Chapter 2

Microbial biodiversity in Mediterranean soils, a challenge in a changing world

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Introduction

Soils represent the most biologically diverse and important ecosystem on the planet (Roger-Estrade et al. 2010). Most of the biodiversity of ecosystems is found in the soil (Young and Crawford, 2004), about one gram of soil may typically contain one billion bacterial cells, that corresponds to about ten thousand different bacterial genomes, up to one million individual fungi, about one million cells of protists, and several hundred of nematodes (EU, 2010a). In this sense, soils are a key reservoir of global biodiversity, yet little is known about them as only 1% of soil microorganism have been identified (FAO et al. 2020) compared to 80% of plants (Jeffery et al. 2010). Soils are remarkably complex and dynamic environments and hence typically comprise a wide range of habitat types for organisms over a range of dimensions from micrometre to the landscape scale. The highly heterogeneous nature of the soil, particularly at the microhabitat level, is responsible for the considerable biodiversity (Jeffery et al. 2010).

The functions performed by soil biota have considerable direct and indirect effects on crop growth and quality, nutrient cycle, quality, and soil sustainability. Moreover, the biodiversity of soil is vital as it is the engine driving soil-based ecosystem services such as carbon sequestration, soil formation, decontamination and bioremediation of pollutants, control of pest outbreaks, and water purification (Dominati et al. 2010, Turbé et al. 2010). Soils with higher biodiversity express more resistance and resilience to perturbations, thus a loss in biodiversity can lead to lower resistance to a perturbation and reduced capacity to recover, affecting the ability of soil to function normally (Brussaard et al. 2007, Allison and Martiny, 2008).

The major threats for soil biodiversity loss include deforestation, urbanization, agricultural intensification, soil organic matter decline, soil compaction, surface sealing, soil acidification, nutrient imbalance, contamination, salinization, sodification, land degradation, fire, erosion, and landslides (FAO et al. 2020). Those threats are of major relevance under Mediterranean conditions based on a strongly seasonal climate with scarce and irregular rainfall and frequent drought periods, which can trigger these processes accelerating the loss of biodiversity.

There is not a specific European regulation or legislation about soil biodiversity conservation, but the European Commission acknowledged the importance of soil biodiversity in the role of ecosystem functioning, stating that "these functions are worthy of protection because their socio-economic as well environmental importance" (Stone et al. 2016). Moreover, "biodiversity pool, such as habitats, species and genes" soil functions are collected in the SDG number 15 of the UN "Sustainable Develop Goals (SDGs)" for the period 2015-2030, by relating the topics "ensure healthy lives and promote well-being for all at all ages" and protect, restore, and promote sustainable use of terrestrial ecosystem, sustainable manage forest, combat desertification, and halt and reverse land degradation and halt biodiversity loss (Keesstra et al. 2016).

The mediterranean characteristics

The Mediterranean-type climate regions are distributed over five continents: Africa, Australia, Europe, North America, and South America. The most representative is the Mediterranean basin, extended over 3800 km from east to west in south Europe, and whose environmental and climatic special factors contribute to exposing soils to adverse processes that make them very vulnerable and condition the microbial biodiversity. The long periods of droughts interrupted by heavy, occasionally torrential rainfall (200-700 mm), together with high annual mean temperatures (16º-19ºC) and high radiation (EU, 2010b), increases rates of erosion in soils prone to degradation.

Soil degradation would be exacerbated in the most vulnerable Mediterranean ecosystems according to the climatic predictions, the arid and semi-arid areas, which especially represented in SE Spain. In those regions, the most widely represented lithological substrates are carbonate rocks, quaternary sediments, and loams, altogether materials easily eroded. Moreover, agriculture has been practised in the semi-arid Mediterranean for ca. 10 000 years, thus centuries of ploughing, burning, and grazing have resulted in the clearing of natural potential vegetation. This combination results in large areas of low-quality soils with little plant cover. Furthermore, the low organic matter content of large extensions, a key factor describing the Mediterranean soils, determines the quality and fertility of soils.

The combination of all these factors implies Mediterranean soils are especially sensitive and vulnerable to soil erosion, and little changes could trigger several degradation processes that ultimately affect soil biodiversity. In this sense, keep the soil biodiversity in Mediterranean soil is a real challenge in a scenario of climate change, fire-recurrence increment, and land-use intensification in the Mediterranean basin.

Millennian Mediterranean agriculture

The Mediterranean basin has a millenary history of intensive and extensive land use, many cultures have evolved in the area generating socioeconomic and land-use changes. Agriculture has been practised for more than one millennium, transforming large extensions of landscapes in terraced slopes for agricultural purpose and natural vegetation to crops lands (Pausas et al. 2009).

Agricultural management influences soil microorganisms and soil microbial processes through changes in the quantity and quality of plant residues entering the soil, altering their spatial distribution and through physical changes (Christensen, 1996). It has been reported intensive arable farming causes a progressive decline in soil organic matter levels (Caravaca et al. 2002), which affects physical, chemical, biochemical and biological soil properties. Intensively used agricultural lands treated with soil tillage often receive high levels of mineral fertilizers and pesticides. The excessive use of herbicides can modify drastically the function and structure of soil microbial communities, thus altering the normal functioning of terrestrial ecosystems, which in turn has important implications for soil fertility and quality (Pampulha and Oliveira, 2006). Since many of the agricultural practices commonly used provoke negative impacts on microorganisms and their processes, the application of a conservation management system tries to minimize soil disturbance.

