

The effect of fire on the dormancy break of three annual legume seeds

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Abstract

Fire is a common phenomenon in the Mediterranean environment and strongly influences vegetal population dynamics through its impact on vegetation and the soil seed bank. Fire is able to break down the seed coat of hard-seeded legumes within the soil and trigger germination. To evaluate the effect of fire on the dormancy break in *Medicago ciliaris*, *Medicago rugosa* and *Scorpiurus muricatus* subsp. *subvillosus*, the seeds were placed at three different depths (surface, 25 mm and 50 mm) and subjected to fires at two different intensities (high and low). As a control sample, a batch of seeds was buried at 25 mm for the duration of the trial and not subjected to fire. Soil temperatures during the fire were compared directly to stubble quantity and indirectly related to soil depth. The two *Medicago* species survived exposure to 90°C for a few minutes and displayed a significant increase in germination with exposure to high temperatures (over 70°C) for several minutes. On the other hand, no germination occurred in *Scorpiurus*, irrespective of treatment. In conclusion, fire had a significant and positive effect in triggering germination of the *Medicago* species, but the dispersal strategies of these hard-seeded legumes are only partially interrupted by fire as a large number of seeds (>50%) remained non-germinated in the soil.

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Introduction

Fire exerts an enormous influence on vegetation in the Mediterranean environment, together with other important variables (Geerling, 1985; Kozłowski, 2000) such as rainfall and herbivores (Mapaure and Campbell, 2002; Wiegand *et al.*, 2006). Vegetation fires, both of natural or anthropogenic origin, occur not only in natural habitats but also in cultivated areas, and can have a great impact on the soil seed bank. The seed bank plays an essential role in the dynamics of plant communities (Leck *et al.*, 1989), providing an immediate source of new vegetation after fire disturbances. In this context, the probability that a seed from the soil seed bank will germinate depends, directly and/or indirectly, on a number of fire-related factors such as heat shock (Keeley, 1994; Núñez and Calvo, 2000), changes in light regimes (Bell *et al.*, 1999; Trabaud and Renard, 1999), smoke (Keeley and Fotheringham, 1998; Staden *et al.*, 2000), ash (Henig-Sever *et al.*, 1996; Reyes and Casal, 1998) and the release of nitrogen compounds (Henig-Sever, 1997; Keeley and Fotheringham, 1998). Exposure to elevated temperatures has been demonstrated to activate seeds that have a primary dormancy (Bell and Williams, 1998; Williams *et al.*, 2004) and to break the physical barrier of hard-seeded legumes, releasing the seeds from tegumental dormancy (Bradstock and Auld, 1995; Smith *et al.*, 2000). Heat diffusion into the topsoil depends on the fire intensity (magnitude and duration) and the soil characteristics (Bradstock and Auld, 1995; Williams *et al.*, 2004). Fire intensity plays a key role in the rupturing of the seed coat (Auld and O'Connell, 1991); however, high intensity fire may burn the seed while low intensity fire may not be hot enough to break the dormancy of hard-seeded legumes (Saharjo and Watanabe, 1997). Soil characteristics also regulate the temperature and heat penetration, decreasing as depth increases and reacting differently depending on soil moisture and texture (Busse *et al.*, 2005). The major part of the research on this topic was carried out in laboratory conditions rather than in field situations, or was focused on bush-woody legume species (Danthu *et al.*, 2003; Walters *et al.*, 2004; Shackleton 2007), while less information is available on the effect of fire on the dormancy break in herbaceous hard-seeded legumes. Within this context, our paper explores the impact of fire intensity and seed position in the soil on the dormancy break of three annual legume species.

