.1 | Soil sampling

To investigate the effects of *V. vinifera* L. variety, soil chemical properties, vineyard location, and grapevine maturity on soil fungal communities, a survey was conducted across 34 vineyards (referred to as 1 to 34) in the primary grape wine-producing valleys of Chile during September to December 2021. These vineyards spanned a 1,000 km area, crossing various Chilean regions, including Coquimbo (29° 54' S), Valparaíso (33° 02' S), O'Higgins (34° 10' S), Maule (35° 25' S), Biobío (36° 49' S), and La Araucanía (38° 44' S). These areas experience an average annual rainfall of 1 to 1,246 mm, predominantly in winter. A total of 11 vine varieties were observed in these vineyards, including "Cabernet" (Cb), "Cabernet Sauvignon" (CS), "Carménère" (Ca), "Chardonnay" (Ch), "Malbec" (Ma), "Merlot" (Me), "Pedro Jimenez" (PJ), "Pinot Noir" (PN), "Riesling" (Ri), "Sauvignon Blanc" (SB), and (Pi) "Pinot". The term "Pinot", as used by communities and farmers in the La Araucanía Region, denotes landrace cultivars that have undergone mass selection over time. Further information about these sites can be found in a companion article, Aguilera et al. (2024) and in the dataset provided in Supporting Information, which includes vineyard variety, age, and standard soil chemical analyses. The vineyards' varieties ranged from 1 to 41 years of age. For each of the 34 vineyards, bulk soil samples were collected. A 30 × 30 m plot was marked for sampling in each vineyard. Three subsamples were gathered from each plot to create a composite vineyard sample. These samples were collected between rows and diagonally across the plot, covering two edges and a central point. To collect each subsample, nitrile gloves were used, the leaf litter was cleared, and around 1 kg of soil was taken with a 20 × 20 cm shovel previously cleaned and disinfected with 70% alcohol. The three subsamples were combined in a plastic bag, the roots were removed, and the big soil aggregates were dissembled. Then, the bag was gently shaken by hand to homogenize the soil. The samples were taken to a – 80°C freezer under cold conditions. In a companion study (Aguilera et al., 2024), we investigated the relative abundance and diversity of arbuscular mycorrhizal fungi (AMF) based on morphological analyses of spores.

### 2.2 | Soil DNA extraction and amplicon sequencing

A total of 34 soil samples were delivered under cold conditions to the Unidad de Genómica y Bioinformática, BIOREN-Universidad de La Frontera (Temuco, Chile), where DNA extraction and amplicon library preparation were carried out. DNA was extracted from the soil samples (0.25 g) using the PowerSoil® DNA Isolation Kit (Mo Bio Laboratories), according to the manufacturer's instructions. A blank sample was included as a negative control for PCR.

The amount of isolated DNA was quantified using 1X dsDNA HS Assay Kit in Qubit 4.0 (Invitrogen). DNA samples were then

prepared using barcoded library adaptors (400 nM) containing the fungal primers ITS1F (CTTGGTCATTAGAGGAAGTAA) and ITS2 (GCTGCGTTCTTCATCGATGC) (Ihrmark et al., 2012). PCR reactions were performed in triplicate and pooled, and the amplicon libraries were purified using the Agencourt® AMPure XP bead protocol (Beckmann Coulter, USA). Concentration was measured using a 1X dsDNA HS Assay kit in Qubit 4.0 (Invitrogen), and the quality was assessed using a DNF-935 kit in the Fragment Analyzer instrument (Agilent). Sequencing libraries were then pooled in equimolar concentrations and sequenced on an Illumina MiSeq platform (v.3) with 15% PhiX spike-in and a read length of 300 bp paired-end.

