



Effect of fire on viability and germination behaviour of *Cistus ladanifer* and *Cistus salvifolius* seeds

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Abstract Fire is a natural phenomenon that is very frequent in ecosystems of the Mediterranean basin. *Cistus ladanifer* and *C. salvifolius* (*Cistaceae* family) are evergreen shrubs and components of plant communities found in this region. The objective of this study was to evaluate the effect of fire on the germination of *C. ladanifer* and *C. salvifolius* seeds. For this purpose, seeds of both species were subjected to different heat-shock treatments simulating natural fire conditions (50–175°C for 1 and 5 min). To elucidate the ecological significance of the effects of fire, viability and different traits informative about the dynamics of the germination process, such as final germination percentage (FGP), first day of germination (FDG), last day of germination (LDG), time spread of germination (TSG) and coefficient of

velocity of germination (CVG), were determined. Seeds of both species exhibited fire-related behaviour, as they showed seed heat resistance and heat-stimulated germination. Germination in control, untreated seeds was very low, with 10 and 0% FGP for *C. ladanifer* and *C. salvifolius*, respectively. Thermal treatments significantly increased germination in both cases, with optimal results achieved at 100–125°C for 1 min and 75–100°C for 5 min. Under these conditions, greater germination percentages were obtained in *C. salvifolius* (92–95%) than in *C. ladanifer* (64–68%). Temperature proved to have a greater influence on seed viability and germination than the accumulated heat dose. The pattern of seed germination varied depending on the species and the heat shock treatments. In general, *C. ladanifer* exhibited earlier and faster germination. These characteristics, along with the high number of seeds produced per plant, make *C. ladanifer* a more competitive species for colonizing empty spaces after fire.

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Introduction

Fire is a natural phenomenon present in many ecosystems worldwide (Zupo et al. 2016). Similarly to other Mediterranean-type climate regions, such as California, the Cape region of South Africa, South

and Southwestern Australia, and Central Chile (Keeley 2012), ecosystems in the Mediterranean Basin are prone to fires (Daibes et al. 2019; Valbuena et al. 2019). They constitute one of the most predictable disturbances faced by plants (Di Castri et al. 1981). Rural exodus and the establishment of forest and shrubland on abandoned land have contributed to an increase in fire incidence in Southern Europe (Mouillot and Field 2005). Moreover, an increase in fire frequency is expected in the future as a result of climate change (Valbuena et al. 2019).

Fire causes a multitude of changes in habitat conditions, such as an increase in soil fertility (Calvo et al. 2008), greater availability of space and light, and lower seed predation risk (Hernández-Serrano et al. 2013). Fire cues and changes provoked in the environment can influence plant vital processes such as flowering, fruiting and seed germination (Zirondi et al. 2019). In consequence, fire produces alterations in plant competitive interactions (Valbuena et al. 2019), thus affecting the structure and dynamics of plant populations and communities (Whelan 1995; Bond and van Wilgen 1996).

Apart from being an important ecological factor, fire may also act as a selective pressure in fire-prone habitats (Valbuena et al. 2019). Therefore, in Mediterranean ecosystems, numerous species have acquired fire-related traits and mechanisms, which can confer an adaptive advantage in repeatedly burnt ecosystems (Saracino et al. 2017; Valbuena et al. 2019; Zirondi et al. 2019). Different strategies have been observed in plants adapted to fire regimes related to both vegetative and reproductive features. According to Valbuena et al. (2019), in these ecosystems, regeneration of plant species after fire largely depends on both their capacity to withstand high temperatures during fire and their strategy for persisting after fire. Traits that contribute to bear the fire include protection of meristems, usually located at the ground level (e.g. leaf sheaths) or below the soil surface (Zirondi et al. 2019), and the production of seeds resistant to fire, which contributes to the formation of persistent soil seed banks (Valbuena et al. 2019). In relation to regeneration after fire, plants have two basic mechanisms for subsistence in fire-prone ecosystems: by resprouting from plants that survived the fire and by recruiting new individuals from a fire-resistant seed bank (Paula and Pausas 2008). In this line, plants adapted to fire present reproductive mechanisms

linked or influenced by fire, such as flowering stimulation, opening of fruits and seeds release and breaking of seed dormancy (Zirondi et al. 2019).

