



## Soil microbial community composition and functions are disrupted by fire and land use in a Mediterranean woodland

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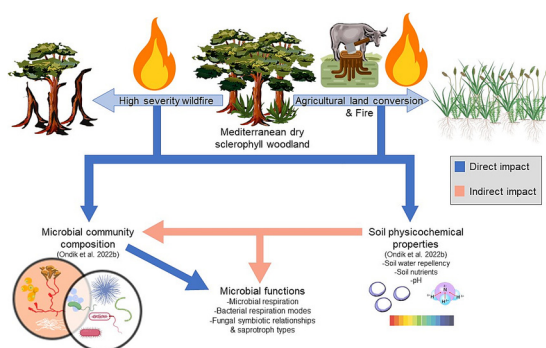
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### HIGHLIGHTS

- Fire and land use can negatively impact soil microbial communities.
- Impacts on soil microbial function were inferred by genus via traits databases.
- Fire reduced anaerobic, microaerophilic, and symbiotic bacteria and symbiotic fungi.
- Agricultural land use promoted soil and litter saprotrophs over wood saprotrophs.
- Soil disturbances drove microbial communities via soil pH, water, and nutrients.

### GRAPHICAL ABSTRACT



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### ABSTRACT

The intersection of fire, land use transformations, and climate change is putting Mediterranean climate-type ecosystems at risk of soil degradation and loss of ecosystem services. Ondik et al. (2022b) showed that in a Mediterranean dry sclerophyll woodland of South Australia, high severity fire and clearing and grazing practices impacted both physicochemical and biological soil quality indicators. Building upon the work of Ondik et al. (2022b) this study aims to 1) identify soil physicochemical properties impacted by fire and land management that are indirect drivers of changes to soil microbial community composition and 2) determine whether the observed changes to soil microbial community composition affect soil microbial functions. Via a redundancy analysis, we identified fire and management-induced changes to pH, soil water repellency, nutrient stoichiometry, and total nutrient content as significant drivers of the composition of soil microbial communities. We then measured basal respiration, substrate induced respiration, and the carbon mineralisation quotient, and calculated functional trait distributions among microbial communities by linking 16S and 18S rRNA sequences to respiration modes and functional guilds, respectively. We found that fire reduced soil microbial respiration and the relative abundance (RA) of microbial symbionts, anaerobic bacteria, and microaerophilic bacteria, while increasing the RA of aerobic bacteria. Furthermore, management increased the RA of post-fire ectomycorrhizal fungi and may have reduced pathogenic load, microbial efficiency, and wood saprotrophs, while increasing litter, soil, and other saprotrophic species that are adapted to grasslands. This study shows that, through changes to microbial community composition, high severity wildfire and land management affected soil respiration rates, bacterial modes of respiration, prevalence of symbiotic bacteria and fungi, and microbial substrate preference. Having identified the main physicochemical drivers of changes to microbial community composition, we provide valuable insights into how fire and land management can impact soils in Mediterranean woodland.

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## 1. Introduction

The intersection of wildfire and land use conversion from woodland to pastures or cropland is becoming more common globally, putting sensitive soils in Mediterranean climate-type regions and the copious ecosystem services they support at risk (Gauquelin et al., 2018; Moreira et al., 2020). In response to drought and climate change-induced temperature rises, high intensity wildfires are causing negative soil outcomes (Lucas-Borja et al., 2021; Staal et al., 2020), including soil erosion, changes to soil hydrology, and loss of soil microbial abundance, microbial function, and soil nutrients (Collins et al., 2021; McIntosh et al., 2005). Furthermore, research shows that anthropogenic conversion of land affects many of the same soil components as wildfire, e.g., soil nutrient content, microbial community composition and diversity, pH, and soil stability (Baldock et al., 2013; Miralles et al., 2012; Ondik et al., 2022b; Sangha et al., 2006). Mediterranean climate-type regions of Australia exemplify fire-prone land that has been heavily impacted by anthropogenic land conversion. In Australia, over 45 % of the country's land has been converted from native vegetation to pastures for grazing livestock (Australian Bureau of Agricultural and Resource Economics and Sciences, 2016). During the summer of 2019–20, Australia experienced unprecedented wildfires of high severity that burned large swaths of land including areas previously converted to agricultural land (Collins et al., 2021; Ondik et al., 2022b). However, it is not well understood how the interaction between widespread land conversion and high severity fires impacted soil microbial communities and their functions.

Soil microbes are an essential component of soil health, fertility, and overall function, and as such, they are an important target of study. Soil microbial functions, such as symbiotic interactions with plants, can protect plants from pathogens (Wu et al., 2020; Zhou et al., 2022), and organic matter decomposition and nitrogen fixation can create a healthy and fertile soil bed (Saccá et al., 2017). In doing so, soil microbes promote carbon and water storage, biodiversity, habitat, and local climate mitigation (Bardgett and van der Putten, 2014; Griscom et al., 2017). However, not all microbes make positive contributions. For example, oomycete pathogens can infect and cause harm to Eucalyptus trees (Zhou et al., 2022). Thus, changes to native microbial communities, e.g., an increase in abundance of pathogens or a decrease in plant symbionts, can have consequential effects on soil health and dependent ecosystems.

Microbes require certain environmental parameters to perform functions in the soil, and changes to these parameters can drive changes to microbial community composition and functions. For example, fire can change the quantity and quality of soil carbon, impact total soil carbon and nitrogen content, increase soil pH, and reduce soil moisture, all of which are known to influence microbial communities (Ahlgren, 1974; Borgogni et al., 2019; D'Ascoli et al., 2005; Moyano et al., 2013; Rutigliano et al., 2013). Land use conversion has also been shown to impact microbial communities through changes to soil nutrient content, nutrient storage capacity, and pH (Muñoz-Rojas et al., 2015; Ondik et al., 2022b). Bacteria and fungi are known to react differently to changes in soil components. For instance, bacteria have been shown to proliferate better under alkaline conditions and recover more quickly after fire than fungi (Bárcenas-Moreno et al., 2016; Muñoz-Rojas and Bárcenas-Moreno, 2019; Rodríguez et al., 2017; Rousk et al., 2009). Yet, there is still much to be learned about the impacts of land management and fire on soil microbial functions and how it relates to overall soil health.

Due to their sensitivity to disturbance, Mediterranean ecosystems have become a target for many restoration efforts around the world to try and prevent loss of soil function and dependent ecosystem services they support (Bateman and Muñoz-Rojas, 2019; IRP, 2019; Lucas-Borja et al., 2021). Identifying drivers of soil microbial communities and disturbance-induced changes to their functions may help direct post-disturbance restoration efforts to counteract the negative impacts of disturbance on soil health in Mediterranean climate-type regions. In Ondik et al. (2022b), both fire and land management practices, i.e., historical clearing and grazing, were shown to alter microbial community compositions, with the strongest changes occurring in fungal communities. However, it was not

clear whether the changes observed in taxonomic composition resulted in differences to microbial function.

This study aims to answer the questions, a) what soil physicochemical properties impacted by fire and land management are indirect drivers of changes to soil microbial community composition and b) do these changes to microbial community composition impact soil microbial function? To answer these questions, we performed a redundancy analysis and assessed changes to rates of microbial activity and metabolic efficiency, bacterial respiration modes, and fungal functional guilds.

## 2. Materials and methods

### 2.1. Study area description

The study area is located in Western River, Kangaroo Island, South Australia (35°42' S, 136°53' E to 35°45' S, 137°1' E) and includes both historically cleared and grazed pastures (hereafter “managed”) and dry sclerophyll woodland (hereafter “non-managed”). The elevation and slope of the study area range from 181 to 298 m and 5–48° (9–110 %), respectively. The region experiences warm/cool summers, with a mean annual rainfall of 553 mm and a mean annual temperature of 11.6–19.2 °C and has been classified as a Mediterranean, Csb, climate (Commonwealth Australia Bureau of Meteorology, 2022; Peel et al., 2007). Loam and sandy loam are the dominant soil textures in the area, with soils consisting of rocky Sodosols and Tenosols and ferric brown-red Chromosols and Kurosols (Department for Environment and Water, 2009a, 2009b; DEWNR Soil and Land Program, n.d.).

Management practices in pastures were abandoned in 2008 upon change of ownership, and in December 2019, a crown fire burned all managed and most non-managed zones within the study area (Ondik et al., 2022b). The study area had no history of fire in the 60 years prior to the 2019–2020 bushfire season (Department for Environment and Water, 2020). In non-managed areas, *Eucalyptus baxteri*, *Eucalyptus obliqua*, and *Eucalyptus cladocalyx* dominated the overstory vegetation, and *Allocasuarina striata* and *Banksia marginata* dominated the understory vegetation. Managed areas were instead dominated by weeds, *Romulea* spp., *Medicago* spp., and *Arctotheca calendula* and the native grass, *Rytidosperma geniculatum*.

### 2.2. Experimental design and soil sampling

In the study area, there were two treatments, 1) fire and 2) historical clearing and grazing (hereafter “management”), and three experimental groups, unburnt non-managed (UNM; n = 4), burnt non-managed (BNM; n = 4), and burnt managed (BM; n = 4), each replicated in four different sites for a total of twelve sites. Because all historically grazed pastures were burnt by the fire event and there were no unburnt managed areas, all analyses considered treatments, fire (UNM v. BNM sites) and management (BNM v. BM sites), separately.

Soil samples were collected six months post-fire, from burnt sites on June 28th, 2020, and from unburnt sites on October 1st, 2020. Within each site, a 10 m × 10 m plot was measured out, and nine soil core samples, 20 cm in diameter and 0–5 cm in depth, were taken from the topsoil at 3 m intervals around the perimeter of the plot. Then, 50 mL samples were aseptically collected into sterile Falcon tubes to be used for microbial analyses. Soil samples were transported, with 50 mL samples on ice, to the University of New South Wales (UNSW) to be processed and analysed. For soil nutrient measures, soil cores were air dried and passed through a 2 mm stainless steel sieve. For each of the twelve sites, the nine cores per site were separated into three groups, pooled into three replicate samples (n = 36), and stored at room temperature. The 50 mL soil samples were kept wet, pooled into three replicate samples per site (n = 36) and stored at 4 °C for biological analyses.

### 2.3. Soil properties

As reported in Ondik et al. (2022b), pH and electrical conductivity (EC) were measured at a 1:5 soil to deionized water ratio using pH and EC probes

(Instrument Choice, SA, Australia), while total carbon (TC) and total nitrogen (TN) were measured on a vario MACRO cube (Elementar, Sydney, NSW, Australia) at the Mark Wainwright Analytical Centre (UNSW Sydney). Then, the carbon to nitrogen ratio (C/N) was calculated, and soil water repellency (SWR) was measured by the water droplet penetration time (Dekker et al., 2001; Ritchie et al., 2020). Raw data can be found at <https://doi.org/10.6084/m9.figshare.21277659.v1>.

Total organic carbon (TOC) was measured by potassium dichromate ( $K_2Cr_2O_7$ ) and sulfuric acid ( $H_2SO_4$ ) extraction (Mingorance et al., 2007). Calibration standards of 0–12 mg/mL of carbon were made by dissolving 7.1204 g of sucrose in 250 mL distilled water and diluting into distilled water. Then, 1 mL of each standard underwent the following protocol alongside soil samples. For each soil sample, 3 mL of 1 N  $K_2Cr_2O_7$  and 6 mL of 99 %  $H_2SO_4$  were added to 100 mg of soil. Samples were loosely capped and gently swirled, left standing for 3 h, and then, 10 mL of distilled water was added to each sample. Samples were tightly capped, shaken on an orbital shaker for 15 min at 100 rpm, then left standing for 48 h. Absorbance of sample supernatant was then measured by an Epoch2 Microplate Spectrophotometer (BioTek Instruments, VT, USA) at the 590 nm wavelength.

