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Short-term microbial response after laboratory heating and ground mulching addition

Elisabeth Jiménez-Compán (1), Nicasio T. Jiménez-Morillo (2), Marco A. Jiménez-González (2), Jose A. González-Pérez (2), Antonio Jordán (1), Gema Bárcenas-Moreno (1*)

(1) Med_Soil Research Group. Department of Crystallography, Mineralogy and Agricultural Chemistry, University of Seville, Chemistry building, c/ Profesor González Perez, n1º, 41012. Sevilla. Spain.
(2) Isntituto de Recursos Naturales y Agrobiolgía. IRNAS-CSIC, Sevilla. Spain
*Corresponding author: gbarcenas@us.es

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Abstract

Fire alters soil organic matter inducing quantitative and qualitative changes that presumably will affect post-fire soil microbial recolonisation. Several studies have evidenced marked soil organic carbon reduction after moderate and high intensity fire, which limit the total recovery of microbial biomass during years.
In order to evaluate the role of soil organic matter alteration in short-term microbial colonization process, we perform a preliminary experiment where unaltered soil from Sierra Nevada Natural Park was heated at 300 ºC during 20 minutes in a muffle furnace (H300) to simulate a medium-high intensity fire. After heating, soil samples were inoculated with unaltered fresh soil, rewetted at 55-65% of water holding capacity and incubated during 3 weeks. At the same time, unheated soil samples were incubated under the same conditions as control (UH). In addition, trying to partially alleviate soil organic matter fire-induced alterations effects on microbial colonization, we include an organic amendment treatment (M^+). So, part of heated and unheated samples were amended with a mix of ground alfalfa:straw (1:1) and soil microbial abundance and activity were monitored together with soil organic matter changes. Heating process reduces total organic carbon content. After one week of incubation carbon content in heated samples was lower than the control one, in both, amended and un-amended samples.
Microbial biomass and respiration were negatively affected by heating. Ground mulching addition increase microbial biomass and respiration but was not enough to reach control values during the whole study. Nevertheless, viable and cultivable fungi and bacteria showed different pattern. After two weeks of incubation both, fungi and bacteria were higher in heated samples. Ground mulching addition appears to stimulate fungal response in both, heated and unheated samples.
Preliminary results of this experiment evidence the transcendence of soil organic matter fire-induced changes on microbial colonization process and the importance to determine several microbial parameters to obtain a more faithful conclusion about microbial response. The organic amendment appears to alleviate partially heated-induced damage, highlighting the positive stimulation on fungal abundance in both, heated and unheated samples.

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1 INTRODUCTION

Fire alters soil organic matter inducing quantitative and qualitative changes that presumably will affect post-fire soil microbial recolonisation. Several studies have evidenced marked soil organic carbon reduction after moderate and high intensity fire (Fernández et al., 1997; González-Pérez et al., 2004), which limit the total recovery of microbial biomass during years (Bárcenas-Moreno et al., 2011b). Immediately after moderate soil heating we can find an increment in carbon availability accompanied by an increase in some microbial parameters as microbial respiration, bacterial growth or fungal and bacterial CFU (Bárcenas-Moreno & Bååth, 2009; Bárcenas-Moreno et al. 2011ab), but long-term effect on soil ecosystem derivates

in an important quantitative and qualitative changes in soil organic matter which will condition microbial colonization and ecosystem post-fire recovery.

Post-fire managements focus on protect soil against erosion-risk can include organic amendment as mulching, logging litter, seeding application, etc. This kind of post-fire management to protect soil can influence soil microbial response as well, since these practices could be modifying post-fire condition to start microbial recolonisation.

In this preliminary study we compare microbial response after soil laboratory heating to simulate a medium-high intensity fire with and without the influence of ground mix of alfalfa:straw (1:1), trying to isolate the possible nutritional influence of mulching application on microbial response