Agricultural land management is one of the most significant anthropogenic activities that greatly alter soil characteristics, including physical, chemical, and biological properties (Jangid et al. 2008, García-Orenes et al. 2010). This fact is particularly relevant in Mediterranean environments, where unsuitable land management along with climatic constraints (scarce and irregular rainfall and frequent drought periods) can contribute to increased rates of erosion and other soil degradation processes in agricultural land (Caravaca et al. 2002). These conditions can lead to a loss in soil fertility and a reduction in the abundance of biodiversity in soils. This is of especial importance in Europe, where more than 45% of the land is for agricultural production (EUROSTAT, 2019).

Differences in agricultural production systems, such as integrated, organic, or conventional, have also reported affecting the soil biota, both the overall biomass as well as biodiversity. Indeed, soils subjected to disturbance by tillage can be more susceptible to reductions in soil biodiversity due to desiccation, mechanical destruction, soil compaction, reduced pore volume, and disruption of access to food resources (Giller, 1996). Some organic fertilisers, such as manure and sewage sludge, promote the activities of soil microbial communities (Enwall et al. 2007); however, repeated application of manures may pose environmental hazards, as they introduce faecal microbial flora into the soil, and have the potential to alter the endogenous microbial structure (Soupir et al. 2006). In many cases, the agricultural activity implicates the use of water for irrigation without enough quality that contains a high amount of salt and organic pollutants, which ultimately has an important effect on soil biodiversity (Friedel et al. 2000, Mangkoedihardjo, 2006). A major threat is salinization, the accumulation of soluble salts of sodium-calcium, potassium and magnesium in soils causing deterioration or loss of one or more soil functions. Salinization of soils occurs either as a result of natural processes or as a consequence of mismanagement of irrigation practices or poor drainage conditions.

On the other hand, soil management implicates in many cases the loss of organic matter, especially relevant in Mediterranean soils that are submitted to semi-arid conditions (Novara et al. 2011, Laudicina et al. 2015). A reduction in soil organic matter is generally associated with a lower soil organism abundance and diversity. Soil biodiversity is intimately bound to soil organic matter content (García-Orenes et al. 2013), each type of soil organism occupies a different niche within the food web of life and favours a different substrate and nutrient source. Consequently, a large, varied source of organic matter will generally support a wider variety of organisms due to the greater range of substrates and nutrients content, for example, functional microorganisms related to N cycling. The use of various types of organic amendments in agricultural soils can lead to an improved potential for N capture and release and therefore contribute to the conservation of soil biodiversity in agricultural land (Pereg et al. 2018a). Organic agriculture management promotes soil structure and fertility as well as increases water infiltration and storage, of major importance in the degraded Mediterranean soils.

The Mediterranean wildfire

Forest fires are a common natural disturbance in the Mediterranean ecosystems, the marked seasonality with long droughts periods and frequent temperatures above 30ºC provides ideal conditions for fuel accumulation and the ignition and fire spread. However, anthropogenic impacts in the last decades in the Mediterranean basin such as changes in land use, agricultural abandonment, and fire suppression policies, have led to an increase in fire severity regimens aggravated in a context of climate change (Pausas and Keeley, 2009). Wildfires have become one of the most critical threats to the Mediterranean forests in Spain, in which the predicted climate change scenario, with expected rainfall decrease and drought risk increase and consequently forest wildfires, make them especially vulnerable. The loss of organic matter through fire and the diminution of vegetation cover contribute to increasing rates of erosion and other soil degradation processes, resulting in the loss of soil ecosystem functionality.

Fire effects on soil biodiversity can vary widely, depending on fire severity, the changes in some soil properties and post-fire environmental conditions. Each of these factors has a range of effects depending on the type of soil organism being considered (Certini et al. 2021). Despite the protective mechanisms, fire can lead to major shifts in the fire-adapted Mediterranean ecosystems. Fire can affect soil microbes directly through heating and indirectly by modifying soil properties. Microbes will also be affected by post-fire environmental factors and the re-establishment of vegetation. The most important factor affecting soil biodiversity seems to be the burn severity, which is controlled by such factors as fire intensity, duration, and soil properties, which normally causes a decrease in the numbers of microbes. The temperatures reached in the topsoil are often sufficient to affect soil microorganisms and other soil properties related to the post-fire microbial recolonization. In extreme cases, the topsoil can undergo complete sterilization. Fungi seem to be more sensitive to heat than bacteria, and a higher impact under wet soil conditions has been reported. In the case of fungi that form arbuscular mycorrhizas, almost all the studies show a negative influence resulting in a reduced number of propagules (Mataix-Solera et al. 2009).

The current level of knowledge sets that although burning strongly reduces belowground biomass and its functioning, it may recover within a few years after burning. However, previous and subsequent forest management practices to the wildfire are of major importance affecting fire impacts in soils and soil microorganisms (Lucas-Borja et al. 2019, Mediavilla et al. 2020). Especially, post-fire management on burned soils can be a key to promote soil biodiversity recovery or, on the other hand, a threat that hinders the restoration of the belowground functions. Several studies have reported salvage logging, the most common post-fire strategy carried out in fire-affected areas in Europe performed to recover timber values, could hamper the soil biodiversity recovery via soil compaction (Hartmann et al. 2014, García-Orenes et al. 2017, Pereg et al. 2018b). However, the effects of post-fire management on microbial community composition and structure and its consequences for ecosystem recovery remain partially unknown, being these of especial importance in Mediterranean forest where soils are prone to degradation. Moreover, the functional resilience and sustainability in fire-prone ecosystems are threatened by the increasing predictions of fire danger and burned areas, due mainly to global warming, hence in need to be addressed (FAO et al. 2020).

