Federico Romeo, Pasquale A. Marziliano, Marìa Belén Turriòn, Adele Muscolo, 2019. Short-term effect of different fire severities on soil properties and Pinus halepensis regeneration. J. For. Res., 31, 1271-1282

Short-term effect of different fire severities on soil properties and *Pinus halepensis* regeneration

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Abstract

Considering that diverse fire severities can differently affect soil properties the aim of this study was to examine at what extent changes in soil properties caused by fire could condition seedling establishment. This new approach is for identifying a new fire cause-effect chain to qualify the impacts of fire on soils with the propose of using fire as a tool in forest management to favour *Pinus halepensis Mill.* regeneration. The study area was located in Astudillo, 'Laderas de Alcubilla' in the south-east of Palencia, in Castilla y León Region (Spain) in a reforested *P. halepensis* area. The pine stand under study was crossed by fire for 78.83 ha, in July 2013, with various degrees of gravity damages. The whole forest affected by fire was subdivided into 3 large areas according to the gravity of crown scorch, (low (LS), medium (MS) and high (HS) severity) on the basis of needle yellowing which usually occurs after exposure to direct flames. Results evidenced significant differences in soil properties in respect to the different fire severities. In HS area total nitrogen and carbon were considerably reduced while ash and phosphorus contents were significantly increased. The changes caused to the soil properties, in particular to the nutrient amount, influenced *P. halepensis* regeneration, mainly during the first year after the passage of fire. The greater pine regeneration was observed in area affected by moderate than low and high fire severity, in which the temperature reached, increased the mineralization of soil organic matter with the consequent release of nutrients right available for seedling growth. Additionally, moderate fire severity contrasted the regeneration of graminaceous, reducing the interspecific competition. The height of pine seedlings was inversely proportional to the numerousness of graminaceae where the number was abundant (LS), the height was modest, conversely, where the number was low (HS), the greater ipsometric differentiation of pine seedlings was observed. These results suggested that moderate fire severity represents a moderate environmental stress (hormesis) that favouring the microscale conditions, is able to increase pine germination and establishment. The exposure of *Pinus halpensis* to a moderate environmental factor, that is damaging at higher doses, induces an adaptive beneficial effect on seedling regeneration. This data can reevaluate the assertion that coniferous burned areas, left unmanaged, would remain unproductive for some indefinite period.

Keywords Ecophysiology, Fire severity, *Pinus halepensis*, Post-fire regeneration, Soil chemical properties

Introduction

In the Mediterranean environment, forest fire can be a significant disturbing factor able to influence, especially if recurrent, the forest ecosystem functioning (Attiwill 1994; Van Lierop et al. 2015; FAO 2016). Recently, the number of fires enhanced in most Mediterranean regions (JRC 2018), with significant impact not only on forest stands, but also on chemical, physical and biological soil properties (Certini 2005; Mataix-Solera et al. 2009; Šimanský et al. 2012). The way fire affects soils, and how strong the impact of fire is on an ecosystem, depend primarily on how the fire burns.

Fire severity is the result of length and quantity of energy that is released (Urbanek 2013), and of temperature interval reached at different soil depths (De la Rosa et al. 2012). Despite the heat in the moist soil moved quickly and penetrates into depth, the latent heat of vaporization prevents that the soil temperature exceed 95°C, up to the water doesn't evaporate completely (Campbell et al. 1994), then the temperature increases up to 200-300°C (Franklin et al. 1997). Under severe fire, the temperature can reach 500-700°C on the soil surface (DeBano et al. 1998), and sometimes values of 850°C (DeBano 2000) can be reached. The residence time is probably the element of fire severity more harmful for the subsoil. Indeed, severe fires that move quickly in the well-powered sites convey heat mainly to the first centimeters under soil surface, conversely, slow moving fires radiate heating in deeper layers (Gorbett et al. 2015). After the fires, soil temperatures can remain elevated since few minutes to several days. As consequence, a forest fire results in prominent changes in soil characteristics including: increase or decrease in soil aggregate stability, increase in bulk density (Rastad 2009; Arocena and Opio 2003), increase in soil cation amounts (Franklin et al. 2003; Liechty et al. 2005) and decrease in carbon (C) and nitrogen (N) stocks in surface soils (Johnson and Curtis 2001).

The major changes in soils, during burning, are the loss of organic matter and modifications in its composition (Neff et al. 2005). The impact of fire on the organic matter depends on the degree of its severity and consists of slight distillation (volatilisation of minor constituents), charring, or complete oxidation. As a result of changes in soil organic matter, fire may also influence soil aggregate stability (Kavdir et al. 2005), causing indirectly short or long-term changes in the environment which in turn can influence the health of biological organisms.

Short term effect of fire significantly alters microbial community and consequently large-scale processes such as nutrient cycling (Neary et al. 1999). The prompt effect of fire on soil microorganism is the decrease in their biomass. The intense fire can reduce a significant amount of microbial biomass as peak temperature considerably exceed those required, killing the majority of living being (DeBano et al. 1998).

A part the effects on soil, some latest studies showed that fire plays an important role in the dynamics, stand structure and species composition of specific forest type (Brazhnik et al. 2017), as reported in Rodrigo et al. (2004) and Marzano et al. (2012), post-fire recovery is generally accomplished by direct regeneration, i.e., the quick recovery of a plant community composed by the same pool of species that existed before the disturbance. Two main plant regeneration strategies attributing resiliency to Mediterranean ecosystems are the ability to resprout after fire (resprouter species), and the stimulation of the recruitment by fire (seeder species).

Pinus halepensis is one of the most widespread trees in the Mediterranean Basin (Quezel 2000; Maestre and Cortina 2004) occupying an area of about 7.5 million hectares, both in natural stands and in plantations (Quezel 2000). However, many of these forests and plantations were abandoned and over time this has negatively affected their structure and their stand density, leading to a loss of ecosystem functionality and resilience (Coello et al. 2015). In the Mediterranean forested landscapes, degradation results in a reduced capacity of natural ecosystems to face increasing external disturbances (i.e., wildfires). The lack of silvicultural activities caused an increased risk of fire, due to the great amount of biomass that remain in forest and that is susceptible to inflame (Baldock et al. 1996), therefore in the last decades extreme forest fires occurred, especially in pine forests. Additionally, the abandonment of agricultural lands with the consequent recolonization by shrub and forest species also caused a high risk of fires for forest (Fernandez Ales 1991; Gonzalez 1991; Hubert 1991; Höchtl et al. 2004; Sanesi et al. 2013; Marziliano et al. 2015; Marziliano et al. 2017). A recent study including all fires greater than one hectare that occurred in Spain over the past 42 years, between May and November (the 'vegetative' or growing season) and between December and April (the dormant season) in three different regions (Northwest, Hinterland and

Mediterranean) representative of the whole Spanish territory evidenced that wild fire increased in the last years and showed contrasting characteristics between regions and seasons, the Northwest region represents a paradigmatic example of the impact from human factors, especially during winter, whereas Hinterland and Mediterranean regions are mostly dependent on climate conditions (Moreno et al. 2014; Jiménez-Ruano et al. 2017). In any case, the fire in the forest has the non-sustainable disturbance characteristics because it is able to cause damage in the absolute sense, so that in European Community seat, it has been defined as a real social aggression to forests.

