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Abstract

Forest management aims to maintain sustainable production of quality wood while limiting increased competition between trees for light, water, and nutrients. Thinning is a widely used silvicultural practice to reduce plants competition for resources while still exploiting the wood. The investigation of the effects of forest management on stand functioning typically centers on the above-ground compartment, overlooking the alterations and influences exerted on below-ground biotic factors. Within the soil matrix, biological mechanisms are mainly governed by microbial communities. Many studies have focused on the effects of thinning on soil microbial communities (SMC), evidencing contrasted effects. Conversely, stand density effects on SMC are less documented. The aim of this study is therefore to focus on the effects of stand density (SD) on SMC biomass, gene abundance, functional diversity, and activity, according two sylvicultural practices: dynamic (low SD) and conservative (medium SD) in a temperate *Quercus petraea* Stand (QS) in Europe Forest. We hypothesized that dynamic silviculture (low-SD) could promote soil SMC biomass, abundance, functional diversity, and activity. Our results showed that dynamic silvicultural practices in oak forests reduced the abundances of bacteria, archaea and fungi were reduced by 43%, 29% and 34%, respectively. SMC functional diversity was reduced by 10% in dynamic forestry stands. On the contrary, dynamic silvicultural practices increased soil microbial activity by 13 to 47%, depending on the carbon source added, compared with conservative silviculture. Our results were incremented with an extensive number of biotic and abiotic environmental variables that had contrasting effects on SMC, and there is no single factor, which alone can explain all the SMC responses. Our results seem to advocate dynamic silvicultural practices in oak forests to promote soil microbial activity. However, it remains to be seen what the long-term effects will be of the reduced abundance and functional diversity of SMCs observed jointly in low-SD.

Key words: forest stand density, soil microbes, biomass, abundance, functional diversity, activity, environmental factors

Abbreviations

*Q*S	*Quercus* Stand
L-SD	Low Stand Relative Density
M-SD	Medium Stand Relative Density
MSIR	Multiple Substrates Induced Respiration
SMB	Soil Microbial Biomass
SMC	Soil Microbial Community
SOM	Soil Organic Matter

1. Introduction

Forests provide large ecosystem services, *i.e.* providing timber, food, habitat for biodiversity, regulating water resources, allowing recreational opportunities (Ding et al., 2011) and have a fundamental role in carbon sequestration (Duncker *et al.* 2012; Huang *et al.* 2020). Facing the continuous increase of atmospheric CO_2, research has focused on how forests can limit the CO_2 level on Earth's surface, through photosynthetic activity of trees and soil ability to store carbon (Peng *et al.* 2008). Nowadays, one of the major challenges of forest management is the balance trade-off between wood production and carbon sequestration potential (Favero *et al.* 2020). Increase in tree biomass has produced more litter, leading to soil accumulated carbon (Bolte *et al.* 2019), and estimations suggest that it will increase further in the coming years to the point that soil carbon storage may become more important than tree carbon storage, which appears to be the case already in the aging Central European forests (Liski *et al.* 2002; Jonard *et al.* 2017). Consequently, promoting litter production from living trees through forest management could better regulate soil carbon stocks. The global mean soil-derived respiratory of CO_2 emissions to the atmosphere overshadows by tenfold the annual CO_2 emissions from fossil fuel emissions (Oertel *et al.* 2016). The significance of soil lies in the fact that it is considered the most complex biomaterial and, at the same time, the most diverse and important ecosystem on Earth. On average, within a fertile soil, there will be more individual organisms than the total number of human beings who have ever lived: 1 trillion bacteria, 10,000 protozoa, 10,000 nematodes, 25 kilometers of fungi, and countless other species (Young and Crawford 2004). While most studies focus on forest management's impact on aboveground compartments for increased soil carbon sequestration potential, some research emphasizes the urgent need of understanding soil microbial ecology's role in carbon exchange between land and the atmosphere within the framework of climate change (Bardgett *et al.* 2008). Maximizing multiple benefits such as carbon sequestration from forest ecosystem

services requires better knowledge of the dynamics of biological soil functioning (Noormets *et al.* 2015). In the context of rapid global change, it is therefore essential to understand the influence of forest management on soil microbial communities, that contribute to the mineralization of organic matter (Ontl *et al.* 2020; Wang and Huang 2020; Dinca *et al.* 2021).

Reducing forest stand density, through thinning, is one of the main management strategies in temperate forest allowing to enhance wood production (Gauthier *et al.* 2015), and increase forest resilience to environmental disturbances such as drought (Sohn *et al.* 2016; Bastida *et al.* 2019). Lower stand density also increases stand sustainability through reduction of tree competition, and thus controls the maintenance of carbon storage in soil (Jandl *et al.* 2007). Canopy gap caused by thinning leads to changes in the microclimate with higher radiation that directly impact soil functioning, and indirectly through the development of understory vegetation. It is now well established that soil functioning will be mainly impacted by changes in abiotic parameters such as water content (Wang *et al.* 2018), C:N ratio (Masyagina *et al.* 2010), pH, organic carbon (Wu *et al.* 2019), fine root density and nutrient balances (Dang *et al.* 2018; Wang *et al.* 2019; Trentini *et al.* 2020; S Liu *et al.* 2021) but also biotic parameters: the soil microbial communities (SMC), which are responsible for a large part of the belowground activities. It is estimated that 80 to 90% of soil biological activity is carried out by bacteria and fungi on the topsoil (Gupta *et al.* 1997). By mineralizing most of the soil organic matter (SOM, mostly derived from residuals plant tissues and rhizodeposition, Chaparro et al., 2013), they contribute to the maintenance of soil functioning and regulate the nutrients cycling (Tefs and Gleixner 2012; Adeleke *et al.* 2016; Lladó *et al.* 2018). Moreover, beyond influencing these key ecological processes, SMC interact reciprocally with plants to the point of being an integral part of their functioning as resource acquisition strategy (Fernandez *et al.* 2022; Han *et al.* 2023). More than just a tool, the SMC has thus become a relevant component to be integrated to optimize forest management strategies like thinning (Staddon *et al.* 1999; Lladó

and Baldrian 2017), in the context of climate change. It is well established that tree species impact SMC, notably by releasing specific chemical composition of carbon substrates (Jiang *et al.* 2012; Prescott and Grayston 2013; Gartzia-Bengoetxea *et al.* 2016; Khlifa *et al.* 2017), but effect of thinning on microbial biomass, abundance, respiration, and catabolic profiling is less understood (Dang *et al.* 2018; Kim *et al.* 2019). A short review of the literature on forest management reveals that thinning could have contradictory impacts on the microbial community. **Table 1** presents the response (increase, decrease or no significant effect) to thinning of four SMC parameters such as (i) total microbial biomass C and N, (ii) microbial abundance, (iii) diversity and (iv) activity. The table clearly illustrates that the responses of the SMC to thinning are most often variable within the same study. Responses to thinning also vary according to the SMC domain considered: bacteria, archaea and fungi. Concerning soil basal respiration, variable responses to thinning were measured: increase (Zhang et al., 2018: in broadleaves and mixed forest), decrease or stable (Zhang et al., 2018: in coniferous forest). Several additional factors have been recognized as influencing SMC, including precipitation, temperature, season, forest site exposure and litter amount, composition, and decomposition stage (Nave *et al.* 2010; Jonard *et al.* 2017; Lladó and Baldrian 2017; Richter *et al.* 2018; Xiao *et al.* 2018). It is commonly understood that soil microbial biomass, abundance, diversity, and activities depend on environmental variations (Bolat 2014; Yang *et al.* 2017). Moreover, as thinning induced a decrease of the stand density, this led to changes of the microclimate in the understory and in the soil. Trees, through their species or age for instance, can also introduce changes to both physicochemical and biological soil characteristics. Therefore, changes of SMC parameters can be attributed to change in soil pH, water content, organic matter, moisture, nutrient availability, temperature, litter characteristics, understory plants, radiation, microclimate, tree roots traits and rhizodeposits etc. (Lladó *et al.* 2018; Wu *et al.* 2019). Contribution of environmental factors can have different importance on SMC, for

instance, Chodak and Niklińska (2010) showed that soil texture had more effect than planted vegetation on SMC parameters.

The diverse effects of thinning on SMC result from the interplay between biotic and abiotic factors that shape forest soil microbial structure and activity (Mabuhay *et al.* 2006; Griffiths and Philippot 2013; Simonin and Richaume 2015). This complex assemblage makes it challenging to understand how silvicultural practices impact SMC.

Many studies, including those referenced in **Table 1**, primarily examine the immediate impact of forest thinning intensity, while the longer-term influence of stand density is less explored and documented. Regarding the stand density, Wang et al., (2021) wrote that "no comprehensive analysis of soil enzyme activities and microbial compositions, nor any detailed observations of correlations between biological and physicochemical properties, have been performed". The overarching goal of this study was therefore to move beyond the examination of thinning as a transient disturbance, and to focus on the effect of stand relative density (SD) on SMC. We compared the effects of two silvicultural practices: dynamic *i.e.*, low stand relative density (L-SD) and conservative *i.e.*, medium stand relative density (M-SD) on microbial community after one year since the last tree cut. We measured the effect of these two SD on (i) soil basal respiration, (ii) the soil microbial biomass carbon (SMB-C) and nitrogen (SMB-N), (iii) the bacterial, archaeal, and fungal gene abundance (by quantitative PCR), and (iv) the SMC functional diversity and activity. The project was conducted on an experimental device (OPTMix), for which abiotic environmental data (rainfall, water table depth, temperature, etc.) and biotic data (vegetation cover of understory species) were measured. We hypothesized that dynamic silviculture (low-SD) could promote soil SMC biomass, abundance, functional diversity, and activity. We also expected biotic and abiotic environmental factors, such as precipitations, would influence SMC, but to a lesser extent compared to SD. Lastly, we expected the results to highlight the significance of extending

analysis beyond the short-term effects of thinning. We aimed to emphasize the enduring impact of SD on SMC, particularly in perennial ecosystems like forests. This perspective could play a pivotal role in advancing our understanding of soil biological processes.

2. Materials and methods

2.1.Study area

Sampling and measurements were done at the OPTMix (Oak Pine Tree Mixture) experimental site in the Forêt d'Orléans, France (47.82717°N, 2.45313°E, **Figure 1**). OPTMix consists of a network of even-aged adult forest plots (33 plots over 40 ha) that have been managed by the INRAE Forest Ecosystems Research Unit (Nogent-sur-Vernisson, France) to isolate and study the effects of various forest management strategies (tree densities, pure and mixed stands, presence/absence of large wild ungulates thanks to enclosures that exclude deer and wild boar) on ecosystem functioning. Each plot is about 0.5 ha and tree populations are 60-80 years old. Soils are composed of a sandy loam top layer (0-50 cm depth) with an increasing gradient of clay below and are classified as planosols (Lamotte et al., 1988, **Table 2**). Common understory vegetation includes purple moor grasses (*Molinia caerulea* (L.) Moench), ferns (*Pteridium aquilinum* (L.) Kuhn) and heath (*Calluna vulgaris* (L.) Hull). We focused on 3 mono-specific oak (*Quercus petraea* (Matt.) Liebl), one of the most widespread tree species in France) stands, each composed of 2 plots with different stand density. Stand density index measures the density of a stand of trees based on the number of trees per unit area and diameter at breast height (DBH) of the tree of average basal area (Reineke 1933). For each *Quercus* Stand (*Q*S), there are one plot in low stand density (L-SD) and another in medium stand density (M-SD). Plots density was evaluated using Relative Density Index (SD): 0.4 for L-SD and 0.7 for M-SD that were achieved by thinning between 2012 and 2017. The mean oak diameter in the 6 plots was 24.4 cm. The intensity of thinning varies according to the

plots insofar as they aimed to achieve a specific SD. The plot characteristics were presented in **Table 3**.

2.2.Sampling design

Soil samples were taken from the 6 forest plots (3 *Q*S x 2 SD) in June 2018. In each plot, a total of ten soil cores of the top 10 cm of soil (litter layer excluded) were randomly collected within the plot and pooled together to form a single composite sample. We therefore had one soil sample per plot, for a total of six soils. Each of the six composite soil samples was then sieved in a 2 mm sieve to homogenize and remove roots and rock fragments. After this step, each of the six samples was split into 4 subsamples for technical replicates, for a total of 24 soils. For these 24 soil samples, a portion of each sample was flash-frozen for molecular biology experiments to avoid DNA damaging. Another portion of each sample was used for microbial biomass C and N extraction and water content estimation. The remaining soil was stored in a freezer at -20°C for two years for MicroResp analyses.

2.3.Soil basal respiration

Soil basal respiration was measured *in situ* by a closed dynamic system, composed from a portable infrared gas analyser (EGM4, PPsystems, Hitchin, UK), connected to a soil respiration chamber (SRC1, PPsystems, Hitchin, UK). The chamber (100 mm diameter, 150 mm high) was set up directly on soil for measurement. In each plot, 30 measures were conducted in June 2019.

2.4.Soil microbial biomass C and N

Soil microbial biomass C (SMB-C), N (SMB-N) and microbial C:N ratio were estimated by determining and comparing the carbon and nitrogen contents in unaltered and treated samples by fumigation with chloroform. Fumigation method is presented in **Appendix 1**.

2.5.Microbial gene abundance

The gene abundance of total bacterial, archeal and fungal microbial communities were estimated by quantitative PCR (qPCR) assays ($n = 4$ technical replicates for each plot). Total bacterial and archaeal communities were targeted using 16S rDNA genes and fungal communities by using 18S rDNA genes (**Table 4**). DNA extraction and gene amplification methods are presented in **Appendix 2**.