2 METHODS

In order to evaluate the role of soil organic matter alteration in short-term microbial colonization process, we perform a preliminary experiment where unaltered soil from Sierra Nevada Natural Park located at 2000 m altitude at "El Posteruelo" in Nigüelas municipal boundary (30457332E 4094725N). The vegetation is composed by alpine vegetation formed by creeping bearing shrubs with *Genista versicolor* Boiss., *Cytisus oromediterraneus* (Boiss.) Ribas Martínez and *Berberis hispanica* Boiss. & Reut. as main plant species . Soil in this area is classified as Cryochrept developed on mica schist as parent material and sandy loam texture according to U.S.D.A. classification. Soil was heated at 300 ºC during 20 minutes in a muffle furnace (H300) to simulate a medium-high intensity fire. After heating, soil samples were inoculated with unaltered fresh soil (1%), rewetted at 55-65% of water holding capacity and incubated during 3 weeks. At the same time, unheated soil samples were incubated under the same conditions as control (UH). In addition, trying to partially alleviate soil organic matter fire-induced alterations effects on microbial colonization, we include an organic amendment treatment (M^+). So, part of heated and unheated samples were amended with a mix of ground alfalfa:straw (1:1) (2mg g^{-1} fresh soil) and soil samples were collected 7, 15 and 21 days after inoculation.

The soil water content (SWC) was estimated by drying at 105º C overnight each sampling in order to espress the result per g of dry soil. Particle size distribution was determined with a Bouyoucos hydrometer (Bouyoucos 1951) and the water-holding capacity was determined according to Foster (1995). The soil organic C (SOC) was analysed using rapid dichromate oxidation of organic C (Walkley and Black 1934), and the dissolved organic C (DOC) was measured in non-fumigated soil samples, as reported for microbial biomass C

Microbial activity was estimated measuring soil basal respiration by static incubation-titrimetric determination using NaOH trap and microbial biomass carbon (C_{mic}) was determined using fumigation-extraction (FE) (Vance et al. 1987; Sims and Haby 1971). The number of viable and cultivable aerobic bacteria and fungi was estimated using the plate count method following Zuberer (1994). Tryptic Soy Agar (TSA) was used to isolate both total aerobic bacteria and fungi were grown on Rose Bengal Chloramphenicol (0.1 g l-1) Agar. Different dilutions were used in triplicate. The incubation temperature was 28º C for all groups, but different incubation times were used: 3 days for aerobic bacteria and 4 days for fungi.

3 RESULTS

Heating process reduces total organic carbon content from 7 ± 0.1 %C in UH to 3.7 ± 0.3 %C in H300. In spite of ground mulching addition was applied to partially relieve this organic carbon reduction, heated and amended samples showed slight higher values of total organic carbon than the heated and unamended ones after one week of incubation (4.4%).

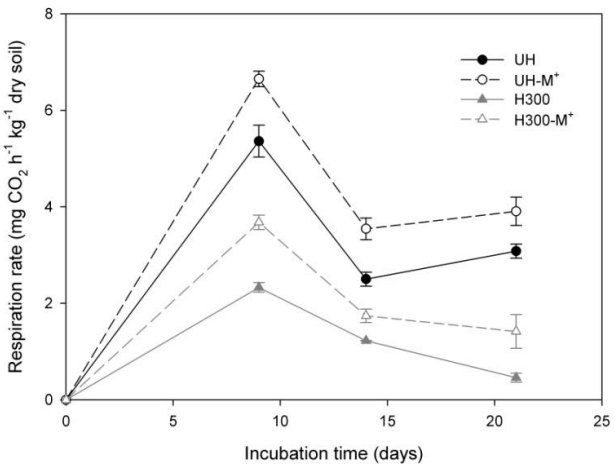

Figure 1. Microbial respiration rate measured 7, 15 and 21 days after inoculation with fresh soil. UH= Unaltered-control; UH-M^+= Unaltered-control amended with ground mulching; H300= heated at 300 ºC 20 min; H300-M^+= heated at 300 ºC 20 min. amended with ground mulching. Mean values ± standard error.

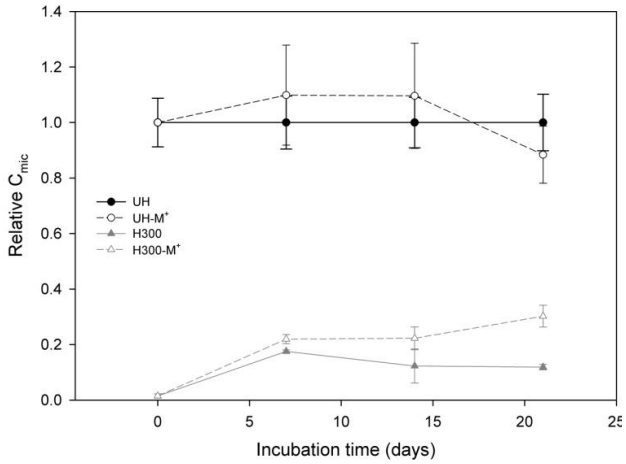

Figure 2. Relative microbial biomass (Cmic) measured immediately after heating and before inoculation with fresh soil and 7, 15 and 21 days after inoculation. UH= Unaltered-control; UH-M+= Unaltered-control amended with ground mulching; H300= heated at 300 ºC 20 min; H300-M+= heated at 300 ºC 20 min. amended with ground mulching. Mean values ± standard error.