Many researchers demonstrated that forest ecosystem differently adapt to fire mainly because no species is 'fire adapted' but rather is adapted to a particular fire regime, but not to all (Keeley et al. 2011). There are many plant traits that are of adaptive value in the face of recurrent fire and these vary markedly with fire regime, highlighting that species exhibit traits that are adaptive under a particular fire regime and can be threatened when that regime changes. Many Mediterranean species and in particular *P. halepensis* and *Pinus pinaster Aiton* are resilient to periodic high-intensity crown fires only if they occur at intervals of several decades or more, showing also a great variability in serotiny driven by different fire regimes (Hernández-Serrano 2013, 2014).

Based on the above consideration, we hypothized that the response of *P. halepensis* to fire can be different and dependent on fire regimes and that post-fire regeneration of *P. halepensis* can be conditioned by fire severity not only because it controls serotinous cones opening, seed release (Saracino 1997) and mortality, but also because fire can affect organic matter trend and soil nutrient availability which in turn influence seed dormancy and seedling development (Osuna et al. 2015). On this regards, the main aim of this study was to evaluate and quantify the effects of different levels of fire severity, in a *P. halepensis* stand, located in Astudillo (Palencia-Spain), with the specific objectives of verifying 1) how fire influenced selected chemical and biochemical soil properties, 2) at what extent fire severity affected *P. halepensis* regeneration, 3) if changes in soil properties caused by fire could condition seedling establishment, and 4) if fire could be used as a tool in forest management to favour pinus regeneration.

Materials and Methods

Study area

The study area was located in Astudillo, 'Laderas de Alcubilla' (42º 11' 27" N; 04º 13' 34" W) at an altitude of 870 m a.s.l. in the south-east of Palencia, in Castilla y León Region (Spain). In 1952 the area was included among the areas of public interest and some years later reforestation activities started and were completed in March 1962. The species mostly used was *P. halepensis*, with a planting density of 2 x 3 m (1667 trees per hectare). Branches pruning was the only silvicultural management. No cutting and/or thinning were performed. The reforestation was very homogeneous in the whole area, with no difference in the forest structure.

The climate is typically Mediterranean. The annual rainfall is 428 mm, occurring mainly in the autumn and winter seasons. The mean annual temperature is 12.0°C. Average temperature of the coldest month is 3.8°C, while the temperature of the warmest ones is 21.5°C. The summer aridity is between June and September. The above reported climatic parameters are referred to 1980-2010 time span. According to climate classification by De Martonne (1926), the climate is "Mediterranean-type semi-arid". The area is also constantly lashed by strong winds (>50 km/h). Soils are calcareous with clay loam textures originated on carbonate-rich parent materials, and they can be classified within Inceptisol order (Llorente and Turrión 2010) as Calcixerepts.

The native vegetation in the studied area is Holm-oak wood (*Quercus ilex* subsp ballota). Currently, almost half of Castilla y León's regional territory is covered by forests (De la Mano Fernández 2007). However, in this region the fires represent the greatest danger and the major disturbing factor for these forest ecosystems. In this region in the last decade, in average, there have been 1955 fires per year, with a total burned area of 29952 ha per year (De la Mano Fernández 2007). On the other hand, the weather conditions of the Region greatly favor the propagation of the fire.

An area of 78.83 ha in the pine stand under study was swept by fire, at 5.07 pm of 31st July 2013, with various degrees of gravity damages. The causes of the fire derived from rural works in a wheat field in the valley. Fire rapidly spread between the crowns. Its extinguishing was communicated on 2nd August 2013, at 10 pm. Weather conditions were characterized by a very hot day (air temperature at 5pm was 33.7°C), low atmospheric humidity (relative humidity was 22.11%) and hot wind from south (wind speed was 1.07 m/s). The effects of fires in the stand were heterogeneous, some areas were wholly burnt, some areas underwent lesser damage. The whole forest swept by fire was therefore subdivided into 3 large areas according to the gravity of crown scorch, considered on the basis of needles yellowing which usually occur after exposure to direct flames.

In the present study, we classified fire severity following the indications of Pausas et al. (2003) modified by Turrión et al. (2012), who provided a threefold classification of fire severity according to pine canopy damage. Low severity (LS): light fire where canopy trees retain >20% of green leaves (top of the canopy), trees remain mainly green after the fire with 1/3 of crown scorch (visual estimation) and absence of post-fire mortality. Moderate severity (MS): where most leaves (>80%) of canopy trees are scorched (dead) but not consumed, the green leaves occur on the top (<5%), trees are mainly brown (retained scorched leaves) after the fire with 1/3-2/3 of crown scorch (visual estimation) and presence of post-fire mortality. High severity (HS): severe fire where canopy trees have >80% of burned leaves and the rest (if any) scorched (top), no green leaves are present and trees are brown with 2/3 of crown scorch (visual estimation) and presence of post-fire mortality. Close to these three burned areas, an unburned pine stand was identified as our control (C). The pine stand originated from a reforestation activity and it was 52 years old at the moment of the fire event.

Dendrometric assessments

The quantification of natural *Pinus halepensis* regeneration was carried out one year after the passage of wildfire (May and June 2014). Within each area with different level of fire severity, 3 randomly selected square-shaped plots were chosen, for a total of 12 plots (3 area for low severity fire, 3 for medium severity fire, 3 for high severity fire, 3 unburned area as control). Each plot had an area of 625 m^2. In each plot the number of live trees (NT-L) and the number of dead trees (NT-D) were counted. In addition, the diameters at breast height (DBH) of all trees (alive and dead) were measured. For the dendrometric and structural characterization of forest stands, in each plot covered by fire and in control plot, we have built the distribution of the trees in diameter classes. In order to monitor and quantify natural post-fire regeneration, a line transect of 1m x 20m was drawn in each plot. In addition, each transect was subdivided into 20 square areas, each of 1 m^2, and within each square area we counted all seedlings (herbaceous and arboreal vegetation), but only the height of *P. halepensis* seedlings was measured.