The genus *Cistus* contains 25 species with a circum-Mediterranean distribution, mainly in the Western part of the region (Pérez-García and Escudero 1997). In this area, *Cistus* species are prominent components of many ecosystems such as ‘maquis’, ‘garri-gue’ or ‘jaral’ (Ferrandis et al. 1999), *Cistus ladanifer* and *Cistus salvifolius* being among the most representative species in these plant formations (Núñez-Olivera et al. 1995).

As characteristics of species originating from fire-prone ecosystems, *Cistus* species present a series of features which reflect their adaptation to this ecological factor. Most of them behave as obligate seeders (Trabaud 1970), accumulating long-lived persistent seed banks in the soil (Thanos et al. 1992), and exhibit fire stimulated germination (Ferrandis et al. 1999; Nadal et al. 2002; Scuderi et al. 2010; Tavsanoglu 2011). Increased success in burnt environments achieved by this trait has led to *Cistus* species to act as typical post-fire pioneer plants (Tavşanoğlu and Gürkan 2005). These species play a relevant role in the recolonization of burnt areas (Thanos and Georgiou 1988).

Germination is a very important developmental step in the life cycle of a plant, as it is the first step for the establishment of a new generation (Roeder et al. 2019). Therefore, proper timing of seed germination is critical for survival and proliferation of seedlings, and plants have evolved diverse strategies to ensure successful germination, including seed dormancy (Nonogaki 2014). Seed dormancy has a clear biological significance, playing an important role in the adaptation and evolution of seed plants (Nonogaki 2014).

Seeds of *Cistus* species exhibit primary dormancy (Pérez-García and González-Benito 2005; Zirondi et al. 2019), meaning the existence of a condition at seed shedding, which prevents immediate germination even under conditions usually permissive for germination (Hartmann et al. 2014). This dormancy is exogenous, as it is imposed upon the seed by factors outside the embryo (Hartmann et al. 2014), specifically by the seed coat (Murdoch and Ellis 1992; Thanos et al. 1992).

A hard seed coat is one of the main characteristics of *Cistus* seeds (Pérez-García and Escudero 1997).

This structure plays a key role in seed biology because it protects the embryo, thus making seed persistence in the soil possible (Thompson and Booth 1993). Apart from this, it constitutes an impermeable barrier to water uptake and gaseous exchange and can inhibit embryo expansion (Bradbeer 1988; Bewley and Black 1994). Consequently, the seed coat also provides an efficient mechanism of physical dormancy (Ferrandis et al. 1999) that prevents the seeds from germinating until the seed coat is broken (Baskin and Baskin 2014). Dormant seeds contribute to the formation of long-lived soil seed banks, until environmental conditions are suitable for seedling establishment.

In fire-prone ecosystems, germination of some species is stimulated by fire cues such as heat, ash, charcoal or smoke produced during fires (Riveiro et al. 2019). In this way, fire can influence the recovery of systems after burning (Riveiro et al. 2019), significantly impacting plant population dynamics (Keeley et al. 2011a; Keeley et al. 2011b).

The aim of this work was to study the effect of fire on germination of *C. ladanifer* and *C. salvifolius* seeds by subjecting them to different heat shock treatments simulating natural fires. Apart from final germination, parameters related to time, rate and synchrony were also analysed, due to the importance of germination kinetics on the species potential for post fire establishment and their consequent ecological implications. The effect on seed viability, an essential requirement for germination, was also investigated.

Material and Methods

Plant Species

Ripe fruits of *Cistus salvifolius* L. and *Cistus ladanifer* L. were collected from natural populations in the Montes de Málaga Natural Park (Málaga, Spain, approximately 36°41'40" N, 5°02'09" W, 820–915 m a.s.l.). The climate of the study area is typically Mediterranean and is classified as Csa using the Köppen classification system (Instituto de Meteorología, AEMET 2011), with a mean annual temperature of 17.5°C and a mean annual precipitation of 500 mm (Malaga Airport, meteorological station) (AEMET, Spain).