Basal respiration (BR) and substrate induced respiration (SIR) rates, representing microbial activity and microbial biomass, respectively (Muñoz-Rojas et al., 2016; Santorufo et al., 2021), were measured using the MicroResp™ system (Campbell et al., 2003, 2008). First, water holding capacity (WHC) was determined using pressure plates at 1 bar (NSW Department of Sustainable Natural Resources, n.d.). Then, the moisture content of all samples was adjusted to  $\leq 45$  % WHC by incubating at 15 °C and mixing each sample at daily intervals to prevent uneven drying (Creamer et al., 2016). For each soil sample, an equal volume (215  $\mu$ L) of soil was added to a deep well plate, the soil was weighed, and moisture content was adjusted to 45 % WHC. Then, samples underwent a 72-h pre-incubation period at 25 °C. Next, 25  $\mu$ L of water was added to each sample, the indicator plate was sealed onto the sample wells, and samples were incubated for 6 h at 25 °C. Using an Epoch2 Microplate Spectrophotometer (BioTek Instruments, VT, USA), the absorbance of the indicator plate was measured before and after the 6-h incubation at 570 nm (Creamer et al., 2016). To measure SIR, the above protocol was repeated with a treatment of 25  $\mu$ L of glucose solution adjusted to 30 mg/g of soil water for each soil sample. The difference in  $CO_2$  percentage, as measured by change in absorbance, was equal to  $CO_2$  produced during respiration, i.e., BR or SIR. Calibration of the spectrophotometer (BioTek Instruments, VT, USA) was performed via gas chromatography–mass spectrometry (GC/MS) using a Focus DSQ II GC/MS instrument (Thermo Fisher Scientific, Bremen, Germany), known  $CO_2$  standards and soil produced  $CO_2$  samples under the supervision of the Bioanalytical Mass Spectrometry Facility (BMSF) of the Mark Wainwright Analytical Centre (UNSW, Australia). SIR and BR were then converted from percentages to  $mg\ C\ g^{-1}\ soil\ h^{-1}$  by following the MicroResp™ manual instructions (Cameron, 2007).

The carbon mineralisation quotient (qM) was calculated to determine how efficiently microbes metabolize organic matter (Francaviglia et al., 2017) by dividing cumulative BR over a 6-h period (Ccum) by TOC percentage (Dommergues, 1960; Francaviglia et al., 2017; Kneller et al., 2018).

$$qM = \frac{Ccum \left( \frac{g\ CO_2}{100g\ soil} \right)}{TOC \left( \frac{g\ C}{100g\ soil} \right)} * 100 = \frac{BR \left( \frac{g\ CO_2}{100g\ soil * 6\ hours} \right)}{TOC (\%)} * 100 \quad (1)$$

Raw data can be found at <https://doi.org/10.6084/m9.figshare.21671891.v1>.

#### 2.4. Soil microbial taxonomy and traits

Raw sequencing data, produced by targeting the V1–V3 region of the 16S rRNA gene, via 27F and 519R primers, and the hypervariable region of the 18S rRNA gene, via 1391F and EukB primers (see Ondik et al., 2022b for further details), were analysed in the following ways. All reads

from 16S and 18S datasets were used to identify relationships between soil physicochemical properties and microbial communities. Then, all 16S and only fungal 18S reads were analysed to assess the distribution of soil respiration modes and fungal guilds, respectively, across experimental groups. Raw reads are available at the NCBI Sequence Reads Archive (SRA) under BioProject ID, PRJNA847045.

Taxonomy was successfully assigned to 99.9 % of 16S rRNA reads and 67.6 % of 18S rRNA reads using the SILVA database v132 (Quast et al., 2013). To ensure 18S OTUs that were unassigned using the SILVA database (32.4 %) were not from fungal species, taxonomy of all 18S reads was reassigned using the PR<sup>2</sup> database (Vaulot et al., 2021). Comparisons made between the two datasets (Fig. S3) showed that 39.8 % of all 18S reads were fungal, 49.1 % were non-fungal eukaryotic, and 11.1 % of all reads remained unassigned. All fungal reads (39.8 % of all reads) were then isolated from the 18S dataset, and all non-fungal eukaryotic (49.1 %) and unassigned OTU reads (11.1 %) from the 18S dataset were excluded from analysis of the distribution of fungal taxa and traits across experimental groups. All unassigned 16S reads were also excluded from the analysis of bacterial taxa and traits. For all identified bacterial and fungal genera, the mean and standard error in relative abundance (RA) by experimental group was then calculated (Table S4; Table S5), and for dominant genera, i.e., those making up at least 1 % RA of their respective microbial community, large differences in RA due to fire or management were identified.

At the genus level, functional guilds from the FUNGuild database (Nguyen et al., 2016) were matched with fungal 18S OTUs. Also at the genus level, respiration modes from the BactoTraits database (Cébron et al., 2021) were matched with 16S OTUs. Dominant genera that did not match with the BactoTraits database, were manually researched (Table S6), and respiration mode was assigned to each genus individually. As a result, 62.9 % of 16S OTUs and 58.8 % of fungal OTUs matched with genera entries in the BactoTraits database plus manual assignments and FUNGuild database, respectively. After matching genera to functional traits, relative abundance (RA) plots and heat maps of each functional trait, i.e., bacterial respiration mode (Fig. 4) and fungal functional guild (Fig. 5), were produced. For all RA plots, all possible trait values per genus were concatenated and separated by a dash, e.g., Animal Pathogen-Fungal Parasite-Undefined Saprotroph, and read count per genus was counted once for the concatenated trait value matched to that genus. Then, individual trait values per genus were represented separately via heat maps, and read count per genus in each experimental group, e.g., 2315, was counted once for each unique trait value matched to that genus, e.g., 2315 reads for Animal Pathogen, 2315 for Fungal Parasite, and 2315 for Undefined Saprotroph, so that all heatmap values represent the maximum possible read count per trait.

#### 2.5. Statistical analyses

The open-source software, R, version 4.0.4, was used to carry out all following analyses (R Core Team, 2021). The TOC, BR, SIR, and qM data satisfied assumptions of normality and homogeneity of variance, as tested by the Kolmogorov-Smirnov test and the Levene test, respectively, and were modelled using a Generalized Linear Mixed model (GLMM) with a Gaussian error distribution (Brooks et al., 2017). Data models considered two treatments, fire and management, as independent variables and site as a random variable. Using the contrasts of estimated marginal means, a significance test ( $p < 0.05$ ) was carried out on modelled data to determine differences in soil properties and parameters due to fire (UNM v. BNM) or management (BNM v. BM).

Previously collected data on soil physicochemical properties, i.e., pH, electrical conductivity (EC), total carbon (TC), total nitrogen (TN), total carbon to total nitrogen ratio (C/N), and soil water repellency (SWR) (Ondik et al., 2022a), along with TOC, BR, and SIR were correlated with fire and management treatments by performing a principal component analysis (PCA) using the “ggbiplot” package (Vu, 2011). Then, transformation based redundancy analysis (RDA) plots, created via the “vegan” package

(Oksanen et al., 2022), were used to derive relationships between soil physicochemical properties, experimental group, and soil microbial community composition at the OTU level. Unlike the analysis of soil microbial traits, both the PCA and RDA analyses were done using all 16S (bacterial) and all 18S (soil eukaryote) rRNA reads data, including fungal, non-fungal, and unassigned reads. RDA plots were done as described in Machado De Lima et al. (2021). For RDA plots, OTU RAs were normalized using the Hellinger method (square root), and then soil physicochemical properties with a  $vif.cca$  value greater than ten, indicating multicollinearity, were removed. Forward selection was performed on the remaining soil properties to identify the variables that may be significantly correlated with the microbial communities of each sample, i.e., TN, pH, EC, SWR, and C/N (see results Section 3.3 for full details). Significance ( $p < 0.05$ ) was determined using an ANOVA test, and the  $Pr(>F)$  of each variable was adjusted using the Benjamini and Hochberg method to control for false discovery (Benjamini and Hochberg, 1995). Then, EnvFit was performed to calculate the  $Pr(>r)$  of each variable. One of the twelve replicates from BNM sites in the 16S OTUs dataset was identified as an outlier by non-metric multi-dimensional scaling and excluded from the RDA.

### 3. Results

#### 3.1. Soil organic carbon, respiration rates, and microbial quotient

Fire caused significantly lower TOC, SIR, and BR and significantly higher qM in BNM compared to UNM sites (Fig. 1). Furthermore,

management resulted in significantly higher post-fire TOC and SIR and significantly lower qM in BM sites compared to BNM sites but caused no significant difference in BR (Fig. 1).

#### 3.2. Correlations between soil physicochemical properties and microbial community composition

The first two principal components (PC) of the principal component analysis (PCA) (Fig. 2) accounted for a total of 77.2% and 68.1% of the variation in measured soil properties in non-managed sites (UNM and BNM; Fig. 2A) and burnt sites (BNM and BM; Fig. 2B), respectively. In the first PC of non-managed sites (64.9% of variation), there was a positive correlation between TC, TN, TOC, SIR, BR, EC, and SWR and UNM sites, while pH was strongly negatively correlated with these properties and positively correlated with BNM sites. In the first PC of burnt sites (52.2% of variation), there was a positive correlation between TC, TN, TOC, SWR, and SIR and BM plots, while pH and C/N were strongly negatively correlated with these properties and positively correlated with BNM sites. Furthermore, BR was correlated with SWR but was not correlated with burnt sites, BNM and BM.

The soil properties, TN, pH, EC, SWR, and C/N, were chosen by forward selection for the RDA (Fig. 3). Due to multilinearity, TN and total carbon (TC) were identified as redundant constraints, so TC was removed from the RDA model. The properties, pH and SWR, were selected for all microbial communities and all experimental groups, and EC was selected for all microbial communities and experimental groups except soil eukaryotic