### 2.3 | Bioinformatic analyses

ITS reads were quality-filtered with the gDAT pipeline (Vasar et al., 2021). There were 2 × 5,628,134 raw sequences. Multiplexed paired-end reads were analyzed in the following way: barcode and primer sequences were matched, allowing 1 mismatch for both pairs and reads were trimmed to a 200 bp length to remove low-quality sections at the end. Only pairs where both reads had an average quality score of >30 were retained (after removal of barcode and primer sequences). Demultiplexed and quality-filtered paired-end reads were combined using FLASH (v1.2.10; Magoč & Salzberg, 2011) with the default parameters (10–300 bp overlap with at least 75% identity). Orphan reads (paired-end reads that did not meet the conditions for combination) were removed from the analyses. The VSEARCH (v2.14.1, Rognes et al., 2016) chimera filtering algorithm was used to remove putative chimeric reads in the de novo mode. ITSx (v1.1.3, Bengtsson-Palme et al., 2013) was used to obtain the region of the ITS1 sequences, resulting in 1,888,654 ITS1 reads. Cleaned reads were clustered with VSEARCH at 97% identity into 8,957 OTUs (excluding singletons). Representative sequences (OTU centroids) for each non-singleton OTU were taxonomically classified using a BLASTn (v2.11.0+; Camacho et al., 2009) search followed by selecting the best hit against the UNITE (v9.0; Abarenkov et al., 2024) database, resulting in 1,874,358 hits (8,050 OTUs). Sequences were assigned to taxonomic levels: orders, families, genera, and species were assigned at >80%, >90%, >95%, and >97% sequence similarity, respectively, using an 80% alignment length to discard sequences with partial alignment. Taxa falling below these thresholds were considered unknowns at particular taxonomic levels and were removed from the analyses. The FungalTraits database was used to assign trophic modes and functional guilds to fungal OTUs (Pöhlme et al., 2020). Both trophic modes and functional guilds were analyzed, as the former specifies the type of nutrition and is based on a broader taxonomic resolution, while the latter assess the specific fungal guild, which is classified at the genus level (Pöhlme et al., 2020). The raw sequence data obtained in this study are deposited in the Sequence Read Archive (NCBI), under BioProject number PRJNA1104547.

## 2.4 | Statistical analyses

Data was analyzed using R Studio (RStudio Team, 2020), and figures were produced using the package *ggplot2* (Wickham et al., 2016). The fungal OTU table was Hellinger-transformed before downstream analysis using the *decostand* function from the *vegan* package (Oksanen et al., 2013). Vineyard age was transformed into three maturity categories according to the following criteria: young ( $\leq 7$  years;  $n = 11$ ), medium ( $> 7$  to  $\leq 17$  years;  $n = 12$ ), and old ( $> 17$ ;  $n = 11$ ). Fungal community structure (beta diversity) was analyzed using the *metaMDS* function from *vegan* to perform nonmetric-dimensional scaling (NMDS) of the Bray–Curtis dissimilarity matrix. The *envfit* function from the *vegan* package was applied for correlation analysis of the community matrix and soil chemical variables.

Bray–Curtis dissimilarity was further related to temporal, chemical, and geographical distances in bivariate plots to assess the importance of those factors on fungal community dissimilarity. Euclidean distances were calculated for temporal and chemical differences using the base R *dist* function. The *distm* function from the *geosphere* package (Hijmans et al., 2017) was used to derive the Haversine distances from longitude and latitude. The *mantel* function from the *vegan* package (Oksanen et al., 2013) was used to test these relationships via Mantel tests.

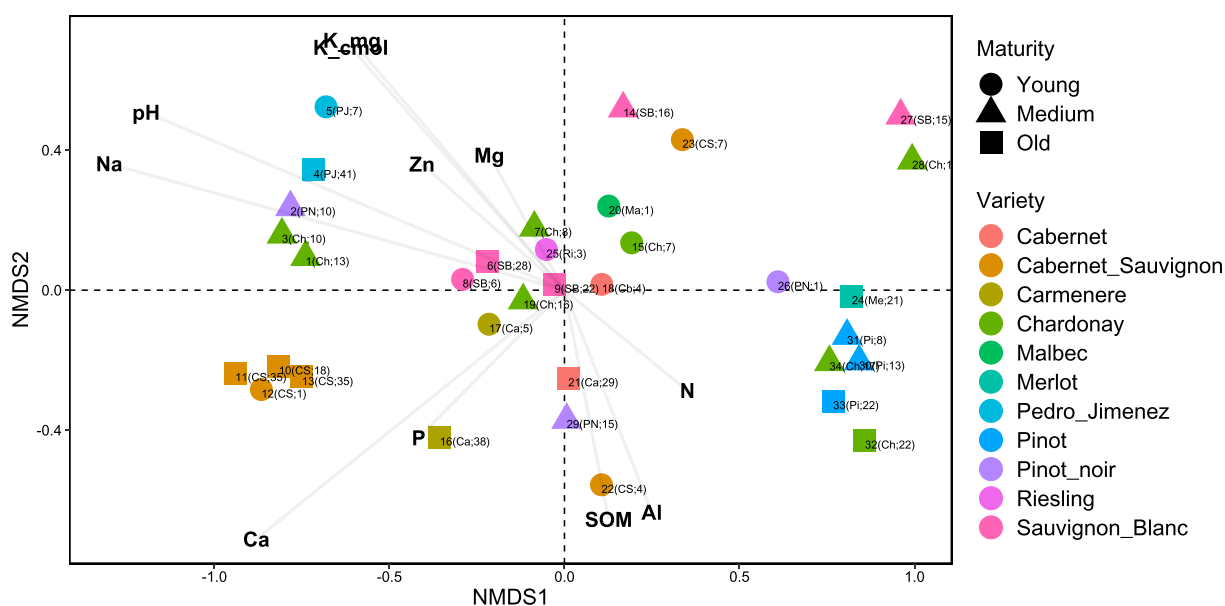
Alpha diversity indices were obtained using the *plot\_richness* function from the *phyloseq* package (McMurdie & Holmes, 2013). To