Materials and Methods

The trial was carried out in August 2007, in experimental plots of 4

m² (2 m × 2 m). Mature seeds of *Medicago ciliaris* L. All., *Medicago rugosa* Desr. and *Scorpiurus muricatus* subsp. *subvillosus* (L.) Thell., closed in wire mesh envelopes, were placed in clay soil at depths of 0 mm, 25 mm and 50 mm, to simulate the stratification of seeds in field crops owing to the tillage. Two intensity controlled fires (low, LIF; high, HIF) were set up, created by burning two different amounts of wheat straw (corresponding to 2 kg in LIF and 4 kg in HIF per plot) to reproduce two field fire conditions. As a control, for each species a batch of seeds enveloped in the same manner was placed on the surface of the soil throughout the experiment, in a plot not subjected to fire. For each treatment (fire intensity and depth of burial), four envelopes, each containing 25 seeds, were used. Temperatures were recorded by thermocouples buried at tested depths and connected to a data logger. Immediately after the fire treatments, the envelopes were taken from the soil and the burnt seeds were counted and removed; those still intact were placed in 90-mm diameter Petri dishes, on filter-paper soaked with 3 mL of distilled water. The Petri dishes were randomly arranged in a dark incubator (MCT 200, Angelantoni Scientifica, Perugia, Italy) set at 20±1°C. Seeds were checked daily for 15 days, and those showing 2 mm of radicle emergence were considered to have germinated and removed from the Petri dishes. To check the viability of the non-germinated seeds (seeds that had not imbibed water), they were scarified with sandpaper at the end of the experiment.

Statistical analyses

The total percentage of seeds that germinated was calculated as the total number of seeds germinated divided by the total number of seeds taken for germination studies × 100. Seed vigour was evaluated on the basis of the speed of germination, and the mean germination time (MGT) was calculated using the formula: $MGT = \frac{\sum(fx)}{\sum x}$, where x is the number of daily germinated seeds, and f is the number of days after the trial started (Ellis and Roberts, 1980). Both total percentage of germinated seeds and MGT were compared by using a nested ANOVA, considering the main effect of fire and of the depth within each level of fire. A graphical analysis of residuals showed that the basic assumptions for linear models were not severely violated and, thus, no stabilising transformation was adopted. In respect of marginality restriction, means for significant effects were compared by using the simulation-based procedure of Edwards and Berry (1987), which is practically equivalent to the Tukey test in the case of balanced data. Treatments in which none of the replicates showed any germination were excluded from the analysis because they would have hindered the data homoscedasticity.

Results

Soil temperatures and their temporal patterns varied widely with treatments during the fires (Figure 1). As expected, temperatures were higher at the surface of the soil, reaching a maximum of 95.3°C and 91.3°C in HIF and LIF fires, respectively, but with a shorter duration in LIF compared to HIF. In this last treatment, a temperature over 70°C was maintained for 20 min, while in LIF this temperature was kept for 5 min only. At 25 mm depth, in HIF the maximum temperature (60°C) was two thirds of that reached at the soil surface and almost one third of that shown in LIF (31.6°C); moreover, temperatures over 50°C were maintained for 40 min. At 50 mm, the temperatures did not vary widely for either fire intensity, and ranged from 36.6°C of HIF to 30.4°C of LIF. The low germination of the seeds of the three studied species detected in the control group, ranging from 38% of *M. ciliaris* to 0% of *S. muricatus*, confirms the high levels of hard-seeded quality frequently encountered in legume species. The effect of fire on seed germination differed among the species and was particularly influenced by

whether the seeds were buried or on the soil surface, except for *Scorpiurus* in which the seeds did not germinate, irrespective of treatments. The seeds of all species recovered from the soil surface when exposed to HIF fire were burnt and, consequently, after having been submitted to the germination trial, were classified as charred, non-viable seeds. In *M. ciliaris*, the HIF treatment significantly increased the germination of buried seeds, irrespective of the soil depth (Figure 2). This treatment induced a significantly greater germination (51% in an average of 25 mm and 50 mm depth) than found in the LIF (44%) and the control (38%) treatments. No significant difference was observed in relation to soil depth. In *M. rugosa*, LIF significantly increased the germination of seeds recovered from the soil surface (45%), when compared to the control (25%) and LIF × 25 mm (27%) treatments (Figure 3). Scarification of non-germinated seeds, done after the field experiment, resulted in full germination (data not reported), showing the presence of a consistent number of hard seeds. The lowest MGT values were recorded when the seeds were buried at 50 mm soil depth for *M. ciliaris*, irrespective of fire treatments, and at 50 mm in LIF treatment in *M. rugosa* (Figure 4).