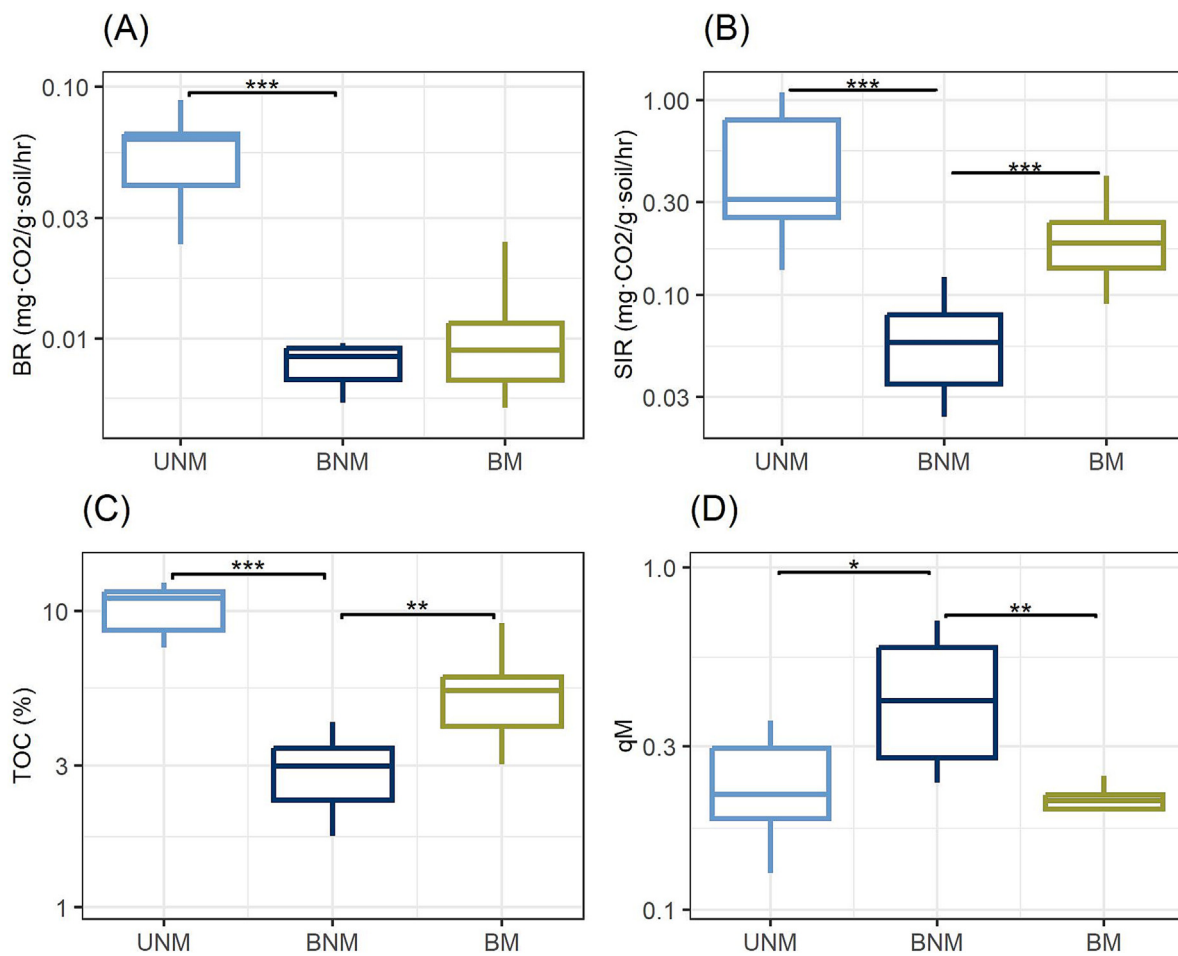
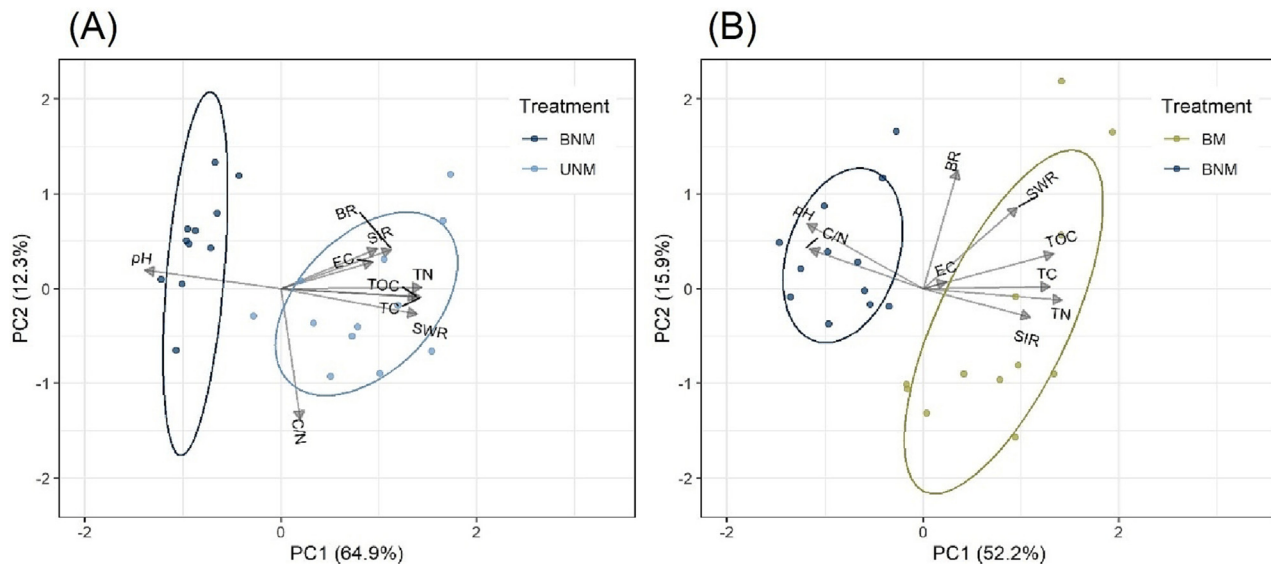


Fig. 1. Differences in soil properties due to fire and management. (A) BR: Basal respiration ( $\text{mg C g}^{-1} \text{ soil h}^{-1}$ ), (B) SIR: substrate induced respiration ( $\text{mg C g}^{-1} \text{ soil h}^{-1}$ ), (C) TOC: total organic carbon (%), and (D) qM: carbon mineralisation quotient by experimental group, UNM: unburnt non-managed; BNM: burnt non-managed; BM: burnt managed. All soil properties were measured from samples of the topsoil (0–5 cm). Significant differences between experimental groups due to fire (UNM vs. BNM) and management (BNM vs. BM) are designated as follows:  $p < 0.05$  (\*),  $p < 0.01$  (\*\*) and  $p < 0.001$  (\*\*\*).



**Fig. 2.** Principal component analysis (PCA). PCA of disturbance-affected soil properties among differing (A) fire treatment (UNM vs. BNM) and (B) management (BNM vs. BM). Variance of soil properties along the first two principal components is given in parentheses in axes labels. pH: pH; EC: electrical conductivity; BR: basal respiration rate; SIR: substrate-induced respiration rate; SWR: soil water repellency; TC: total carbon; TN: total nitrogen; TOC: total organic carbon; C/N: carbon to nitrogen ratio. UNM (light blue): unburnt non-managed; BNM (dark blue): burnt non-managed; BM (green): burnt managed. Each point represents one soil sample.

communities of non-managed sites. Then, TN was selected for only eukaryotic communities and all experimental groups, and C/N was only selected for bacterial communities of burnt sites.

The RDA factors 1 and 2 explained 40.8 % and 4.79 %, respectively, of variance in bacterial community composition in non-managed sites, with pH ( $R^2 = 0.95$ ), SWR ( $R^2 = 0.86$ ), and EC ( $R^2 = 0.60$ ) significantly correlated with factor 1 ( $p \leq 0.001$ ). The RDA factors 1 and 2 explained 33.65 % and 6.93 %, respectively, of variance in bacterial community composition in burnt sites, with SWR ( $R^2 = 0.91$ ), pH ( $R^2 = 0.79$ ), and C/N ( $R^2 = 0.65$ ) significantly correlated with factor 1 ( $p \leq 0.001$ ). The RDA factors 1 and 2 explained 40.81 % and 7.37 %, respectively, of variance in soil eukaryote community composition in non-managed sites, with pH ( $R^2 = 0.93$ ), TN ( $R^2 = 0.86$ ), and SWR ( $R^2 = 0.89$ ) significantly correlated with factor 1 ( $p \leq 0.001$ ). The RDA factors 1 and 2 explained 40.76 % and 6.88 %, respectively, of variance in soil eukaryote community composition in burnt sites, with pH ( $R^2 = 0.75$ ), TN ( $R^2 = 0.87$ ), and SWR ( $R^2 = 0.80$ ) significantly correlated with factor 1 ( $p \leq 0.001$ ). (Supplementary materials Section 1.2.)

### 3.3. Soil microbial community composition, bacterial respiration modes, and fungal guilds

Fire and management impacted the relative abundance of dominant taxonomic genera within both bacterial (Table S2) and fungal communities (Table S3). Fire increased the RA of bacteria, including *Massilia* and *Pseudarthrobacter*, which were absent in UNM plots, and *Streptomyces*, and fungi, including *Penicillium*, *Umbelopsis*, and *Rhynchosporium*. Fire also reduced the RA of bacteria, including *Roseiarcus*, Xanthobacteraceae, *Acidipila*, Acetobacteraceae, and *Acidothermus*, and fungi, including Agaricales, *Mortierella*, *Clavulina*, and Herpotrichiellaceae. Management increased the RA of bacteria, including *Conexibacter*, *Bacillus*, and Gemmataceae, and fungi, including *Mortierella* and Agaricales. Management also reduced the RA of bacteria, such as *Pseudarthrobacter*, *Massilia*, and *Streptomyces*, and fungi, including *Umbelopsis* and *Penicillium*.

By experimental group, 73.91 % (UNM), 66.48 % (BNM), and 56.50 % (BM) of 16S OTUs matched with respiration modes in the BactoTraits database or by manual assignment. Among assigned respiration modes of bacterial communities, fire and management resulted in the following trends (Fig. 4; Table S2). Fire at non-managed sites caused the RA of purely aerobic bacteria, e.g., *Streptomyces*, to increase and the RA of purely anaerobic,

e.g., *Acidipila*, and purely microaerophilic bacteria, e.g., *Roseiarcus*, to decrease. Fire also caused a slight increase in the RA of mixed respiration modes, i.e., taxonomic groups such as Burkholderiaceae that contain species with differing respiration modes. Furthermore, management caused higher post-fire RA of purely anaerobic bacteria and may have decreased the RA of purely aerobic bacteria in BM compared to BNM sites, but BM and BNM sites shared a similar variety of mixed respiration modes.