assess fungal trophic modes and guilds via *FungalTraits* (Pölme et al., 2020), we restricted the analyses to data entries marked with the confidence ranking “Highly probable”. One-way analysis of variance (ANOVA) was applied to test for statistical differences between differing stages of vineyard maturity using the base R *aov* function, and TukeyHSD was implemented as post-hoc tests.

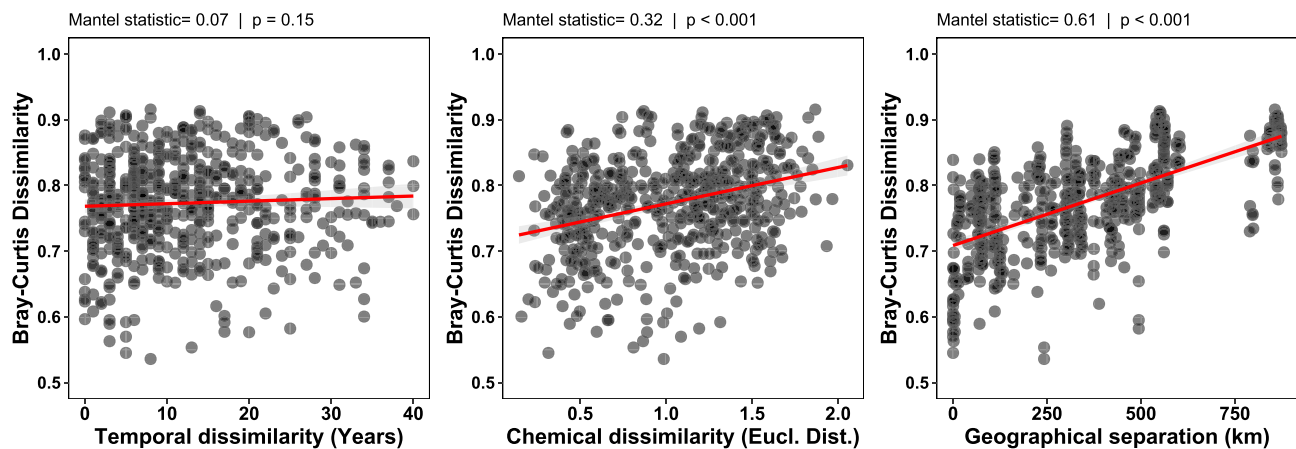
## 3 | RESULTS

Vineyard maturity and grape variety had a negligible impact on soil fungal community structure, as revealed by an NMDS ordination analysis; instead, sample divergence was more strongly affected by vineyard location (Figure 1). Nevertheless, some clustering was found between some varieties, like Pinot and Cabernet Sauvignon (Figure 1). Soil Na, pH, Ca, and K were most significantly ( $p < 0.05$ ) related to the NMDS ordination (Figure 1).