Microbial gene abundances were expressed as gene copy numbers per gram of dry soil.

2.6. Microbial functional diversity and activity

Activity was measured by assessing the Multiple Substrate-Induced Respiration. MSIR was determined with the MicroResp™ method using the functional capacities of carbon sources mineralization (Campbell *et al.* 2003). Soil samples were first incubated in a 96 deep-well plate for 2 weeks at 25°C to stabilize the microbial communities (Lerch *et al.* 2011) before substrate addition. Fifteen different substrates belonging to 3 different molecular families were selected: 5 sugars (D-fructose, D-glucose, D-galactose, L-arabinose and D-(+)-trehalose dehydrate); 6 amino acids (L-alanine, N-acetylglucosamine, L-lysine-HCl, L-proline, L-cysteine-HCl monohydrate and γ-aminobutyric acid), and 4 carboxylic acids (citric acid, ascorbic acid, L-malic acid, and α-ketoglutaric acid). Final substrates concentration was 30 mg C mL^{-1} and substrates addition brought the water content to 60 % of water holding capacity. Thereafter, the soils were incubated for 6 h at 25 °C and the absorbance of each well was measured at a wavelength of 570 nm using a microplateplate reader (BioTek Eon™). After conversion of absorbance into CO_2 flux, MSIR was calculated for each substrate by subtracting the respiration of the control (without substrate) to that of the total respiration. Total substrate mineralization was calculated as the sum of CO_2 evolved for each substrate and the functional diversity of SMC based on MSIR was estimated using the Shannon index calculated as followed:

$$H = -\Sigma\, pi \times ln\, pi$$

where pi is the respiration response to the substrate i as a proportion of total substrate mineralization. We then search among the different biochemical classes of substrate (carbohydrates, amino acids, and organic acids), through an analysis of variance, if one or more of these classes are more specifically used by SMC).

2.7.Environmental parameters

The environmental parameters in each plot, were obtained from the OPTMix dataset. A total of 24 environmental parameters were tested but only those with the highest number of significant correlations ($n>8$, $r>|0.3|$ and $p<0.05$) with the SMC parameters were presented in the results. The 24 environmental parameters have been divided into 5 categories: (i) stand characteristics (*SD*, *final volume of standing trees* in the plots after thinning and *total volume of cutting trees*, representing thinning intensity), (ii) water properties (sum of *precipitation* under the tree canopy, *soil water content*, *depth of the water table during the last 30 days before the soil harvest* and *depth of the water table on the day of the soil harvest*), (iii) the physicochemical properties of the soil (soil texture including *clay*, *sand* and *silt*, percentage of *organic matter*, *organic carbon*, *nitrogen*, *calcium*, *potassium*, *magnesium*, *pH*, *thickness of the organic horizon* (OH), *cation exchange capacity*, *average soil temperature during the last 30 days prior to soil harvest*), (iv) litter mass (*average of leaf litter mass during the last 30 days prior to soil harvest*) and (v) the average vegetation cover of understory species (*Calluna vulgaris*, *Molinia caerulea* and *Rubus fructosa*) within each of the six plots. The different methodologies used to obtain the different environmental parameters in OPTMix forest are detailed in Bello et al., 2019, Korboulewsky et al., 2015 and Perot et al., 2019.

2.8.Statistical analysis

Statistical analyses were performed using R software (Version 3.4.1.). The MSIR data were log-transformed before the statistical analyses for the normalization. The data were tested for normality and homoscedasticity using Shapiro-Wilk test and using Levene test, respectively. ANalysis Of VAriance (ANOVA, $\alpha = 0.05$) was performed to assess the effects of thinning on SMB-C, SMB-N, microbial gene abundance, functional diversity of SMC, MSIR and soil basal respiration. For each SD, there were 3 biological replicates. Correlation coefficient with environmental factors and associate p-value were performed using the correlation function from the easystats {correlation} package. Pearson correlations between microbial and environmental parameters were considered significant at $p\text{-}value \leq 0.05$ and non-significant data were identified as "*ns*". We logically did not compare environmental data measured exclusively in 2018 with soil basal respiration data measured in 2019.

3. Results

3.1.Soil basal respiration

The soil basal respiration was not significantly impacted by forest SD (**Figure 2**), with a value of 3.64 ± 0.17 and 3.54 ± 0.15 µmol CO_2 m^{-2} s^{-1} in L-SD and M-SD, respectively. Correlation analysis also shows no relationship between SD and soil basal respiration (**Table 5 –** SD column).

3.2. Soil microbial biomass C and N

SMB-C mean tended to be higher in L-SD (1.3 times) compared to M-SD ($p = 0.07$, **Figure 3.a**), with a value of 858.47 ± 105.78 and 644.41 ± 36.23 $mg.kg^{-1}$ in L-SD and M-SD, respectively. However, although Anova's analysis showed a trend, no correlation was found between SMB-C and SD (Pearson analysis) (**Table 5 –** SD column). SMB-N was not significantly affected by SD, with a value of 58.86 ± 9.64 and 50.81 ± 5.06 $mg.kg^{-1}$ in L-SD and M-SD, respectively (**Figure 3.b**). Similarly, microbial C:N ratio was not significantly

affected by SD, with a value of 16.57 ± 1.30 and 13.80 ± 1.19 mg.kg^{-1} in L-SD and M-SD, respectively (**Figure 3.c**). Biomass C tended to be higher in L-SD (ANOVA analysis).

3.3. Soil microbial gene abundance

Regarding the microbial gene abundance, SD showed significant impact on the three SMC domains. M-SD led to a higher gene abundance compared to L-SD for each group: bacteria ($p = 0.005$), archaea ($p = 0.04$) and fungi ($p = 0.007$), corresponding to an increase of 43%, 29% and 34% respectively (**Figure 4.a, b** and **c.**). The Archaea:Bacteria ratio (A:B ratio) was significantly higher ($p = 0.02$) under L-SD compared to M-SD, but there was no effect of SD on the F:B ratio (**Figure 4.e** and **f.**). According to correlation analysis, there was no strong effect of SD on SMC gene abundance (positive correlation coefficients are lower than 0.5, **Table 5 –** SD column).

3.4.Microbial functional diversity and MSIR

Functional diversity of SMC was significantly lower (1.1 times) in L-SD than in M-SD ($p < 0.001$) evidencing clear differences in the microbial functional diversity composition between L-SD and M-SD (**Figure 5**).

The CO_2 rate was significantly lower in M-SD for all the substrates ($p < 0.02$) excepted galactose ($p = 0.15$, **Figure 6**). Ketoglutaric acid stood out with the highest increase between L-SD and M-SD (47%). For the other substrates, the increase varied between 13% (water) and 33% (alanine). The substrates nature also influenced respiration as carboxylic acid led to higher CO_2 rate, especially ascorbic, malic and ketoglutaric acid (> 0.19 µg C-CO_2 g^{-1} h^{-1}) while amino acids cause the lower, especially lysine, proline, and cysteine (< 0.09 µg C-CO_2 g^{-1} h^{-1}). Water caused the lowest functional activity (< 0.05 µg C-CO_2 g^{-1} h^{-1}).

3.5.Correlations between environmental factors and microbial parameters

Table 5 shows the significant correlation coefficients between the 26 SMC parameters (rows in the table) presented previously and the environmental parameters (columns in the table) that showed the highest number of correlations ($n \geq 8$, $r \geq |0.3|$ and $p < 0.05$). Of the 24 environmental parameters studied, 15 corresponded to the above criteria. The correlations were variable depending on the SMC parameters (biomass, gene abundance, functional diversity and activity).

Stand characteristics

SD was positively correlated with the functional diversity of SMC ($r = 0.57$ and $p < 0.001$) and negatively with the respiration induced by α-ketoglutaric acid ($r = -0.5$ and $p < 0.001$) and citric acid ($r = -0.51$ and $p < 0.001$). The correlation between SD and respiration induced by other substrates was globally negative and moderate ($-0.5 < r < -0.3$ and $p < 0.05$). The final volume of standing trees per hectare presented correlation coefficients logically similar to those of the SD with globally higher values. The total volume of cut trees per hectare had a significant positive correlation with microbial C:N ratio ($r = 0.59$ and $p < 0.001$) and negative correlations with bacterial ($r = -0.62$ and $p < 0.001$), archaeal ($r = -0.58$ and $p < 0.001$), and fungal ($r = -0.62$ and $p < 0.001$) gene abundances. However, the total volume of cutting trees did not exhibit any correlation with the SMC (except for α-ketoglutaric acid, $r = 0.79$ and $p < 0.001$), unlike the SD.

Hydric properties

The environmental parameter that exhibited the highest number of correlations with the different microbial parameters was the average precipitation of the last 30 days before soil harvesting. A total of 17 microbial parameters had a significant correlation greater than |0.5| with precipitation. Precipitation had significant positive correlations with microbial biomass C ($r = 0.71$ and $p < 0.001$) and N ($r = 0.84$ and $p < 0.001$) but negative with microbial C:N ratio

(r = -0.74 and p < 0.001). Precipitations also had significant positive correlations with the gene abundance, especially with archaea (r = 0.54 and p < 0.001), and MSIR (except α-ketoglutaric-induced respiration for which the correlation coefficient was negative, r = -0.77 and p < 0.001, and there was no correlation with citric acid). Contrary to the precipitations, soil water content of the last 30 days before soil harvesting showed a significant negative correlation with each bacterial (r = -0.63 and p < 0.001), archaeal (r = -0.61 and p < 0.001), and fungal (r = -0.65 and p < 0.001) gene abundance and did not have correlation with MSIR (except α-ketoglutaric-induced respiration, r = 0.82 and p < 0.001). The average water table depth during the 30 days prior to soil harvesting had a significant (p < 0.001) negative correlation with microbial C:N ratio (r = -0.71) and positive correlations with each bacterial (r = 0.63), archaeal (r = 0.64), and fungal (r = 0.67) gene abundance. On the contrary, the average depth on the day of harvesting did not exhibit correlation with microbial gene abundance and had negative correlation with microbial biomass C (r = -0.53 and p < 0.001). The average depth on the day of harvesting had positive correlation with functional diversity of SMC (r = 0.62 and p < 0.001) and negative correlations with MSIR.

Soil physicochemical properties

Soil organic matter had negative correlations with soil basal respiration (r = -0.70 and p < 0.001) and the microbial C:N ratio (r = -0.83 and p < 0.001). Conversely, it had positive correlations with microbial biomass C (r = 0.57 and p < 0.001) and N (r = 0.85 and p < 0.001) and each bacterial (r = 0.58 and p < 0.001), archaeal (r = 0.71 and p < 0.001), and fungal (r = 0.56 and p < 0.001) gene abundance. The correlation between SOM and respiration induced by substrates was globally positive and moderate (0.3 < r < 0.7 and p < 0.01, except with α-ketoglutaric acid which exhibited negative correlation, r = -0.67 and p < 0.001, and there was no correlation with citric acid). Cation exchange capacity correlation was positive with soil basal respiration (r = 0.59 and p <0.001) and was globally negative and moderate with

microbial biomass C and N, and MSIR (-0.6 < r < -0.4 and p < 0.001, except with α-ketoglutaric acid which exhibited positive correlation, r = 0.75 and p < 0.001, and there was no correlation with citric acid). Soil temperature mean the 30 days prior to soil harvest had negative and moderate correlation with SMC gene abundance (-0.5 < r < -0.3 and p < 0.05) and with functional diversity of SMC (r = - 0.55 and p < 0.001). On the contrary, it had globally positive and moderate correlation with MSIR (0.3 < r < 0.6 and p < 0.05).

Litter mass

Leaf litter mass had negative correlations with microbial biomass C (r = -0.66 and p < 0.001) and N (r = -0.54 and p < 0.001), and MSIR (except α-ketoglutaric-induced respiration for which the correlation coefficient was positive, r = 0.54 and p < 0.001, and there was no correlation with citric acid).

Understory species cover

Calluna vulgaris cover in the understory had a negative correlation with functional diversity of SMC (r = -0.6 and p < 0.001) while it exhibited a positive correlation positive with MSIR (0.5 < r < 0.9 and p < 0.001). *Molinia caerulea* cover had positive correlations with soil basal respiration (r = 0.5 and p < 0.001), microbial C:N ratio (r = 0.62 and p < 0.001) and moderate with MSIR. On the contrary, it had negative correlation with each bacterial (r = -0.52 and p < 0.001), archaeal (r = -0.48 and p < 0.001), and fungal (r = -0.54 and p < 0.001) gene abundance. Among the understory species, *Rubus fructosus* cover had the greatest number of coefficient correlation with microbial parameters. The correlations between R. *fructosus* and soil basal respiration, microbial biomass and gene abundance were similar to those obtained with *M. caerulea*. In contrast, there was a global negative correlation between *R. fructosus* and MSIR (-0.6 < r < -0.4 and p < 0.001, except α-ketoglutaric-induced

respiration for which the correlation coefficient was positive, $r = 0.79$ and $p < 0.001$, and there was no correlation with citric acid).

4. Discussion

In line with the short review of forest thinning research (**Table 1**), our results highlight the diverse influence on soil microbial communities, depending on the specific stand characteristics and environmental factors considered.

4.1.Soil basal respiration

In our study, soil basal respiration was not influence by stands characteristics, but we noted a moderate positive correlation with thinning intensity (*i.e.* total volume of cutting trees per hectare). Soil basal respiration can exhibit differing trends based on the specific study conditions. For instance, consistently with our finding, thinning intensity has been linked to an increase in soil respiration (Lee *et al.* 2023). However, Liu et al., (2021) showed that soil respiration was higher in stands with medium density when compared to those with low density, but it also depends on the age of the stand. These outcomes suggest that soil respiration is likely influenced by stand characteristics, including temporary disturbances (such as thinning) and SD. Impacts on soil basal respiration are thus diverse and seem to arise from a combination of multiple factors.