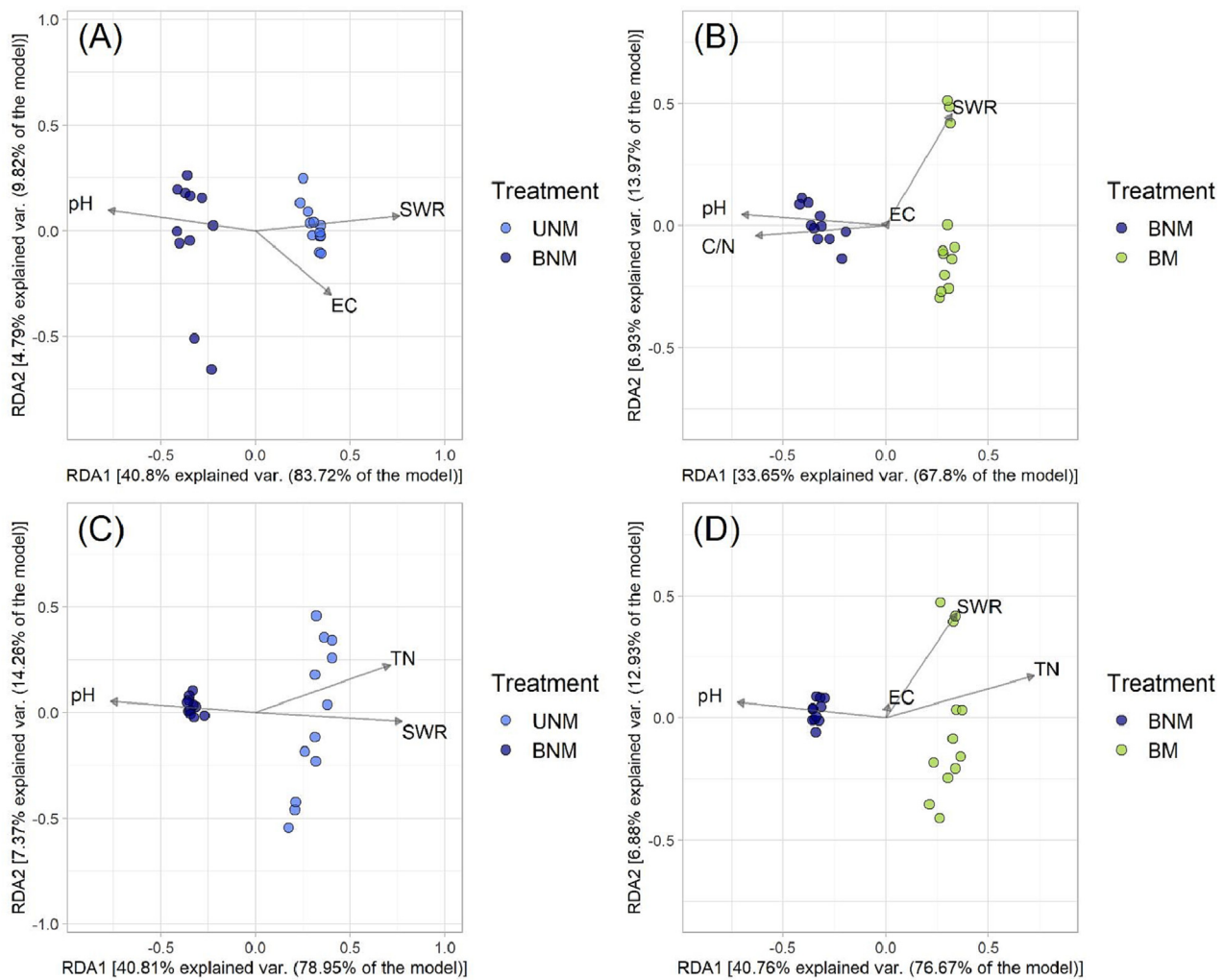
By experimental group, 52.08 % (UNM), 51.44 % (BNM), and 63.53 % (BM) of all fungal 18S reads matched with fungal guild entries in the FUNGuild database. Regarding the distribution of assigned functional guilds within soil fungal communities (Fig. 5; Fig. S2; Table S3), fire caused a reduction in saprotroph-symbiotrophs, e.g., *Mortierella*, symbiotrophic ectomycorrhizal fungi, e.g., *Clavulina* and *Endogone*, and parasite containing pathotroph-saprotrophs, e.g., Herpotrichiellaceae, and pathotroph-saprotroph-symbiotrophs, e.g., Agaricales. Fire also caused an increase in the pathotroph, *Rhynchosporium*, pure saprotrophs, e.g., *Umbelopsis*, and the pathotroph-saprotroph-symbiotroph, *Penicillium*. Management caused an increase in the RA of saprotroph-symbiotrophs, e.g., *Mortierella*, a reduction in pathotroph-saprotroph-symbiotrophs, e.g., *Penicillium*, and pure saprotrophs, e.g., *Umbelopsis*, but had little impact on parasite containing genera.

## 4. Discussion

Here, we identified important physicochemical drivers of bacterial and fungal community composition from soil samples collected over the winter of 2020 and assessed whether changes to microbial community composition impact the functions microbes perform. Fire and management-induced changes to soil water repellency (SWR), pH, total nitrogen (TN) and total carbon (TC), and the total nitrogen to total carbon ratio (C/N) were identified as indirect drivers of microbial community composition. Furthermore, our results confirm that, in modifying the composition of soil microbial communities (Ondik et al., 2022b), fire and management impacted the distribution of fungal function guilds and bacterial respiration modes, as well as microbial biomass, activity rates, and efficacy of carbon mineralisation.

### 4.1. Drivers of microbial community composition and function

Fire and agricultural land use have been shown to change physicochemical properties in the soil (Ondik et al., 2022b), and in consequence, they



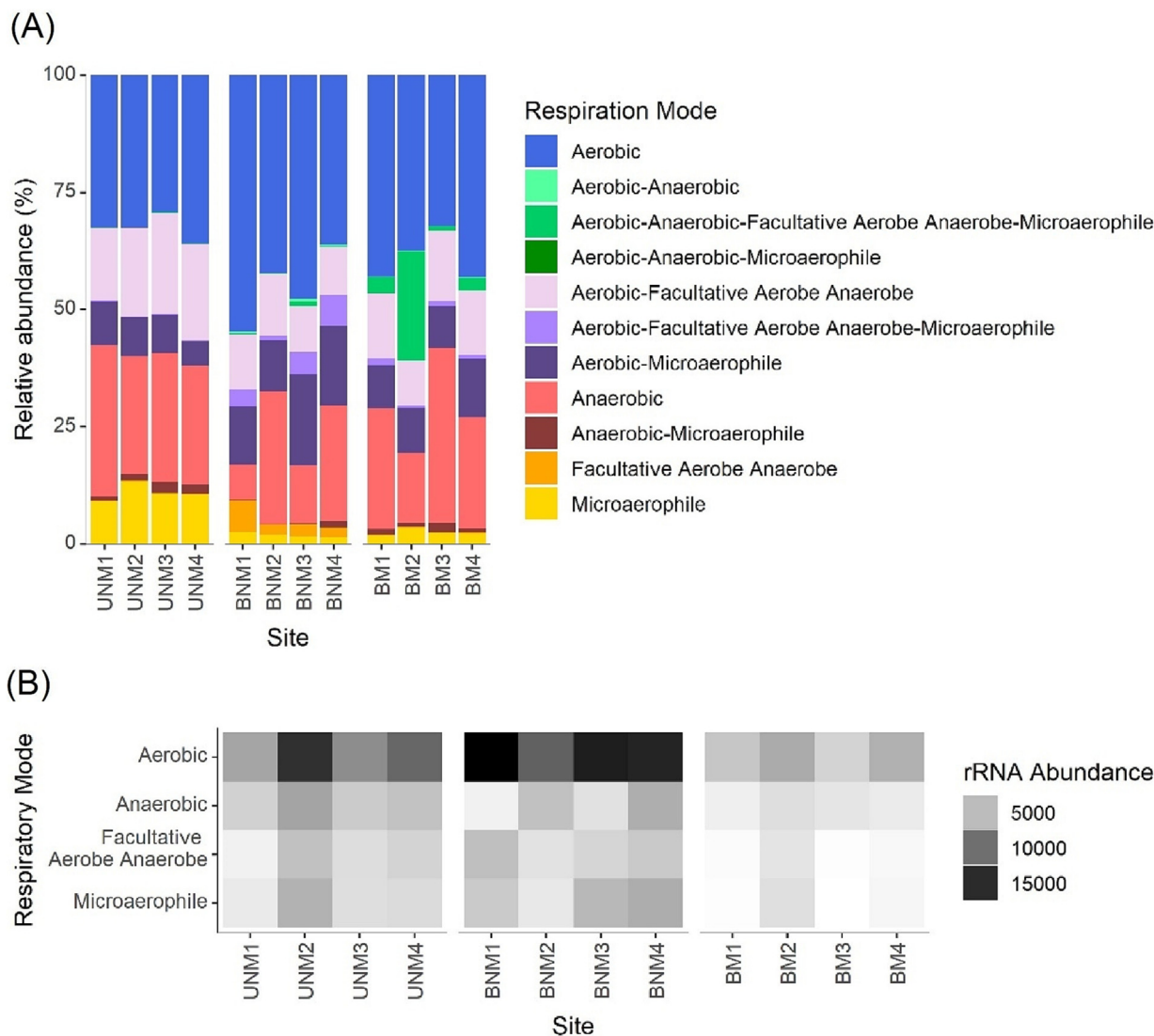
**Fig. 3.** Redundancy analysis (RDA). The environmental loading scores plot of the soil properties highly correlated with community composition of, A&B: bacteria under different (A) fire treatment (UNM v. BNM) and (B) management (BNM v. BM) and C&D: eukaryotes under different (C) fire treatment (UNM vs. BNM) and (D) management (BNM vs. BM). Variance of soil microbial communities along the first two principal RDA components is given in parentheses in axes labels. pH: pH; EC: electrical conductivity; SWR: soil water repellency; TN: total nitrogen; C/N: carbon to nitrogen ratio. UNM (light blue): unburnt non-managed; BNM (dark blue): burnt non-managed; BM (green): burnt managed. Each point represents one soil sample.

can cause indirect changes to soil microbial communities (Franco et al., 2018; Xiang et al., 2014). For example, after disturbance, changes to soil physicochemical properties, including pH, soil water repellency, and soil nutrient levels and ratios have been shown to impact soil bacteria and fungi (Ammitzboll et al., 2022; Lucas-Borja and Delgado-Baquerizo, 2019; Tian et al., 2021). Here, we interpret soil physicochemical properties that are known to be directly impacted by fire and agricultural land use and are correlated with soil microbial communities as indirect drivers of the microbial community composition.

The soil properties, pH and SWR, were identified as significant drivers of both eukaryotic and bacterial communities (Fig. 3). Bacterial abundance has been observed to recover as quickly as one-month post-fire (Pietikäinen and Fritze, 1993), while fungal eukaryotic communities are known to take years if not decades after an event such as fire to return to pre-disturbance levels (Kasel et al., 2008; Xiang et al., 2014). This may be largely because bacterial communities can proliferate better under alkaline conditions than fungi (Ahlgren, 1974; Bárcenas-Moreno et al., 2016; Muñoz-Rojas and Bárcenas-Moreno, 2019; Rodríguez et al., 2017; Rousk et al., 2009). However, some bacteria, e.g., *Acidipila*, *Acidotherrmus*, Gemmataceae, WD2101 soil group, and Micropepsaceae, are known to thrive in acidic soils and react negatively to alkaline conditions (Bräuer et al., 2018; Chen et al., 2021; Dedysh et al., 2021; Hiraishi, 2019; Šimonovičová et al.,

2019). In consequence, the relative abundance (RA) of these bacteria was reduced after the fire increased pH levels but was higher post-fire in the more acidic managed sites (Table S2). Microbes are also sensitive to soil moisture content (Ahlgren, 1974; Moyano et al., 2013). Consequently, fire and management-induced changes to SWR impacted both soil bacteria and eukaryotes.

The high severity fire and management practices resulted in widespread impacts on organic matter degrading bacteria, as well as the soil physicochemical properties, TC, TN, TOC, and C/N (Ondik et al., 2022b). However, C/N was the only nutrient related variable found to be a significant driver of soil bacterial communities (Fig. 3), indicating that the ratio between nutrients was a more important driver of the composition of bacterial communities than nutrient levels alone. Furthermore, C/N has also been identified as a driver of bacterial communities in alpine grassland ecosystems of China indicating a relationship between C/N and bacterial community composition across ecosystem types (Wang et al., 2023). Eukaryotic communities on the other hand were found to be driven by the total amount of nutrients, i.e., nitrogen or carbon, present in the topsoil, but not C/N. Correlation between total nutrient levels and eukaryotic communities has also been observed in other ecosystem types including a subtropical peatland in North Carolina, USA (Shen et al., 2013; Tian et al., 2021; Xiang et al., 2014). Sensitivity of the fungal eukaryotic community to total nutrient levels in



**Fig. 4.** Respiration modes of soil bacteria. (A) Relative abundance plot of the total read count per unique concatenated value of all possible respiration modes per genus. (B) Heatmap of the maximum possible read count per individual respiration mode. Sites by experimental group, UNM: unburnt non-managed ( $n = 4$ ); BNM: burnt non-managed ( $n = 4$ ); BM: burnt managed ( $n = 4$ ). Shown here are all 16S reads that matched with respiration mode entries at the genus level, and unmatched OTUs are not shown.

particular, including complex carbon substrates such as lignin and cellulose (Ammitzoll et al., 2022), may be another reason bacteria are known to recover more quickly after fire events than fungi.