Soil and litter analysis

Three samples of soil were taken from 0-2 and 2-5 cm depth in each study area. Each soil sample consisted of a mixture of three sub-samples taken at random. At each point, one disturbed and one undisturbed mineral sample were taken by using a steel cylinder (5 cm diameter and 5 cm height). The samples were used to determine the bulk density. Two litter layers undecomposed litter (5-2 cm) and decomposed litter (2-0 cm) were also sampled at the same three points where soil samples were taken. A 25x25 cm wooden frame was used and each litter sample was a composite sample obtained mixing three subsamples. Soil samples were air-dried, sieved (< 2 mm) and percentages of fine earth and gravels were determined. Litter samples were dried at 70ºC for 48 hours, weighed in the laboratory and an aliquot was ground by a mill to plant samples. Soil surface temperature was estimated as suggested by De Bano et al. (1977; 1998; 2000); pH was measured in distilled water (weight:volume ratio 1:2.5) with a glass electrode; electrical conductivity (EC) was determined by a soil/water suspension 1:2.5 and subsequent reading by using the conductimeter which directly measures conductivity in µS/cm (micro-Siemens/centimeter); available phosphorus (Pav) was extracted by using ionic exchange membranes (Turrión et al. 1997) and colorimetric determination of phosphorus in the extracts (Murphy and Riley 1962) was carried out; total carbon and nitrogen, in the soil sample were measured with the LECO CHN-2000 analyzer. Soil total carbonates were determined using 1M HCl titrated with 0.5M NaOH (FAO 2007). Soil respiration was determined by using the Isermeyer method (1952). Microbial biomass carbon (MBC) was determined in moist samples by following Vance et al. (1987) method. Soluble organic carbon was detected on soil extracts of both fumigated and unfumigated samples by using the methods of Walkley and Black (1934). MBC was estimated on the basis of the differences between the organic carbon extracted from the fumigated and unfumigated soil, and an extraction efficiency coefficient of 0.38 was used to convert soluble carbon into biomass carbon (Vance et al. 1987); soil organic carbon density (SCD), and change in organic carbon stock (ΔSOC) were estimated as reported in (Molla et al. 2014); soil bulk density as weight of dry soil dived by the volume of soil have been detected. Litter samples were analysed with Leco CHN 2000 element analyser to determine total C and total N concentrations. Carbon to nitrogen ratio was calculated. Plant and litter analyses were done in duplicate.

Statistical analysis

For each undecomposed and decomposed litter layer, analysis of variance (ANOVA) was carried out to test if there was a significant effect of the fire severity on litter amount (L), total organic carbon (TOC), total nitrogen (TN) and carbon nitrogen ratio (C/N). As ANOVA indicated an overall significant effect of the main factors, with the Tukey's test we examined which pairs differed significantly. In order to explore the relationships among *P. halepensis* post fire regeneration, soil parameters, at two soil depth (0-2 cm and 2-5 cm) and three different fire severities (LS, MS, HS), datasets were analyzed using Principal Component Analysis (PCA), Multivariate Analysis of Variance (MANOVA) and T test for paired values. PCA multivariate approach and the results were summarized in an ordination diagram. PCA was carried out using the soil parameters to weight the presence of natural regeneration in plots under different fire severity, using the software PAST (Hammer et al. 2001). Because of the data are expressed in different units, we have standardized the results with the following formula:

$$z = \frac{(x_i - \bar{x})}{SD}$$

where x_i is the individual value of each parameter, $\bar{x}$ is the mean and SD is the standard deviation. We have carried out MANOVA analysis for evaluating the effects of two factors (fire severity, soil depth and their interaction) on the set of soil parameters and natural regeneration. Finally, since there are only two soil depth (0-2 cm and 2-5 cm), a T test was used for paired values to evaluate the significant differences between the two soil depth. This last analysis allowed us to verify if the severity of the fire affected the properties of the soil according to the depth and if the soil properties impacted by fire in the first layer (0-2 cm) were the same or different from those influenced by fire in the underlying soil layer. For each parameter analysed with ANOVA and t-test, the data matrix (sample size) derived from the average of the values per sub-plot, and from the average per plot, we obtained three values for each experimental condition (Control, Low severity, Moderate severity, High severity). Anova, Manova models and t-test were carried out using R statistical software version 2.3.0 (R Development Core Team 2008).

Results

Dendrometric assessment

Under different fire severities, the distribution of live and dead trees in respect to the diameter classes, one year after the passage of fire was different in HS plot compared to the other plots (C, LS, MS) (Fig. 1). In the plots crossed by fire, the total number of live trees ranged from 987 to 1205 per hectare. The number of dead trees increased when increased fire severity, and was more abundant in the class diameter with low DBH (diameter at breast height; 5 and 10 cm in LS; 5, 10 and 15 cm in MS and 5, 10, 15 and 20 cm in HS) showing a positive relationship between fire severity and DBH (Fig. 1). *P. halepensis* regenerated better in the burned area, but at different extent depending on fire severity. Regeneration ranking was as follows MS>HS>LS>C (Figs. 2 and 3). The highest regeneration was observed in MS area with a number of 3.2 seedlings per m² (32000/ha), with very significant differences ($F_{(3,6)} = 230.3$; $p < 0.001$) compared to unburned (C) and LS area, but not to HS area (2.8 seedlings per m^2) (Fig. 2). *Graminaceae* were the most abundant undergrowth vegetation that regenerated better in LS than in the other sites (Fig. 2), in inverse proportion to *P. halepensis*. The height of pine seedlings was inversely proportional to the numerousness of *Graminaceae*: where the number was abundant (LS) the height was modest. Conversely, where the number was low (HS), the greater ipsometric differentiation of pine seedlings (Figs. 2 and 3) was observed.

Soil chemical analysis

One year after the passage of fire, the amount of undecomposed litter (g/cm^2) in LS and MS area was similar to the control. No undecomposed litter was present in HS (Table 1), because the high temperatures have totally transformed it into ashes. A greater amount of total organic carbon and total nitrogen was found in MS affected area in respect to the other ones. Conversely, a lower C/N ratio in comparison to the control and to the other fire affected area was instead observed. In the decomposed litter layer, increasing fire severity, TOC and TN decreased with the lowest amounts in HS (Table 1). Fire severity and soil depth significantly affected soil parameters and natural regeneration (Table 2) evidencing also a significant effect of their interaction (fire severity-soil depth) on soil parameters (Table 2). This means that differences in soil parameters between plots with different fire severities were not constant at the two different soil depths. At both soil depths, only C/N ratio did not show significant differences between plots with different fire severity (Fig. 4, Table 3). The MBC and C/N parameters were not significantly different between the two different soil depths (Fig. 4, Table 3). The "fire severity-soil depth" interaction did not cause significant differences in MBC, C/N and CO_2. Figure 5 shows the PCA diagram for soil depth of 0-2 cm and 2-5 cm. In both soil depths the first two components (Eigenvalues >1) have been extracted. The explained variance was higher at soil depth 2-5 cm (80.6%), compared to a depth of 0-2 cm (75.6%). At both soil depths, component 1 explains about 50% of all the variability in the analysed parameters, while component 2 explains about 25% of the variance (Fig. 5). In the soil layer 0-2 cm, the values of P and EC (located in the quadrant with both positive components) increased (Fig. 4, Table 3**)**, especially in MS plot, with significant differences compared to the other plots (C, LS, HS). Electric conductivity enhanced proportionally increasing fire severity. The highest value (1026 µS/cm) was observed in the MS area. Available P concentration was significantly higher in soils affected by fires than in control soils. The highest value was detected in MS. This could justify the increased electrical conductivity observed in the area affected by medium-fire severity. After the fire, the increase in P and especially in EC didn’t seem to prevent the post-fire natural regeneration. While at the soil depth of 0-2 cm P and EC increased compared to control plot and were collocated in the same quadrant, in the layer 2-5 cm, P and EC were far apart (Fig. 4, Table 3), indicating their different variation at this soil depth. In this layer EC was collocated in the first quadrant (top left), where MS plots were located. Furthermore, in the MS plots, the EC showed the highest values (1800 µS/cm), not only in relation to the other plots with different fire severity (Fig. 4, Table 3), but also in respect to EC values found in the same plots but at the soil depth of 0-2 cm (Fig. 4, Table 3). It is interesting to observe that this parameter increased at the soil depth of 2-5 cm only in MS plots, showing significant differences. The high EC values in MS plots, seemed to have a positive effect on natural post-fire regeneration. At the soil depth of 2-5 cm, phosphorus was located at bottom right, opposite to the soil depth 0-2 cm (Fig. 5). It decreased in all plots compared to soil depth 0-2 cm, with significant differences in LS, MS and HS (Fig. 4, Table 3). Additionally, in this layer P concentration decreased in burned areas compared with the control, but without significant differences (Fig. 4, Table 3). The fire severities didn’t affect soil bulk density values in comparison with control, except for HS. When the fire severity increased, in the soil layer 0-2 cm, the pH increased (Fig. 4,