4.2. Soil microbial biomass C and N

Global mean of microbial biomass in this study was similar to those measured in *Quercus sessiflora* Morvan forest (Lejon *et al.* 2005) and in *Quercus petraea* forest in Turkey (Bolat and Şensoy 2019). Our results showed that SMB-C and SMB-N were not significantly impacted by SD, but we did observe that biomass C tended to be higher in low-density stands. However, neither SD nor thinning (total volume of cutting trees per hectare) showed any correlation with SMC, contrary to studies on *Quercus* forests (Kim *et al.* 2018, 2019) and *Pinus*

forests (Bolat 2014; Wu *et al.* 2019), that observed an increase with thinning intensity. Through global meta-analysis, Zhou et al., (2020) demonstrated that thinning does not affect microbial biomass, highlighting a discordance regarding the effects of thinning on the SMC biomass. Is our study, we can conclude that SD and, more broadly, the characteristics of the forest stand, have not significant impact on SMB-C and SMB-N.

4.3. Soil microbial gene abundance

The effect of SD on microbial gene abundance can be interpreted differently depending on whether ANOVA or Pearson correlation analysis is considered. According to the ANOVA, microbial gene copies number was lower in L-SD for each SMC domains, but there was no effect through correlation analysis. The ANOVA clearly showed that microbial gene abundance was higher in medium-density stands than in low-density stands. Pearson's correlation coefficients indicate moderate positive correlations between SD and microbial gene abundance, except for archaea where there is no correlation. Yet, the correlation matrix showed a negative correlation between thinning intensity (total volume of trees cut per hectare) and microbial gene abundance, reinforcing the idea that decreasing tree density in the forest stand induced a decrease in microbial gene abundance. Cai et al., (2020) and Wu et al., (2019) results revealed that effect of thinning on relative abundance of the soil dominant bacterial taxa varied according to thinning intensity. Medium-intensity thinning tended to increase of some bacterial taxa (*e.g.* Gram-positive and Gram-negative) relative abundance. On the contrary, low-intensity thinning, which leads to higher stand density than medium-intensity thinning, caused a decrease of bacterial taxa relative abundance (*e.g.* *Gemmatimonadetes* and *Nitrospirae*), which was the opposite of our results. At this point, we can hypothesize that both thinning and SD affect SMC abundance. Nevertheless, the direction of the effect (positive or negative) varies according to the studies, which supports the idea that

the response of SMC is subject to a combination of factors, including the distinct influences of SD and thinning.

4.4. Microbial functional diversity and MSIR

Two key findings stand out from the MicroResp™ analyses: (i) the higher microbial functional diversity and (ii) the lower microbial respiration, in M-SD compared to L-SD. SMC were thus more efficient for mineralize all C-substrates in plots with dynamic silviculture (L-SD) despite there was less functional diversity.

Carboxylic acids induced the highest CO_2 rate whatever the SD, while amino acid had led to a lower CO_2 rate, which is commonly observed in studies (Banning *et al.* 2012; Gartzia-Bengoetxea *et al.* 2016; Xu *et al.* 2019). Exudates and decomposition of plant tissues contain a significant portion of low molecular weight carboxylic acids (Strobel 2001; Macias-Benitez *et al.* 2020) that constitutes an important source of labile C for SMC (Van Hees and Clerkx 2003; Fujii *et al.* 2010). Klimek et al., (2016) demonstrated that carboxylic acids contributed the most to differences in SMC functional diversity between forest types, underlying that forest soil bacteria preferentially use this substrate category. Interestingly, the analysis of correlations between microbial functional activity and environmental parameters highlights similar coefficients, except for citric acid and ketoglutaric acid. Specifically, microbial respiration induced by α-ketoglutaric was strongly and positively correlated with total volume of cutting trees per hectare and soil water content, but negatively with precipitations, contrary to other substrates that were positively correlated with precipitations (except respiration induced by citric acid that was not correlated). Regarding stand density, the coefficients were notably most negative with citric acid and ketoglutaric acid. This finding demonstrated that dynamic silvicultural practices enhance microbial activity. Ritz et al., (2006) also showed that citric acid and α-ketoglutaric acid were the substrates that allowed to establish differences in SIR between coniferous woodland soils and the others, including deciduous woodland. Thus, our

results corroborate studies that emphasize citric acid and ketoglutaric acid as the primary substrates for identifying functional differences in SMC, considering various factors studied, such as stand density and vegetation type. Overall, it has been frequently observed that the types of microbial carbon sources utilized vary among thinning treatments, with a significantly increased of some enzyme activities with thinning intensities (Tan *et al.* 2008; Wu *et al.* 2019; Zhou *et al.* 2020). Conversely, Kim et al., (2018) demonstrated that thinning had no significant effect on activities of all enzymes although microbial biomass was generally higher with thinning, again highlighting the variability of SMC response to forest harvesting.

It clearly appears from both combined ANOVA and correlation analyses that SD alone is insufficient as an explanatory factor to describe the soil microbial community. Therefore, it is necessary to consider other environmental factors and forest management parameters.

4.5. Multifactorial responses of soil microbial community

Overall, the results of our study showed that SD impacts SMC differently depending on the parameter considered (*i.e.,* SMC biomass, gene abundance, functional diversity or activity). A dynamic sylviculture (L-SD) led to a lower functional diversity of SMC but tend to favor soil microbial mineralization than a conservative sylviculture (M-SD). The correlation analysis further underscored the significance of various environmental factors in influencing SMC.

Depending on the stand characteristic parameters studied, although they are partly linked, the correlation coefficients with the microbial parameters were different. To our knowledge, there is limited existing research that comparatively examines the impact of thinning and the SD on SMC. Most of the available literature primarily focuses on the influence of thinning practices on soil functioning and the associated microbial communities. Thinning is a one-off forest management method which provides presumably temporary

information on the SMC while the SD could provide a more lasting representation of the structure and functioning of the SMC. These hypotheses could be supported by providing more study on the effects in stand density on SMC (Wang *et al.* 2021). Nevertheless, it is worth noting that none of the three stand characteristic parameters exhibited a correlation with all of the microbial parameters when considered individually.

Incorporating additional environmental factors, such as hydric properties, revealed that precipitation stood out as the primary factor exhibiting the highest number of robust and statistically significant correlations with microbial parameters. Zhao et al., (2016) also demonstrated a positive correlation between precipitations and microbial biomass. Overall, shifts in microbial community composition could be largely attributed to changes in soil water and nutrient availability (Ma *et al.* 2012), but surprisingly, only negative correlation was found in soil water content with the microbial gene abundance, and one positive correlation was found with microbial respiration for α-ketoglutaric acid. Difference of precipitation and soil water content effects on SMC can be attributed to the fact that soil water content is not only dependent on precipitation but is a result of interactions including also soil texture, litter and understory species (Dodd and Lauenroth 1997; Cubera and Moreno 2007; Xiong *et al.* 2008). The analysis of perched water table depth at two different time scales, long term (mean over the 30 days prior to soil harvest) and short term (day of harvest) shows contrasting effects on SMC. Logically, soil water content and perched water table depth mean the 30 days prior to soil harvest have opposite effects on SMC. On the other hand, on the day of harvest, we observed that the higher the water table, the more active the SMC was, corroborating the correlations with precipitations. The results of our study support the widely supported consensus that soil water properties and SMC are closely interacting.

Regarding soil physicochemical properties, we found that temperature was negatively correlated with functional diversity of SMC suggesting that increase in temperature decrease

SMC functional diversity, but this does not necessarily affect its activity. Chen et al., (2015) described soil temperature as one of the major factors affecting the functional diversity of the SMC, underlying the need to analyze effect on soil surface temperature on microbial biomass, abundance, and activities (Mateos-Rivera *et al.* 2016). Soil temperature and moisture also depend on the quantity of fresh litter, which play a crucial role in shaping the temporal variation in the microbial community on a month to season scale (Chemidlin Prevost-Boure *et al.* 2011). Numerous studies have also shown that litter properties and SMC are closely linked. We found that leaf litter mass average the 30 prior to harvest was mostly negatively correlated with SMC parameters which was not in line with Q. Wang et al., (2014) that demonstrating a positive effect of leaf litter addition on soil organic carbon mineralization. The multiplicity of litter properties directly or indirectly influencing SMC (*e.g.* changes in soil temperature) may explain differences in effects between studies. For instance, Tan et al., (2008) demonstrated that a numerous SMC response, including biomass, respiration, or mineralization, clearly depends on the stage of litter decomposition. The role of litter and particularly its stage of decomposition may also be one reason why the effects of thinning on SMC may differ from one study to another. Our results also showed that O horizon thickness increase led to decrease in microbial functional diversity that does not corroborate Cartwright et al., (2016). However, a thick O horizon was observed to enhance microbial activity, aligning with the common findings in the existing literature (Hellwig *et al.* 2018).

Regarding the biotic factors, few studies described crucial role of understory species on SMC in forest ecosystems. Understory removal could significantly reduce soil microbial biomass C (Xiong *et al.* 2008) and change microbial community composition (Wu *et al.* 2011), leading to decreased respiration. To our knowledge, no study described effect of the presence of *R. fructosus* and *C. vulgaris* on soil microbial community. Our study shows the importance of considering understory vegetation, especially *C. vulgaris* and *R. fructosus*, in the analysis of

microbial communities and especially their activity. Further studies on these understory species should be conducted to understand the extent to which their rhizodeposits influence the soil microbial community, as is the case with different forest species (Philippot *et al.* 2013; Fu *et al.* 2015; Yang *et al.* 2018).

Finally, precipitations, and to a lesser extent soil organic matter and H horizon thickness, are the three factors that favor both SMC biomass, gene abundance, and MSIR. In contrast, functional diversity appears to be positively influenced by higher stand density and greater standing tree volume. Perched water table depth, leaf litter mass and *R. fructosus cover* were the environmental parameters that were overall negatively correlated with microbial biomass, gene abundance, functional diversity and/or activity. Other environmental parameters, including SD, had contrasting correlations with microbial parameters.

Our study reflects the great complexity of interactions between abiotic and biotic factors in the soil ecosystem. In addition to considering a multitude of hydrological, chemical, and physical factors, it appears that the time frame over which these factors are assessed holds paramount significance. Indeed, we note that within the same forest, the differences between the environmental factors measured can be significant although stands are separated of a maximum of 30 km. The stand *Q*S3, particularly the L-SD plot, is clearly different from the others regarding the studied factors (**Figure A.1**). A probable reason for such a difference is that the thinning intensity was on average 1.8 and 2 times higher in plots *Q*S1 L-SD and *Q*S2 L-SD, respectively. However, thinning intensity is not a sufficient explanatory factor either, as shown by the correlation analysis. Besides, variation of precipitations under the canopy were observed between the 6 plots (**Table A.6**). Such differences could thus be explained by (i) geographical distance, (ii) the position of the rain gauges in the plots, and/or (iii) the canopy density of each stand. Grayston and Rennenberg, (2006) study demonstrated that forest stand fine local characteristics (*e.g.* geographical exposure) could have strong effects on

SMC and interfered with thinning effect. For instance, soil microbial activity was significantly higher in the plots of the northeast-facing compared with the site southwest-facing and was significantly reduced by heavy thinning only on the northeast-facing site. Furthermore, Liu et al., (2019) study focusing on fungal community, established that geographic location was a determining factor for differential fungal diversity patterns. Previous studies also observed an altitudinal, latitudinal, and longitudinal gradient of microbial biomass responses but this has rarely been observed on such a small scale (Van Horn *et al.* 2013; Ren *et al.* 2018; Xu *et al.* 2018; Liu *et al.* 2019). The fact that *QS*3 is situated more to the northwest than the other two stations could lead to differences in certain abiotic variables (*e.g.*, soil history, wind, etc.). None of the data from our studied database allows us to support this hypothesis or establish a particular factor to explain the uniqueness of the results obtained in *QS*3 compared to *QS*1 and *QS*2.

It is therefore important to maximize the number of technical and biological replicates to characterize with more precision the environmental properties of each forest plot. We also wish to emphasize the importance of the forest metric data (*e.g.* stand characteristics parameters) as well as the duration (*e.g.* point parameters such as thinning, or longer term parameters such as stand density) considered in the study of factors impacting SMC. These considerations are in line with recent studies highlighting the need to improve current practices in hypothesis generation, modeling, and visual representation of interactions in ecology (Spake *et al.* 2023).

5. Conclusions and outcomes

Microbial biomass and gene abundance seem to depend more on forest local environmental characteristics than forest plot density or even thinning, contrary to our initial expectations. An important consideration is that although the forest stands where supposed to be similar (same pedological station, trees age, size and composition, and understory characteristics) and

within 30 km of each other, the differences observed in abiotic factors (*e.g.* precipitation under canopy, soil properties) explained better the microbial biomass and gene abundance than forest relative density index. Conversely, it appears that SD exerts a more significant influence on the functional diversity and activity of SMC. A dynamic silvicultural practice negatively affected SMC functional diversity but favored their activity, partly validating our initial hypotheses. A noteworthy aspect of our study is that our primary focus was to analyze the impact of SD, whereas many other studies typically investigate the effects of thinning intensity. Thinning represents a temporary disturbance, yet our findings emphasize the importance of considering thinning post-effects, taking into account in particular the stand SD, which provides a good indicator of SMC in the longer term. Recent studies also support the idea that understanding these effects in the context of a longer timeframe is crucial (Lee *et al.* 2023), especially in perennial ecosystems like forests. Thus, longer-term studies should be conducted to characterize the effect of forest plot density on soil microbial community. An acceptable conclusion which is in line with Bolat (2014) is that the influence of forest thinning on the SMC parameters result in the combination of multiple biotic and abiotic factors including soil properties, understory species and environmental conditions, one influencing the other. Additional research efforts should be directed towards investigating various environmental parameters across forest stands, with a particular emphasis on stand density, which has received comparatively less attention than thinning in previous studies. Furthermore, a thorough environmental characterization should be carried out, involving the interactions between different these factors, to provide a more holistic understanding of the soil microbial communities. This could also help to fill an important gap in our understanding of forest soil ecosystem dynamics.