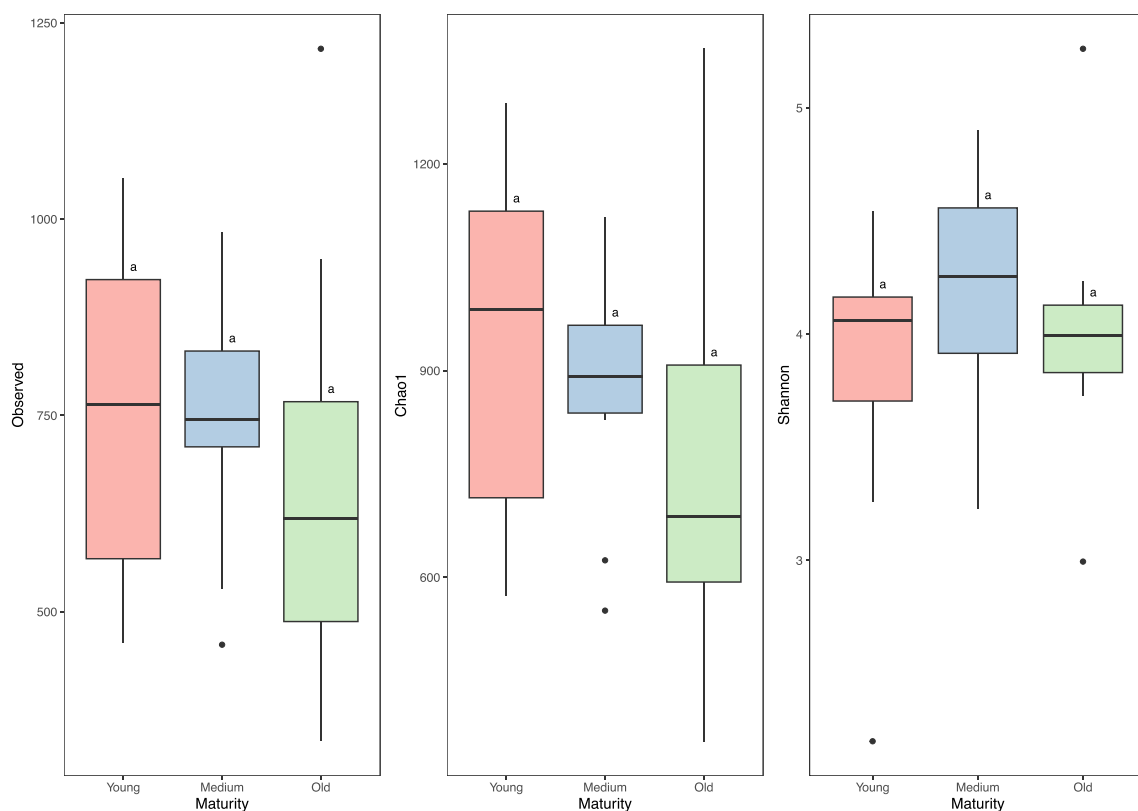
Vineyard maturity (temporal dissimilarity) did not significantly affected the total fungal community dissimilarity (Figure 2). Thus, when vineyard maturity was treated as either a categorical variable (Figure 1) or a continuous variable (Figure 2), it did not affect soil fungal community structure; instead, soil chemical differences ( $r = 0.32$ ;  $p < 0.001$ ) and geographical distances ( $r = 0.61$ ;  $p < 0.001$ ) between the sampled vineyards were positively and significantly related to fungal community dissimilarity, as revealed by Mantel tests (Figure 2). No alpha diversity pattern existed for all soil fungi across the 11 grapevine varieties



**FIGURE 1** Ordination of fungal community structure derived from ITS sequences based on non-metric multidimensional ordination (NMDS) of the Bray–Curtis dissimilarity. Symbols and colors represent vineyard maturity and grape variety, respectively. The number (before brackets) of the assigned vineyard identifier represents a longitudinal gradient, being the higher the number the further south the vineyard. Radial lines are derived from the “*envfit*” function and delineate relationships between edaphic soil variables and the fungal community matrix. The model output ( $r^2$  and  $p$ ) was as follows: N (0.06/0.487), P (0.10/0.224), K<sub>mg</sub> (potassium in mg/Kg) (0.24/0.012\*), pH (0.46/0.001\*\*\*), SOM (soil organic matter) (0.12/0.135), K<sub>cmol</sub> (potassium in cmol+kg) (0.23/0.015\*), Na (0.50/0.001\*\*\*), ca (0.35/0.002\*\*), Mg (0.05/0.423), AI (0.13/0.114), and Zn (0.08/0.267). For maturity: ANOSIM statistic R: 0.039, significance: 0.173. For variety: ANOSIM statistic R: 0.087, significance: 0.149. Stress: 0.115.



**FIGURE 2** Bivariate relationships between fungal community dissimilarity and temporal, soil chemical, and geographical vineyard dissimilarity.