#### 4.2. Microbial abundance, activity rates, and efficacy

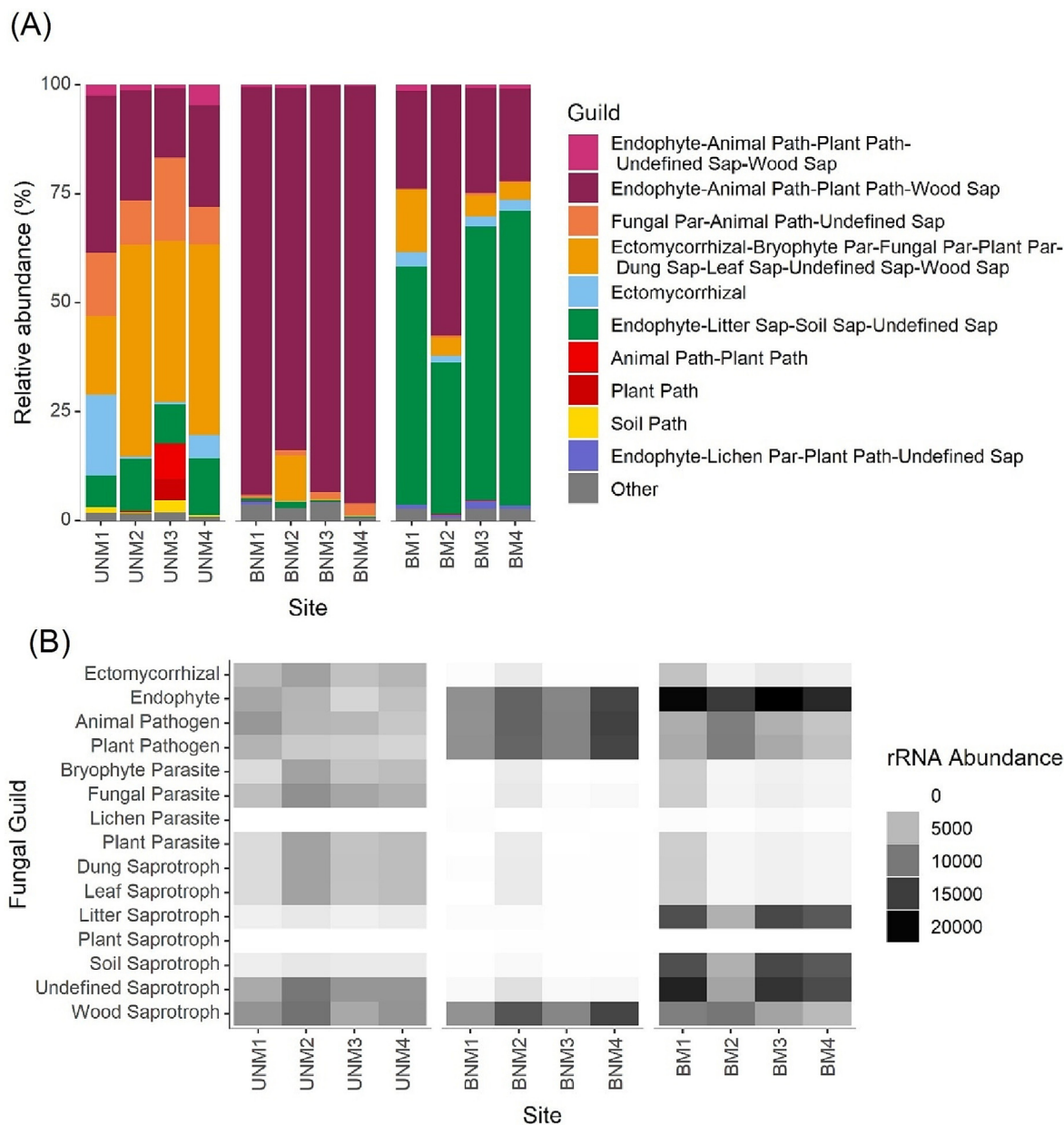
Fire is known to reduce microbial activity and biomass, as observed here despite the three-month difference in sampling of burnt and unburnt soil samples (Fig. 1a, b). Fire does so by causing microbe mortality in the topsoil (Bárceñas-Moreno and Díaz-Raviña, 2013). Fire is also capable of stimulating post-fire microbial activity through the addition of easily decomposed organic matter to the topsoil (Pietikäinen and Fritze, 1993; Poth et al., 1995). However, the high intensity of this crown fire and consequent complete combustion of aboveground and belowground biomass, including microbes, may explain why we did not observe post-fire stimulation of microbial activity six months post-fire. Interestingly, we observed that fire increased the microbial mineralisation quotient (qM), which indicates how efficiently microbes metabolize organic matter (Fig. 1d). Increased pH after fire has been reported to reduce acid stress on microbes and increase substrate bioavailability (Kemmitt et al., 2006).

Thus, the post-fire increase in pH observed here likely increased carbon mineralisation despite the decrease in soil TOC.

Contrastingly, management caused higher post-fire total organic carbon (TOC) and microbial biomass, i.e., substrate induced respiration (SIR), but not microbial activity, i.e., basal respiration (BR). This indicates less efficient metabolism of organic matter (qM) in BM sites compared to BNM sites (Fig. 1d). On average, BM sites were significantly more acidic compared to BNM sites (Ondik et al., 2022b), further indicating that acidic conditions inhibit carbon mineralisation. Furthermore, the raised SIR in pastures may be correlated with the observed reduction in C/N in BM plots (Ondik et al., 2022b), as soil respiration is known to be negatively correlated with the C/N of soils of Mediterranean climates (Lucas-Borja and Delgado-Baquerizo, 2019).

#### 4.3. Distribution of fungal guilds and bacterial respiration modes after fire and management

By using databases that describe unique functional traits of fungal and bacterial taxa, we were able to describe the link between community composition of bacteria and fungi and distribution of functional traits. Using



**Fig. 5.** Functional guilds of soil fungi. (A) Relative abundance plot of the total read count per unique concatenated value of all possible fungal guilds per genus. Par: Parasite; Path: pathogen; Sap: saprotroph; Other: functional guilds making up <1% of all fungal read counts. (B) Heatmap of the maximum possible read count per individual guild. Site descriptions, same as Fig. 4. Shown here are all fungal 18S reads that matched with functional guild entries at the genus level, and unmatched OTUs are not shown.

DNA sequencing and the FUNGuild fungal traits database, we observed that fire changed the kinds of saprotrophic, pathogenic, and parasitic fungi identified in soil samples (Table S3). Fire has been shown to reduce symbionts and increase saprotrophs in boreal coniferous forests (Ammitzboll et al., 2022), and Ondik et al. (2022b) reported that fire reduced the diversity of ectomycorrhizal fungi in Mediterranean dry sclerophyll woodland. Here we went beyond the work of Ondik et al. (2022b) and performed an in depth analysis of potential functions of fungal taxa at the genus level.

This analysis revealed that the high severity fire event reduced the RA of ectomycorrhizal (ECM) fungi, i.e., *Clavulina* and *Endogone*, and taxonomic groups that contain symbiotic species, i.e., Agaricales and Mortierella (Fig. 5; Table S3). Fire also caused a three-fold increase in the pathogen, endophyte, and wood saprotroph containing genus, *Penicillium* ( $12.92 \pm 1.51\%$  to  $36.6 \pm 3.91\%$ ) and an eight-fold increase in saprotrophic *Umbelopsis* ( $1.49 \pm 0.4\%$  to  $11.85 \pm 2.63\%$ ) in BNM sites compared to

UNM sites (Table S3). Saprotrophs, such as certain *Penicillium* spp. and *Umbelopsis*, provide benefits to soil by participating in post-fire nutrient cycling and organic matter decomposition (Orumaa et al., 2022). It has been shown that the RA of post-fire *Penicillium* is positively correlated with fire severity (Azaz and Pekel, 2002; Whitman et al., 2019), and post-fire *Penicillium* are often reported as saprotrophs associated with positive effects on vegetation (Reazin et al., 2016; Zhang et al., 2021). By resulting in a ten-fold decrease in the RA of Herpotrichiellaceae ( $7.21 \pm 1.1\%$  to  $0.69 \pm 0.22\%$ ) and a sixteen-fold decrease in the RA of Agaricales ( $19.75 \pm 4.44\%$  to  $1.17 \pm 1.04\%$ ) (Table S3), fire caused an overall decrease in parasitic load on fungi, plants, and bryophytes (Fig. 5). However, fire also caused the appearance of the plant pathogen, *Rhynchosporium* ( $1.29 \pm 0.73\%$ ) (Table S3). *Rhynchosporium*, which is primarily studied within the context of agriculture, is known to cause the disease, scald, in barley but can also infect native grasses (King et al., 2013). *Rhynchosporium* was absent from unburnt sites (Table S3), but its presence in BNM sites may



negatively impact post-fire recovery of native grasses. To our knowledge this is the first report of the relationship between *Rhynchosporium* abundance and fire, and more research is needed to understand why fire promoted *Rhynchosporium* in the soil.

Management caused post-fire dominant fungi to shift from wood saprotrophs to litter and soil saprotrophs (Fig. 5; Fig. S2), increased the total DNA reads of saprotrophs and potentially symbiotrophic fungal genera, and may have reduced the load of fungal plant pathogens. In BM compared to BNM sites, we identified a much lower RA of genera containing pathogenic species, e.g., *Penicillium*. Due to management, BM sites also had a much higher amount of genera containing only endophytes and soil, litter, and other saprotrophs (35.49 %), e.g., *Mortierella*, which are highly adaptable litter decomposing fungi (Dong et al., 2021; Zhang et al., 2021). Because the genus, *Mortierella*, is absent of pathogenic species, BM sites likely have a lower amount of plant pathogens compared to BNM sites, which contain a high RA of *Penicillium*. However, since many *Penicillium* spp. are wood saprotrophs, the reduced RA of *Penicillium* in BM sites is likely due to the lack of wood present in pastures.

In contrast to the impacts of fire, management resulted in elevated post-fire RA of identified taxonomic groups containing ECM fungi, e.g., Agaricales and *Endogone*, and endophytic fungi, e.g., *Mortierella*. Agaricales and *Mortierella* have been shown to be elevated in pastures (Praeg et al., 2020), suggesting that these fungal groups were also high in RA prior to the fire event. Fire has been shown to significantly decrease Agaricales RA in forests. For example, Meng et al. (2021) showed a high severity fire drove Agaricales from 15.6 %  $\pm$  13.0 % to 1.4 %  $\pm$  1.4 %, which is comparable to the observed change reported here (19.75 %  $\pm$  4.44 % to 1.17 %  $\pm$  1.04 %). *Mortierella* has also been shown to decrease after both high and low intensity burns in Ponderosa pine forest (Reazin et al., 2016). This is consistent with what was observed in the BNM sites.

Fire also decreased the RA of the plant symbiont, *Roseiarcus*, one of the dominant bacterial genera present in UNM sites (Table S2). Like ECM fungi, *Roseiarcus* is a symbiont, known to promote plant growth through fungal associations in the mycosphere (Yu et al., 2020). Fire-induced reduction of *Roseiarcus* is likely related to the reduction in ECM fungi and may negatively impact post-fire recovery of dependent plant species on Kangaroo Island. For example, Orchidaceae is a prevalent family of plants on Kangaroo Island (Stonor et al., 2023) that contains many species that are highly dependent on microbial associations throughout their life-cycle (Waterman et al., 2011). It is known that a large proportion of Orchidaceae species are under threat of extinction due to changing fire regimes (Wraith and Pickering, 2019), which could be related to our observation of this crown fire's ability to reduce plant symbionts in soils.

