

**Recovering soil ecosystem services and functions at a burned  
native forest in the Mediterranean zone of central Chile by  
the use of organic amendments**

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**Abstract summary**

Global climatic conditions have exacerbated the occurrence of fires, particularly in Mediterranean biomes. In Chile, during the 2016-2017 summer, a megafire affected more than 5,000 km<sup>2</sup> in the nation's south-central area, compromising sclerophyllous forests characteristic of the Chilean mediterranean ecosystem. This work aims to evaluate the effect of organic amendments (swine manure, poultry manure and compost) on the recovery of soil biological conditions including basal respiration, microbial biomass, microbial metabolic coefficient and the coefficient of carbon mineralization, and some physicochemical conditions. Results indicated that soils microbial related process responded differently following eight month of treatment establishment. Related to the type of organic amendment, soils receiving swine manure evidenced the highest microbial basal respiration and microbial biomass, followed by those receiving poultry manure. However, greater mineralization rates and thus presumably shorter periods of C sources consumption were related to such organic amendments. Soils treated with compost accumulated the most organic carbon and nitrogen, ensuring a long-term nutrient release, thus a longer time of C storage.

*Keywords: restoration ecology, soil respiration, microbial biomass, wildfires.*

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**Introduction, scope and main objectives**

Fires have historically played an important role in the composition and distribution of terrestrial ecosystems (Pausas and Keeley, 2009). However, these events also represent an important pressure that induce land degradation worldwide, especially in Mediterranean biomes (D'Ascoli et al., 2005). These ecosystems are recognized as one of the world's most susceptible environments to fire occurrence, principally due to their climatic conditions (Bodí et al., 2012). They have also been indicated as environments highly susceptible to the effects of Climate Change, which make them one of the most fragile biomes on Earth (Fletcher and Zielhofer, 2013). The most devastating fires ever recorded in Chile occurred during the 2016-2017 season, affecting a close to 600,000 ha in central Chile (CONAF, 2017).

Land burning can compromise belowground conditions that are essential to support aboveground life (Certini, 2005). This is critical if the edaphic properties altered are precisely those enhancing plant establishment and successions after the occurrence of these events (Pérez *et al.*, 2004). Soil biological conditions including diversity, functions, and their services are all affected by land burning. Thus, understanding how biotic properties affected by fires can be recovered in degraded lands is central to the restoration and resilience of forest ecosystem functions (D'Ascoli *et al.*, 2005). This work aims to evaluate the effect of organic amendments (swine manure, poultry manure, and compost) on the recovery of soil biological conditions including basal respiration, microbial biomass, microbial metabolic coefficient and the coefficient of carbon mineralization, along with some physicochemical properties. We hypothesize that biological conditions in soils receiving fresh manures will show an immediate, but transient responses to these amendments as compared to those in soils amended with compost.

## Methodology

### *Research site*

the study was conducted at a private land (34°36.502'S; 71°42.281'W) in the east front (dryland zone) of the coastal mountain range of the O'Higgins region. This is covered by native sclerophyllous forests, with the canopy dominated mainly by *Quillaja saponaria*, *Lithraea caustica*, and *Peumus boldus*, followed by *Trevoa trinervis*, *Azara serrata* and *Colliguaja odorifera* in the understory. According to local residents, in the last 30 years this site has been mainly used by small farmers for livestock pasture and wood charcoal production and had not experienced fire events until January of 2017.

### *Experimental design*

In early June 2018, the application of organic amendments and vegetation establishment were implemented to evaluate the effect of them on soil biological conditions. Organic amendments were manually incorporated over the upper 30 cm of soil (Figure 1), following rototilling at the same depth. All treatments, except the control and reference, were covered with a mulch layer (1 cm approx.) consisting of a wheat and oat straw mix. For the present work, plots established in 2018 with organic amendments and seedlings were revisited for further study. Thus, there were six treatments evaluated, five at a burned area (50 x 50 m) and one located at an unburned area (20 x 20 m) within the research site at a distance of approximately 500 m. Treatment consisted of: T0, reference (unburned area); T1, control (with no treatment); T2, no amended (only mulch cover); T3, compost (200 m<sup>3</sup> ha<sup>-1</sup>, plus mulch); T4, poultry manure (200 m<sup>3</sup> ha<sup>-1</sup>, plus mulch); T5, swine manure (200 m<sup>3</sup> ha<sup>-1</sup>, plus mulch), each of them distributed in four parcels (3 x 1.5 m).