Table 3) in all the sites, except for MS plots, with significant differences between HS, control and MS plots. The collocation of pH at the negative dial (bottom left), where HS areas were located (Fig. 5a), suggested a negative weight of pH in the post-fire natural regeneration in this plot. In the layer 2-5 cm pH increased in all plots (Fig. 4, Table 3) compared to the more superficial layer, showing significant differences between the two soil depths, only in the control plot (Fig. 5).

TN, TOC, MBC, C/N ratio and CO_2 parameters were on the right side of the PCA diagrams at both depths (Fig. 5), indicating similar variations after the passage of fire, at both depths. As fire severity increased, a progressive decrease in the content of total nitrogen, total carbon, microbial biomass C, C/N ratio was observed. For these parameters, no significant differences were found between control, LS and MS plots, conversely, their values decreased under high fire severity, with significant differences compared to control and to the others fire affected area, especially in the layer 0-2 cm (Fig. 4, Table 3). TN, TOC and CO_2 parameters decreased with increasing soil depth, regardless of fire severity (Fig. 4, Table 3), with significant differences compared to minor soil depth, except for HS plot (Fig. 4, Table 3). For MBC and C/N ratio, no significant differences were observed between the soil depth of 0-2 and 2-5 cm (Fig. 4, Table 3). Control and LS plots had the same variations in TN, MBC, TOC, C/N and CO_2 parameters at both soil depths (Fig. 4, Table 3), with significant differences between 0-2 cm and 2-5 cm (Fig. 4, Table 3). MS and HS plots were characterized by significant differences in P values between the two soil depths (significantly absent in control areas and LS). MS area showed a considerable increase in EC at the soil depth of 2-5 cm (Fig. 4, Table 3). In the PCA diagram, the number of seedlings was always in the top left quadrant, collocated near the EC (positive weight) in MS plots, and near pH (negative weight) in HS plots (Fig. 5). However, in these plots, these negative aspects were probably compensated by the simultaneous decrease in the C/N ratio, CO_2 and MBC values (Fig. 4, Table 3). The decreased concentration of these parameters in HS plots seemed to have favoured the natural post fire regeneration. Probably these plots, were subjected to the negative effect of pH, that affected the natural regeneration. Instead, it seems plausible to assume that the natural regeneration in MS plots might be favoured mainly by the quantitative increase in P and EC, followed by TN and TOC. Our results (Table 4), evidenced that in the soil layer 0-5 cm, the SOC content didn’t change in LS and MS compared to control area, while strongly decreased in HS. These results are confirmed by soil carbon density, values evidencing that the greater accumulation of carbon per meter square was observed in MS followed by LS, C and HS area (Table 4). Soil respiration decreased increasing fire severity in both soil layers. Soil organic carbon, and soil bulk density values detected in the soil layer 0-5 cm one year after the passage of fire in LS and MS area were similar to the unburned area. In HS area, SOC value was reduced to 50% of unburned area, while the value of bulk density was significantly higher (+ 25%) than in the other sites. The relationship among soil properties changed in respect to the levels of fire severity. In control stand we observed only negative correlation between nitrogen and CO_2 and between pH, MBC and P, instead in LS only positive correlation between CO_2 and TN was detected. Increasing fire severity increased also the number of correlations among soil properties. In MS, important correlations were found between MBC, P, and TN, and between CO_2, P, TN and MBC. In HS stand only SCD was inversely correlated to SBD.

Discussion

This study, in agreement with previous findings of Busse et al. (2005), demonstrated a relationship between fire severity and soil properties confirming that modifications in soil characteristics related to fire can be highly dependent on fire temperature and duration. Soil responses can in fact compass from positive increases in plant nutrient availability in area with moderate fires to detrimental loss of nutrients, soil physical structure, and soil biota in area with more severe fires, suggesting that the exposure to an environmental factor (fire) that is damaging to high doses (temperature or duration), can induce beneficial effects at low doses.

Our data evidenced a dose-response manifestation in response to fire, suggesting a hormetic response of soil and seedlings to fire, in agreement with numerous published articles showing a biphasic dose response curve of organisms to changing environmental conditions (Calabrese et al., 2007; Calabrese and Blain 2011; Calabrese and Mattson 2017; Kim et al. 2018).

Our results demonstrated that soils in area passed by fire had the same amount of carbon in the first 5 cm except for the area under HS, suggesting that the duration and the severity of fire in LS and MS area were not so intense to transform/destroy the soil aggregates, clay minerals and biomass. As reported by Giovannini et al. (1988) and Ando et al. (2014), degradation of organic matter starts at 200–250°C and begin to be complete at ca. 460°C. Depending on the fire's severity and duration, organic matter can be subject to slight distillation, volatilization of minor constituents, carbonization or complete oxidation. One year after the passage of fire, organic matter and total nitrogen were significantly lower in area with high fire severity, indicating that the elevated temperature reached (690°C) increased the carbonization or complete oxidation of soil organic matter. This is in accordance with results obtained by other authors Certini (2005) and Smithwick et al. (2005) showing that in coniferous forests the content of carbon and nitrogen often lowered significantly as result of direct volatilization after a severe fire. Nitrogen is the nutrient most subject to volatilization into the atmosphere, it begins to volatilize at more than 200°C (Neary et al. 1999) and its volatilization increases with the duration of fire. In our study, the temperature reached during the passage of fire ranged from 250 (LS) to 690°C (HS). Changes in TN and TOC in the upper layers of soils were evident only in the area passed by fire with high severity, suggesting that the duration of the fire in HS area was so prominent to cause a significant increase in the temperature than in LS and MS area. The decrease in organic matter in HS stands, caused an increase in SBD that appeared straight correlated with fire severity. SBD values increased in response to the collapse of soil organic-mineral aggregates (Miesel et al. 2015) and to the sealing due to the obstruction of soil pores by ashes or freed clay minerals (Durgin and Vogelsang 1984; Muráňová 2015). All this was responsible for the decrease in soil water holding capacity (Wieiting et al. 2017), which in turn resulted in an accentuation of soil runoff and surface erosion (Martin and Moody 2001). In particular, the soil organic matter reduction and the contemporary increase in SBD found in our sites, were in perfect agreement with findings of other authors (Badia and Marti 2003; Seymour and Tecle 2004; DeBano et al. 2005) showing that high level of fire severity in *Ponderosa* pine or *Quercus coccifera L.* stands, caused the deterioration of soil structure and enhanced the bulk density in the upper soil layer. The marked increase in P and ash, accompanied also by an increase in the EC values, can be recognized as the principal cause of the fast and plenty development of *P. halepensis* on the MS sites one year after the passage of fire. The enhancement of vegetation cover, in burned forest, is generally related to high amount and availability of nutrients (Sitlhou et al. 2014). Hille and den Ouden (2004) demonstrated that more seeds of pine germinated on severely burned plots where litter and humus had been burnt, and evidenced that the seedlings were remarkably taller than those grown on lightly burned plots where only the litter layer was burned. Our observations were similar.