Examples of research work carried out in the Mediterranean area related to soil biodiversity

Since the late 1980s, soil biological parameters have been assessed in an increasing number of studies (Pulleman et al. 2012). The study of soil microbial structure, function, and biodiversity has been considered essential to understand the soil ecosystem and keep its quality and fertility in the last decade. In this sense, several experiments developed in the Mediterranean area under semiarid conditions have been selected as examples of the effect of different human activities on soil biodiversity.

Agricultural management effect on soil microbial properties

The different agricultural practices can produce important changes in microbial structure, functionality, and biodiversity. In general, sustainable agriculture practices enhance soil microbial biodiversity in contraposition to conventional practices. As an example, an experiment developed on erosion plots in an agricultural area of the Spanish Mediterranean (38º50´N; 0º42´W) discerned the effect of five different treatments on soil microbial structure (Garcia-Oerenes et al. 2013). The treatments tested were based on some of the most common practices used by farmers in the east of Spain, as residual herbicides, tillage, tillage with oats and oats straw mulching, practices which were evaluated against an abandoned land after farming and an adjacent long term wild forest coverage. The results showed a substantial level of differentiation in the microbial community structure in terms of management practices (*Figure 1*), which were highly correlated with the soil organic matter content. The application of oat straw to soil increased the organic carbon content and, as a result, a significant increase of the microbial biomass was registered. The total PLFAs were highly correlated with the microbial biomass carbon, determined by the fumigation-extraction method. The correlation obtained in this research between these parameters (r= 0.80) was similar to the reported in previous works (Zornoza et al. 2009, Hackl et al. 2005). These results corroborate the notion the microbial community structure is a good indicator of soil quality, perturbations, and the effects of different management practices (Zornoza et al. 2009) since the microorganisms respond against changes in soil management more rapidly than chemical or physical soil properties.

In the same area, a field experiment in a vineyard was performed after 10 years to assess the medium-term effect of mineral fertilizer and two organic fertilization systems with different nitrogen sources on the soil microbial community biomass, structure, and composition (phospholipid fatty acids, pattern, and abundance), and microbial activity (basal respiration, dehydrogenase, protease, urease, β-glucosidase, and phosphomonoesterase activity) (*Figure 2*) (García-Orenes et al. 2016). The three fertilization systems assayed were: inorganic fertilization, the addition of grapevine pruning with sheep manure (OPM), and addition of grapevine pruning with a legume cover crop (OPL). The organic fertilization systems increased microbial biomass, shifted the structure and composition of the soil microbial community, and stimulated microbial activity, compared with inorganic fertilization (García-Orenes et al. 2013, Zhang et al. 2005). The abundances of fungi and Gram+ bacteria were increased by treatments OPM and OPL. The total replacement of inorganic by organic fertilization in a semiarid agroecosystem had a significant medium-term effect on the biomass, composition, and function of the microbial community, which may be attributed to the organic carbonaceous substrates and nutrients introduced into the soil. In particular, the addition of grapevine pruning combined with sheep manure, or a legume cover crop promoted the proliferation of fungal and Gram+ bacterial populations in the soil microbial community. Shifts in soil microbial populations and microbial processes related to nutrient cycling promoted by the use of organic fertilizers improved soil fertility, maintaining crop yields at levels similar to those of the inorganic fertilization system. The effectiveness of the organic fertilization systems, for promoting the sustainability and soil biological and chemical fertility of an agroecosystem under semiarid conditions, was dependent on the organic N source.