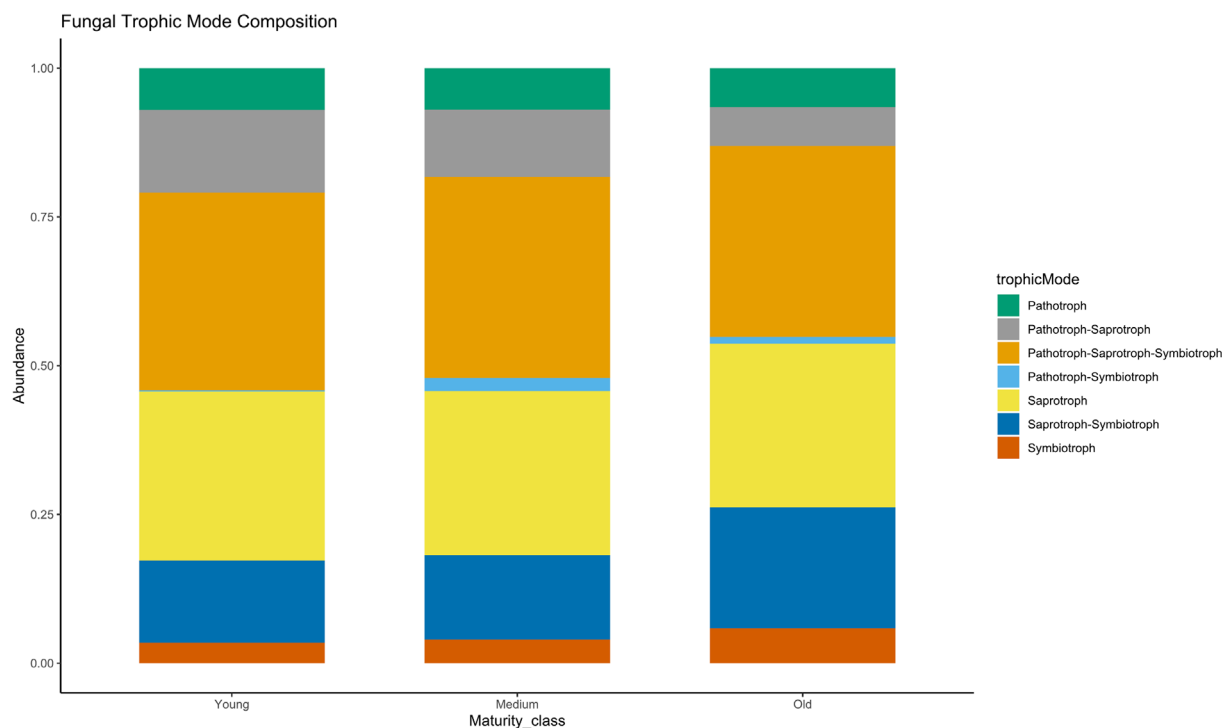


**FIGURE 3** Box plots indicating responses of fungal alpha diversity indices per vineyard maturity class. Letters indicate significant differences according to TukeyHSD tests.

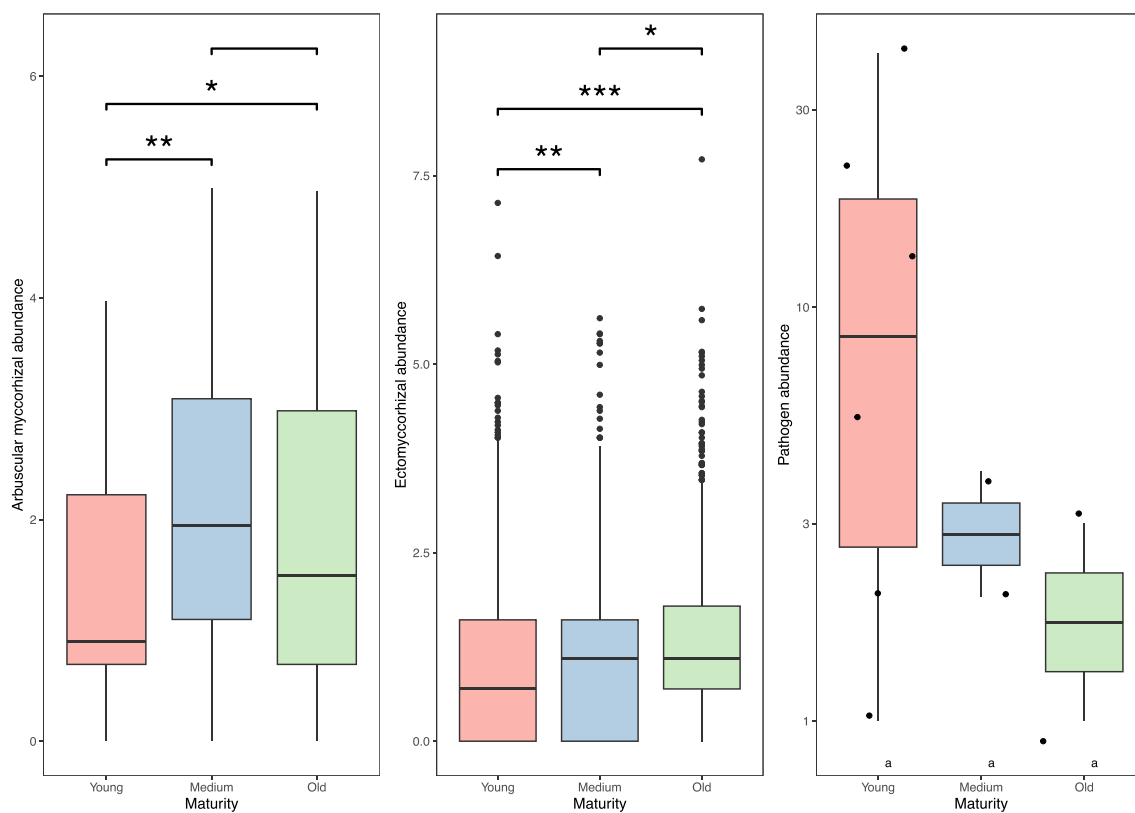
examined (Figure S1; Supporting Information). There were no significant statistical differences on either alpha diversity index between vineyard maturity classes (Figure 3), which varied from 335 to 1,217 (Observed richness), 362–1,367 (Chao1), and 2.2–5.3 (Shannon).