A year after the passage of fire, more pines grew in the area under medium fire severities rather than in slightly burned plots, or in very burned plots suggesting that in low severity fire an incomplete combustion of upper layers can produce a murky, porous stratum at the soil surface that being incline to rapid change in temperature and quick soil surface exsiccation doesn’t favor seed germination. Our results, were in agreement with data of Johnstone and Chapin (2006) showing that severe burning, at sites with intermediate drainage strongly favored coniferous seed germination and seedling growth because highly severe fire combusted the organic matter producing ash rich in nutrients. In our study, the abundance of *Graminaceae* sprouted on the slightly burned plots, was also an important factor which contributed to limit tree seedling establishment. No great damage to established vegetation was observed in LS and MS burned area, where only a minor percentage of adult tree were burned.

Conclusions

This investigation confirmed that are mainly the characteristics of fire disturbance to influence and define early patterns of plant community assemblage rather than the disturbance as such. Pine regeneration was markedly higher in medium than in low and high fire severity highlighting the dual effect of this severity with the creation of favourable forest soil conditions that caused less damage to the seeds interfering with understory regeneration, underlying that variations in fire severity are able to drive the success of natural regeneration, the distribution of vegetation and the shape of the community assemblage. In short, in this study we provided evidence of environmental hormesis induced by fire, and we can assume that if wildfires burn under secure conditions at moderate severity may be an effective tool to restore landscape heterogeneity and conserve forest biodiversity, representing an useful management practice to ameliorate poor soil conditions and in turn the tree natural regeneration. These results on the adaptive responses of soil and *P. halepensis* seedlings to different fire severities within a hormetic perspective can have important implications to vegetation environmental change biology and can provide further insight to manage *P. halepensis* stand in a sustainable way.

Acknowledgements

We are very grateful to Teresa Bueis for her support during laboratory experiments at University of Valladolid-INIA, Spain.

Conflict of interest statement

None declared.

Funding

This work was supported by Mediterranea University of Reggio Calabria Italy as part of the Lifelong learning Program ERASMUS Placement 2013-2014.

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Figure Captions

Fig. 1 Number of living (red) and dead (blue) trees per hectare according to the diameter classes. C = unburned plot; LS = low fire severity; MS= medium fire severity; HS= high fire severity. Bars represent standard errors and mean values

Fig. 2 *P. halepensis* regeneration and undergrowth vegetation (Graminaceae) one year after the passage of fire at different severities. C = unburned plot; LS = low fire severity; MS= medium fire severity; HS= high fire severity

Fig. 3 Seedling distribution according to height class of *P. halepensis* one year after the passage of fire at different severities. LS = low fire severity; MS= medium fire severity; HS= high fire severity

Fig. 4 Variation of the main chemical parameters of soil at 2-5 cm depth compared to the same parameters measured at a depth of 0-2 cm. The lines are the standard error of the parameters analysed, in each area with different fire severity.

Fig. 5 PCA (principal component analysis) diagram for areas with different fire severities at 0-2 cm (A) and 2-5 cm (B) soil depth

Table 1 Mean values and standard deviations of litter amount (L), total organic carbon (TOC), total nitrogen (TN) and carbon nitrogen ratio (C/N) in undecomposed and decomposed litter layers of *Pinus halepensis* forest, one year after the passage of fire. In HS the values are missing because the high temperatures have totally transformed undecomposed litter it into ashes. For each parameter the sample size was n = 3 for treatment (unburned area (C), low fire severity (LS), medium fire severity (MS) and high fire severity (HS)). Means in the same column followed by the same letter are not statistically different at $p \leq 0.05$ (Tukey test)

Undecomposed Litter (depth 5-2 cm)

Fire severity	**L**	**TOC**	**TN**	**C/N**
	(g/cm^2)	(%)	(%)	-
C	$0.07^{a}\pm0.02$	$47.88^{ab}\pm0.67$	$1.00^{b}\pm0.08$	$48.1^{a}\pm3.54$
LS	$0.04^{a}\pm0.01$	$43.67^{b}\pm2.44$	$0.97^{b}\pm0.04$	$45.2^{ab}\pm3.35$
MS	$0.07^{a}\pm0.01$	$48.38^{a}\pm1.59$	$1.34^{a}\pm0.17$	$36.4^{b}\pm4.08$
HS	-	-	-	-
p-value	0.072	0.029	0.013	0.019

Decomposed Litter (depth 2-0 cm)

Fire severity	**L**	**TOC**	**TN**	**C/N**
	(g/cm^2)	(%)	(%)	-
C	$0.45^{b}\pm0.23$	$39.20^{a}\pm3.30$	$1.36^{a}\pm0.11$	$28.73^{a}\pm1.06$
LS	$0.29^{b}\pm0.10$	$25.87^{b}\pm6.40$	$1.00^{b}\pm0.24$	$25.03^{b}\pm0.38$
MS	$0.16^{c}\pm0.02$	$26.38^{b}\pm3.31$	$1.07^{b}\pm0.85$	$24.86^{b}\pm4.48$
HS	$0.74^{a}\pm0.75$	$17.41^{c}\pm2.91$	$0.64^{c}\pm0.10$	$27.42^{a}\pm1.88$
p-value	0.011	0.002	0.002	0.024

Table 2 Statistical parameters of Factorial Multivariate Analysis of Variance (MANOVA) to evaluate the effects of fire severity, soil depth and their interaction on the set of soil parameters and natural regeneration

Effects		**Value**	**F**	**Hypothesis Df**	**Error df**	**Sig.**
Fire severity	Pillai's Trace	2.50	5.56	27.00	30.00	<0.001
	Wilks' Lambda	0.00	26.61	27.00	24.01	<0.001
	Hotelling's Trace	241.02	59.51	27.00	20.00	<0.001
	Roy's Largest Root	182.31	202.56	9.00	10.00	<0.001
Soil depth	Pillai's Trace	0.99	61.95	9.00	8.00	<0.001
	Wilks' Lambda	0.01	61.95	9.00	8.00	<0.001
	Hotelling's Trace	69.69	61.95	9.00	8.00	<0.001
	Roy's Largest Root	69.69	61.95	9.00	8.00	<0.001
Interaction between fire severity and soil depth	Pillai's Trace	2.43	4.70	27.00	30.00	<0.001
	Wilks' Lambda	0.00	8.63	27.00	24.01	<0.001
	Hotelling's Trace	49.77	12.29	27.00	20.00	<0.001
	Roy's Largest Root	37.21	41.34	9.00	10.00	<0.001

Table 3. Soil parameters at two soil depth, assessed by paired-sample t test. *p*-values < 0.05 indicate significant differences between 0-2 cm and 2-5 cm depth.