**Figure 15: treatment establishment**

### *Sample collection and analysis*

In January 2019, sampling was conducted. At each parcel, five sub-samples were taken at the corners and the centre of them, at a 6 cm depth following the removal of organic debris (1 Kg each sub-sample approx.). Soil sub-samples were carefully mixed in the field to obtain a composite sample per plot. For every sample an aliquot was kept under 4 °C to measure microbiological parameters, and the rest was dried at room temperature. Air dry samples were used to determine: Aggregate Stability (AS) (samples sieved between 4.00 and 0.25mm) and physicochemical analyses (samples sieved at 2 mm) including: EDTA-extractable trace elements, pH, electrical conductivity (EC), soil organic carbon (SOC), carbohydrates (Ch), total N, available P. Microbial measurements included Basal Respiration (BR) and Microbial biomass (MB), determined by substrate-induced respiration (SIR) with glucose. Both were performed in an automated impedance-meter (BacTrac 4200 Microbiological Analyser, Sylab, Austria). These values were used to calculate: 1) the metabolic coefficient ( $qCO_2 = BS/MB$ ) and 2) the coefficient of carbon mineralization ( $C \text{ min.coef} = BR/\text{soil organic carbon}$ ).

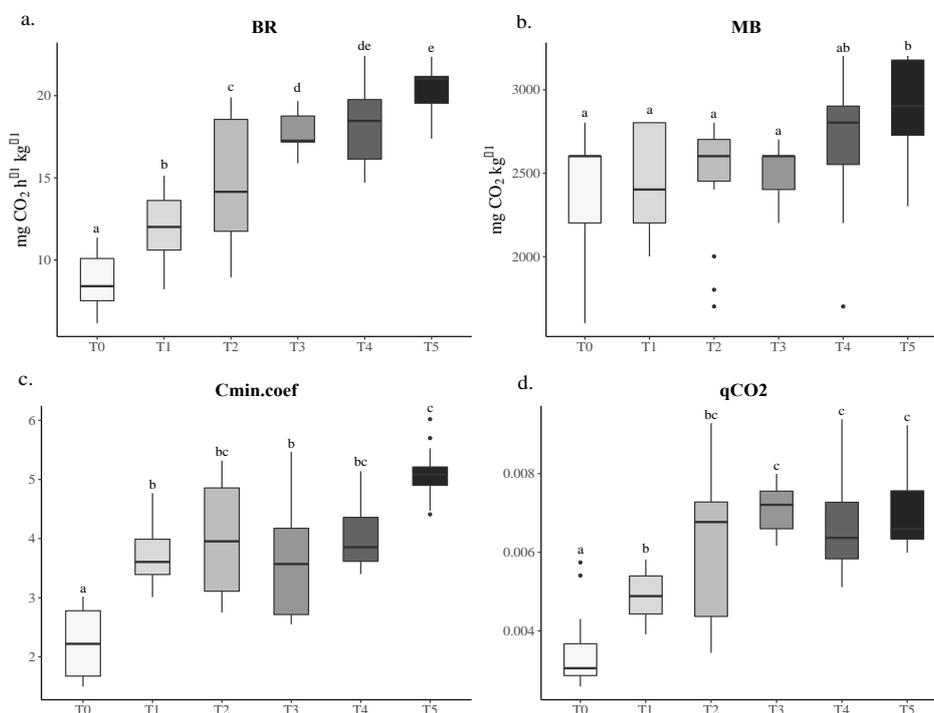
### *Statistical analysis*

To test whether the soil characteristics differed among treatments, we fitted Generalized Linear Mixed Models (GLMM) using the soil properties analysed as response, followed by multiple comparisons performed with the Tukey test ( $p < 0.05$ ). The relationship between the soil parameters measured regarding the treatments applied was analysed using principal component analysis (PCA). Two steps were taken to test the effects of soil physicochemical parameters and the different treatments on BR, MB, Cmin.coef and  $qCO_2$ . First, stepwise regressions with all physicochemical parameters (but excluding treatments) in both directions were performed. Then, in new models, the treatment was included as a predictor before the model resulting from the stepwise regressions. All analyses were performed in RStudio v.3.6.2 (RStudio Team, 2019)

## Results

The soils under study showed variable responses in physicochemical conditions following fire occurrence and the incorporation of treatments. Amended plots showed the highest OC contents, with those receiving compost having the greatest values (5.19 percent), followed by poultry manure (4.73 percent) and swine manure (4.14 percent) plots. The same pattern was observed for N contents, where soils amended with a more stable material, such as compost, had significantly higher values (0.54 percent) than those receiving fresh amendments in the form of poultry manure (0.47 percent) and swine manure (0.36 percent).

Soils microbial related process responded differently following eight months of treatment establishment (Figure 2). Based on the comparison between soils from the ecosystem of reference and those from the burned area, it is evident that basal respiration (Figure 2.A) was significantly disturbed by fires, while microbial biomass carbon (Figure 2.B) resemble to pre-fire conditions even without the application of an organic amendment. Soils receiving swine manure showed the highest microbial basal respiration and microbial biomass, followed by those receiving poultry manure. Soils amended with compost showed values similar to poultry manure amended soils. The later resulted in a significant increase of respiration as compared to control plots and plots covered with mulch, but not in microbial biomass.