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Figure 1. Geographic map of the Orleans forest (France) showing the location of the 3 *Quercus* stands (*Q*S). The inset shows the Low (L-SD) and Medium SD (M-SD) plots of QS.

Figure 2. Soil basal respiration in June 2019 in L-SD (gray bar) and M-SD (dark bar) plots of the 3 *Quercus* stands. Values are reported as means ± SE (n = 40).

Figure 3. Biomass C (a), biomass N (b) and microbial C:N ratio (c) in L-SD (gray bar) and M-SD (dark bar) plots of the 3 *Quercus* stands. Values are reported as means ± SE (n = 4). **.** corresponds to p less than 0.1.

Figure 4. Microbial genes abundances (number of gene copies per gram of soil) of bacteria (a) of archaea (b), fungi (c), (d) Archaea:Bacteria ratio, and (e) Fungi:Bacteria ratio in L-SD (gray bar) and M-SD (dark bar) plots of the 3 *Quercus* stands. Values are reported as means ± SE (n = 4). * and ** correspond to p less than 0.05, and 0.01, respectively.

Figure 5. Functional diversity of SMC (calculated using Shannon index) in L-SD (gray bar) and M-SD (dark bar) plots of the 3 *Quercus* stands. Values are reported as means ± SE (n = 24). *** corresponds to p less than 0,001.

Figure 6. CO_2 production (MSIR) in L-SD (gray bar) and M-SD (dark bar) plots of the 3 *Quercus* stands for each substrate. Values are reported as means ± SE (n = 24). *, **, *** corresponds to p less than 0.1, 0.05, 0.01 and 0,001 respectively.

Table 5. Correlation coefficients less than -0.5 (red shading according to correlation intensity) and greater than 0.5 (blue shading) with p-value < 0.05 between microbial parameters (row) and environmental parameters (column). The coefficients between -0.5 and 0.5 are uncolored. "*ns*" indicate that there is no significative correlation between the 2-to-2 parameters. Given that soil basal respiration was assessed in 2019, we do not present correlations with variable environmental parameters measured in 2018.

Figure S1. Individual principal component analysis (PCA) of the SMC and environmental parameters according to the plots (L-SD: empty symbols, M-SD: full symbols, *Q*S1: circle, *Q*S2: diamond, *Q*S3 : triangle).

Table 1. Short review of studies of forest thinning on the soil microbial communities, by measuring microbial biomass, microbial abundance, diversity, and activity. Studies are classified according to responses of microbial parameters (increase, decrease, no effect or variable effect) to thinning. In some cases, we specify which SMC domain (bacteria, archaea or fungi) was affected by forest thinning in the cited study.

Microbial parameters	Responses to thinning			
	Increase	Decrease	No effect	Variable effect
Biomass	(Lei et al., 2021)	(Geng et al., 2012)	(Tan et al., 2008); (Maassen et al., 2006); (Purahong et al., 2014): fungi	(Grady and Hart, 2006): no effect except a **decrease** in July for microbial C; (Chen et al., 2016):**increase** under high intensity thinning but no effect of light intensity; (Thibodeau et al., 2000): depends on horizon and C or N biomass; (Kim et al., 2018): **increase** in only one of the two sites; (Chen et al., 2015): depends on thinning intensity; (Grayston and Rennenberg, 2006): no effect or decrease, depends if forest site faces to North East or South West; (Lin et al., 2016): **increase** of fungi only in April, no effect for others months
Abundance				(Cai et al., 2020; Wu et al., 2019): **increase** or no effect, depends on thinning intensity; (Chen et al., 2015): **decrease** or not effect, depends on thinning intensity; (Dang et al., 2018); (Bastida et al., 2019; Purahong et al., 2014); **increase** in bacteria only in spring

Diversity	(Trentini et al., 2020) : bacteria and archaea	(Dang et al., 2018); (Trentini et al., 2020): fungi	(Wu et al., 2019): **increase** or no effect, depends on thinning intensity and season; (Collado et al., 2021): depends on fungal species
Activity		(Kim et al., 2018); (Tan et al., 2008); (Maassen et al., 2006); (Ntoko et al., 2018); (Purahong et al., 2014)	(Geng et al., 2012): depends on enzyme and soil depth; (Chen et al., 2016): **decrease** or not effect, depends enzyme; (Wu et al., 2019); **increase** or no effect, depends on date and thinning intensity; (Yang et al., 2017): depends on enzyme and thinning intensity; (Grayston and Rennenberg, 2006): depends on C-substrates, thinning intensity and exposure to North or South; (Bastida et al., 2019): **increase** or no effect, depends on enzyme and season; (Xiao et al., 2018): **increase** or no effect, depends on enzyme and litter decomposition phase

Table 2. Means of soil chemical properties for each *Quercus* stand (*Q*S) according to the density; L-SD: Low Stand Density, M-SD: Medium Stand Density

	Clay (%)	**Sand (%)**	**Silt (%)**	**OM (%)**	**C (%)**	**N (%)**	**pH water**	**pH KCl**	**CEC meq.100g**	**Ca (mg.kg^{-1})**	**K (mg.kg^{-1})**	**Mg (mg.kg^{-1})**

*Q*S1	L-SD	9,1	59,5	29,4	2,09	1,22	0,06	4,95	4,45	2,00	20	50	21
	M-SD	11,6	61,5	24,9	2,01	1,17	0,05	4,86	4,36	2,17	80	46	25cv
*Q*S2	L-SD	9,6	67,5	21,4	1,56	0,91	0,04	4,97	4,47	2,00	20	38	25
	M-SD	10,1	69,1	19,0	1,94	1,13	0,06	4,89	4,39	2,52	60	46	23
*Q*S3	L-SD	13,7	63,6	21,8	0,93	0,54	0,04	4,95	4,45	4,19	260	57	86
	M-SD	8,2	76,1	15,0	0,74	0,43	0,03	5,17	4,67	2,00	60	26	14

Table 3. Plot characteristics at the end of the 2017 growing season after the last thinning. Density: plot density. BA tot.: total stand basal area, Dg: quadratic mean diameter, Ho: dominant height, BA exp. 2017: exported basal area (m^3/ha) in the last thinning, BA exp. tot.: exported basal area (m^3/ha) since 2012, V.cut tot.: exported volume (m^3) since 2012, SD: Stand Density index after thinning.

Plot	Dimension (m)	Density	SD	V tot (m^3/ha)	BA tot. (m^2/ha)	Dg oak (cm)	Ho oak (m)	BA exp. tot.	V exp. tot.
*Q*S1	50 x 100	L-SD	0.35	153.75	12.8	23.6	20.2	7.2	78.3
	70 x 70	M-SD	0.59	263.12	21.5	23.4	21.5	0	0
*Q*S2	50 x 100	L-SD	0.35	145.97	12.8	24.1	18.6	7.4	68.6
	50 x 100	M-SD	0.53	218.67	19.9	20.5	18.6	0.6	0
*Q*S3	60 x 80	L-SD	0.35	167.81	12.6	28.9	22.0	11.4	138.4
	60 x 80	M-SD	0.60	273.76	21.9	25.6	21.0	1.6	18.2

Table 4. Primers, sequences of total bacterial, archeal and fungal communities using targets (16S rDNA or 18S rDNA primers) according to cited references.

rimer	Sequence	Target	Reference
C341F:	5’ CCT ACG GGA GGC AGC AG 3’	Bacterial 16S rRNA gene	López-Gutiérrez et al., 2004
C515R:	5’ ATT ACC GCG GCT GCT GGC A 3’		López-Gutiérrez et al., 2004
rch519F	5'-CAG CCG CCG CGG TAA-3'	Archaeal 16S rRNA gene	Øvreås et al., 1997
rc915R	5’-GTGCTCCCCCGC CAATTCCT-3’		Casamayor et al., 2000
FR1	5'-AIC-CAT- TCA-ATC-GGT-AIT-3'	Fungal 18S rRNA gene	Vainio and Hantula, 2000
F390	5’-CGA-TAA-CGA-ACG-AGA-CCT-3’		Vainio and Hantula, 2000

Appendix

n_{bio} Number of biological replicates

n_{tech} Number of technical replicates

Appendix 1. Soil fumigation

Fumigation was done by exposing 5 g of fresh soil to chloroform vapors for 24 hours in a sealed vacuum. Chloroform vapors act as solvent extracting polar lipid molecules that compose microbial cell membranes, degrading the cell walls and releasing internal organic compounds into the soil (Vance *et al.* 1987). For both fumigated and non-fumigated samples, organic C and N were extracted from a 5 g soil sample into solution using 20 ml of a K_2SO_4 buffer (0.5 M). Samples were placed on a shaking table at 250 rpm for 30 minutes to thoroughly mix the soil with the solvent and dissolve all organic C and N. The solution was passed through a Whatman GF/C glass microfiber filter into a Falcon tube to remove any soil particles and impurities, and the clear solution was frozen and sent to the INRAE Agronomy and Environment Lab in Nancy (France) for quantification of organic C and total N (TOC analyzer, (TOC-VCSH CSH/CNS, Shimadzu, Champs-sur-Marne, France) connected online to a N analyzer (TNM-1, Shimadzu)). The calculations of soil microbial biomass C and N were revised by a conversion factor of 2.22 (Jenkinson *et al.* 2004).

Appendix 2. DNA extraction and PCR

Total DNA was extracted and purified from 500 mg of soil using the NucleoSpin Soil kit and the NucleoSpin gDNA clean-up kit (Macherey-Nagel, NucleoSpin Soil and NucleoSpin gDNA clean-up, 2017), according to manufacturer's instructions. The DNA quality was assessed by spectrophotometry (Biotek Eon spectrophotometer and Take3 plate), and DNA concentration was assessed by fluorimetry (QuBit dsDNA BR Assay Kit, Thermofisher).

Reactions were carried out in a Applied Biosystems Step One Plus qPCR System, with a 20 µL reaction volume containing 10 µl of 2X SsoAdvanced Universal SYBR Green Supermix (Biorad), 1 µL of each primer (at 10 µM for bacteria and archaea and 20 µM for fungi) 1.25 µl of BSA (2 mg ml−1), and 2 µl of template DNA at 0.2 ng/µL, so 0.4 ng of DNA. At least four independent runs were performed for each qPCR assay. Standard curves were obtained using serial dilutions of linearized plasmids containing the studied genes respectively amplified from Pseudomonas fluorescens and Nitrososphaera viennensis (16SDNA sequences), and Trametes versicolor (18SDNA sequence). PCR efficiency for the different assays ranged from 85 to 102% with R2 > 0.9. No-template controls gave null or negligible values. The specificity of amplified products was verified by melting curves from 65 °C to 95 °C at 0.5 °C. Inhibition in qPCR assay was tested by using 10-fold serial dilutions of the DNA template, from 2ng to 0.02 ng.

Table A.1 (**Figure 2**). Means ± SE of soil respiration *in situ* (µmol CO_2 m^{-2} s^{-1}) in low stand density (L-SD) and medium stand density (M-SD) for each *Quercus* stand (*Q*S). Test statistic (F-value and t-value), statistical significance (p-value), and degrees of freedom (DF) assessing the effect of density on the microbial parameters (ANOVA, $\alpha = 5\%$, $n_{bio} = 3$, $n_{tech} = 4$).

	Soil respiration	
	L-SD	M-SD
*Q*S1	3.17 ± 0.24	3.14 ± 0.20
*Q*S2	3.65 ± 0.22	3.75 ± 0.30
*Q*S3	4.07 ± 0.37	3.70 ± 0.27
*Q*S mean ± SE	3.64 ± 0.17	3.54 ± 0.15
F-value	0.28	
p-value	0.6	

DF	1	

Table A.2 (**Figure 3**). Means ± SE of soil microbial carbon (SMB-C), nitrogen (SMB-N) biomass and microbial C:N ratio in low stand density (L-SD) and medium stand density (M-SD) for each *Quercus* stand (*Q*S). Test statistic (F-value and t-value), statistical significance (p-value), and degrees of freedom (DF) assessing the effect of density on the microbial parameters (ANOVA, $\alpha = 5\%$, $n_{bio} = 3$, $n_{tech} = 4$).