	Control		**Low Severity**		**Medium Severity**		**High Severity**	
Parameters	t-value	p-value	t-value	p-value	t-value	p-value	t-value	p-value
pH	-4.00	0.057	-4.00	0.057	-1.73	0.225	-4.00	0.057
EC	3.21	0.085	2.36	0.142	-80.16	0.001	0.64	0.584
P	2.47	0.132	2.02	0.180	6.11	0.026	4.58	0.044
TN	10.36	0.009	32.82	0.001	8.19	0.015	2.60	0.121
MBC	-5.91	0.027	-2.34	0.143	3.10	0.090	-1.46	0.279
TOC	9.02	0.012	13.26	0.006	8.79	0.013	2.78	0.108
C/N	6.86	0.021	-0.05	0.964	0.95	0.442	1.10	0.384
CO_2	7.70	0.016	6.68	0.022	7.80	0.016	1.16	0.363

Table 4 Mean values and standard deviations of soil organic carbon (SOC), soil bulk density (SBD), variation of soil carbon density (ΔSCD) and soil surface temperatures in 0-5 cm soil depth of unburned (C), low fire severity (LS), medium fire severity (MS) and high fire severity (HS) in *Pinus halepensis* affected areas. For each parameter the sample size was n = 3 for treatment (four different fire severity). Means in the same column, followed by the same letter, are not statistically different at $p \leq 0.05$ (Tukey test)

Fire severity	**SOC** (%)	**SBD** (kg soil/m^3)	**ΔSCD** (kg C/m^2)	**Surface Soil Temperatures** (°C)
C	$16.09^{a} \pm 1.19$	$0.97^{b} \pm 0.22$	$24.59^{ab} \pm 10.29$	± 24
LS	$15.34^{a} \pm 2.50$	$1.02^{b} \pm 0.31$	$35.47^{ab} \pm 7.85$	± 240
MS	$14.38^{a} \pm 0.50$	$0.94^{b} \pm 0.29$	$45.26^{a} \pm 12.41$	± 410
HS	$7.07^{b} \pm 2.68$	$1.22^{a} \pm 0.20$	$19.31^{b} \pm 3.39$	± 690
p-value	0.002	0.045	0.034	-

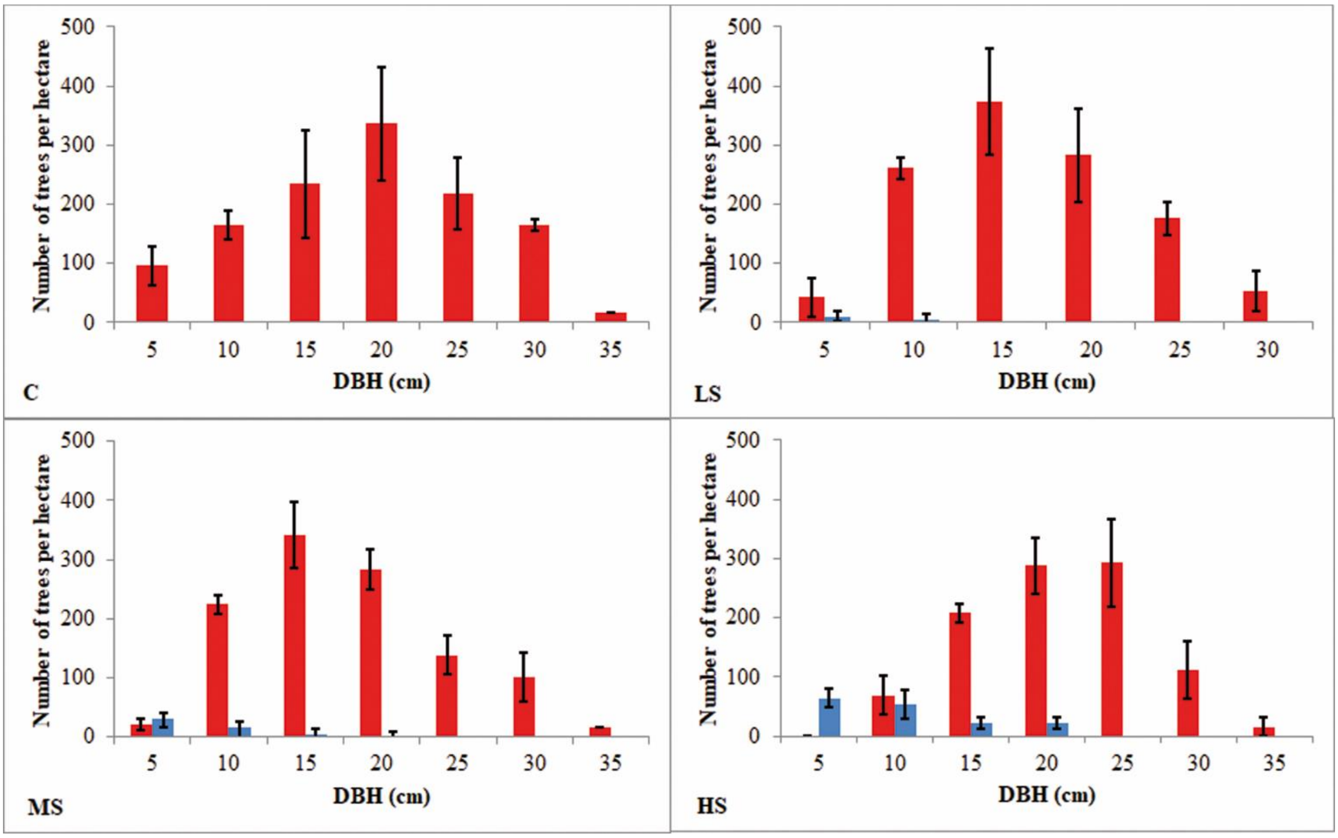
C
LS
MS
HS
Number of trees per hectare
DBH (cm)
0
100
200
300
400
500
5
10
15
20
25
30
35