Discussion and Conclusion

As expected, the temperatures caused by the fire vary greatly in magnitude and extent in relation to the soil depth considered. After the stubble fire, the temperatures peaked on the soil surface and decreased

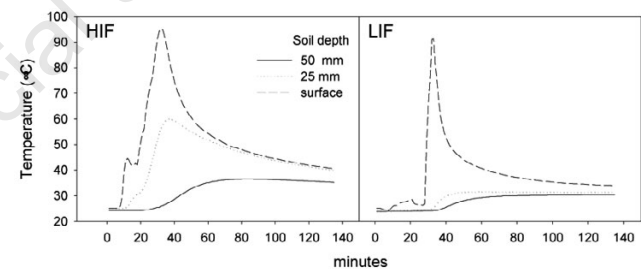


Figure 1. Temperatures during the fire experiment in relation to fire intensity and soil depth. HIF, high intensity fire; LIF, low intensity fire.

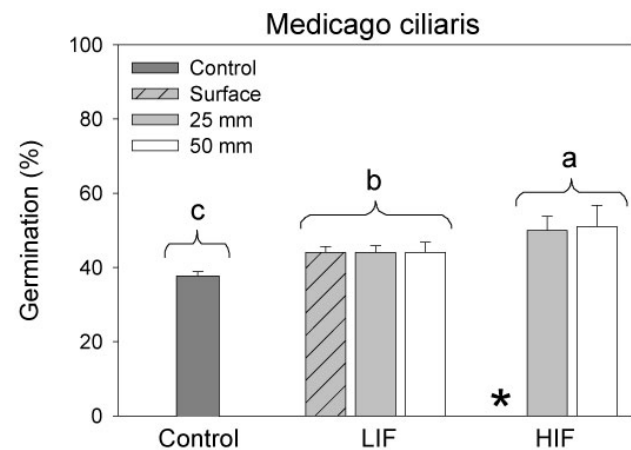


Figure 2. Total germination percentage in *M. ciliaris* and in the control, at the surface and at 25 and 50 mm soil depth. HIF, high intensity fire; LIF, low intensity fire; *no germination occurs owing to the burning of seeds. Different letters indicate a significant difference for $P < 0.05$.

as the depth in the soil increased (Patten and Cave, 1984; Bradstock *et al.*, 1992). The patterns of soil temperatures during the fires appear to be directly related to stubble quantity and indirectly related to depth.

Legume seeds frequently have hard, impermeable, seminal teguments, which ensure long-term survival and dispersal strategies (Danthu *et al.*, 1996; Vassal, 1998). Many studies have confirmed a release of seed coat-induced dormancy in legume seeds after fire

(Sabiiti and Wein, 1987; Mbalo and Witkowski, 1997). Heat shock, in fact, provoked by the passage of fires is one of the natural factors that trigger seed germination (Oba, 1990; Tarrega *et al.*, 1992; Cox *et al.*, 1993). However, the seed coat fracture as a result of fire and the following germination depend on the fire's intensity (temperature, duration) and on the species of seed. Conversely, some studies indicated the negative impact of fire on seed germination while others reported minimal or no effect (Brown, 1993; Van Staden *et al.*, 1994). Thus, the questions are: i) do different hard-seeded legume species have the same response to heat shock? ii) which combination of temperature and soil depth is needed to perpetuate seed germination?