	SMB-C		SMB-N		Microbial C:N	
	L-SD	M-SD	L-SD	M-SD	L-SD	M-SD
*Q*S1	1197.78 ± 172.02	772.34 ± 32.23	97.91 ± 5.63	66.11 ± 2.42	12.08 ± 1.18	11.71 ± 0.51
*Q*S2	916.24 ± 28.63	603.35 ± 17.29	56.25 ± 5.55	57.19 ± 5.16	16.66 ± 1.30	10.79 ± 0.75
*Q*S3	461.40 ± 24.94	557.54 ± 67.58	22.40 ± 2.24	29.30 ± 2.08	20.97 ± 1.53	18.89 ± 1.27
*Q*S mean ± SE	858.47 ± 105.78	644.41 ± 36.23	58.86 ± 9.64	50.81 ± 5.06	10.60 ± 0.89	10.39 ± 0.66
F-value	3.67		0.55		0.03	
p-value	0.07 .		0.47		0.86	
DF	1		1		1	

Table A.3 (**Figure 4**). Means ± SE of microbial abundance (gene copies) in low stand density (L-SD) and medium stand density (M-SD) for each *Quercus* stand (*Q*S). Test statistic (F-value and t-value), statistical significance (p-value), and degrees of freedom (DF) assessing the effect of density on the microbial parameters

	Bacteria		Archaea		Fungi	
	L-SD	M-SD	L-SD	M-SD	L-SD	M-SD
*Q*S1	2151501302 ± 199764267	3293263912 ± 311980198	1221883845 ± 69613400	1613868051 ± 102785864	45296161 ± 3765312	56889559 ± 7721492
*Q*S2	1956832371 ± 331210533	4230976158 ± 933578277	1167612591 ± 153491846	1999268248 ± 230888870	41448723 ± 6177293	81238894 ± 6744637
*Q*S3	1222727816 ± 195485152	1777285000 ± 153689256	746004078 ± 218229599	803024671 ± 74483305	32064082 ± 5977364	40676771 ± 1127581
*Q*S mean ± SE	1777020496 ± 177803981	3100508357 ± 427947657	1045166838 ± 104997837	1472053657 ± 170020804	39602988 ± 3288411	59601742 ± 5910509
F-value	9.57		4.56		8.74	
p-value	0.005 **		0.04 *		0.007 **	
DF	1		1		1	

(ANOVA, $\alpha = 5\%$, $n_{bio} = 3$, $n_{tech} = 4$).

	A:B ratio		F:B ratio	
	L-SD	M-SD	L-SD	M-SD
*Q*S1	0.58 ± 0.05	0.50 ± 0.02	0.02 ± 0.0003	0.02 ± 0.0009
*Q*S2	0.61 ± 0.03	0.52 ± 0.07	0.02 ± 0.0007	0.02 ± 0.005
*Q*S3	0.58 ± 0.07	0.46 ± 0.04	0.03 ± 0.002	0.02 ± 0.003
*QS m*ean ± SE	0.59 ± 0.03	0.49 ± 0.03	0.023 ± 0.001	0.021 ± 0.001
F-value	6.52		0.83	
p-value	0.02 *		0.3	
DF	1		1	

Table A.4 (**Figure 5**). Means ± SE of functional diversity of SMC in low stand density (L-SD) and medium stand density (M-SD) for each *Quercus* stand (*Q*S). Test statistic (F-value and t-value), statistical significance (p-value), and degrees of freedom (DF) assessing the effect of density on the microbial parameters (ANOVA, α = 5%, n_{bio} = 3, n_{tech} = 6).

	Functional diversity of SMC (Shannon index)	
	L-SD	M-SD
*Q*S1	1.98 ± 0.05	2.39 ± 0.04
*Q*S2	1.78 ± 0.03	2.20 ± 0.05
*Q*S3	2.30 ± 0.05	2.45 ± 0.03
*Q*S mean ± SE	2.02 ± 0.04	2.34 ± 0.03
F-value	52.11	
p-value	< 0.001 ***	
DF	1	

Table A.5 (**Figure 6**). Means ± SE of CO_2 rate in low stand density (L-SD) and medium stand density (M-SD) for each *Quercus* stand (*Q*S). Test statistic (F-value and t-value), statistical significance (p-value), and degrees of freedom (DF) assessing the effect of density on the microbial parameters (ANOVA, α = 5%, n_{bio} = 3, n_{tech} = 24 and 36).

		Fructose		**Glucose**		**Galactose**		**Arabinose**		**Trehalose**	
		L-SD	M-SD	L-SD	M-SD	L-SD	M-SD	L-SD	M-SD	L-SD	M-SD
	*Q*S1	0.11 ± 0.01	0.06 ± 0.00	0.14 ± 0.01	0.08 ± 0.00	0.09 ± 0.00	0.08 ± 0.01	0.09 ± 0.00	0.05 ± 0.00	0.14 ± 0.01	0.08 ± 0.00
	*Q*S2	0.14 ± 0.01	0.08 ± 0.00	0.18 ± 0.01	0.11 ± 0.01	0.12 ± 0.01	0.08 ± 0.00	0.12 ± 0.00	0.08 ± 0.00	0.17 ± 0.01	0.10 ± 0.00
	*Q*S3	0.04 ± 0.00	0.06 ± 0.00	0.05 ± 0.00	0.07 ± 0.00	0.04 ± 0.00	0.06 ± 0.00	0.04 ± 0.00	0.05 ± 0.00	0.05 ± 0.00	0.07 ± 0.00
Sugar	*Q*S mean ± SE	0.1 ± 0.006	0.07 ± 0.002	0.12 ± 0.009	0.09 ± 0.003	0.08 ± 0.005	0.07 ± 0.003	0.08 ± 0.005	0.06 ± 0.002	0.12 ± 0.008	0.08 ± 0.002
	F-value	7.66		5.44		2.11		8.65		6.63	
	p-value	0.006 **		0.02 *		0.15		0.004 **		0.01 *	
	DF	1		1		1		1		1	

		Alanine		**Acetylglucos-amine**		**Lysine**		**Proline**		**Cysteine**		**Aminobutyric**	
		L-SD	M-SD	L-SD	M-SD	L-SD	M-SD	L-SD	M-SD	L-SD	M-SD	L-SD	M-SD
	*Q*S1	0.12 ± 0.01	0.05 ± 0.00	0.08 ± 0.00	0.05 ± 0.00	0.08 ± 0.00	0.04 ± 0.00	0.07 ± 0.00	0.04 ± 0.00	0.07 ± 0.00	0.04 ± 0.00	0.07 ± 0.00	0.05 ± 0.00
	*Q*S2	0.13 ± 0.00	0.08 ± 0.00	0.13 ± 0.00	0.07 ± 0.00	0.07 ± 0.00	0.05 ± 0.00	0.09 ± 0.00	0.06 ± 0.00	0.09 ± 0.00	0.06 ± 0.00	0.10 ± 0.00	0.07 ± 0.00
	*Q*S3	0.04 ± 0.00	0.05 ± 0.00	0.04 ± 0.00	0.05 ± 0.00	0.03 ± 0.00	0.04 ± 0.00	0.04 ± 0.00	0.05 ± 0.00	0.04 ± 0.00	0.05 ± 0.00	0.04 ± 0.00	0.05 ± 0.00
Amino acid	*Q*S mean ± SE	0.09 ± 0.005	0.06 ± 0.002	0.08 ± 0.005	0.06 ± 0.002	0.06 ± 0.003	0.04 ± 0.001	0.06 ± 0.003	0.05 ± 0.001	0.06 ± 0.003	0.05 ± 0.001	0.07 ± 0.003	0.05 ± 0.002
	F-value	20.23		13.49		16.78		11.98		13.3		11.07	
	p-value	< 0.0001 ***		0.0003 ***		< 0.0001 ***		0.0007 ***		0.0004 ***		0.001 **	
	DF	1		1		1		1		1		1	

		Citric		**Ascorbic**		**Malic**		**Ketoglutaric**		**Water**	
		L-SD	M-SD	L-SD	M-SD	L-SD	M-SD	L-SD	M-SD	L-SD	M-SD
	*Q*S1	0.08 ± 0.00	0.05 ± 0.00	0.35 ± 0.01	0.24 ± 0.01	0.28 ± 0.02	0.21 ± 0.01	0.12 ± 0.01	0.06 ± 0.00	0.05 ± 0.00	0.03 ± 0.00
	*Q*S2	0.12 ± 0.00	0.07 ± 0.00	0.38 ± 0.01	0.31 ± 0.01	0.37 ± 0.02	0.22 ± 0.01	0.15 ± 0.00	0.09 ± 0.01	0.06 ± 0.00	0.05 ± 0.00
	*Q*S3	0.09 ± 0.00	0.08 ± 0.00	0.18 ± 0.01	0.17 ± 0.00	0.15 ± 0.01	0.18 ± 0.01	0.29 ± 0.01	0.16 ± 0.01	0.04 ± 0.00	0.04 ± 0.00
Acid citric	*Q*S mean ± SE	0.1 ± 0.003	0.07 ± 0.002	0.3 ± 0.01	0.24 ± 0.009	0.27 ± 0.01	0.05 ± 0.002	0.19 ± 0.001	0.1 ± 0.006	0.05 ± 0.002	0.04 ± 0.001
	F-value	55.68		11.71		8.41		70.73		7.56	
	p-value	< 0.0001***		0.0008 ***		0.004 **		< 0.0001 ***		0.007 **	
	DF	1		1		1		1		1	

Table A.6. Means ± SE of environmental factors in low stand density (L-SD) and medium stand density (M-SD) for each *Quercus* stand (*Q*S). Precipitation data showed the sum of rainfall for the last 30 days before harvest, not the mean. Test statistic (F-value and t-value), statistical significance (p-value), and degrees of freedom (DF) assessing

	Stand density						**Hydric properties**							
	SD		Final volume of standing trees.ha^{-1}		Total volume of cutting trees. ha^{-1}		Precipitations (-D30)		Soil water content (-D30)		Perched water table depth (D-30)		Perched water table depth (D0)	
	L-SD	M-SD	L-SD	M-SD	L-SD	M-SD	L-SD	M-SD	L-SD	M-SD	L-SD	M-SD	L-SD	M-SD
*Q*S1	0.35	0.59	153.75	263.12	78.27	0	274.0	268.6	31.09	27.38	60.37	60.4	44.21	44.35
*Q*S2	0.35	0.53	145.97	218.69	68.61	0	260.8	224.0	38.26	21.71	20.5	71.64	11.95	65.89
*Q*S3	0.35	0.60	167.81	273.76	138.38	18.23	73.8	152.8	56.51	29.03	37.99	36.3	51.46	71.27
*Q*S mean ± SE	0.35 ± 0.00	0.57 ± 0.00	156.07 ± 1.19	251.86 ± 2.83	96.90 ± 4.00	6.08 ± 1.02	195.21 ± 11.63	215.13 ± 5.66	43.12 ± 1.33	26.04 ± 0.37	37.39 ± 1.95	56.11 ± 1.75	34.97 ± 2.23	60.50 ± 1.38
F-value	3860		903.9		530.2		2.52		167.4		51.38		98.73	
p-value	<0.001 ***		<0.001 ***		<0.001 ***		0.12		<0.001 ***		<0.001 ***		<0.001 ***	
DF	1		1		1		1		1		1		1	

	Soil physicochemical properties								**Litter**		**Vegetation cover of understory species**					
	Organic matter		OH thickness		Cation Exchange Capacity		Soil temperature (-D30)		Leaf litter mass (-D30)		*Calluna vulgaris*		*Molinia caerulea*		*Rubus fructose*	
	L-SD	M-SD	L-SD	M-SD	L-SD	M-SD	L-SD	M-SD	L-SD	M-SD	L-SD	M-SD	L-SD	M-SD	L-SD	M-SD
*Q*S1	2.09	2.01	8.0	0.6	2.00	2.17	15.55	14.95	3.43	4.67	0.27	0.16	0.06	0.41	0.8	0.10
*Q*S2	1.60	1.96	15.7	16.9	2.00	2.00	15.84	15.01	1.93	7.12	19.40	1.33	6.06	0.09	0.03	0.03
*Q*S3	0.93	0.75	0	0	3.31	2.00	15.79	14.82	10.35	4.57	0.73	0.81	3.67	2.10	0.58	0.12
*Q*S mean ± SE	1.48 ± 0.06	1.57 ± 0.07	7.89 ± 0.84	5.83 ± 0.93	2.48 ± 0.08	2.06 ± 0.01	15.75 ± 0.01	14.93 ± 0.009	5.43 ± 0.48	5.45 ± 0.14	7.50 ± 1.14	0.76 ± 0.06	3.61 ± 0.29	0.87 ± 0.10	0.25 ± 0.03	0.08 ± 0.005
F-value	1.03		2.64		31.8		2267		0.002		38.72		83.37		27.36	
p-value	0.31		0.11		<0.001 ***		<0.001 ***		0.96		<0.001 ***		<0.001 ***		<0.001 ***	
DF	1		1		1		1		1		1		1		1	

1 the effect of density on the microbial parameters (ANOVA, α = 5%, n_{bio} = 3, n_{tech} = 24 and 36