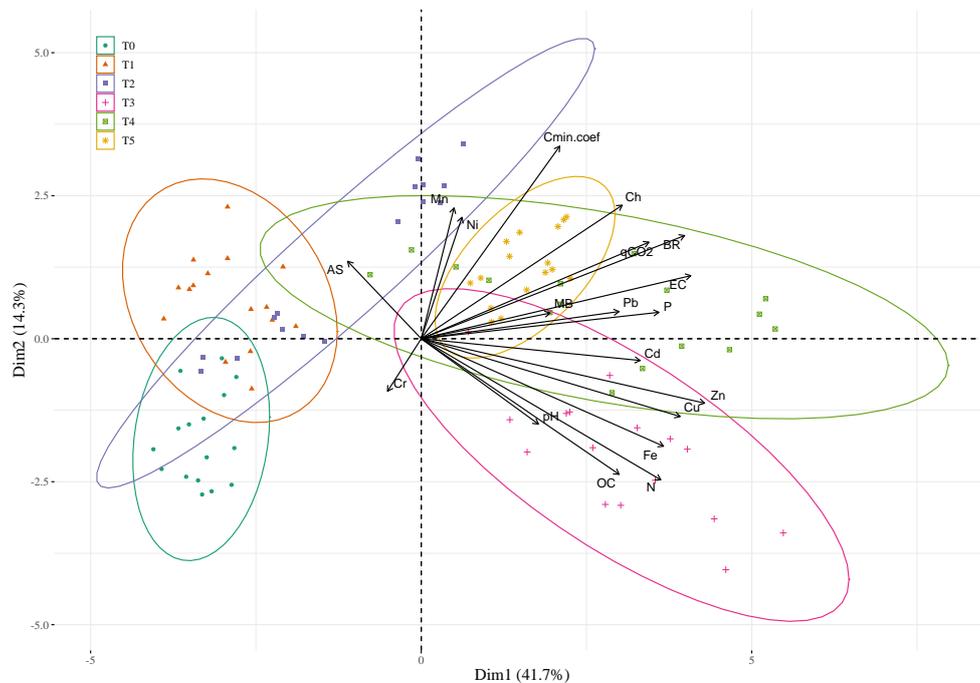


**Figure 2: Soil biological conditions studied**

Metabolic coefficient and carbon mineralization rates were significantly lower in soils from the reference site than burned soils (Figure 2.C and 2.D). Out of burned soils, all of those receiving organic amendments had significantly greater metabolic coefficient values than control soils. Carbon mineralization rates and microbial

metabolic coefficient varied among burned soils, with the greatest value observed in soils amended with swine manure.

PCA analyses (Figure 3), based on soil physicochemical variables, showed amended soils clearly separated apart from those at the ecosystem of reference and burned soils without amendment. Micronutrients including Zn, Cu and Fe along with N were the variables that explained the most this ordination.



**Figure 3: PCA analysis performed for soil physicochemical variables**

The organic treatment had a significantly higher effect than the soil physicochemical parameters on these four microbial parameters. The EC had an effect on all measurements except for qCO<sub>2</sub>, while some micronutrients (Fe and Mn) and potential pollutants (Pb and Cd) also affected some of the microbial parameters.

## Discussion

Soil microbial biodiversity and functions are recognized as the main drivers of post-fire ecosystem recovery: however, the effects of fires on them are less understood than those on soil physicochemical properties -which are less sensible to these events (Prendergast-Millerr *et al.* 2017). In this study, we clearly found higher effects on biological than in physicochemical characteristics. Out of the organic amendments used, those based of rather fresh materials (manure) as compare to more stable materials (compost) resulted in more dynamic conditions in soils, well above the ecosystem of reference, that represent a more stable (indeed, less disturbed) environment.

The incorporation of carbon sources at a fire affected soil enhanced the presence of heterotrophic microorganisms, as observed by CFU counts (Marín and Rojas, 2020) and stimulated microbial activity and

abundance, as observed here via respiration and biomass. Soil *priming* effect has been shown to positively respond to organic inputs in arid environments, which in turn influence soil C cycling (Bastida *et al.*, 2019). This influence likely contributed to plant colonization and succession at amended plots (data not shown). Under a global change scenario, soil restoration should consider the effect of proposed practices on soil ecosystem services as C sequestration, directly in its matrix and indirectly promoting vegetation reestablishment. Thus, future work should contemplate the type of organic inputs utilized, to precisely enhance soil functioning as a C reservoir in the long term.

## Conclusions

Out of the organic amendments evaluated, fresh materials (manure) showed greater influence in the biological conditions studied, as compared to compost amendment. Following eight months of treatment establishment, all amended soils appeared to remain in an active transition phase above the ecosystem of reference. Future studies, considering global change climatic conditions, should consider the effect of diverse organic inputs to promote, in addition to short-term reestablishment of soil functions and services, the effect of them on soil ecosystem services as important as C sequestration.

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