From our study, a clear indication has emerged of how fire breaks the seed dormancy in the two studied *Medicago* species but not in *S. muricatus*, which is in agreement with other authors (Herranz *et al.*, 1998). Fire causes an increase of germination of 90% in *M. rugosa* and 26% in *M. ciliaris* after exposure to temperatures between 70°C and 90°C when compared to the control, which is consistent with the findings of other researchers (Auld and O'Connell 1991; Jhurree *et al.*, 1998). Exposure to temperatures between 80°C and 100°C has been shown to break the seed dormancy of several legumes of eucalypt savannas in north-eastern Australia, but temperatures greater than 100-120°C are shown to be lethal to seeds of these species (Williams *et al.*, 2003). In our trial, fire intensity and consequently high temperature duration greatly influenced the chance of seed survival during fires. The two *Medicago* species survived exposure to 90°C for a few minutes and exhibited a significant increase in germination with exposure to high temperatures (over 70°C) for several minutes. This suggests the ability of seeds to tolerate the passage of fire, and provides evidence of a stimulatory effect of the heat on germination. In *Scorpiurus*, the lack of an effect of fire in triggering germination is probably a result of a greater hardness of the seed coat (Gresta *et al.*, 2007; Abbate *et al.*, 2010), which is not fissured by a high intensity fire when it is buried and which is burnt when it is on the surface of the soil. All seeds on the soil surface were destroyed during exposure to HIF fire. Clearly, the burial of seeds reduces the negative impact of fires, as well as of predation (Miller, 1994). In conclusion, the fire had a significant and positive effect in triggering germination of *Medicago* species, but the dispersal strategies of these hard-seeded legumes is only partially interrupted by fire as a great number of seeds (>50%) remained non-germinated in the soil.

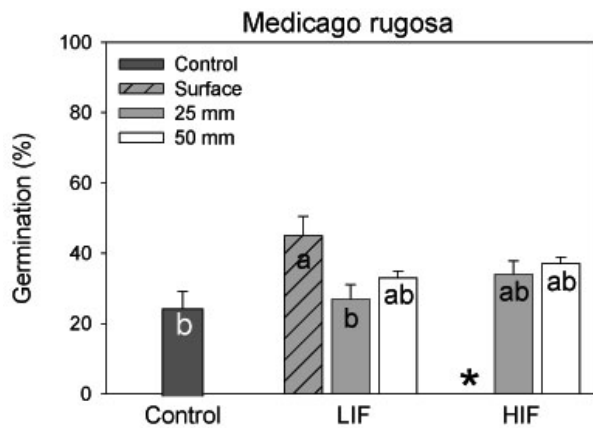


Figure 3. Total germination percentage in *M. rugosa* and in the control, at the surface and at 25 and 50 mm soil depth. HIF, high intensity fire; LIF, low intensity fire; *no germination occurs owing to the burning of seeds. Different letters indicate a significant difference for $P < 0.05$.

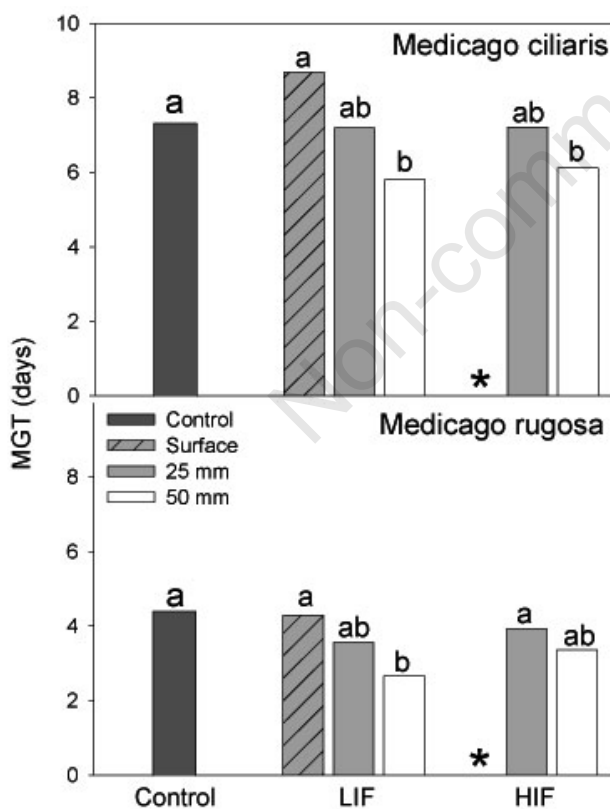


Figure 4. Mean germination time of the studied species in relation to the control, at the surface and at 25 mm and 50 mm soil depth. HIF, high intensity fire; LIF, low intensity fire. Different letters indicate a significant difference for $P > 0.05$.

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