Figure 1

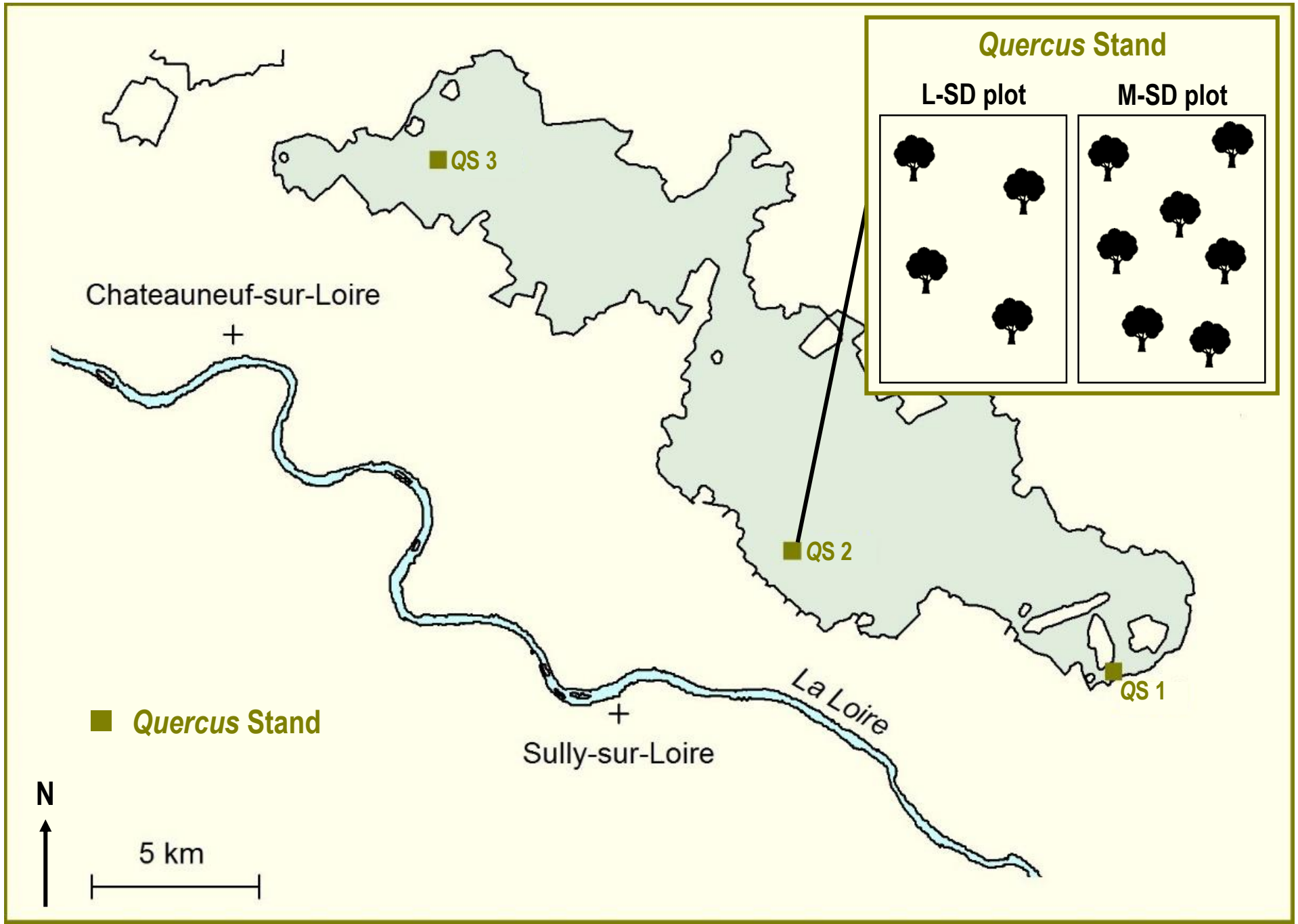

Quercus Stand
L-SD plot
M-SD plot
QS 3
QS 2
QS 1
Chateauneuf-sur-Loire
Sully-sur-Loire
La Loire
Quercus Stand
N
5 km

Figure 2

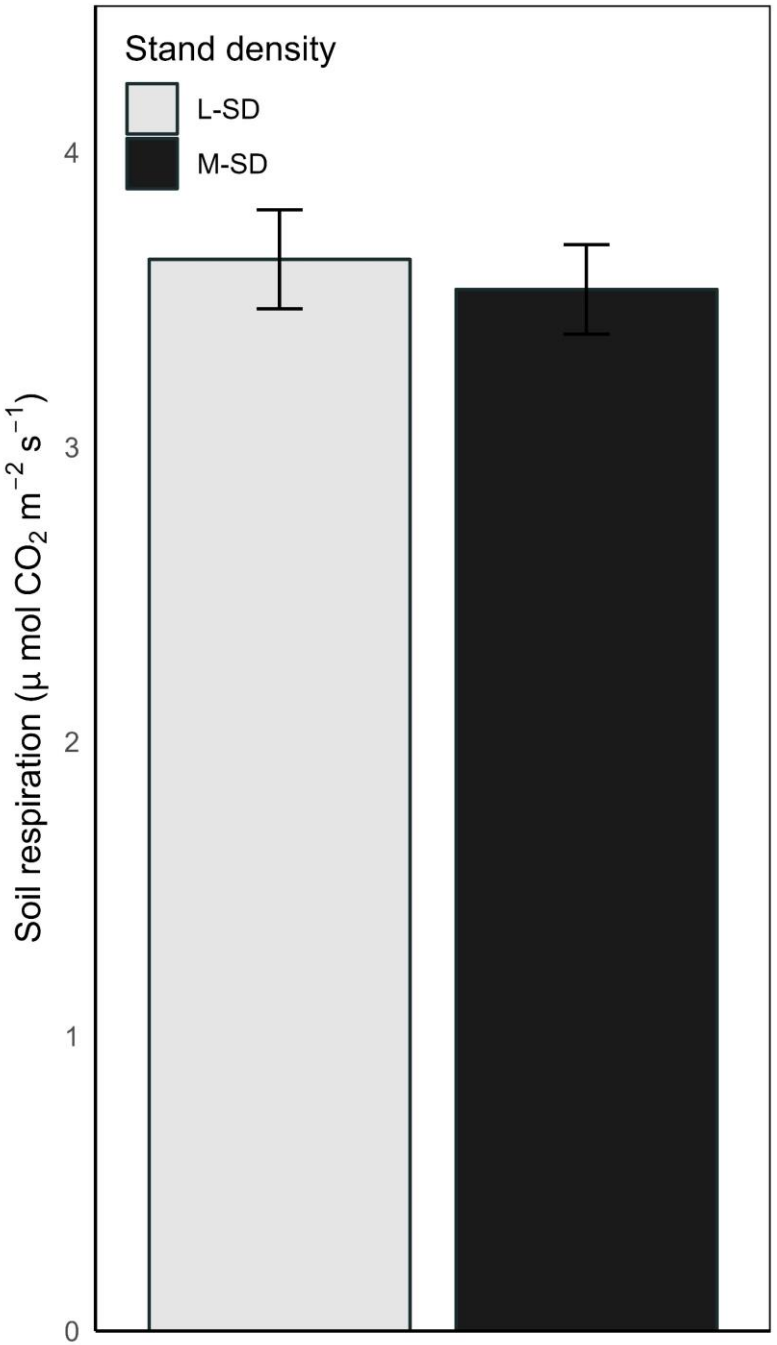


Figure 3

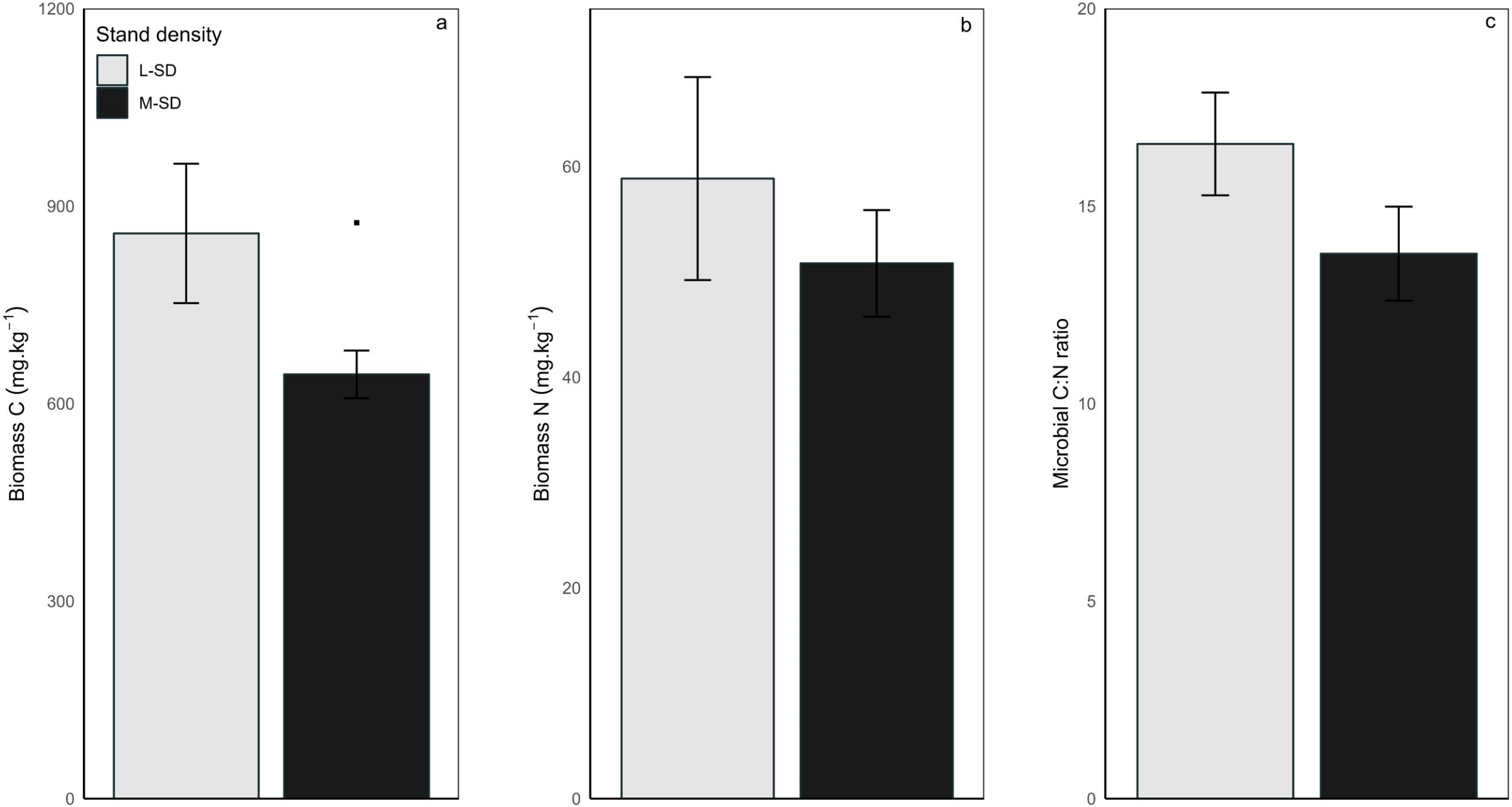

a
b
c
Stand density
L-SD
M-SD
Biomass C (mg.kg^{-1})
Biomass N (mg.kg^{-1})
Microbial C:N ratio
0
300
600
900
1200
20
40
60
5
10
15
20