Capsule harvesting was carried out at the time of seed dispersal, from approximately ten randomly

selected individuals of each species. Seeds were removed from the capsules, pooled and kept in the dark under low temperatures (7–9°C) for several days until the start of the experiments.

Heat Shock Treatments

Fire is a very variable disturbance, being temperature and exposure time the main factors determining fire intensity (Herrero et al. 2007). To investigate the effect of fire on seed germination and viability, seeds of *C. ladanifer* and *C. salvifolius* were subjected to heat shock treatments representing different levels of fire severity (low, moderate and high intensity) found in Mediterranean ecosystems (Trabaud 1979; Bradstock and Auld 1995; Odion and Davis 2000; Tavşanoğlu et al. 2015; Valbuena et al. 2019). Specifically, seeds were heated at 50, 75, 100, 125, 150 and 175°C for two exposure times, 1 and 5 min. Untreated seeds were used as a control. For the application of dry heat treatments, seeds were spread into aluminium foil envelopes and introduced in the centre of a forced-air oven with controlled temperature (mod. SFB-400, Memmert, Germany).

Seed Viability Analysis

The effect of thermal shock treatments on seed viability was analysed using the tetrazolium test, carried out according to Witty (2012). Three replicates with ten seeds each, were analysed per species and heat treatment.

Germination Tests

Seeds subjected to heat shocks were tested for germination and their behaviour compared to that of untreated seeds. Germination was carried out under aseptic conditions to avoid contamination. For this purpose, seeds were surface sterilized by immersion in a 0.5% (v/v) sodium hypochlorite solution supplemented with Tween 20 (1 drop/100 ml) for 10 min and subsequently rinsed three times for 5 min with sterile distilled water. For germination induction, they were placed, under aseptic conditions, onto two sterile filter paper discs in 5.5 cm sterile Petri dishes containing 3 ml of deionized sterile water. Petri dishes were sealed with Parafilm™ and incubated in a growth chamber at 25 ± 1°C under a 16/8 h light/

dark photoperiod ($40 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ irradiance level), provided by GroLux lamps (Sylvania, Erlangen, Germany), conditions that had been previously shown to be adequate for seed germination of different *Cistus* species (Sánchez-Romero, personal communication).

Five independent replicates of ten seeds each were carried out per treatment and species. Seeds were considered germinated when shoot and/or root elongation was ≥ 2 mm. Data on germinated embryos were recorded periodically. To keep the seeds moistened, additional distilled water was added to the filter papers when needed.

To investigate the ecological significance of the effect of fire, different parameters were determined according to Kader (2005): (1) final germination percentage (FGP): the final number of seeds germinated in a seed lot $\times 100$; (2) first day of germination (FDG): the day on which the first germination event occurred; (3) last day of germination (LDG): the day on which the last germination event occurred; (4) time spread of germination (TSG): the time in days between the first and last germination event in a seed lot (therefore, the smaller TSG, the more synchronous germination in a seed lot); (5) and coefficient of velocity of germination (CVG): estimation of germination speed:

$$\text{CVG} = \frac{\sum_{i=1}^k ni}{\sum_{i=1}^k niti} \times 100,$$

where: ni = number of seeds germinated each day, ti = number of days from seeding corresponding to n .

Statistical Analysis

Data of viability and the germination parameters were analysed by three-way ANOVA in order to test the effects of the predictor variables species, temperature and exposure time. When a statistically significant influence of the species was found, the effect of the investigated fire cues (temperature and exposure time) were analysed separately for *C. ladanifer* and *C. salvifolius* by two-way ANOVA. Following ANOVA, significant differences among treatments were determined by the least significant difference (LSD) post hoc test.

To analyse the effect of the accumulated heat dose received by the seeds, discrete time and temperature combinations were converted into a 'heat

index' (Wiggers et al. 2017). According to Paula and Pausas (2008), the heat index was calculated by multiplying the temperature ($^{\circ}\text{C}$) by the natural logarithm of the exposure time (min) plus one.