Microbial survival after soil heating was evaluated immediately after heating and before the establishment of treatments and incubation by microbial biomass carbon estimation, resulting in no surviving microorganisms in heated samples. H300 treatment hardly showed some recovery during the experiment. Ground mulching appears to stimulate microbial biomass in both, heated and unheated samples, although microbial stimulation in heated samples was not enough to reach the unaltered values in 3 weeks (Fig.1).

During the first week microbial activity showed the highest values of the whole experiment in all treatments, decreasing with time. Heating treatment causes a marked decrease in microbial respiration. Mulching treatment induce an increment in soil microbial respiration in both, heated and unheated samples, although heated samples amended with mulching did not reach the respiration rate values of unaltered samples (Fig. 2), evidencing that the organic amendment applied was not enough to solvent soil organic matter fire-induced changes.

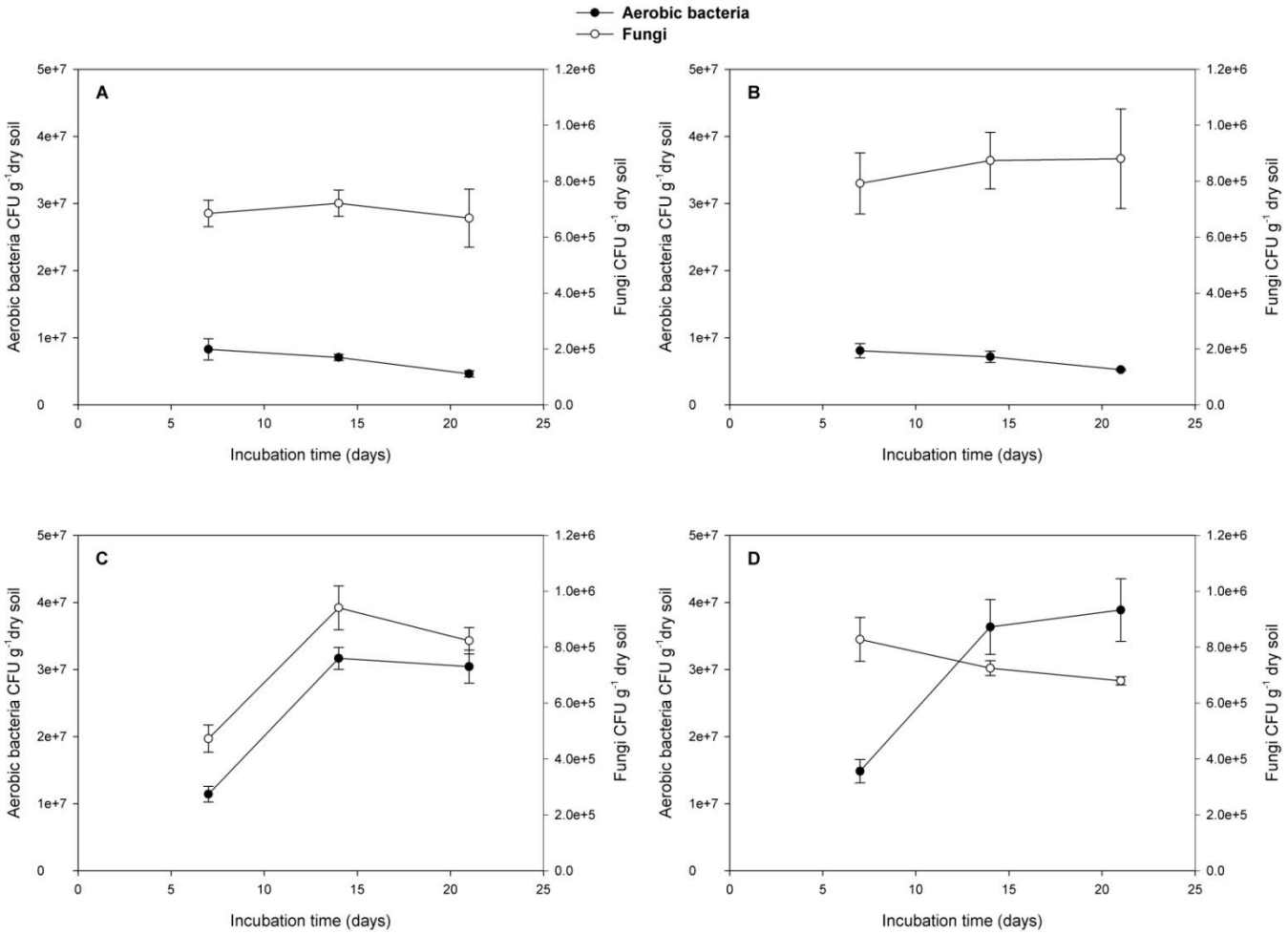

Figure 3. Viable and cultivable fungi and bacteria quantified 7, 15 and 21 days after inoculation. A) Unaltered-control; B) Unaltered-control amended with ground mulching; C) heated at 300 ºC 20 min; D) heated at 300 ºC 20 min. amended with ground mulching. Mean values ± standard error.

The microbial abundance of viable and cultivable fungi and bacteria showed a different pattern depending on the treatment. Heated samples showed higher abundance of viable and cultivable bacteria compared to the unheated ones, showing a marked increasing with incubation time. Mulching addition did not show marked differences in unheated samples, while appears to stimulate slightly bacterial proliferation in heated samples. On the other hand, fungal abundance showed different behavior. Unamended samples showed a marked decreased of fungal abundance due to heating process during the first week of incubation, increasing until reach the unheated samples values at the end of the experiment. Unheated samples amended with ground mulching mix showed higher values than the unamended ones during the whole experiment. While, heated and amended samples showed higher values than the control during the first week of incubation but it was decreasing slightly until the end of the incubation coinciding with the increase of bacterial abundance (Fig. 3).

4 DISCUSSION