Management counteracted fire-induced changes to most dominant bacterial taxa (Table S2). Fire reduced the RA of symbiotic *Roseiarcus* (Yu et al., 2020), nitrogen fixing Xanthobacteraceae (Oren, 2014), and organic matter decomposing Xanthobacteraceae, *Acidipila*, Acetobacteraceae, and *Acidothormus* (Gołębiewski et al., 2019; Hiraishi, 2019; Oren, 2014; Šimonovičová et al., 2019). Fire also increased aromatic carbon degrading *Massilia* and *Streptomyces* (de Lima Brossi et al., 2014; Whitman et al., 2019) and plant growth promoting *Pseudarthrobacter* (Zhang et al., 2022). Contrastingly, management slightly increased the post-fire RA of *Roseiarcus* but significantly decreased the RA of *Pseudarthrobacter* (4.37  $\pm$  0.55 % to 0.02  $\pm$  0.01 %). Management also increased the post-fire RA of organic matter degrading Acetobacteraceae and Gemmataceae but decreased the RA of *Massilia*. These observations are consistent with Ondik et al. (2022b), which showed that historical clearing and grazing reduced fire severity and counteracted impacts of fire on soil properties.

Among 16S OTUs with matched respiration modes, the fire event also increased the RA of aerobic bacteria and reduced that of purely anaerobic and microaerophilic bacteria, while management had the opposite effect (Fig. 4). Aerobic respiration occurs in the presence of oxygen, microaerophilic respiration in microoxic conditions, and anaerobic respiration in the absence of oxygen. Here we show that fire caused a large increase in the RA of genera containing aerobic bacteria, indicating

that fire may have decreased the number of anoxic microsites present in the topsoil of non-managed sites. Leaf litter is known to create anaerobic pockets in the topsoil, particularly in the presence of bacteria undergoing high rates of aerobic respiration (Reith et al., 2002; Santiago and Geisseler, 2022). This evidence suggests that the combustion of leaf litter and organic matter and the consequent reduction in soil respiration we observed in BNM sites (Fig. 1) increased the diffusion rate and concentration of oxygen in the topsoil, thus favouring aerobic bacteria. Contrastingly, anaerobic bacteria made up slightly more of the bacterial communities of BM compared to BNM sites, but both fire and management caused an increase in the RA of mixed respiration modes (Fig. 4). Thus, diversification of respiration forms could be one way soil bacterial communities respond to the threat of disturbances, such as fire and anthropogenic activities.

## 5. Conclusion

Here we show that fire and management-induced changes to soil physicochemical properties indirectly impact soil microbial community composition, which in turn, affects microbial respiration and distribution of functions. Global increases in fire severity and extent threaten many above-ground vegetative species (Le Breton et al., 2022), and the impacts of high severity fire on soils identified here may have additive negative impacts. As shown above, the high severity fire event decreased soil microbial symbionts, which are important for post-fire establishment of plant groups, such as Orchidaceae, that depend on microbial associations throughout multiple life-stages. Furthermore, land conversion from woodland to pasture resulted in a complete transformation of the aboveground vegetation profile but also modified the post-fire substrate preference of the organic matter degrading microbes in the soil. The interactions between climate-driven extreme fires and anthropogenic land conversion in Mediterranean climate-type regions emphasizes the need for studies like this that explore the interactive effects of land conversion and wildfire on soils.

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## CRediT authorship contribution statement

Mercedes Ondik: Conceptualization, Methodology, Investigation, Software, Formal analysis, Data curation, Visualization, Writing – original draft, review & editing; Mark Ooi: Conceptualization, Methodology, Writing – review & editing, Funding acquisition, Supervision; Miriam Muñoz-Rojas: Conceptualization, Resources, Methodology, Writing – review & editing, Funding acquisition, Supervision.

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## Data availability

All data used in this paper has been made publicly available at URL links provided in the Materials and Methods section within the manuscript.

## Declaration of competing interest

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests: Mark Ooi reports financial support was provided by Australian Research Council. Miriam Munoz-Rojas reports financial support was provided by Australian Research Council.

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## References

- Ahlgren, I.F., 1974. The effect of fire on soil organisms. In: Kozłowski, T.T., Ahlgren, C.E. (Eds.), *Fire and Ecosystems*. Academic Press, pp. 47–72 <https://doi.org/10.1016/B978-0-12-424255-5.50008-0>.
- Ammitzbohl, H., Jordan, G.J., Baker, S.C., Freeman, J., Bissett, A., 2022. Contrasting successional responses of soil bacteria and fungi to post-logging burn severity. *For. Ecol. Manag.* 508, 120059. <https://doi.org/10.1016/j.foreco.2022.120059>.
- Australian Bureau of Agricultural and Resource Economics and Sciences, 2016. Land use of Australia 2010–11 summary statistics. <https://www.awe.gov.au/sites/default/files/abares/aclump/documents/2010-11%20nlum%20summary%20statistics.pdf> accessed 6 May 2021.
- Azaz, A.D., Pekel, O., 2002. Comparison of soil fungi flora in burnt and unburnt forest soils in the vicinity of Kargıcak (Alanya, Turkey). *Turk. J. Botany* 26, 409–416.
- Baldock, J.A., Sanderman, J., MacDonald, L.M., Puccini, A., Hawke, B., Szarvas, S., McGowan, J., 2013. Quantifying the allocation of soil organic carbon to biologically significant fractions. *Soil Res.* 51, 561–576. <https://doi.org/10.1071/SR12374>.
- Bárceñas-Moreno, G., Díaz-Raviña, M., 2013. Efectos del fuego sobre los microorganismos del suelo. *Red Temática Nacional Efectos de Los Incendios Forestales Sobre Los Suelos (FUEGORED)*. Ficha Técnica FGR2013/07.
- Bárceñas-Moreno, G., Bååth, E., Rousk, J., 2016. Functional implications of the pH-trait distribution of the microbial community in a re-inoculation experiment across a pH gradient. *Soil Biol. Biochem.* 93, 69–78. <https://doi.org/10.1016/j.soilbio.2015.10.024>.
- Bardgett, R.D., van der Putten, W.H., 2014. Belowground biodiversity and ecosystem functioning. *Nature* 515, 505–511. <https://doi.org/10.1038/nature13855>.
- Bateman, A.M., Muñoz-Rojas, M., 2019. To whom the burden of soil degradation and management concerns. *Adv. Chem. Pollut. Environ. Manag. Prot.* 4, 1–22. <https://doi.org/10.1016/bs.acpmp.2019.07.001>.
- Benjamini, Y., Hochberg, Y., 1995. Controlling the false discovery rate: a practical and powerful approach to multiple testing. *J. R. Stat. Soc. Ser. B* 57, 289–300. <https://doi.org/10.1111/j.2517-6161.1995.tb02031.x>.
- Borgogni, F., Lavecchia, A., Mastrodonato, G., Certini, G., Ceccherini, M.T., Pietramellara, G., 2019. Immediate- and short-term wildfire impact on soil microbial diversity and activity in a Mediterranean forest soil. *Soil Sci.* 184, 35–42. <https://doi.org/10.1097/SS.0000000000000250>.
- Bräuer, S., Harbison, A., Ueki, A., 2018. Micropepsaceae. *Bergey's Man. Syst. Archaea Bact.* 362, 1–5. <https://doi.org/10.1002/9781118960608.fbm00311>.
- Brooks, M., Kristensen, K., van Benthem, K., Magnusson, A., Berg, C., Nielsen, A., Skaug, H., Maechler, M., Bolker, B., 2017. glmmTMB balances speed and flexibility among packages for zero-inflated generalized linear mixed modeling. *R J.* 9, 378–400.
- Cameron, C., 2007. *MicroResp™ Technical Manual — A Versatile Soil Respiration System*. Campbell, C.D., Chapman, S.J., Cameron, C.M., Davidson, M.S., Potts, J.M., 2003. A rapid micro-plate method to measure carbon dioxide evolved from carbon substrate amendments so as to determine the physiological profiles of soil microbial communities by using whole soil. *Appl. Environ. Microbiol.* 69, 3593–3599. <https://doi.org/10.1128/AEM.69.6.3593-3599.2003>.
- Campbell, C.D., Cameron, C.M., Bastias, B.A., Chen, C., Cairney, J.W.G., 2008. Long term repeated burning in a wet sclerophyll forest reduces fungal and bacterial biomass and responses to carbon substrates. *Soil Biol. Biochem.* 40, 2246–2252. <https://doi.org/10.1016/j.soilbio.2008.04.020>.