In contrast with total soil fungal communities, the relative abundances of fungal trophic modes (Figure 4) and guilds (Figure 5) were affected by vineyard maturity. Relative abundances of symbiotrophic modes increased with maturity, while the opposite pattern was found for pathotrophic modes (Figure 4). While the relative abundance of

arbuscular mycorrhizal fungi was significantly ( $p < 0.05$ ) higher in vineyards of medium and old maturity, younger vineyards showed a higher pathogen relative abundance. However, plant pathogen abundance was markedly but not significantly lower at medium and low maturity (Figure 5). Interestingly, ectomycorrhizal fungal abundance also significantly increased with vineyard maturity, albeit grapevines do not associate with this mycorrhizal type (Figure 5); the abundance of this guild was much lower than those of AMF and plant pathogenic fungi.



**FIGURE 4** Bar plots indicating the relative abundance of fungal trophic modes per vineyard maturity class.



**FIGURE 5** Box plots indicating arbuscular mycorrhizal, ectomycorrhizal, and plant pathogen fungi relative abundance per vineyard maturity class. Asterisks represent significance levels at  $p \leq 0.05 = *$ ,  $p \leq 0.01 = **$  and  $p \leq 0.001 = ***$  (t-test).



## 4 | DISCUSSION

Our study provides one of the most extensive assessments of soil fungal biodiversity in Chilean vineyards (along with Castañeda & Barbosa, 2017; Aguilera et al., 2024; and Aguilar-Paredes et al., 2024). Furthermore, this study significantly adds to the literature on arbuscular mycorrhizal (AM) and total fungal biodiversity of Chile (Marín et al., 2017, 2022). Supporting previous, global-scale research (Gobbi et al., 2022), we found that the soil fungal composition is significantly affected by geographical distance, while the effects of grape variety and maturity are negligible, in accordance with Fors et al. (2023), whom also found negligible effects of grapevine genotype in community structure. Our results contrast with those of Cureau et al. (2021), whom did find strong effects of vineyard genotype/variety on soil fungal communities.

Geographical distance and soil chemical distance (related to each other;  $r^2 = 0.367$ ,  $p < 0.001$ ) affected soil fungal community structure, but this was not the case with vineyard maturity. Nevertheless, a different pattern emerged when specific fungal guilds were analyzed: plant pathogenic fungi markedly decreased while arbuscular mycorrhizal fungi (AMF) significantly increased with vineyard maturity. Several global-scale studies on soil fungal communities have shown marked latitudinal patterns (Mikryukov et al., 2023; Tedersoo et al., 2014, 2022), so our findings showing marked effects of geographical distance on the whole soil fungal community are unsurprising. In a companion study (i.e., in the same plots) analyzing AMF through spore morphological analyses (Aguilera et al., 2024), we also found a marked latitudinal gradient with a higher number of spores and AMF species richness towards the northern vineyards. Previous studies at regional scales have also emphasized the importance of geographical distance in structuring whole soil fungal communities (Zhang et al., 2017) and AMF specifically (Hazard et al., 2013). It is quite possible that the factor 'geographical distance' confounds the effects of several other abiotic and/or climatic factors. Across the Coastal Mountain range of Chile, significant gradients of soil phosphorous (Brucker & Spohn, 2019) and rock weathering (Krone et al., 2021), among others, have been elucidated. Furthermore, Andean and Coastal soils/geological origins have distinct effects on soil (Marín et al., 2023) and AMF (Marín et al., 2017) communities. Thus, in our study, the effect of geographical distance on the whole soil fungal community structure is concordant with previous research but also reflects the marked ecosystem gradient (from arid to Mediterranean to temperate forest ecosystems) of the selected vineyards.

Soil characteristics and grapevine maturity have been shown to affect different soil fungal guilds, like AMF (Betancur-Agudelo et al., 2021). In our study, in particular, soil pH and cations (Na, Ca, K) were found to influence the total soil fungal community structure. Global-scale studies - based on thousands of soil samples - across the years (Mikryukov et al., 2023; Tedersoo et al., 2014, 2022) have shown the crucial role of soil pH in explaining soil fungal alpha diversity and community structure. Interestingly, recently Mikryukov et al. (2023) found quadratic relationships between soil pH and some of the fungal guilds examined in our study: AMF, ectomycorrhizal fungi, and

plant pathogens. Previous Chilean studies targeting whole soil fungal communities (Marín et al., 2023) or specifically AMF (Marín et al., 2017) have also found marked effects of cations in community structure, though in this study this might be related to the agricultural practices implemented at the vineyards.