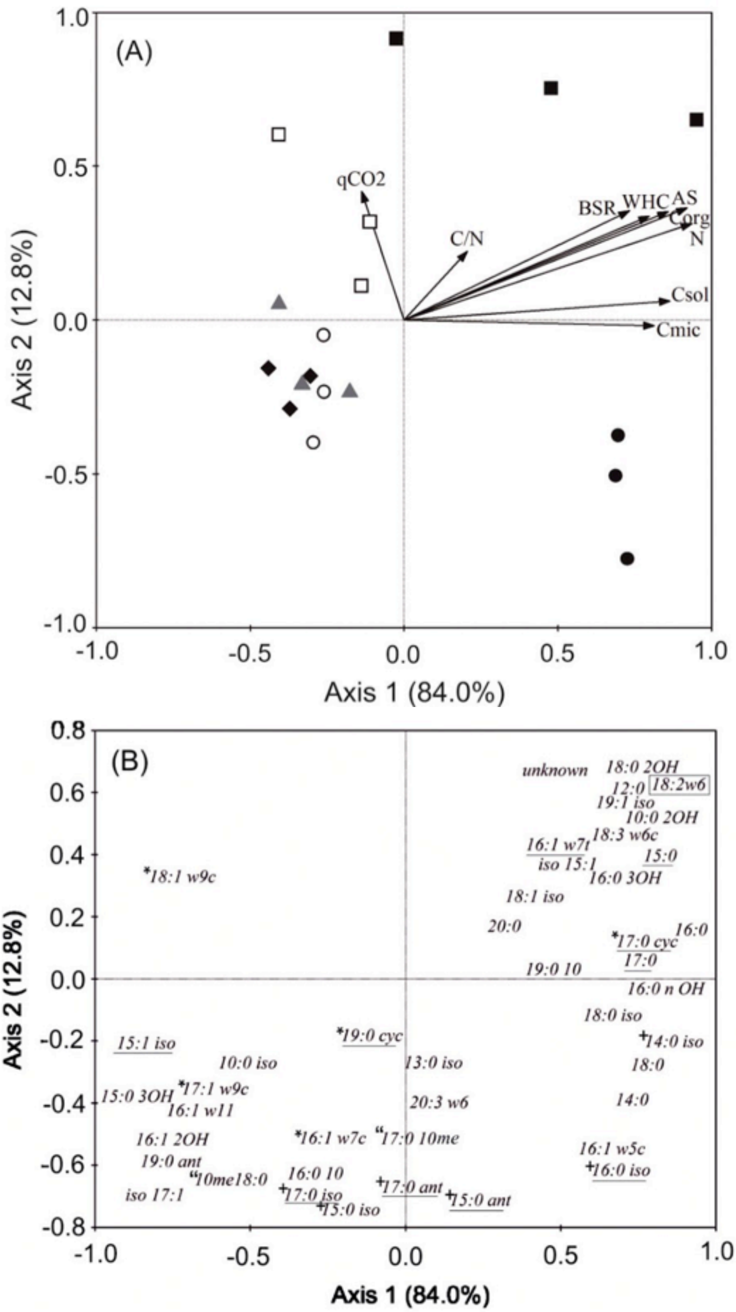

Figure 1. Samples and soil characteristics biplots (A) and loadings plots (B) from RDA performed on the relative concentration of PLFAs in all management practices: residual herbicide (♦), tillage (▲), oats + tillage (○), oats straw (●), land abandonment (□) and wild forest coverage (■). PLFAs used for microbial groups designation are marked as: underlined (bacteria), framed (fungi), * (Gbacteria), + (G+ bacteria) and (actinobacteria). Corg: soil organic carbon; N: total nitrogen; Csol: soluble carbon; WHC: water holding capacity; AS: aggregate stability; Cmic: microbial biomass C; BSR: basal soil respiration; qCO2: BSR/ Cmic. (García-Orenes et al. 2013).

Figure 2. Vineyard crop with organic fertilization situated on Mediterranean area under semiarid conditions (39º49´24"N; 0º48´17"W).

In this agricultural area, the effect of these treatments on the nitrogen cycling microbial community of soil was investigated by quantification of genes involved in key pathways, in particular nitrogen fixation, denitrification and nitrification (Pereg et al. 2018a). The assimilation of inorganic N by soil microorganisms is critical for N retention in soil, thus reducing the loss of N fertilizer to the environment (Tahovská et al. 2013), particularly important in degraded agricultural lands. In addition, N is often applied in forms that are less efficient or unavailable for uptake by plants, such as urea (Witte, 2011) or organically bound N, making microbial N cycling a key process required for N transformation in soils (He et al. 2010). Nitrification and denitrification are major pathways in the soil N cycle, involving ammonia oxidation and nitrate and nitrite reduction to N_2O and N_2 (reviewed by Teixeira et al. 2012), respectively. Elucidating the impact of N application practices on microbial diversity and community structure in general, and on N cyclers in particular, is pivotal to identify agricultural practices that enhance the soil potential for microbial immobilization and transformation of fertilizer N. The results of the study showed the abundances of bacterial *nif*H, *nos*Z, *nir*S and *nir*K genes, these were significantly increased under a decade of organic fertilization when compared to inorganic fertilization (*Figure 3*), linked to an increase in soil organic carbon. The abundance of *nif*H was lower where fertilizers rich in ammonia and nitrate were used, and its increase under organic fertilization was more related to the availability of organic carbon than to the nature of the organic amendment. Our results indicate that soil microbial communities involved in biological nitrogen cycling, in particular nitrogen fixation and denitrification, are more abundant under management practices that include organic fertilization compared to traditional agricultural practices. A decade of organic amendments using grapevine pruning, combined with sheep manure or legume cover crop, resulted in an enhanced grapevine soil potential for N fixation and denitrification, two microbial processes that are essential for input and output of N into the biosphere. The raise in diazotrophic and denitrifying communities, shown by increased *nif*H, *nir*S, *nir*K and *nos*Z gene copy numbers under organic management, comes hand in hand with a significant improvement in biological and physical soil properties. It may therefore be recommended that organic fertilization should be utilized in agricultural systems to assist with sustaining healthy soils.

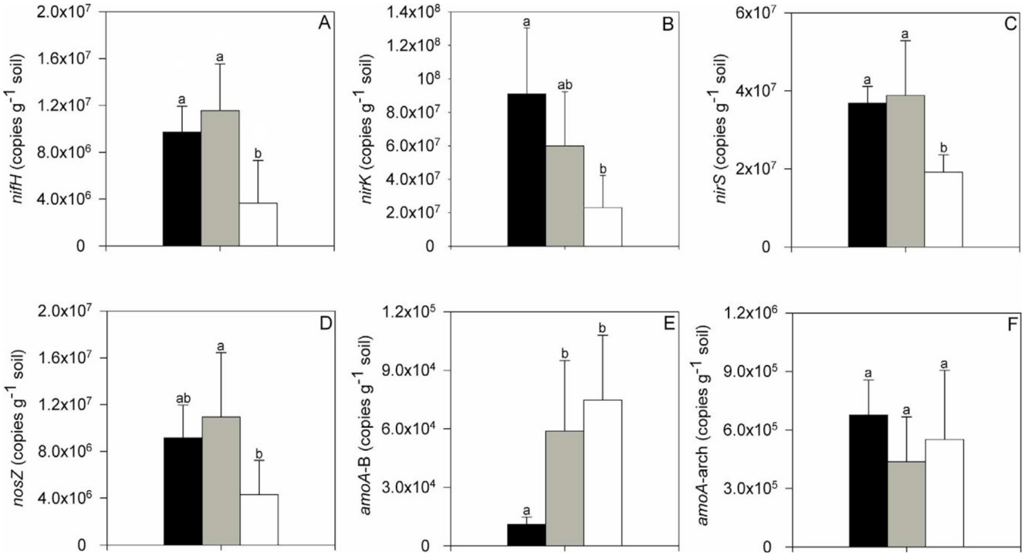

Figure 3. Mean values (±standard deviation) of nitrogen cycle genes in grapevine soil. Different letters indicate significant differences (one-way ANOVA, $P < 0.05$) between treatments. Bars: white (IF), black (OPM) and grey (OPL). (Pereg et al. 2018).