Fig 1

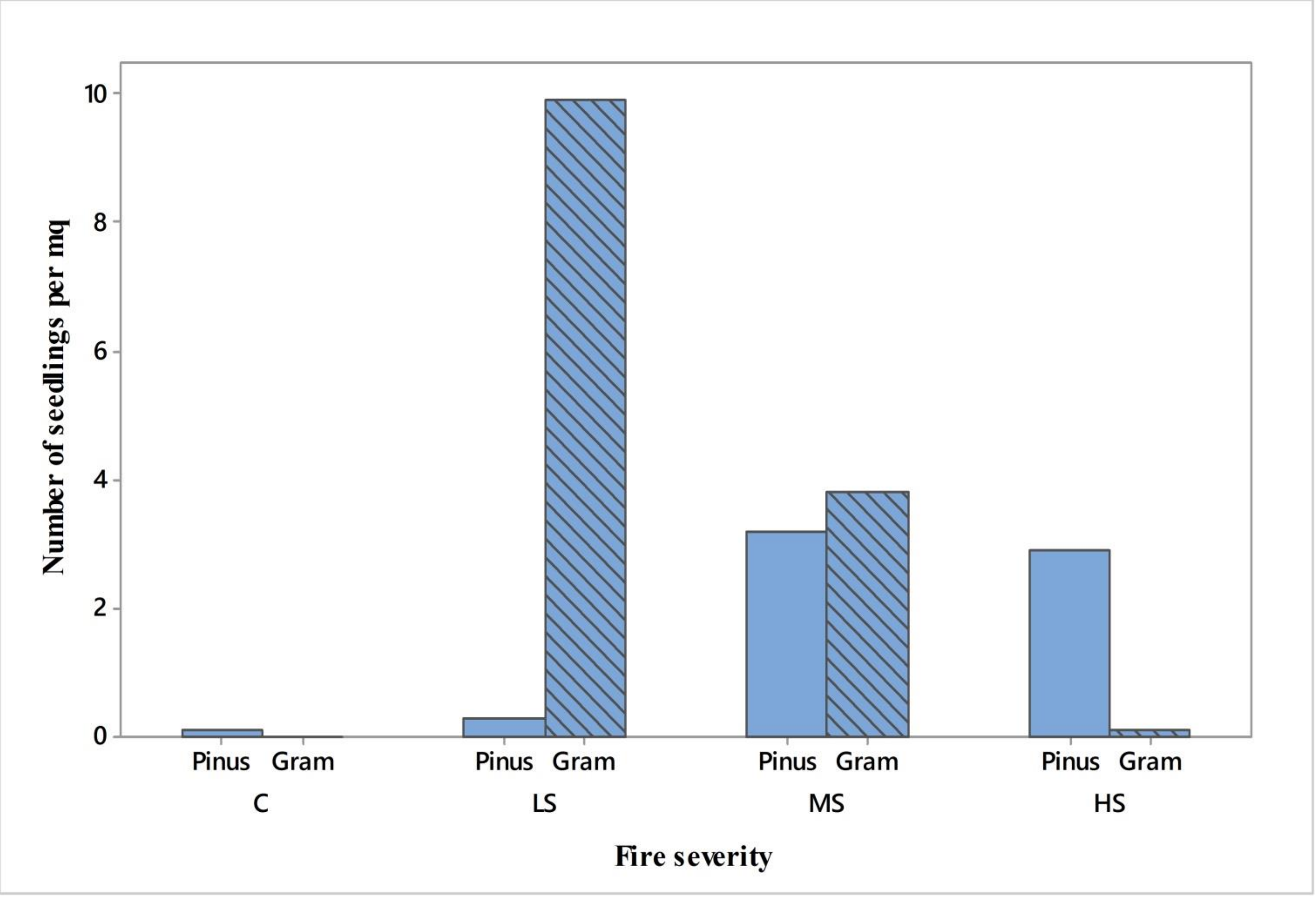
Number of seedlings per mq
10
8
6
4
2
0
Pinus
Gram
Pinus
Gram
Pinus
Gram
Pinus
Gram
C
LS
MS
HS
Fire severity

Fig. 2

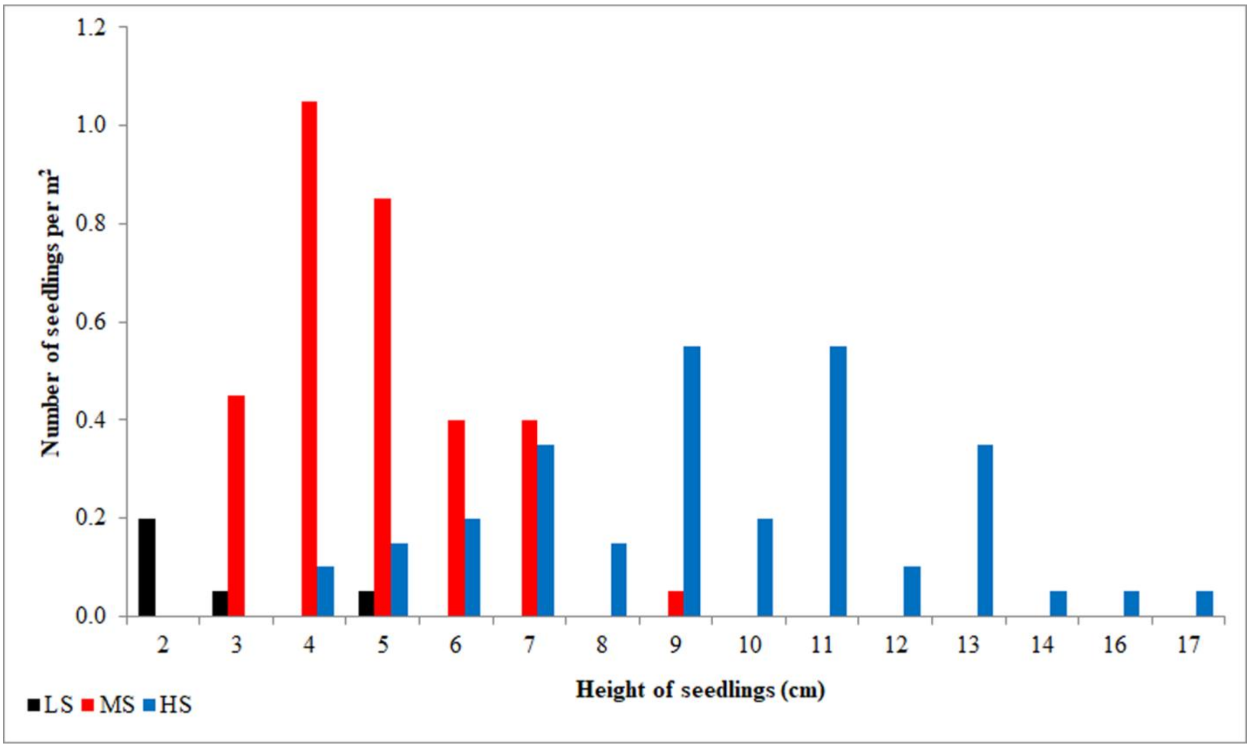
1.2
1.0
0.8
0.6
0.4
0.2
0.0
Number of seedlings per m^2
2
3
4
5
6
7
8
9
10
11
12
13
14
16
17
Height of seedlings (cm)
■LS ■MS ■HS