Regarding the metabarcoding analyses, it is worth noticing that although we did not utilize the most used regions for AMF ecology, like the SSU (Öpik et al., 2010) or the LSU (Delavaux et al., 2021, 2022), the relative abundances of these guilds are comparable to many studies using such a region. Albeit the used primers do have mismatches against some AMF taxa, current global-scale AMF molecular databases are in fact incorporating the ITS2 region (Větrovský et al., 2023). A major caveat of this study is that we did not investigate the AMF inhabiting inside roots - which do select which fungi they allow in. Similarly - as in most, if not all metabarcoding studies - a major caveat of the current study is that trophic modes abundances were relative. Other techniques such as microbial biomass or fatty acids measurements should be used in order to tackle total abundance. In contrast with Betancur-Agudelo et al. (2021) - whom estimated AMF diversity based on morphological data, we found an increased relative abundance of AMF with vineyard maturity. In contrast, plant pathogen fungal relative abundance decreased with vineyard age. Although grapevines exclusively associate with AMF, we found that about 2% of fungal reads were ectomycorrhizal fungi. This might be explained by the surrounding vegetation, at least in some of the vineyards. Particularly, in southern Chile, the 10 species belonging to the genus *Nothofagus* associate with ectomycorrhizal fungi (Godoy & Marín, 2019), while central and northern Chile vineyards are most probably surrounded by *Pinus* and *Eucalyptus* plantations, which associate with invasive ectomycorrhizal fungi (Policelli et al., 2019). Recent hypotheses suggest that plant defense might be more critical in structuring AMF communities than carbon transfer (Frew et al., 2024) - but this needs further empirical evidence. Nevertheless, it is worth noting that plant pathogens were still somewhat prevalent in old vineyards; for example, we found two OTUs of the order *Phaeomoniellales*, which includes species such as *Phaeomoniella chlamydospora*, which is assumed as the primary causal agent of the two most destructive grapevine trunk diseases, Petri disease and esca (Kraus et al., 2020). In the same line of argument, in older vineyards, we found (Aguilera et al., 2024) AMF taxa - like *Gigaspora margarita*, *Funneliformis mosseae*, and *Claroideoglomus etunicatum* - that have been related to an increased biotic stress tolerance (Marro et al., 2022).

Despite its broad geographical range, our study has several limitations. First, it is possible that the different vineyards had different agricultural practices, which have been shown to affect soil microbial communities (Likar et al., 2017; Vega-Avila et al., 2015; Zaller et al., 2018). Second, we sampled composite bulk soil samples, but several studies have shown the significant effect that the grapevine rootstock has on structuring soil microbial communities (Berlanas et al., 2019; Darriaut et al., 2022; Dries et al., 2021; Marasco et al., 2018). Third, we had an uneven design due to restricted access, i.e., not the same number of vineyards for each variety and maturity

category combination. Other caveats include: the molecular marker was not the most commonly used for AMF, and because of the technique constraint, all abundance was relative. Further studies focused on roots and guild microbial biomass might show different patterns.

Across 1,000 km of 34 Chilean vineyards, we found that the total soil fungal composition is mainly affected by geographical and soil chemical distances but not by grapevine maturity and variety. AMF relative abundance increased with vineyard maturity, while the opposite pattern was found regarding plant pathogenic fungi.

## AUTHOR CONTRIBUTIONS

Conceptualization: César Marín, Paula Aguilera, Felix Dittrich, Martti Vasar. Methodology: César Marín, Felix Dittrich, Martti Vasar. Software: Martti Vasar. Formal analysis: Felix Dittrich, Martti Vasar. Investigation: Felipe Gaínza-Cortés, Patricia Silva-Flores, Paula Aguilera. Data curation: Martti Vasar. Writing—original draft: Felix Dittrich, César Marín. Writing—review and editing: César Marín, Felix Dittrich, Martti Vasar, Felipe Gaínza-Cortés, Patricia Silva-Flores, Paula Aguilera. Visualisation: Felix Dittrich. Supervision: César Marín, Paula Aguilera. Project administration: Paula Aguilera. Funding acquisition: Paula Aguilera.