Water shortage is a major problem faced by agricultural industries in the Mediterranean area. With currently available water levels below 300 mm per year, this situation is progressively worsening as a result of climate change (IPCC, 2013). The scarcity of water is problematic in many areas of southern Spain, where groundwater is used for irrigation in 27% of the irrigated agricultural soils and the rest of irrigation is provided by surface water (Lidón et al. 2013). In southern Spain, there is 158,859 ha of citrus orchards, representing 53.5% of the total citrus orchards in Spain, of which about 70,162 ha are sweet orange. The most common irrigation system in sweet orange orchards in this region is drip irrigation (64% is drip irrigation versus 36% flood irrigation) (MAPAMA, 2017).

As a necessary issue to address, it was evaluated how different soil moisture levels, dependent on distance from drip irrigation points, impact the biological, properties of citrus soil under organic and inorganic fertilization, soil microbial community structure (phospholipid fatty acid assay), and abundance of microbial nitrogen cyclers (by quantitative PCR). A field experiment was performed in an orange orchard (Citrus sinensis) (Morugán-Coronado et al. 2019) in southeast Spain. 30 and 18 soil samples were taken from each plot to compare the impacts of soil moisture: near (wet, w) or away (dry, d) from drip-irrigation points, in plots with inorganic fertilizers under intensive ploughing (PI) or organic fertilization (OA) (*Figure 4*).

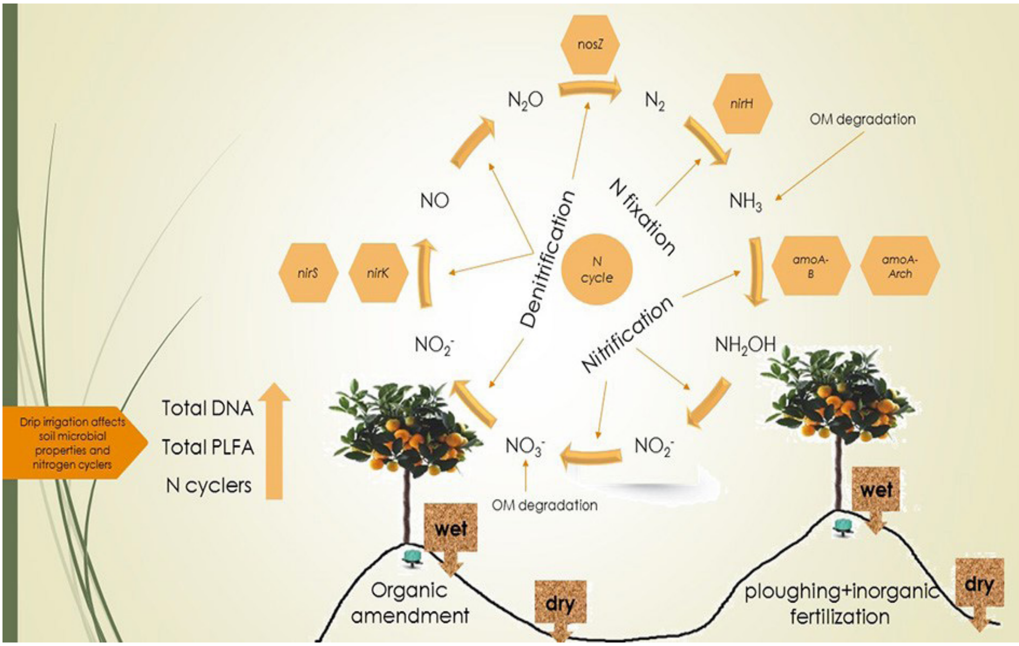

Figure 4. Diagramming of the experiment to evaluate the effect of moisture on soil microbial properties and nitrogen cyclers in Mediterranean sweet orange orchards under organic and inorganic fertilization. (Morugán-Coronado et al. 2019).

The results showed that changes in microbial properties and soil microbial indexes were strongly associated with soil moisture content under both organic and inorganic fertilization, and with organic carbon content. Soil moisture influenced bacterial and fungal load (PLFAs) (*Table 1*), and the abundances of bacterial N cycling genes, including nifH (nitrogen fixation) *nir*S/K and *nos*Z genes (denitrification) and amoA-B (bacterial nitrification) (*Table 2*). The potential for N fixation and denitrification, two microbial processes that are crucial for determining the amount of N in the soil, were improved by increased soil moisture in the proximity of the drip irrigation. Soil organic carbon and total N, which are usually higher under organic fertilization than under inorganic fertilization, were also highly correlated with the abundance of the N cycling genes. By controlling irrigation doses and applying organic amendments, it may be possible to increase the microbial abundance and function in soil, thus supporting greater fertility of soils.