To investigate the possible relationship between the germination performance and different aspects of germination kinetics, the degree of correlation between variables was estimated by using Pearson's coefficient. To study dependence of one variable upon another, curvilinear regression analyses were carried out.

The significance level used was 0.05 in all cases. Statistical analysis was performed using SPSS 24.0 (IBM SPSS Statistics, USA).

Results

Seed Viability

As revealed by the tetrazolium test, all seeds of *C. ladanifer* and *C. salvifolius* used in this investigation were viable before exposure to heat shock treatments (Figs. 1a, 2a, b). Thermal treatments had a significant influence on seed viability, with both species showing the same vulnerability (Fig. 1a). Seeds of *C. ladanifer* and *C. salvifolius* tolerated exposure up to 125°C for 1 min, but the application of higher temperatures for the same time caused the death of all seeds in both species (Fig. 2c, d). Similarly, seed viability virtually did not change when seeds were treated with temperatures up to 100°C for 5 min. However, it drastically decreased to 6.67 and 10% for *C. ladanifer* and *C. salvifolius*, respectively, after 5 min at 125°C . Seeds treated for 5 min at 175°C appeared completely burnt (Fig. 2e, f). As revealed by three-way ANOVA (Table S1), exposure time, temperature and the interaction time \times temperature exhibited a significant influence on seed viability. No significant effects were found related to the species or its interaction with the other independent variables.

Representation of viability as a function of the heat index revealed very different results for similar heat indices, thus revealing an important effect of the temperature (Fig. 3a). Accordingly, a strong negative correlation was observed between seed viability and temperature ($r = -0.812$, $P = 0.000$).

Fig. 1 Viability percentage (a) and germination parameters of *C. ladanifer* (○) and *C. salvifolius* (●) seeds subjected to different heat shock treatments: final germination percentage (b), first day of germination (c), last day of germination (d), time spread of germination (e), and coefficient of velocity of germination (f). Different letters indicate significant differences by the LSD test with a significance level of 0.05.

Seed Germination

Germinability was very low in control seeds, with 10 and 0% FGP for *C. ladanifer* and *C. salvifolius*, respectively (Fig. 1b). Heat shock treatments significantly affected final germination, showing a bell-shaped response to the treatments (1 and 5 min) in both species (Fig. 1b). A minimum temperature was required in order to significantly improve final germination. Thus, 50°C did not significantly modify seed germination, independently of the exposure time, 1 or 5 min, but positive results could be observed with temperatures from 75°C onwards. Negative effects due to treatment severity were evident after incubation at 100–150°C, depending on the species and the exposure time. Nevertheless, significant differences were observed between *C. ladanifer* and *C. salvifolius* seeds. Whereas for *C. ladanifer* optimal heat treatments were 100°C for 1 min and 75°C for 5 min, *C. salvifolius* presented a wider optimum response with better results obtained at 100–125°C for 1 min and 75–100°C for 5 min. FGPs achieved in both species were significantly different with higher percentages obtained in *C. salvifolius* (92–95%) than in *C. ladanifer* (64–68%). Three-way ANOVA results revealed a significant influence of all the independent variables tested (species, temperature and exposure time) as well as of their respective interactions, except species × exposure time (Table S1).

Similarly to these findings for viability percentage, representation of FGP in function of heat index (Fig. 3b) revealed a determinant effect of temperature. Nevertheless, taking into consideration only those heat indices which induce positive effects, a strong relationship between the accumulated heat dose and the FGP of treated seeds could be observed (Fig. S1).

Germination Dynamics

The pattern of seed germination varied depending on the species and the heat shock treatments as revealed by the germination parameters analysed (Fig. 1c–f,