- Cébron, A., Zeghal, E., Usseglio-Polatera, P., Meyer, A., Bauda, P., Lemmel, F., Leyval, C., Maunoury-Danger, F., 2021. BactoTraits — a functional trait database to evaluate how natural and man-induced changes influence the assembly of bacterial communities. *Ecol. Indic.* 130. <https://doi.org/10.1016/j.ecolind.2021.108047>.
- Chen, W.-C., Ko, C.-H., Su, Y.-S., Lai, W.-A., Shen, F.-T., 2021. Metabolic potential and community structure of bacteria in an organic tea plantation. *Appl. Soil Ecol.* 157, 103762. <https://doi.org/10.1016/j.apsoil.2020.103762>.
- Collins, L., Bradstock, R.A., Clarke, H., Clarke, M.F., Nolan, R.H., Penman, T.D., 2021. The 2019/2020 mega-fires exposed Australian ecosystems to an unprecedented extent of high-severity fire. *Environ. Res. Lett.* 16, 044029. <https://doi.org/10.1088/1748-9326/abeb9e>.
- Commonwealth Australia Bureau of Meteorology, 2022. Climate statistics for Australian locations: summary statistics CAPE BORDA. [http://www.bom.gov.au/climate/averages/tables/cw\\_022823.shtml](http://www.bom.gov.au/climate/averages/tables/cw_022823.shtml) accessed 12 January 2022.
- Creamer, R.E., Stone, D., Berry, P., Kuiper, I., 2016. Measuring respiration profiles of soil microbial communities across Europe using MicroResp™ method. *Appl. Soil Ecol.* 97, 36–43. <https://doi.org/10.1016/j.apsoil.2015.08.004>.
- D'Ascoli, R., Rutigliano, F.A., De Pascale, R.A., Gentile, A., De Santo, A.V., 2005. Functional diversity of the microbial community in Mediterranean maquis soils as affected by fire. *Int. J. Wildl. Fire* 14, 355–363. <https://doi.org/10.1071/WF05032>.
- Dedysh, S.N., Beletsky, A.V., Ivanova, A.A., Kulichevskaya, I.S., Suzina, N.E., Philippov, D.A., Rakin, A.L., Mardanov, A.V., Ravin, N.V., 2021. Wide distribution of Phycisphaera-like planctomycetes from WD2101 soil group in peatlands and genome analysis of the first cultivated representative. *Environ. Microbiol.* 23, 1510–1526. <https://doi.org/10.1111/1462-2920.15360>.
- Dekker, L.W., Doerr, S.H., Oostindie, K., Ziogas, A.K., Ritsema, C.J., 2001. Water repellency and critical soil water content in a dune sand. *Soil Sci. Soc. Am. J.* 65, 1667–1674. <https://doi.org/10.2136/sssaj2001.1667>.
- Department for Environment and Water, 2009a. Surface soil texture. [dataset]. NatureMaps [http://location.sa.gov.au/lms/Reports/ReportMetadata.aspx?pu=y&p\\_no=1066+&pa=dewnr](http://location.sa.gov.au/lms/Reports/ReportMetadata.aspx?pu=y&p_no=1066+&pa=dewnr). (Accessed 12 January 2022).
- Department for Environment and Water, 2009b. Soils (soil type) [dataset]. NatureMaps. [http://location.sa.gov.au/lms/Reports/ReportMetadata.aspx?pu=y&p\\_no=1009+&pa=dewnr](http://location.sa.gov.au/lms/Reports/ReportMetadata.aspx?pu=y&p_no=1009+&pa=dewnr) (accessed 21 May 2021).
- Department for Environment and Water, 2020. Burn year. [dataset]. NatureMaps [http://location.sa.gov.au/lms/Reports/ReportMetadata.aspx?pu=y&p\\_no=1268+&pa=dewnr](http://location.sa.gov.au/lms/Reports/ReportMetadata.aspx?pu=y&p_no=1268+&pa=dewnr) (accessed 6 May 2021). NatureMaps.
- DEWNR Soil and Land Program, n.d. Snelling land system report. <https://data.environment.sa.gov.au/Content/Land-System-reports/SNE.pdf> (accessed 1 January 2022).
- Dommergues, Y., 1960. La notion de coefficient de minéralisation du carbone dans les sols. *L'Agronomie Trop.* 15, 54–60.
- Dong, L., Li, J., Sun, J., Yang, C., 2021. Soil degradation influences soil bacterial and fungal community diversity in overgrazed alpine meadows of the Qinghai-Tibet Plateau. *Sci. Rep.* 11, 11538. <https://doi.org/10.1038/s41598-021-91182-7>.
- Francaviglia, R., Renzi, G., Ledda, L., Benedetti, A., 2017. Organic carbon pools and soil biological fertility are affected by land use intensity in Mediterranean ecosystems of Sardinia, Italy. *Sci. Total Environ.* 599–600, 789–796. <https://doi.org/10.1016/j.scitotenv.2017.05.021>.
- Francos, M., Pereira, P., Mataix-Solera, J., Arcenegui, V., Alcañiz, M., Úbeda, X., 2018. How clear-cutting affects fire severity and soil properties in a Mediterranean ecosystem. *J. Environ. Manag.* 206, 625–632. <https://doi.org/10.1016/j.jenvman.2017.11.011>.
- Gauquelin, T., Michon, G., Joffre, R., Duponnois, R., Génin, D., Fady, B., Bou Dagher-Kharrat, M., Derridj, A., Slimani, S., Badri, W., Alifriqui, M., Auclair, L., Simenel, R., Aderghal, M., Baudoin, E., Galiana, A., Prin, Y., Sanguin, H., Fernandez, C., Baldy, V., 2018. Mediterranean forests, land use and climate change: a social-ecological perspective. *Reg. Environ. Chang.* 18, 623–636. <https://doi.org/10.1007/s10113-016-0994-3>.
- Gołębiewski, M., Tarasek, A., Sikora, M., Deja-Sikora, E., Tretyn, A., Niklińska, M., 2019. Rapid microbial community changes during initial stages of pine litter decomposition. *Microb. Ecol.* 77, 56–75. <https://doi.org/10.1007/s00248-018-1209-x>.
- Griscom, B.W., Adams, J., Ellis, P.W., Houghton, R.A., Lomax, G., Miteva, D.A., Schlesinger, W.H., Shoch, D., Siikamäki, J.V., Smith, P., Woodbury, P., Zganjar, C., Blackman, A., Campari, J., Conant, R.T., Delgado, C., Elias, P., Gopalakrishna, T., Hamsik, M.R., Herrero, M., Kiesecker, J., Landis, E., Laestadius, L., Leavitt, S.M., Minnemeyer, S., Polasky, S., Potapov, P., Putz, F.E., Sanderman, J., Silvius, M., Wollenberg, E., Fargione, J., 2017. Natural climate solutions. *PNAS USA* 114, 11645–11650. <https://doi.org/10.1073/pnas.1710465114>.
- Hiraishi, A., 2019. Acidipila. *Bergey's Man. Syst. Archaea Bact.* 1–7. <https://doi.org/10.1002/9781118960608.gbm01666>.
- IRP, 2019. Land restoration for achieving the sustainable development goals: an international resource panel think piece. In: Herrick, J.E., Abrahamse, T., Abhilash, P.C., Ali, S.H., Alvarez-Torres, P., Barau, A.S., Branquinho, C., Chhatre, A., Chotte, J.L., Cowie, A.L. (Eds.), *United Nations Environment Programme, Nairobi, Kenya* <https://doi.org/10.18356/799094c6-en>.
- Kasel, S., Bennett, L.T., Tibbitts, J., 2008. Land use influences soil fungal community composition across central Victoria, south-eastern Australia. *Soil Biol. Biochem.* 40, 1724–1732. <https://doi.org/10.1016/j.soilbio.2008.02.011>.
- Kemmitt, S.J., Wright, D., Goulding, K.W.T., Jones, D.L., 2006. pH regulation of carbon and nitrogen dynamics in two agricultural soils. *Soil Biol. Biochem.* 38, 898–911. <https://doi.org/10.1016/j.soilbio.2005.08.006>.
- King, K.M., West, J.S., Brunner, P.C., Dyer, P.S., Fitt, B.D.L., 2013. Evolutionary relationships between *Rhynchosporium lolii* sp. nov. and other *Rhynchosporium* species on grasses. *PLoS One*, 8. <https://doi.org/10.1371/journal.pone.0072536>.
- Kneller, T., Harris, R.J., Bateman, A., Muñoz-Rojas, M., 2018. Native-plant amendments and topsoil addition enhance soil function in post-mining arid grasslands. *Sci. Total Environ.* 621, 744–752. <https://doi.org/10.1016/j.scitotenv.2017.11.219>.
- Le Breton, T.D., Lyons, M.B., Nolan, R.H., Penman, T., Williamson, G.J., Ooi, M.K.J., 2022. Megafire-induced interval squeeze threatens vegetation at landscape scales. *Front. Ecol. Environ.* 20, 327–334. <https://doi.org/10.1002/fee.2482>.
- de Lima Brossi, M.J., Mendes, L.W., Germano, M.G., Lima, A.B., Tsai, S.M., 2014. Assessment of bacterial *bph* gene in Amazonian dark earth and their adjacent soils. *PLoS One* 9, e99597. <https://doi.org/10.1371/journal.pone.0099597>.
- Lucas-Borja, M.E., Delgado-Baquerizo, M., 2019. Plant diversity and soil stoichiometry regulates changes in multifunctionality during pine temperate forest secondary succession. *Sci. Total Environ.* 697, 134204. <https://doi.org/10.1016/j.scitotenv.2019.134204>.
- Lucas-Borja, M.E., Delgado-Baquerizo, M., Muñoz-Rojas, M., Plaza-Álvarez, P.A., Gómez-Sánchez, M.E., González-Romero, J., Peña-Molina, E., Moya, D., de las Heras, J., 2021. Changes in ecosystem properties after post-fire management strategies in wildfire-affected Mediterranean forests. *J. Appl. Ecol.* 58, 836–846. <https://doi.org/10.1111/1365-2664.13819>.
- Machado de Lima, N., Muñoz-Rojas, M., Vázquez-Campos, X., Branco, L.H.Z., 2021. Biocrust cyanobacterial composition, diversity, and environmental drivers in two contrasting climatic regions in Brazil. *Geoderma* 386, 114914. <https://doi.org/10.1016/j.geoderma.2020.114914>.
- McIntosh, P.D., Laffan, M.D., Hewitt, A.E., 2005. The role of fire and nutrient loss in the genesis of the forest soils of Tasmania and southern New Zealand. *For. Ecol. Manag.* 220, 185–215. <https://doi.org/10.1016/j.foreco.2005.08.028>.