Fig. 3

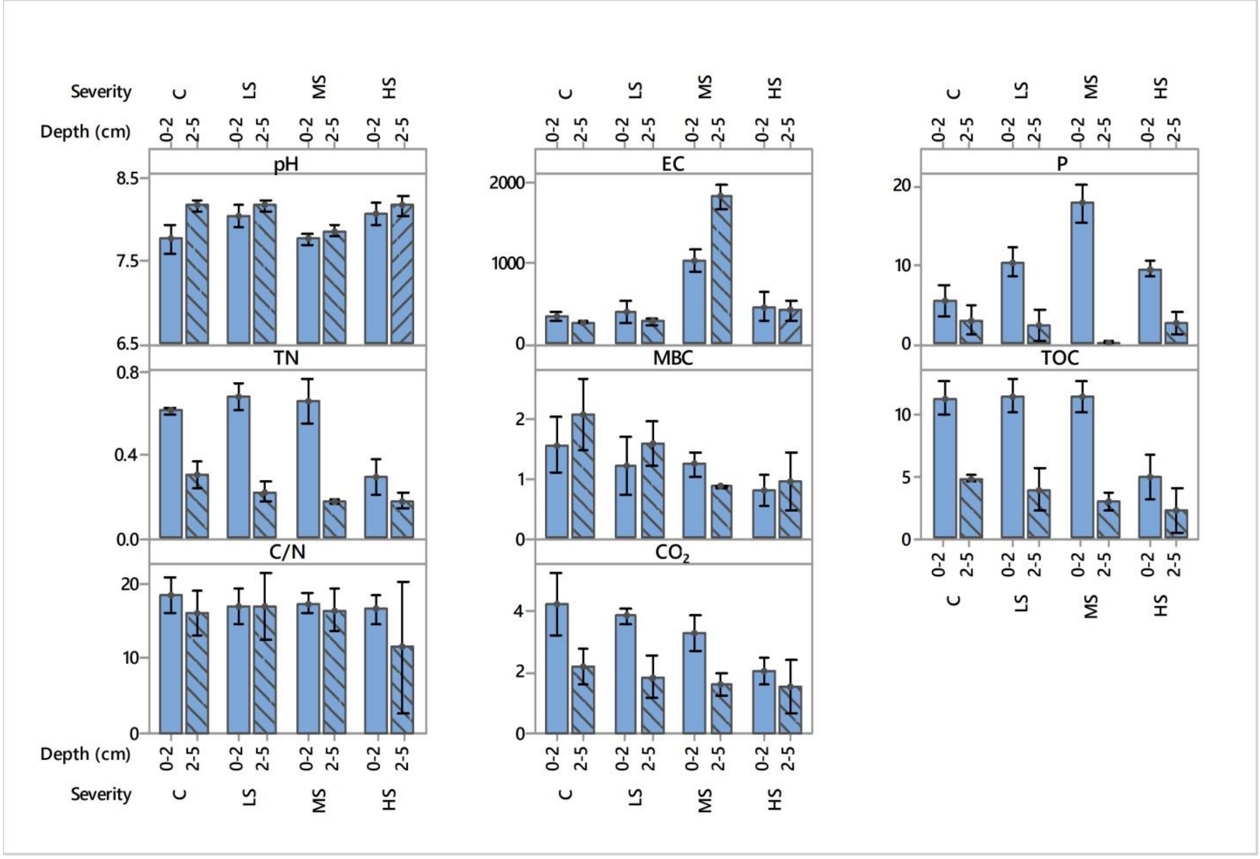
Severity
Depth (cm)
C
LS
MS
HS
0-2
2-5
pH
8.5
7.5
6.5
TN
0.8
0.4
0.0
C/N
20
10
0
EC
2000
1000
0
MBC
2
1
0
CO_2
4
2
0
P
20
10
0
TOC
10
5
0

Fig. 4

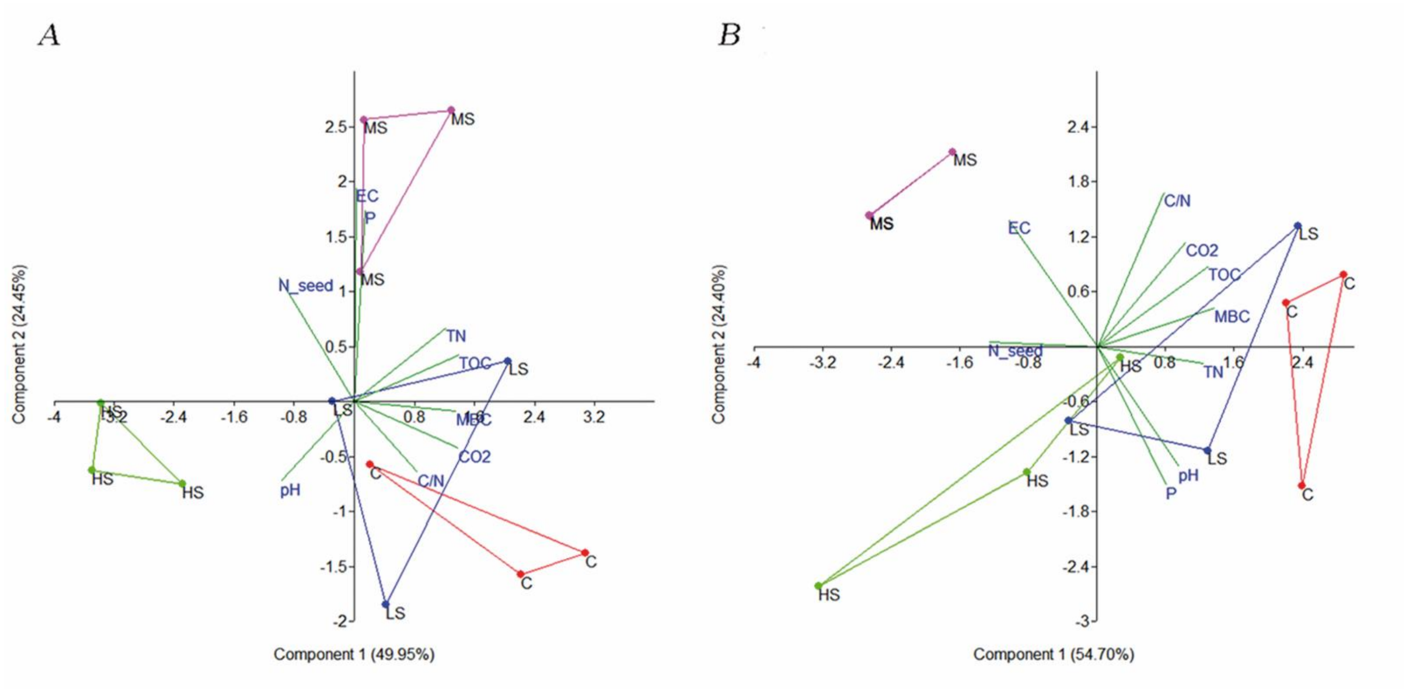
A
MS
MS
MS
EC
P
N_seed
TN
TOC
LS
LS
HS
HS
HS
MBC
CO2
C/N
C
C
C
pH
LS
Component 2 (24.45%)
Component 1 (49.95%)
2.5
2
1.5
1
0.5
-0.5
-1
-1.5
-2
-4
-3.2
-2.4
-1.6
-0.8
0.8
1.6
2.4
3.2
B
MS
MS
C/N
EC
CO2
TOC
MBC
LS
C
C
N_seed
HS
TN
LS
LS
HS
pH
P
C
HS
Component 2 (24.40%)
Component 1 (54.70%)
2.4
1.8
1.2
0.6
-0.6
-1.2
-1.8
-2.4
-3
-4
-3.2
-2.4
-1.6
-0.8
0.8
1.6
2.4

Fig. 5