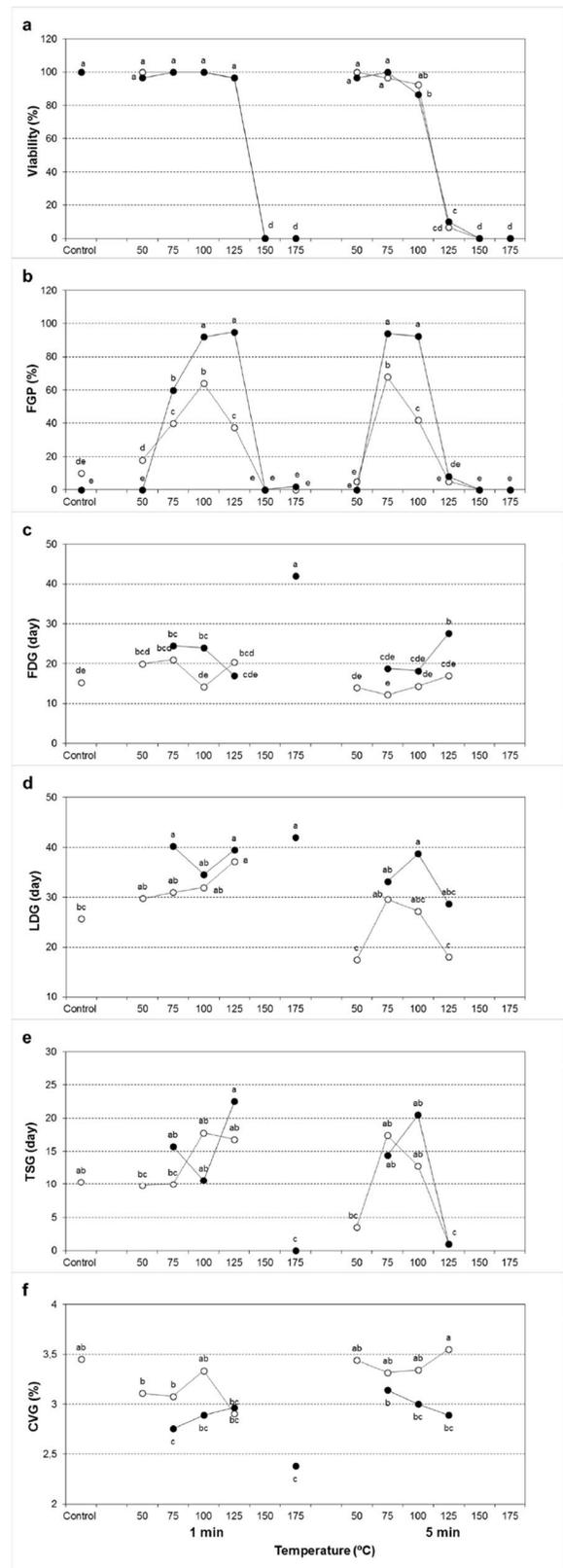


Fig. 2 Tetrazolium test executed in *C. ladanifer* (a, c, e) and *C. salvifolius* seeds (b, d, f). Seeds a, b – untreated; c, d – heat-treated at 150°C for 1 min; and e, f – heat-treated at 175°C for 5 min.