- Meng, M., Wang, B., Zhang, Q., Tian, Y., 2021. Driving force of soil microbial community structure in a burned area of Daxing'anling, China. *J. For. Res.* 32, 1723–1738. <https://doi.org/10.1007/s11676-020-01229-0>.
- Mingorance, M.D., Barahona, E., Fernández-Gálvez, J., 2007. Guidelines for improving organic carbon recovery by the wet oxidation method. *Chemosphere* 68, 409–413. <https://doi.org/10.1016/j.chemosphere.2007.01.020>.
- Miralles, I., Ortega, R., Almendros, G., Gil-Sotres, F., Trasar-Cepeda, C., Leirós, M.C., Soriano, M., 2012. Modifications of organic matter and enzymatic activities in response to change in soil use in semi-arid mountain ecosystems (southern Spain). *Eur. J. Soil Sci.* 63, 272–283. <https://doi.org/10.1111/j.1365-2389.2012.01426.x>.
- Moreira, F., Ascoli, D., Safford, H., Adams, M.A., Moreno, J.M., Pereira, J.M.C., Catty, F.X., Armezzo, J.G., Bond, W., González, M.E., Curt, T., Koutsias, N., McCaw, L., Price, O., Pausas, J.G., Rigolot, E., Stephens, S., Tavsanoglu, C., Vallejo, V.R., Van Wilgen, B.W., Xanthopoulos, G., Fernandes, P.M., 2020. Wildfire management in Mediterranean-type regions: paradigm change needed. *Environ. Res. Lett.* 15, 011001. <https://doi.org/10.1088/1748-9326/ab541e>.
- Moyano, F.E., Manzoni, S., Chenu, C., 2013. Responses of soil heterotrophic respiration to moisture availability: an exploration of processes and models. *Soil Biol. Biochem.* 59, 72–85. <https://doi.org/10.1016/j.soilbio.2013.01.002>.
- Muñoz-Rojas, M., Bárcenas-Moreno, G., 2019. *Microbiology*. In: Pereira, P., Mataix-Solera, J., Úbeda, X., Rein, G., Cerdá, A. (Eds.), *Fire Effects on Soil Properties*. CSIRO Publishing, Clayton South, Victoria, Australia, pp. 157–174.
- Muñoz-Rojas, M., Jordán, A., Zavala, L.M., De la Rosa, D., Abd-Elmabod, S.K., Anaya-Romero, M., 2015. Impact of land use and land cover changes on organic carbon stocks in Mediterranean soils (1956–2007). *L. Degrad. Dev.* 26, 168–179. <https://doi.org/10.1002/ldr.2194>.
- Muñoz-Rojas, M., Erickson, T.E., Martini, D., Dixon, K.W., Merritt, D.J., 2016. Soil physico-chemical and microbiological indicators of short, medium and long term post-fire recovery in semi-arid ecosystems. *Ecol. Indic.* 63, 14–22. <https://doi.org/10.1016/j.ecolind.2015.11.038>.
- Nguyen, N.H., Song, Z., Bates, S.T., Branco, S., Tedersoo, L., Menke, J., Schilling, J.S., Kennedy, P.G., 2016. FUNGuild: an open annotation tool for parsing fungal community datasets by ecological guild. *Fungal Ecol.* 20, 241–248. <https://doi.org/10.1016/j.funeco.2015.06.006>.
- NSW Department of Sustainable Natural Resources, n.d. Soil Survey Standard Test Method Available Water Capacity.
- Oksanen, J., Simpson, G., Blanchet, F., Kindt, R., Legendre, P., Minchin, P., O'Hara, R., Solyomos, P., Stevens, M., Szöcs, E., Wagner, H., Barbour, M., Bedward, M., Bolker, B., Borcard, D., Carvalho, G., Chirico, M., De Caceres, M., Durand, S., Evangelista, H., FitzJohn, R., Friendly, M., Furneaux, B., Hannigan, G., Hill, M., Lahti, L., McGlenn, D., Ouellette, M., Ribeiro Cunha, E., Smith, T., Stier, A., Ter Braak, C., Weedon, J., 2022. *vegan: Community Ecology Package*. R Package Version 2.6-2.
- Ondik, M.M., Bennell, M., Davies, R.J.-P., Ooi, M.K.J., Muñoz-Rojas, M., 2022a. Ondik-et-al-2022-Raw-physicochemical-data.csv. <https://doi.org/10.6084/m9.figshare.21277659.v1>.
- Ondik, M.M., Bennell, M., Davies, R.J., Ooi, M.K.J., Muñoz-Rojas, M., 2022b. Fire and land use impact soil properties in a Mediterranean dry sclerophyll woodland. *J. Environ. Manag.* 324, 116245. <https://doi.org/10.1016/j.jenvman.2022.116245>.
- Oren, A., 2014. The family Xanthobacteraceae. In: Rosenberg, E., DeLong, E.F., Lory, S., Stackebrandt, E., Thompson, F. (Eds.), *The Prokaryotes: Alphaproteobacteria and Betaproteobacteria*. Heidelberg, Berlin, pp. 709–726. <https://doi.org/10.1007/978-3-642-30197-1>.
- Orumaa, A., Agan, A., Anslan, S., Drenkhan, T., Drenkhan, R., Kauer, K., Köster, K., Tedersoo, L., Metslaid, M., 2022. Long-term effects of forest fires on fungal community and soil properties along a hemiboreal Scots pine forest fire chronosequence. *Sci. Total Environ.* 851, 158173. <https://doi.org/10.1016/j.scitotenv.2022.158173>.
- Peel, M.C., Finlayson, B.L., McMahon, T.A., 2007. Updated world map of the Köppen-Geiger climate classification. *Hydrol. Earth Syst. Sci.* 11, 1633–1644. <https://doi.org/10.5194/hess-11-1633-2007>.
- Pietikäinen, J., Fritze, H., 1993. Microbial biomass and activity in the humus layer following burning: short-term effects of two different fires. *Can. J. For. Res.* 23, 1275–1285. <https://doi.org/10.1139/x93-163>.
- Poth, M., Anderson, I.C., Miranda, H.S., Miranda, A.C., Riggan, P.J., 1995. The magnitude and persistence of soil NO<sub>x</sub>, N<sub>2</sub>O, CH<sub>4</sub>, and CO<sub>2</sub> fluxes from burned tropical savanna in Brazil. *Glob. Biogeochem. Cycles* 9, 503–513. <https://doi.org/10.1029/95GB02086>.
- Praeg, N., Seeber, J., Leitinger, G., Tasser, E., Newesely, C., Tappeiner, U., Illmer, P., 2020. The role of land management and elevation in shaping soil microbial communities: insights from the central European Alps. *Soil Biol. Biochem.* 150, 107951. <https://doi.org/10.1016/j.soilbio.2020.107951>.
- Quast, C., Pruesse, E., Yilmaz, P., Gerken, J., Schweer, T., Yarza, P., Peplies, J., Glöckner, F.O., 2013. The SILVA ribosomal RNA gene database project: improved data processing and web-based tools. *Nucl. Acids Res.* 41, D590–D596. <https://doi.org/10.1093/nar/gks1219>.
- R Core Team, 2021. R: a language and environment for statistical computing. R Foundation for Statistical Computing. URL <http://www.r-project.org>.
- Reazin, C., Morris, S., Smith, J.E., Cowan, A.D., Jumpponen, A., 2016. Fires of differing intensities rapidly select distinct soil fungal communities in a Northwest US ponderosa pine forest ecosystem. *For. Ecol. Manag.* 377, 118–127. <https://doi.org/10.1016/j.foreco.2016.07.002>.
- Reith, F., Drake, H.L., Küsel, K., 2002. Anaerobic activities of bacteria and fungi in moderately acidic conifer and deciduous leaf litter. *FEMS Microbiol. Ecol.* 41, 27–35. [https://doi.org/10.1016/S0168-6496\(02\)00236-2](https://doi.org/10.1016/S0168-6496(02)00236-2).
- Ritchie, A.L., Stevens, J.C., Erickson, T.E., 2020. Developing extruded seed pellets to overcome soil hydrophobicity and seedling emergence barriers. *Ecol. Solut. Evid.* 1, e12024. <https://doi.org/10.1002/2688-8319.12024>.
- Rodríguez, J., González-Pérez, J.A., Turmero, A., Hernández, M., Ball, A.S., González-Vila, F.J., Arias, M.E., 2017. Wildfire effects on the microbial activity and diversity in a Mediterranean forest soil. *Catena* 158, 82–88. <https://doi.org/10.1016/j.catena.2017.06.018>.
- Rousk, J., Brookes, P.C., Bååth, E., 2009. Contrasting soil pH effects on fungal and bacterial growth suggest functional redundancy in carbon mineralization. *Appl. Environ. Microbiol.* 75, 1589–1596. <https://doi.org/10.1128/AEM.02775-08>.
- Rutigliano, F.A., Migliorini, M., Maggi, O., D'Ascoli, R., Fanciulli, P.P., Persiani, A.M., 2013. Dynamics of fungi and fungivorous microarthropods in a Mediterranean maquis soil affected by experimental fire. *Eur. J. Soil Biol.* 56, 33–43. <https://doi.org/10.1016/j.ejsobi.2013.02.006>.
- Saccà, M.L., Caracciolo, A.B., Di Lenola, M., Grenni, P., 2017. Ecosystem services provided by soil microorganisms. In: Lukac, M., Grenni, P., Gamboni, M. (Eds.), *Soil Biological Communities and Ecosystem Resilience. Sustainability in Plant and Crop Protection*. Springer, Cham, pp. 9–24. [https://doi.org/10.1007/978-3-319-63336-7\\_2](https://doi.org/10.1007/978-3-319-63336-7_2).
- Sangha, K.K., Jalota, R.K., Midmore, D.J., 2006. Litter production, decomposition and nutrient release in cleared and uncleared pasture systems of central Queensland, Australia. *J. Trop. Ecol.* 22, 177–189. <https://doi.org/10.1017/S0266467405003020>.
- Santiago, S., Geisseler, D., 2022. Effects of moisture contents in incorporated residues and soil on net nitrogen mineralization in a laboratory study. *Agrosyst. Geosci. Environ.* 5, e20268. <https://doi.org/10.1002/agg2.20268>.
- Santorufu, L., Memoli, V., Panico, S.C., Santini, G., Barile, R., Giarra, A., Di Natale, G., Trifuoggi, M., De Marco, A., Maisto, G., 2021. Combined effects of wildfire and vegetation cover type on volcanic soil (functions and properties) in a mediterranean region: comparison of two soil quality indices. *Int. J. Environ. Res. Public Health* 18, 5926. <https://doi.org/10.3390/ijerph18115926>.
- Shen, C., Xiong, J., Zhang, H., Feng, Y., Lin, X., Li, X., Liang, W., Chu, H., 2013. Soil pH drives the spatial distribution of bacterial communities along elevation on Changbai Mountain. *Soil Biol. Biochem.* 57, 204–211. <https://doi.org/10.1016/j.soilbio.2012.07.013>.
- Šimonovičová, A., Kraková, L., Piecková, E., Planý, M., Globanová, M., Paudišová, E., Šoltys, K., Budiš, J., Szemes, T., Gáfríková, J., Pangallo, D., 2019. Soil microbiota of dystic cambisol in the High Tatra Mountains (Slovakia) after windthrow. *Sustainability* 11, 6851. <https://doi.org/10.3390/sul1236851>.
- Staal, A., Flores, B.M., Aguiar, A.P.D., Bosmans, J.H.C., Fetzer, I., Tuinenburg, O.A., 2020. Feedback between drought and deforestation in the Amazon. *Environ. Res. Lett.* 15, 044024. <https://doi.org/10.1088/1748-9326/ab738e>.
- Stonor, M.B., Lapeyre, F., Petit, S., 2023. Orchids of Kangaroo Island. Kangaroo Island Research Station. [WWW document]. URL <https://www.kiresearchstation.org/orchids>.
- Tian, J., Wang, H., Vilgaly, R., Ho, M., Flanagan, N., Richardson, C.J., 2021. Response of fungal communities to fire in a subtropical peatland. *Plant Soil* 466, 525–543. <https://doi.org/10.1007/s1104-021-05070-0>.
- Vaulot, D., del Campo, J., Mahé, F., Morard, R., Sandin, M.M., Schönlé, A., Fiore-Donno, A.M., 2021. PR<sup>2</sup> Version 4.14.0. <https://doi.org/10.5281/zenodo.5031733>.
- Vu, V.Q., 2011. *ggbiplot: A ggplot2 Based Biplot*. R Package Version 0.55.
- Wang, Q., Liu, K., Tao, K., Hou, T., 2023. Biogeographical patterns and drivers of bacterial community in the Qinghai-Tibetan Plateau. *Appl. Soil Ecol.* 183, 104757. <https://doi.org/10.2139/ssrn.4171521>.
- Waterman, R.J., Bidartondo, M.I., Stofberg, J., Combs, J.K., Gebauer, G., Savolainen, V., Barraclough, T.G., Pauw, A., 2011. The effects of above- and belowground mutualisms on orchid speciation and coexistence. *Am. Nat.* 177, E54–E68. <https://doi.org/10.1086/657955>.
- Whitman, T., Whitman, E., Woollet, J., Flannigan, M.D., Thompson, D.K., Parisien, M.A., 2019. Soil bacterial and fungal response to wildfires in the Canadian boreal forest across a burn severity gradient. *Soil Biol. Biochem.* 138, 107571. <https://doi.org/10.1016/j.soilbio.2019.107571>.
- Wraith, J., Pickering, C., 2019. A continental scale analysis of threats to orchids. *Biol. Conserv.* 234, 7–17. <https://doi.org/10.1016/j.biocon.2019.03.015>.
- Wu, L., Yang, B., Li, M., Chen, J., Xiao, Z., Wu, H., Tong, Q., Luo, X., Lin, W., 2020. Modification of rhizosphere bacterial community structure and functional potentials to control *Pseudostellaria heterophylla* replant disease. *Plant Dis.* 104, 25–34. <https://doi.org/10.1094/PDIS-04-19-0833-RE>.
- Xiang, X., Shi, Y., Yang, J., Kong, J., Lin, X., Zhang, H., Zeng, J., Chu, H., 2014. Rapid recovery of soil bacterial communities after wildfire in a Chinese boreal forest. *Sci. Rep.* 4, 3829. <https://doi.org/10.1038/srep03829>.
- Yu, F., Liang, J.F., Song, J., Wang, S.K., Lu, J.K., 2020. Bacterial community selection of *Russula griseocarnosa* mycosphere soil. *Front. Microbiol.* 11, 347. <https://doi.org/10.3389/fmicb.2020.00347>.
- Zhang, M., Riaz, M., Xia, H., Li, Y., Wang, X., Jiang, C., 2022. Four-year biochar study: positive response of acidic soil microenvironment and citrus growth to biochar under potassium deficiency conditions. *Sci. Total Environ.* 813, 152515. <https://doi.org/10.1016/j.scitotenv.2021.152515>.
- Zhang, Y., Cao, H., Zhao, P., Wei, X., Ding, G., Gao, G., Shi, M., 2021. Vegetation restoration alters fungal community composition and functional groups in a desert ecosystem. *Front. Environ. Sci.* 9, 589068. <https://doi.org/10.3389/fenvs.2021.589068>.
- Zhou, X.M., Ranathunge, K., Cambridge, M.L., Dixon, K.W., Hayes, P.E., Nikolic, M., Shen, Q., Zhong, H., Lambers, H., 2022. A cool spot in a biodiversity hotspot: why do tall *Eucalyptus* forests in Southwest Australia exhibit low diversity? *Plant Soil* 476, 669–688. <https://doi.org/10.1007/s1104-022-05559-2>.