Figure 4

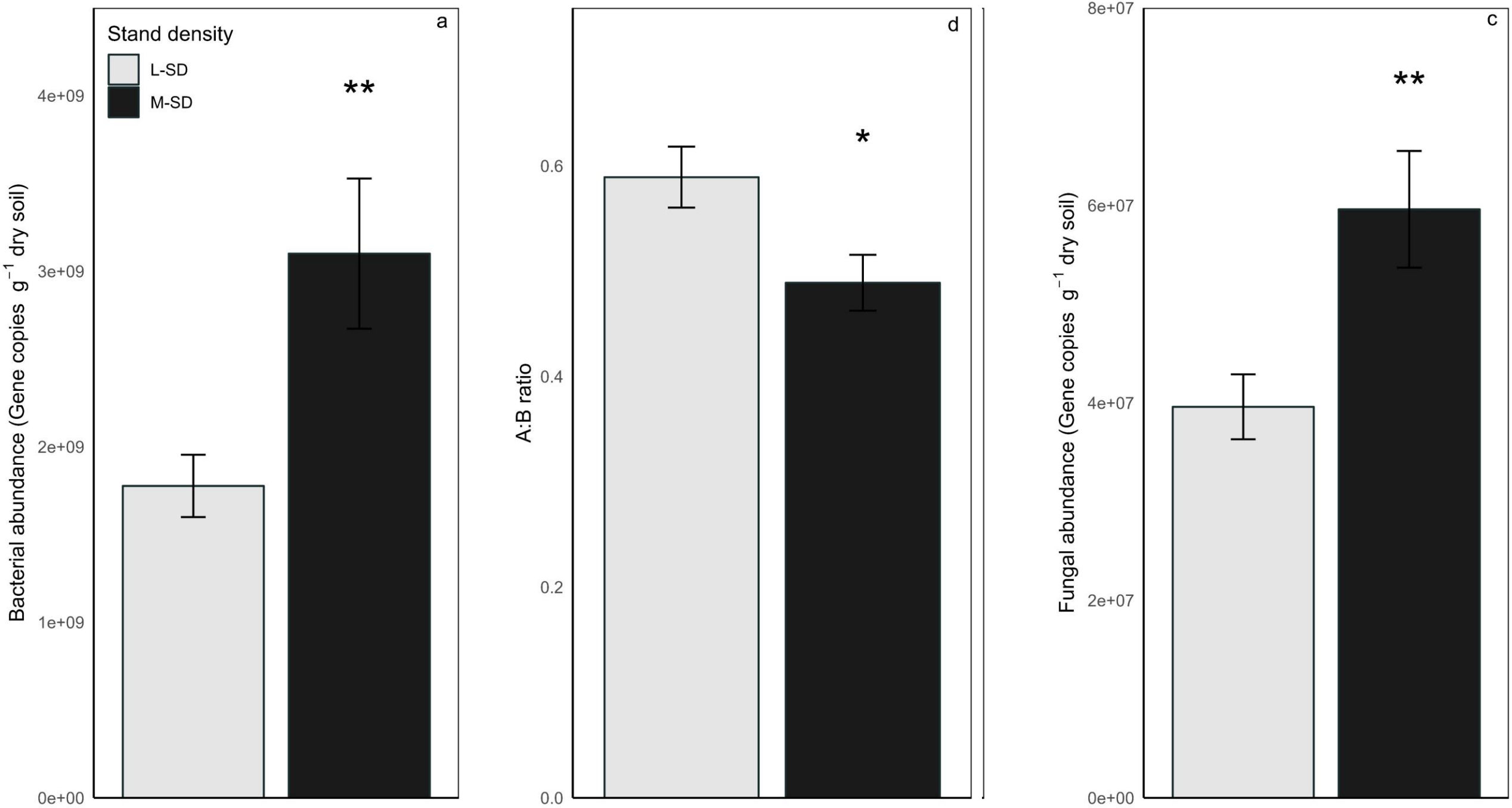
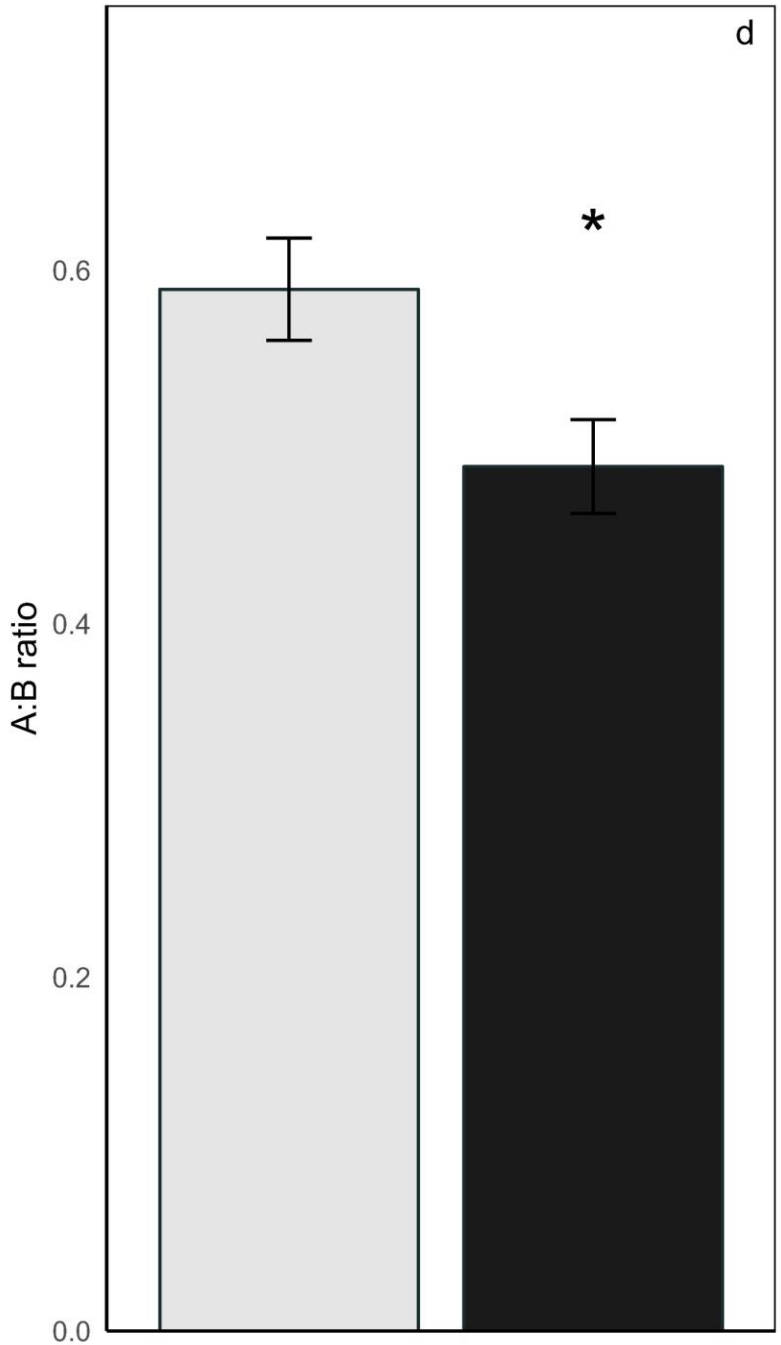

a
Stand density
L-SD
M-SD
**
Bacterial abundance (Gene copies g^{-1} dry soil)
0e+00
1e+09
2e+09
3e+09
4e+09
d
*
A:B ratio
0.0
0.2
0.4
0.6
c
**
Fungal abundance (Gene copies g^{-1} dry soil)
0e+00
2e+07
4e+07
6e+07
8e+07