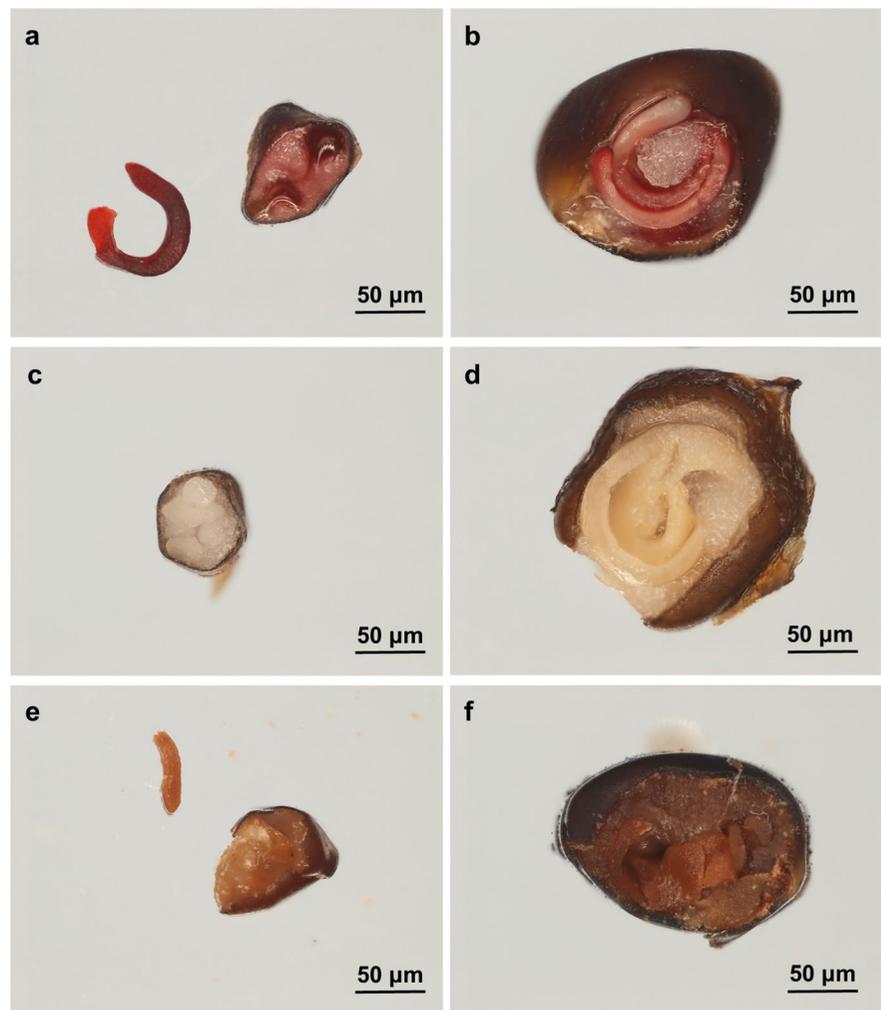


Table S1, S2). Initiation of germination was earlier in *C. ladanifer* than in *C. salvifolius* (Fig. 1c). While, on average, germination in *C. ladanifer* started after 16.65 days, 24.60 days were required to start germination in *C. salvifolius*. Although three-way ANOVA results revealed a significant influence of the temperature on the initiation of germination (Table S1), this effect was restricted to *C. salvifolius*, where a significant interaction of exposure time \times temperature could be observed (Table S2). Time at which the last germination event occurred was also significantly different among the treatments (Fig. 1d). As revealed by three-way ANOVA (Table S1), this germination parameter varied significantly depending on the species. In *C. ladanifer*, the last seeds germinated 27.79 days after the initiation of the experiment, whereas seeds of

C. salvifolius continued germinating for up to 36.71 days. The effect of exposure time and the interaction exposure time \times temperature only was statistically significant in *C. ladanifer* (Table S2). In general in this species, exposing to heat shock treatments for longer times significantly reduced the time at which the last count was attained. The length of time elapsed between the first and the last germination event did not present any significant difference between the species (Fig. 1e, Table S1). Duration of germination only was significantly influenced by the interaction exposure time \times temperature.

Speed of germination was significantly affected by the species (Fig. 1f, Table S1). Faster germination was observed in *C. ladanifer*, with an average CVG value of 3.26 compared to 2.86 in *C. salvifolius*. Speed of germination also varied depending

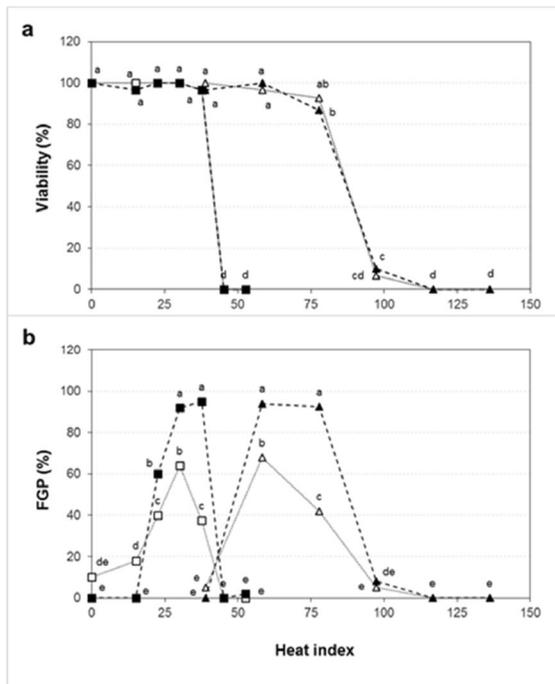


Fig. 3 Viability (a) and final germination percentages (b) of *C. ladanifer* (□—1 min, △—5 min) and *C. salvifolius* (■—1 min, ▲—5 min) seeds subjected to different accumulated heat doses, expressed as heat indexes. Different letters indicate significant differences by the LSD test with a significance level of 0.05.