	OAd	OAw	Pld	Plw
Total PLFA (nmol g^{-1})	71.1±2.0^{a}	260±4.5^{b}	22.5±0.1^{c}	29.2±2.1^{d}
Gram+ PLFA (nmol g^{-1})	12.2±2.3^{a}	26.6±1.7^{b}	bdl	0.4±0.1^{d}
Gram- PLFA (nmol g^{-1})	31.9±2.90^{a}	166.8±2.3^{b}	3.60±0.3^{c}	15.0±2.1^{d}
Fungi PLFA (nmol g^{-1})	25.5±0.9^{a}	55.0±2.4^{b}	18.8±0.4^{c}	13.4±0.5^{d}
Actinobacteria PLFA (nmol g^{-1})	1.4±0.4^{a}	11.7±2.4^{b}	bdl	0.4±0.1^{d}
Fungi/bacteria	0.6±0.1^{a}	0.3±0.1^{b}	5.2±0.6^{c}	0.9±0.1^{d}

Values are mean ± standard deviation (n=36)
A one-way ANOVA (P<0.05) was used to compare differences between treatments. Values in rows sharing the same letters do not differ significantly
bdl: below detected levels (<0.1 nmol g^{-1})

Table 1. Total fatty acid content and abundance of signature phospholipids fatty acids of different treatments (**OAd**: organic amendment management without irrigation; **OAw**: organic amendment management with irrigation; **Pld**: ploughing and inorganic fertilization without irrigation; **Plw**: ploughing and inorganic fertilization with irrigation) (n=36) (Morugán-Coronado et al. 2019).

Code	*nif*H gene copies g^{-1} soil (x10^{5})	*nir*S gene copies g^{-1} soil (x10^{6})	*nos*Z gene copies g^{-1} soil (x10^{6})	*nir*K gene copies g^{-1} soil (x10^{7})	*amo*A-B gene copies g^{-1} soil (x10^{4})	*amo*A-arch gene copies g^{-1} soil (x10^{4})	16S rRNA copies g-1 soil (x107)
OAd	72.7±8.9^{a}	29.5±3.5^{a}	29.0±2.6^{a}	14.5±6.1^{a}	8.5±0.3^{a}	30.0±10.4^{a}	0.1±0.1^{a}
OAw	223.1±20.7^{b}	78.4±9.0^{b}	40.1±3.9^{b}	34.6±12.3^{b}	34.4±1.0^{b}	26.4±9.5^{a}	1.1±8.2^{a}
Pld	3.1±0.9^{C}	2.4±0.4^{c}	1.8±0.2^{c}	0.6±0.3^{c}	3.95±0.7^{c}	3.2±2.2^{b}	1.4±1.9^{a}
Plw	5.5±0.2^{C}	4.9±0.4^{c}	5.1±0.4^{d}	2.8±1.3^{d}	15.1±1.6^{d}	5.4±3.1^{b}	2.9±2.4^{a}

Values are mean ± standard deviation (n=36)
A one-way ANOVA (P<0.05) was used to compare differences between treatments. Values in rows sharing the same letters do not differ significantly.

Table 2. Copy numbers of genes related to the nitrogen cycle in soils under different agricultural managements (**OAd**: organic amendment management without irrigation; **OAw**: organic amendment management with irrigation; **Pld**: ploughing and inorganic fertilization without irrigation; **Plw**: ploughing and inorganic fertilization with irrigation) (n = 36) (Morugán-Coronado et al. 2019).

Post-fire management effects on soil microbial properties

Wildfires can impact soil productivity of burnt areas (Robichaud, 2009), especially when are affected by huge and high-intensity fires such as those that are frequent in summer seasons under extreme weather conditions in the Mediterranean area. The negative impacts can be exacerbated after the post-fire management, which in some cases are even more severe than the fire itself. Salvage logging (SL), one of the most common management techniques in fire-affected areas, which comprise the extraction of the burnt wood and in many cases using heavy machinery and dragging the trunks over the soil, is an example of a strategy that can lead to an increase in soil vulnerability to erosion and degradation (Mataix-Solera et al. 2016).

After a wildfire in “Sierra de Mariola Natural Park” in Alicante (Spain), a study was developed to evaluate the impact of post-fire salvage logging on soils. The implementation of the management triggered severe soil physic-chemical degradation, with a critical decrease in the nutrient content and soil structure deterioration, and a decrease in the microbial biomass and activity (García-Orenes et al. 2017). In addition, impacts of SL on soil microbial communities were assessed, specifically on the abundance of nitrogen cycles and, thus, the potential of the soil for microbial nitrogen cycling (Pereg et al. 2018b). It was demonstrated that salvage logging reduced bacterial load compared to tree retention control, resulting in significant changes to the abundance of functional bacteria involved in nitrogen cycling (*Figures 5 and 6*). Microbial gene pools involved in various stages of the nitrogen cycle were larger in control soil than in soil subjected to post-fire salvage logging and were significantly correlated with organic matter, available phosphorous, nitrogen and aggregate stability (*Table 3*). The impact of post-fire management strategies on soil microbial communities needs to be considered in relation to maintaining ecosystem productivity, resilience, and potential impact on climate.

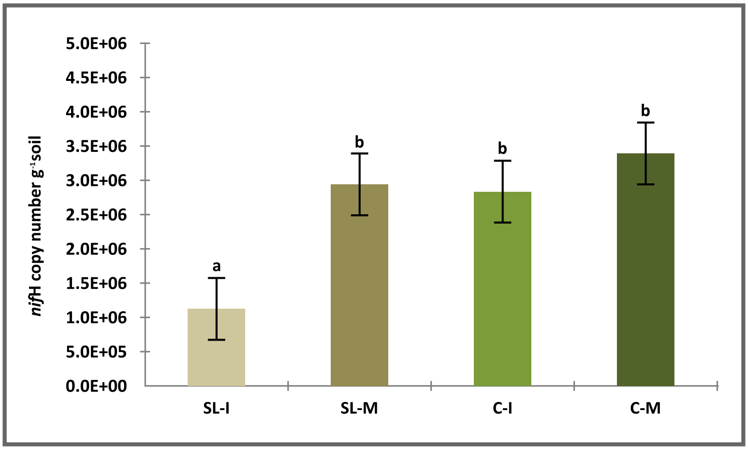

Figure 5. Abundance of nifH gene (mean ±standard deviation) in soil with different treatment after one-way ANOVA. SL-I: soil under salvage logging treatment sieved at 2mm; SL-M: soil under salvage logging treatment sieved between 63 and 250 μm; C-I: control soil with tree retention sieved at 2mm; C-I: control soil with tree retention sieved between 63 and 250 μm. Different letters above the bars indicate significant differences (Pereg et al, 2018b).

Parameter[a]	OM	N	AP	AS	MBS	BSR	*nif*H	*nos*Z	*nir*S	*nir*K	amoA-Arch	amoA-B	16rDNA	Total DNA
OM	1	0.95**	0.91**	0.77**	0.75**	0.78**	0.70**	0.47*	ns	ns	ns	ns	0.76**	0.84**
N		1	0.88**	0.81**	0.80**	0.70**	0.68**	ns	ns	ns	ns	ns	0.79**	0.83**
AP			1	0.78**	0.66**	0.75**	0.74**	0.49*	ns	ns	ns	ns	0.63**	0.70**
AS				1	0.69**	0.54*	0.64**	0.56*	ns	ns	ns	ns	0.76**	0.63**
MBS					1	0.51*	ns	ns	ns	ns	ns	0.62**	0.72**	0.59**
BSR						1	0.53*	0.66**	ns	ns	ns	ns	0.53*	0.71**
*nif*H							1	0.67**	ns	ns	ns	ns	ns	0.67**
*nos*Z								1	ns	ns	0.56*	ns	ns	0.59*
*nir*S									1	ns	0.56*	ns	0.65**	0.66**
*nir*K										1	ns	ns	ns	ns
amoA-Arch											1	ns	ns	ns
amoA-B												1	ns	ns
16rDNA													1	0.77**
Total DNA														1

ns: not significant; 16S rDNA: 16SrRNA gene. The results were confirmed using a non-linear method (Spearman) a similar value was obtained.
a OM: soil organic matter; N: Kjeldahl nitrogen; AP: available phosphorus; AS: aggregates stability; MBC: microbial biomass carbon; BSR: basal soil respiration.
* Significant at P<0.05
**Significant at P<0.01

Table 3. Correlation coefficients (r values) for relationships between the different physico-chemical soil properties determined and genes of soil (n=18).

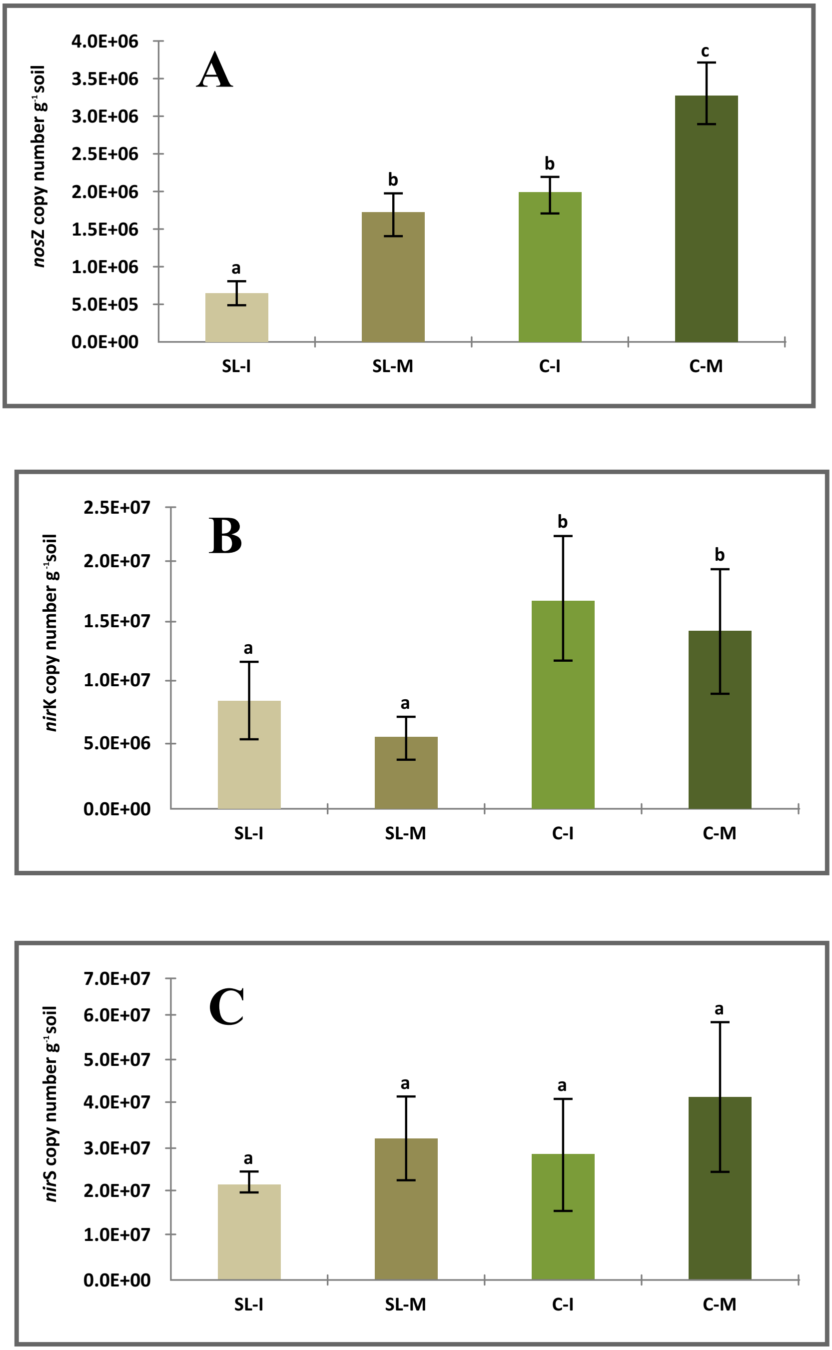

Figure 6. Abundance of *nos*Z gene (A), *nir*K gene (B) and *nir*S gene (C) (mean ± standard deviation) in soil with different treatment after one-way ANOVA. SL-I: soil under salvage logging treatment sieved at 2 mm; SL-M: soil under salvage logging treatment sieved between 63 and 250 µm; C-I: control soil with tree retention sieved at 2 mm; C-I: control soil with tree retention sieved between 63 and 250 µm. Different letters above the bars indicate significant differences. (Pereg et al. 2018b)

The degradation of soil structure- promoted by the decrease in organic matter content and reduced plant development after the wildfire and the post-fire salvage logging- resulted in the main driver in the community composition and structure shifts of the bacterial and fungi in a study of amplicon sequencing (García-Carmona et al. 2021). Soil degradation induced by logging operations resulted in new niches related to anoxic habitats, being Proteobacteria and Firmicutes families capable of anaerobic respiration found in high levels in the affected soils (*Figure 7*). In addition, the depletion in C and N nutrients as a consequence of the soil erosion reduced the microbial populations sensible to substrates availability, e.g., Actinomycetales. Ascomycota increased proportionally in managed soils, which might be due to the removal of host plants dependent on ectomycorrhizal fungi. The study demonstrated that physical soil disturbance related to post-fire management could seriously hamper the resilience of soil microorganisms after a fire perturbation.

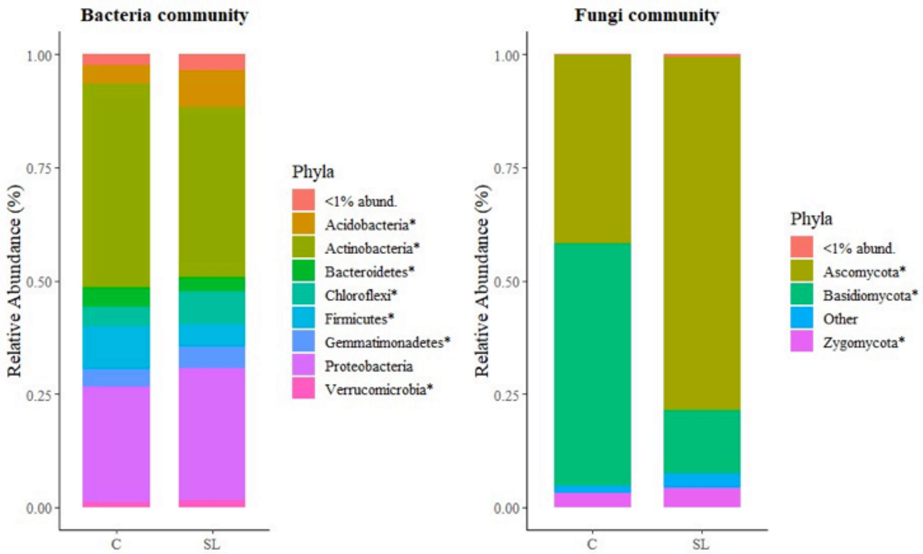

Figure 7. Relative abundances of the dominant bacterial and fungal phyla of control (C) and salvage logging (SL) soils. For each phylum, significant differences were assessed by the t-test calculated at p<0.05 and indicated by an asterisk. (García-Carmona et al. 2021).

The early post-fire vegetation colonizing has special relevance for the Mediterranean soils, the absence of vegetation cover exposes soils to rainfall events, runoff, and erosion processes. Ruderal mosses, forming part of the biological soil crust, have been pointed as early successional species colonizing burned soils in Mediterranean ecosystems, thus revealing important roles such as soil stabilization. A study in the same study area revealed that the presence of mosses played a significant role in soil fertility improvement and microbial activity six years after the wildfire (García-Carmona et al. 2020). In addition, SL management negatively affected the percentage of soil covered by mosses, decreasing 22% in SL soils. Considering the presence of mosses in the post-fire management was reveal of major importance given their role in Mediterranean ecosystem functioning.

Conclusions

The soils in the Mediterranean area extremely sensible to agro-forest management due to their particular environmental conditions. Therefore, agricultural practices or post-fire action can modify the equilibrium of microbial populations that can be reflected in a loss of biodiversity. In the Mediterranean area, it is essential to look for the best sustainable management practices of the soil to keep the fertility, quality, and microbial biodiversity of the same.

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