## ACKNOWLEDGMENTS

This project was funded by the Fondo Nacional de Desarrollo Científico y Tecnológico (Fondecyt) Regular Project No. 1211655 (P.A.) by ANID – Chile. M.V. is supported by the Estonian Research Council (PRG1065). C.M. thanks the ANID – Chile projects SIA No. SA77210019 (2021) and Fondecyt Regular Project No. 1240186 (2024). F.G-C. thanks the Corporación de Fomento de la Producción (CORFO) Project No. PI-4452 (2020). F.D. acknowledges the financial support provided by “Freundeskreis Trierer Universität e.V.”. P.S-F. thanks the ANID – Chile project Fondecyt Iniciación No. 11230870 (2023).

## CONFLICT OF INTEREST STATEMENT

No conflict of interest is declared.

## DATA AVAILABILITY STATEMENT

The raw sequence data (fungal ITS2) obtained in this study are deposited in the Sequence Read Archive (NCBI) (<https://www.ncbi.nlm.nih.gov/bioproject/>), under BioProject number PRJNA1104547.

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#### SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

**How to cite this article:** Marín, C., Dittrich, F., Vasar, M., Gaínza-Cortés, F., Silva-Flores, P., & Aguilera, P. (2025). Vineyard maturity increases arbuscular mycorrhizal and decreases plant pathogen fungal relative abundance in bulk soil across a 1,000 km Chilean gradient. *Plants, People, Planet*, 7(4), 987–997. <https://doi.org/10.1002/ppp3.10598>

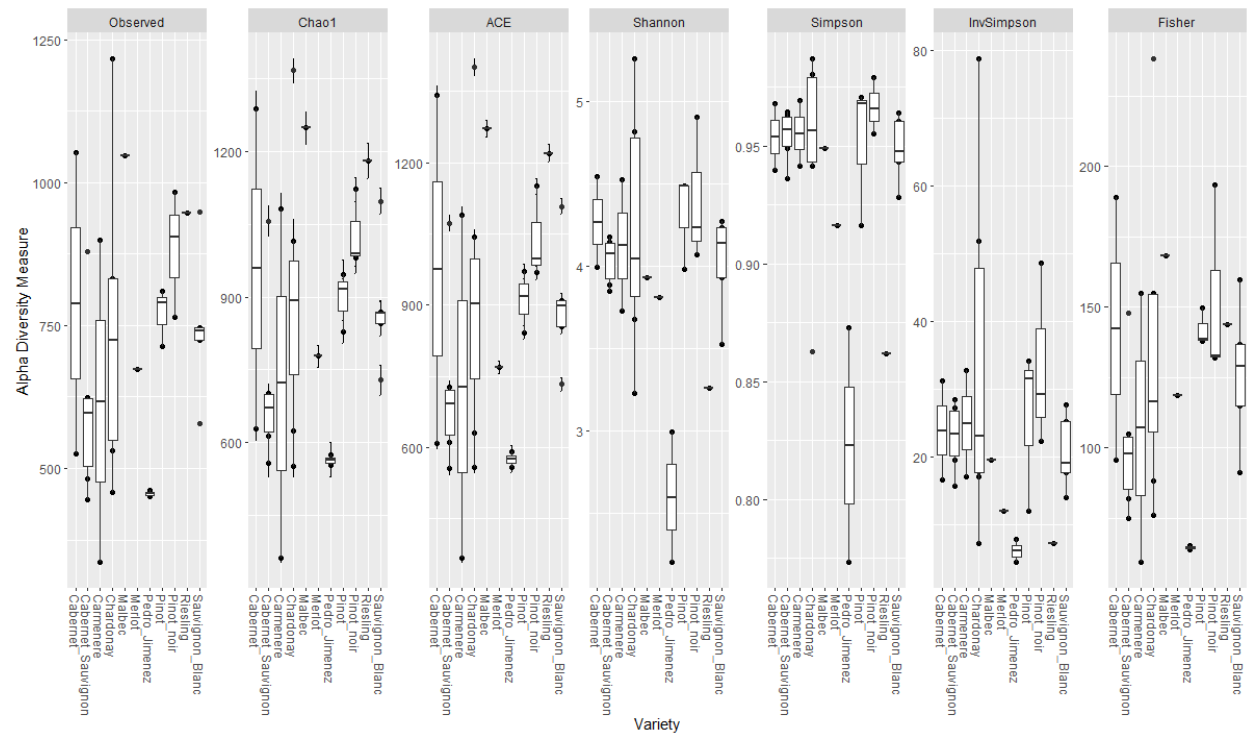
## ***Plants, People, Planet Supporting Information***

Article title: Vineyard maturity increases arbuscular mycorrhizal and decreases plant pathogen fungal abundance across a 1000 km Chilean gradient.

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The following Supporting Information is available for this article:

**Fig. S1:** Different Alpha diversity indices for all soil fungal diversity across the different vineyard varieties.



**Figure S1.** Different Alpha diversity indices for all soil fungal diversity across the different grapevine varieties.