on the exposure time and the interaction species \times exposure time \times temperature. CVG significantly increased when thermal treatments were applied for longer times, from 2.93 in 1-min treatments to 3.24 in 5-min treatments. This time effect was statistically significant in both species (Table S2).

Correlation analysis revealed interesting associations between germination parameters (Table S3, S4). In both species, FGP was negatively correlated with FDG and positively correlated with TSG, meaning that higher germination percentages were associated to early and extended germination. Nevertheless, some differences could be observed in the germination dynamics of *C. ladanifer* and *C. salvifolius*. While in *C. ladanifer* final germination was positively correlated with LDG, in *C. salvifolius* higher germination percentages were associated to faster germination, that is, to higher CVG values.

Discussion

In both species, seed viability presented a similar response to heat-shock treatments, tolerating thermal shocks of moderate severity (Tavşanoğlu et al. 2015). A strong heat resistance has been found in *Cistus* species (Pérez-García and Escudero 1997). The observed tolerance to high temperatures in species of the *Cistaceae* family has been associated with the protection of their lignified seed coat (Ferrandis et al. 1999). Nevertheless, other factors, such as the functional type, phylogenetic relatedness and species microhabitat may also contribute to prevent the death of the embryo by fire (Zirondi et al. 2019).

No or very low germination was obtained in control seeds of *C. ladanifer* and *C. salvifolius*. In *Cistus*, the main reason for no germination of untreated seeds appears to be the presence of a primary dormancy imposed by the seed coat (Thanos and Georghiou 1988; Nadal et al. 2002). The hardness and water impermeability of this covering structure constitutes a physical barrier to water uptake, thus preventing the initiation of the germination process (Pérez-García and Escudero 1997; Nadal et al. 2002).

Nevertheless, some germination capacity in control seeds has been previously reported in *Cistus* species (Thanos and Georghiou 1988; Nadal et al. 2002). This could be explained by seed population heterogeneity in relation to coat hardness, specifically to the existence of a small percentage of seeds with soft, thin or ruptured seed coats, which are therefore ready to germinate (Thanos and Georghiou 1988).

Thermal treatment stimulated germination in both species with optimal germination results at moderate or high heat intensities: 100°C and 100–125°C for 1 min or 75°C and 75–100°C for 5 min, for *C. ladanifer* and *C. salvifolius*, respectively. Similar temperatures have been reported during a fire at depths of the first few centimetres in the soil (Giovannini 1994; Penman and Towerton 2008).

Stimulation of seed germination by thermal treatments is a common effect on species with hard-coated seeds (Riveiro et al. 2019) and they have demonstrated to be effective in different *Cistus* species (Ferrandis et al. 1999; Nadal et al. 2002; Scuderi et al. 2010; Tavşanoğlu 2011). Similar heat shock requirements have been previously reported in *C. creticus* and *C. ladanifer*, with optimum

conditions ranging 90–150°C, for 5–15 min (Delgado et al. 2008; Tavsanoğlu 2011; Silva Dias et al. 2019).

The amplitude of the optimum response also has ecological significance. Presenting optimal response at a wider range of temperatures, as found in *C. salvifolius*, is an important adaptive advantage. Fire is a phenomenon which is extremely variable and heterogeneous, producing a broad variety of heat loads (Silva Dias et al. 2019). Therefore, species exhibiting a wider ‘window’ of germinability can respond to fire in a more flexible way (Silva Dias et al. 2019). The production of seeds with different requirements for breaking physical dormancy is one of the most important survival strategies for species growing under unpredictable environmental conditions (Nadal et al. 2002). In the case of fire, this allows seeds to germinate in a wide range of heat shock conditions.