Heating treatment applied to soil samples (300 ºC during 20 min) was enough to alter carbon content quantitative and qualitatively, as have been evidenced in previous studies (Bárcenas-Moreno & Bååth, 2009; Bárcenas-Moreno et al., 2014). In spite of total carbon content diminution after heating, carbon availability usually increases after low and moderate intensity fire evidenced by increases in DOC and microbial activity immediately after fire (Bárcenas-Moreno & Bååth, 2009; Bárcenas-Moreno et al., 2011ab; Bárcenas-Moreno et al., 2014), nevertheless, in our study both, DOC and microbial respiration were below control values one week after inoculation. One possible explanation is that soil temperature reached during heating could had been higher than in other experiments damaging to a greater extent carbon content. Other possibility is that DOC produced immediately after fire was consumed by a rapid microbial growth of inoculated microorganisms and one week after inoculation DOC increases could have been neutralized and no visible.

Microbial abundance estimated by C_{mic} was drastically reduced by heating and no recovery was evidenced in 3 weeks, since several years can be required to recover the original microbial biomass after fire (Bárcenas-Moreno et la., 2011b). However fungal and bacterial CFU appears to be stimulated by heating treatment and showed higher values to the control in two weeks after inoculation revealing part of microbial community that has the capacity to react and adapt to heating disturbance (Bárcenas-Moreno et al., 2011b, Bárcenas-Moreno et al., 2014).

The application of ground mulching showed partial stimulation of microbial community in both, heated and unheated samples, highlighting the effect on fungal community that appears to be specially stimulated by this organic amendment. The dominance of fungal decomposition of plant residues compared to bacteria has been reported previously (Neely et al., 1991) and evidences the importance of more deep studies to evaluate the ecosystem implication of the different post-fire management.

5 CONCLUSIONS

Moderate intensity soil heating treatment alters soil as growth media for microorganisms, reducing microbial respiration and biomass compared to unaltered soil. Although part of the microbial population appears to be stimulates by heating-induced alteration as evidence fungal and bacterial CFU. The application of ground mulching stimulated soil microorganisms, specially fungal community, but deeper studies with more microbial parameters, different doses and plant residues are need to start to conceive real transcendence of this kind of post-fire management on soil ecosystem functioning.

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