Figure 4



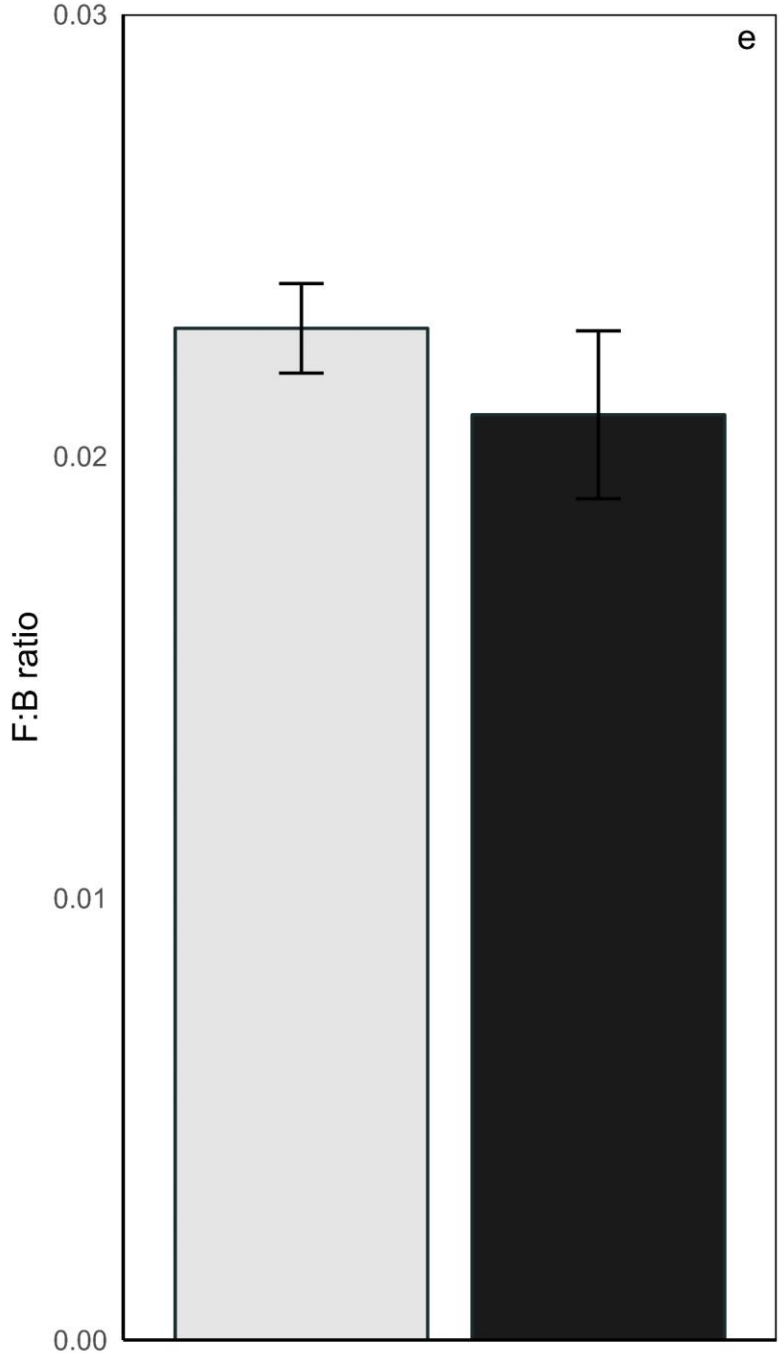


Figure 5

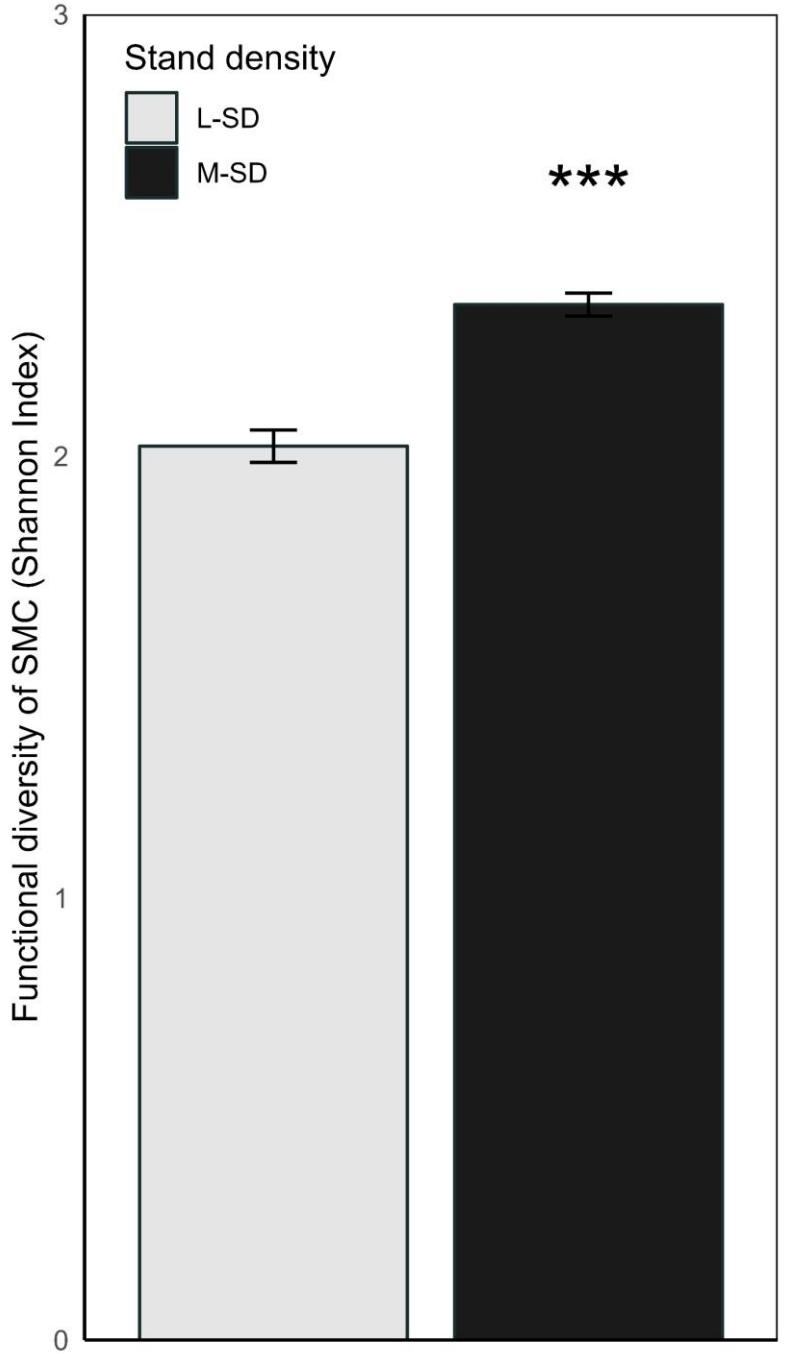

Stand density
L-SD
M-SD
***
Functional diversity of SMC (Shannon Index)
0
1
2
3

Figure 6

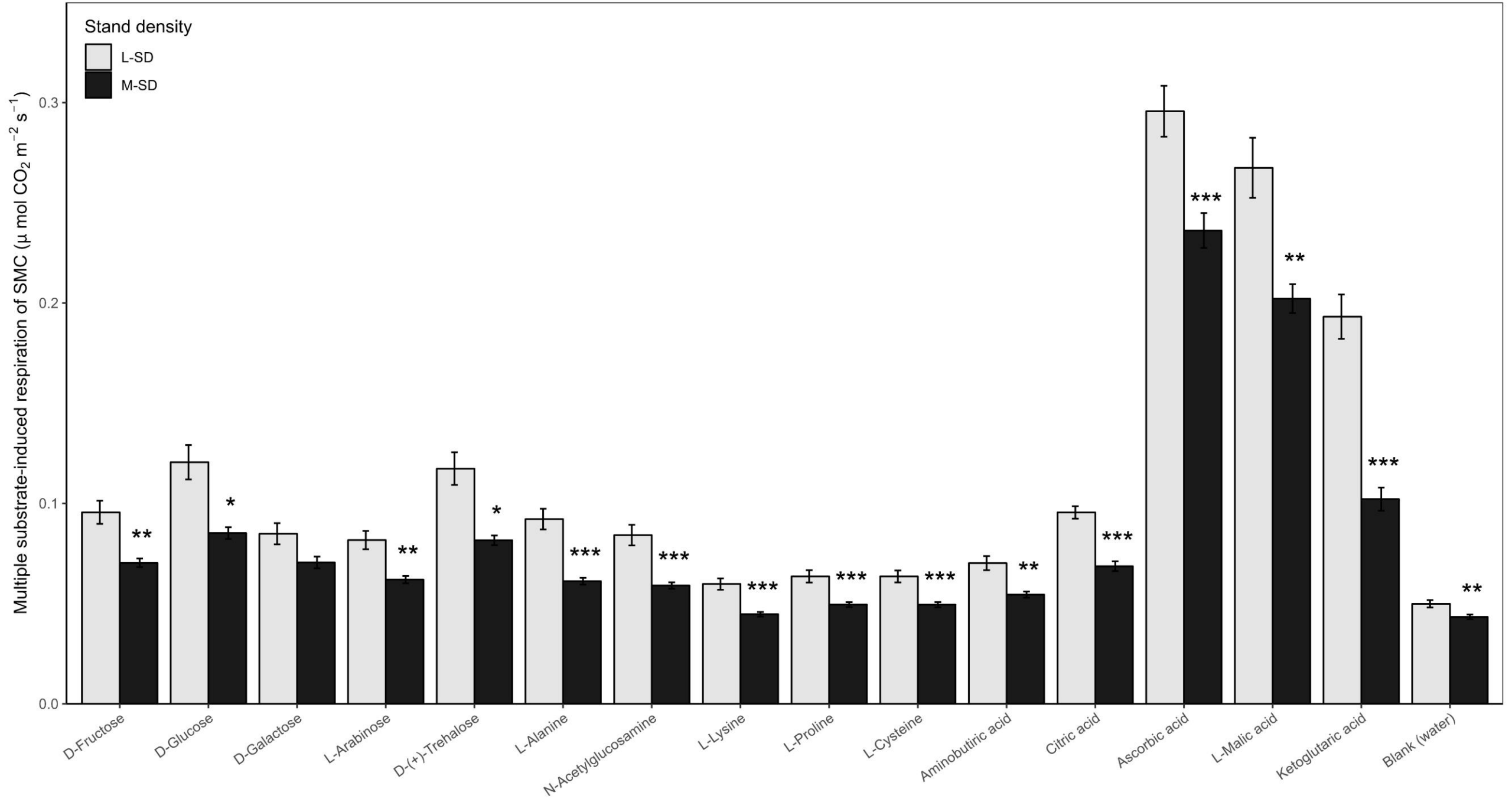
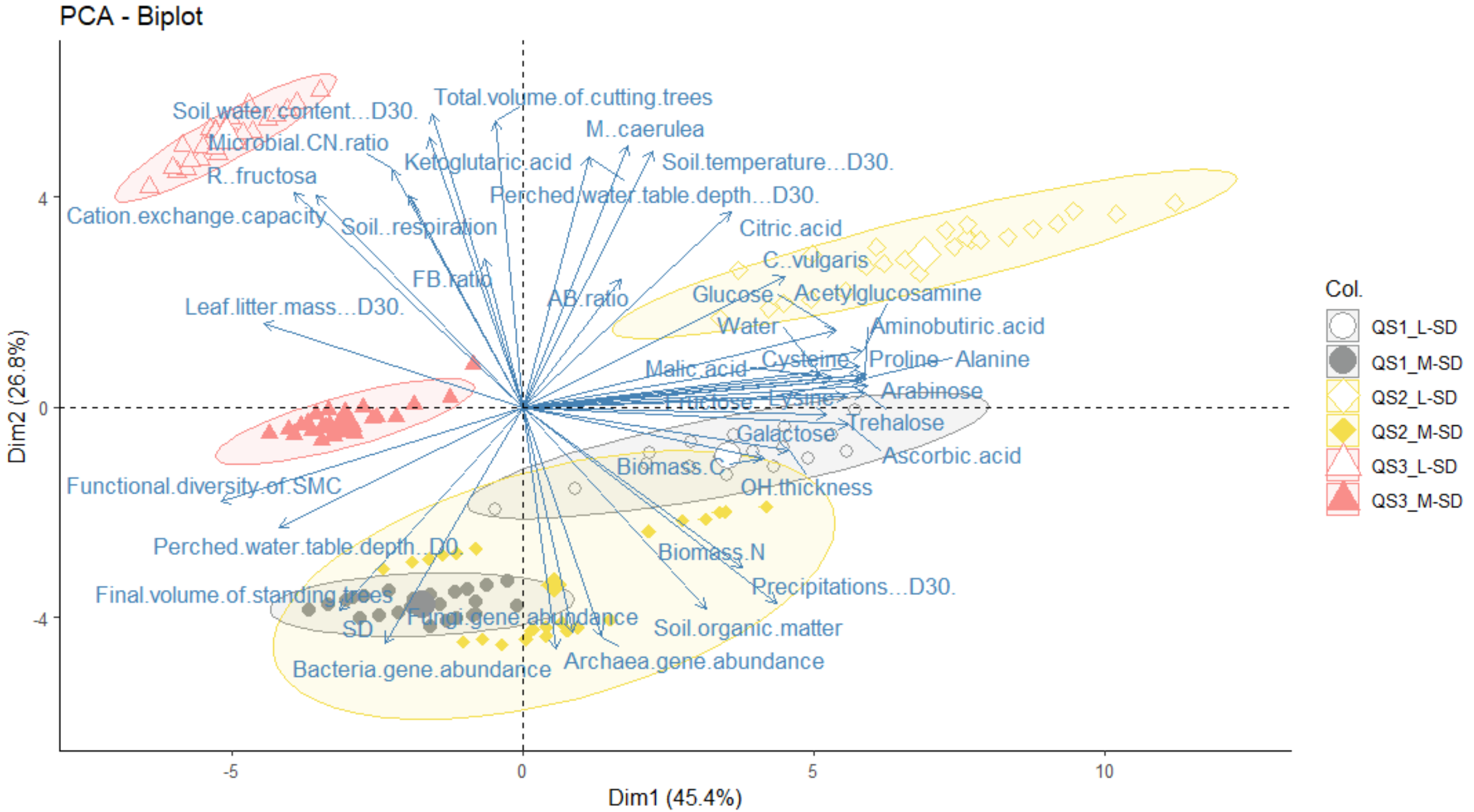

Stand density
L-SD
M-SD
Multiple substrate-induced respiration of SMC (μ mol CO_2 m^{-2} s^{-1})
0.0
0.1
0.2
0.3
D-Fructose
D-Glucose
D-Galactose
L-Arabinose
D-(+)-Trehalose
L-Alanine
N-Acetylglucosamine
L-Lysine
L-Proline
L-Cysteine
Aminobutiric acid
Citric acid
Ascorbic acid
L-Malic acid
Ketoglutaric acid
Blank (water)

Table 5

		Stand characteristics			Hydric properties				Soil physicochemical properties				Litter mass	Vegetation cover of understory species		
		Stand density	Final volume of standing trees.ha^{-1}	Total volume of cutting trees .ha^{-1}	Precipitations (-D30)	Soil water content (-D30)	Perched water table depth (-D30)	Perched water table depth (D0)	Organic matter	OH thickness	Cation Exchange Capacity	Soil temperature (-D30)	Leaf litter mass (-D30)	*Calluna vulgaris*	*Molinia caerulea*	*Rubus fructosus*
Respiration *in situ*	Soil basal respiration (June 2019)	***ns***	*ns*	*0.49****					-0.70***	*ns*	0.59***			*ns*	0.50***	0.60***
Microbial biomass	Biomass C	***ns***	*-0.37***	*ns*	0.71***	*ns*	*ns*	-0.53***	0.57***	*ns*	*-0.47****	*ns*	-0.66***	*ns*	*ns*	*-0.49****
	Biomass N	***ns***		*ns*	0.84***	-0.50***	*0.46****	*ns*	0.85***	*0.39***	-0.55***	*ns*	-0.54***	*ns*	*-0.42***	-0.59***
	Microbial C:N ratio	***ns***	*ns*	0.59***	-0.74***	0.70***	-0.71***	*ns*	-0.83***	*-0.41***	*0.46****	*0.35**	*0.41***	*ns*	0.62***	0.62***
Microbial gene abundance	Bacterial	***0.42****	*ns*	-0.62***	*0.44****	-0.63***	0.63***	*ns*	0.58***	*0.38***	*-0.41****	*-0.45****	*ns*	*ns*	-0.52***	*-0.47****
	Archaeal	***ns***	*ns*	-0.58***	0.54***	-0.61***	0.64***	*ns*	0.71***	*0.49****	*-0.42****	*-0.34**	*ns*	*ns*	*-0.48****	-0.51***
	Fungal	***0.41****	*ns*	-0.62***	*0.39***	-0.65***	0.67***	*ns*	0.56***	*0.47****	*-0.41****	*-0.46****	*ns*	*ns*	-0.54***	*-0.48****
	A:B ratio	***-0.49****	-0.50***	*0.38***	*ns*	*ns*	*ns*	*-0.39***	*ns*	*ns*	*ns*	0.50***	*ns*	*ns*	*ns*	*ns*
	F:B ratio	***ns***	*ns*	*0.34**	*-0.46****	*0.34**	*ns*	*ns*	*-0.40***	*ns*	*ns*	*ns*	*ns*	*ns*	*ns*	*0.35**
Functional diversity and activity (multiple substrate induced-respiration)	Functional diversity	**0.57*****	0.64***	*ns*	*-0.38***	*ns*	*ns*	0.62***	*ns*	-0.58***	*ns*	-0.55***	*0.40***	-0.63***	*-0.39***	*ns*
	Fructose	***-0.37****	*-0.48****	*ns*	0.58***	*ns*	*ns*	-0,61***	*0.37***	0.67***	-0.52***	*0.35**	-0.67***	0.70***	*0.33**	-0.56***
	Glucose	***-0.36****	*-0.46****	*ns*	*0.44****	*ns*	*-0.47****	-0,65***	*ns*	0.63***	*-0.44****	*0.39***	-0.63***	0.83***	0.55***	*-0.48****
	Galactose	***ns***	*ns*	*ns*	0.62***	*ns*	*ns*	-0,59***	*0.43***	0.52***	*-0.49****	*ns*	-0.63***	0.60***	*ns*	-0.54***
	Arabinose	***-0.38****	-0.50***	*ns*	0.60***	*ns*	*ns*	-0,62***	*0.41***	0.73***	-0.53***	*0.36***	-0.65***	0.72***	*0.33**	-0.58***
	Trehalose	***-0.38****	*-0.48****	*ns*	0.64***	*ns*	*ns*	-0,64***	*0.45****	0.63***	-0.52***	*0.35**	-0.68***	0.67***	*ns*	-0.56***
	Alanine	***-0.49****	-0.59***	*ns*	0.62***	*ns*	*ns*	-0,64***	*0.44****	0.69***	-0.51***	*0.44****	-0.67***	0.67***	*ns*	-0.55***
	Acetylglucosamine	***-0.42****	-0.53***	*ns*	0.55***	*ns*	*-0.37***	-0,70***	*ns*	0.72***	*-0.48****	*0.43***	-0.66***	0.83***	*0.49****	-0.54***
	Lysine	***-0.44****	-0.55***	*ns*	0.57***	*ns*	*ns*	*-0,47****	*0.42***	0.66***	-0.53***	*0.35**	-0.62***	0.52***	*ns*	-0.55***
	Proline	***-0.40****	-0.52***	*ns*	0.56***	*ns*	*ns*	-0,60***	*0.37***	0.74***	-0.51***	*0.37***	-0.63***	0.72***	*0.35**	-0.56***
	Cysteine	***-0.39****	-0.51***	*ns*	0.52***	*ns*	*ns*	-0,58***	*ns*	0.71***	-0.51***	*0.37***	-0.64***	0.72***	*0.37***	-0.55***
	Aminobutiric	***-0.39****	-0.51***	*ns*	0.57***	*ns*	*ns*	-0,64***	*0.37***	0.74***	-0.51***	*0.38***	-0.64***	0.77***	*0.39***	-0.57***
	Citric	**-0.51*****	-0.52***	*0.39***	*ns*	*0.32*	-0.59***	*-0,43****	*ns*	*0.34**	*ns*	0.53***	*ns*	0.61***	0.64***	*ns*
	Ascorbic	***-0.40****	-0.53***	*ns*	0.65***	*ns*	*ns*	-0,58***	0.61***	0.74***	*-0.45****	*0.35**	-0.51***	0.56***	*ns*	-0.53***
	Malique	***-0.35****	*-0.43****	*ns*	0.55***	*ns*	*ns*	-0,63***	*0.36***	0.54***	*-0.42***	*0.35**	-0.60***	0.65***	*0.33**	*-0.47****
	Ketoglutarique	**-0.5*****	*-0.39***	0.79***	-0.77***	0.82***	*-0.49****	*ns*	-0.67***	*ns*	0.75***	0.54***	0.54***	*ns*	*0.49****	0.79***
	Water	***ns***	*-0.42***	*ns*	*0.39***	*ns*	*ns*	*-0.38***	*ns*	0.70***	*-0.47****	*ns*	*-0.48****	0.60***	*ns*	*-0.49****

Figure A.1


PCA - Biplot
Soil.water.content...D30.
Microbial.CN.ratio
R..fructosa
Cation.exchange.capacity
Total.volume.of.cutting.trees
M..caerulea
Ketoglutaric.acid
Soil.temperature...D30.
Perched.water.table.depth...D30.
Soil..respiration
Citric.acid
C..vulgaris
FB.ratio
AB.ratio
Glucose
Acetylglucosamine
Leaf.litter.mass...D30.
Water
Aminobutiric.acid
Malic.acid
Cysteine
Proline
Alanine
Fructose
Lysine
Arabinose
Trehalose
Galactose
Ascorbic.acid
Biomass.C
OH.thickness
Functional.diversity.of.SMC
Perched.water.table.depth...D0.
Biomass.N
Precipitations...D30.
Final.volume.of.standing.trees
SD
Fungi.gene.abundance
Soil.organic.matter
Archaea.gene.abundance
Bacteria.gene.abundance
Dim2 (26.8%)
Dim1 (45.4%)
4
0
-4
-5
0
5
10
Col.
QS1_L-SD
QS1_M-SD
QS2_L-SD
QS2_M-SD
QS3_L-SD
QS3_M-SD

Declaration of interests

☒ The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

☐The authors declare the following financial interests/personal relationships which may be considered as potential competing interests: