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Physico-chemical and microbial perturbations of Andalusian pine forest soils following a wildfire



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HIGHLIGHTS

GRAPHICAL ABSTRACT

- Physico-chemical and microbial characteristics of fire affected soils are studied.
 Fire increased soil N. soil OM and phose
- Fire increased soil N, soil OM and phosphatase activity.
- Proteobacteria and Firmicutes phyla increased in burnt soil.
- Acidobacteria and Bacteroidetes phyla decreased in burnt soil.
- Microbes respond to soil OM with higher diversity, activity and biomass production.



A R T I C L E I N F O

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ABSTRACT

Wildfires are a recurrent disturbance in Mediterranean forests, triggered by high fuel load, high environmental temperature and low humidity. Although, human intervention is behind the initiation of most fire episodes, the situation is likely to worsen in the future due to the effects of climate change in the Mediterranean "hotspot". Here we study chemical, physical and microbial characteristics of burnt soils from two well differentiated sites at Sierra de Cazorla, Segura and Las Villas Natural Park, Andalusia, (Spain) affected and unaffected by a wildfire, and followed their evolution for three years. The soils affected by a severe surface burn showed a significant increase in organic matter after 3 years from the fire. Viable bacteria and fungi also increased, especially 2–3 years post-burning. Substrate induced respiration (SIR) also increased significantly in burnt soil from site 1 (rendzina on carbonate) while a significant decrease was observed in the burnt soils sampled from site 2 (calcic luvisols) in samples taken one month after the wildfire. A recovery in both SIR and organic matter was observed after 2 and 3 years. Of seven soil enzymes studied, only phosphatase activity was significantly higher in most burnt soils over the three years. Analysis of bacterial community diversity using clone libraries showed a recovery in the number of phyla in burnt soils after 2 and 3 years in both sites, with an increase in Proteobacteria and Firmicutes and a decrease in Acidobacteria phyla. For Bacteroidetes, the percentages were lower in most burnt samples. This study reveals that if wildfire increases the organic matter availability, then the microbial community responds with increased activity and biomass production. Although fire exerts an initial impact on the soil bacterial community, its structure and functional profile soon recovers (after 2-3 years) contributing to soil recovery.

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1. Introduction

No single indicator that encompasses all aspects related to soil health has been accepted (Kibblewhite et al. 2008). In turn, a combination of soil variables such as soil respiration, physico-chemical characters and microbial biomass have been used to assess soil status. Microorganisms are able to react quickly to changes in soil ecosystem and exhibit a high degree of adaptability to the new environment. Therefore, soil microbial communities and microbial activities are also considered good indicators of soil health (Turco et al. 1994; Pankhurst et al. 1995). Among such indicators are those informing microbial biomass [commonly estimated by substrate induced respiration (SIR), analysis of polar lipid fatty acid (PLFA) and DNA soil quantification)], microbial activity (e.g. enzyme assays, BIOLOG systems) and microbial biodiversity [e.g. denaturing gradient gel electrophoresis (DGGE), terminal restriction fragment length polymorphism (T-RFLP)] (Kaiser et al. 1992; Kirk et al. 2004; Wright et al. 2004; Smith et al. 2008; Islam et al. 2011). Among the physico-chemical soil health indicators pH, conductivity, density, water retention and stability of aggregates are commonly assessed (Arias et al. 2005). The analysis of soil health is often most useful when applied to an environment which is being restored following perturbations such as pollution events and wildfires. Knowledge of soil health enables better restoration management and therefore a more effective recovery of degraded ecosystems (Mataix-Solera and Cerdá 2009b).

Extreme environmental conditions in the summer period (high temperatures and low relative humidity), together with other factors like the accumulation of easily flammable dried biomass and human intervention, responsible for the initiation of most fire episodes, makes wildfires the cause of important perturbations in Mediterranean bush/forest ecosystems. In addition, this situation is likely to worsen in the future due to global warming, with the Mediterranean region widely considered as a "hot-spot" for climate change (Giorgi 2006; Turco et al., 2017). The role of wildfires in the alteration of floral composition, tree regeneration and improving wood production has been widely studied (Mataix-Solera and Cerdá 2009a). Despite the fact that fires are considered a natural ecological factor in the Mediterranean ecosystem, anthropogenic activities in the region over the past 50 years *i.e.* grazing, cinegetic activities and tourism, has led to an increase in the occurrence of these perturbations. Wildfires produce changes in the physical, chemical and biological properties of soil (González-Pérez et al. 2004; Mataix-Solera and Cerdá 2009b; Gómez-Rey et al. 2013). Depending on severity, wildfires may exert soil nutrient depletion through processes including leaching and erosion, partial or complete combustion of organic matter and deterioration of the soil structure (Chandler et al. 1983; Carballas et al. 2009; Díaz-Raviña et al. 2010; Almendros and González-Vila 2012; Vega et al. 2013).

The effects of wildfires on the soil microbial community are also highly variable and dependent on multiple factors *i.e.* severity and intensity of fire as well as on the physico-chemical properties of the soil before and after the fire event. In many studies, it has been found that soil microbial biomass decreases following a wildfire and remains low for many years (Dooley and Treseder 2012). However, in other studies either an increase (Goberna et al. 2012) or no significant changes have been observed when comparing non-burnt and burnt soils (Hamman et al. 2007). In addition to the direct effects of fire and temperature reached in the soil, toxic compounds such as dioxins, hydrocarbons, polycyclic aromatic hydrocarbons (PAHs) and polychlorinated biphenyls (PCBs), which often inhibit the growth and survival of the living soil organisms may be produced (Kim et al., 2004a).

The analysis of the soil microbiota by means of culture-dependent techniques such as isolation plating has in the past provided useful but limited information because only a minor proportion of the soil microbes are cultivable. However, the recent application of cultureindependent techniques to environmental microbiology studies, including analysis of polar lipid fatty acids (PLFA) and molecular techniques based on 16S and 18S rRNA sequences, have greatly improved our understanding of the activity and true diversity of soil microbial communities although molecular approaches also have known weaknesses (Rütters et al., 2002). Among the culture-independent techniques developed, denaturing gradient gel electrophoresis (DGGE) is one of the most widely applied techniques used to profile soil microbial communities (Nakatsu et al., 2000). This method is particularly useful when examining time series and population dynamics (Theron and Cloete 2000). Once the identity of an organism associated with any particular band has been determined, fluctuations in individual components of a microbial population, due to environmental perturbations can be promptly assessed (Head et al. 1998).

Fire not only impacts the soil microbial community quantitatively, but also qualitatively through changes in the composition of the soil microbiota, availability of carbon and nutrients and microbial activity (Hernández et al. 1997; Jensen et al. 2001). Previous research has shown that the most significant differences in microbial diversity are found at lower taxonomical levels (*i.e.* at the species level) rather than at higher taxonomical levels (i.e. phylum, class, order) (Smith et al. 2008). Theodorou and Bowen (1982) observed that bacteria from the Pseudomonas genus were very sensitive to fire (phylum Proteobacteria) while other genera such as Bacillus or Clostridium (phylum Firmicutes) produce resistant spores which allow them to survive at 100-120 °C. In recent work, carried out in a Spanish Mediterranean area (Sierra de Aználcollar, Sevilla) affected by severe wildfires, higher diversity of both Bacteria and Archaea domains was found in burnt soils compared with the control soils, as assessed through DGGE analysis. In addition, based on analysis of 16S rRNA cloned sequences, several variations in the number of the different phyla present in burnt and control soils were reported (Rodríguez et al. 2017).

Most studies on the impact of wildfires have focused on short-term responses of the soil's physico-chemical properties or on its effect on specific organisms (Scharenbroch et al. 2012). Our limited knowledge of the consequences of wildfires on the activity and diversity of soil microbial communities over time, together with the increasing frequency of these events has enhanced research interest in this area. Improved understanding of the microbial community response to wildfires will undoubtedly contribute to improved biorestoration of affected soils and an enhanced understanding of the effects of wildfires on ecosystem services for which the soil microbial community play a vital role (Arias et al. 2005; Thom and Seidi 2016).

The main objective of this study was to assess the effects of a wildfire on the activity and diversity of the soil microbial population as well as following the response of the microbial population after the fire event For this, we studied two well differentiated locations and soils (rendzina on carbonate and calcic luvisol), affected and unaffected by a wildfire that occurred in 2005 in Cazorla, Segura and Las Villas (Jaén, Spain).

2. Materials and methods

2.1. Site description and sampling procedure

The study area is located in Sierra de Cazorla, Segura and Las Villas Natural Park (Jaén province, Andalusia, Spain), an area affected by a wildfire of a high severity that occurred between 7 and 10 August 2005 and burnt an area of >5000 ha, mainly of pine trees (*Pinus pinaster & P. nigra*) and Mediterranean shrubby vegetation. The topography of the area is abrupt with about 50% of the area at >1000 m above sea level (Molina et al. 2014). The area is characterized by Mediterranean climate with a mean annual temperature of 13.5 °C, and a mean annual rainfall of 1209 mm. Rainfall is abundant during the winter but from June to September severe drought and high temperatures raise the wildfire index to a critical level (Rivas-Martínez 1983).

In this area the soils are over a calcareous substratum and two well differentiated sites, 1 and 2 were selected. A map of the study site location is provided in the Supplementary Information (Fig. S1). The burnt

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