Similarly to viability, a more determinant effect of the temperature compared with the heat index was observed in FGP. According to the results obtained, a minimum temperature is required for inducing germination. In the present investigation, 75°C appears to be this threshold temperature. Similarly, in *Halimium lasianthum*, Valbuena et al. (2019) found that light temperatures, such as 60°C for 5 min, were ineffective in promoting germination.

It is generally assumed that high temperatures act as a scarifying agent that breaks physical dormancy by cracking the seed coat (Navarro-Cano et al. 2009). Mechanical fracture mainly affects the palisade layer of macrosclereids, with strongly lignified cell walls. This makes the seed coat permeable to water, thus providing a way for water entry and germination onset (Nadal et al. 2002). Temperature thresholds therefore probably represent the minimum required for inducing physical changes leading to seed coat rupture. Nevertheless, other processes, such as induction of physiological processes and denaturing of seed coat inhibitors, may also be involved in releasing dormancy (Paula and Pausas 2008; Silva Dias et al. 2019).

Germination decline above optimal conditions presented interesting differences between *C. ladanifer* and *C. salvifolius*. Whereas in *C. salvifolius* failure of germination could be explained by embryo loss of viability, in *C. ladanifer*, additional processes must be involved, as germination declined at heat shock treatments without detrimental effects on seed survival.

Germination dynamics have important ecological implications due to their influence on interspecific competition for space or natural resources such as light, water and nutrients. Apart from stimulating germination, fire can affect germination kinetics, thus provoking changes in competition hierarchy and, consequently, altering the stand structure (Zirondi et al. 2019).

As found by Silva Dias et al. (2019) in *C. ladanifer*, germination patterns were affected by heat shocks in both *C. ladanifer* and *C. salvifolius*, although some of the germination parameters were differently influenced.

Earlier germination was in general observed in *C. ladanifer* as compared with *C. salvifolius*. However, differently to *C. salvifolius*, in *C. ladanifer* FDG was not affected by heat. Nonetheless, Silva Dias et al. (2019) found in *C. ladanifer*, a trend for a faster initiation of germination with increasing temperatures in heat treatments below 100°C. In species from other than *Cistaceae*, Riveiro et al. (2019) observed that the length of this period appears to increase proportionally to the intensity of treatment. For species overlapping habitat niches, earlier germination give an advantage compared to gradually recruiting species, and due to its important influence on colonization of burnt spaces, a strong selection pressure for early germination is expected (Delgado et al. 2008). Seed germination also ends earlier in *C. ladanifer* than in *C. salvifolius*. Therefore, no significant differences could be observed in both species in relation to the length of germination, despite a short germination period can also be a very important ecological advantage when occupying an empty space (Grime 2006; Arán et al. 2013). Contrarily to the results reported by Silva Dias et al. (2019), in *C. ladanifer*, the time necessary to attain final germination was significantly affected by heat shock.

Germination speed is an important trait, with decisive implications in plant regeneration after a fire event (Grime 2006; Arán et al. 2013). Differences in germination velocity have been reported in species of the *Cistaceae* family (Navarro-Cano et al. 2009). Higher germination speeds were in general found in *C. ladanifer* compared with *C. salvifolius*. The influence of thermal treatments on this parameter was species dependent, as previously observed by Navarro-Cano et al. (2009) and Delgado et al. (2008). Nevertheless, in both cases the germination

speed increased when heat treatments were applied for longer times.

As previously described in *C. ladanifer* (Silva Dias et al. 2019), in both species, earlier germination was associated with higher final germination rates. Germination duration also contributed to attain higher germination percentages. Nevertheless, some particularities were found related to their germination pattern. Thus, while in *C. ladanifer* higher germination was achieved by prolonging the germination process, in *C. salvifolius*, it was associated to a higher germination speed.

In conclusion, the results obtained reveal that both species exhibited fire-adaptive traits, such as seed heat resistance and heat-stimulated germination (Kealey et al. 2011b), which favour seedling establishment after a fire event. *Cistus* species are considered the most common post-fire colonizers in the Mediterranean basin (Tavşanoğlu and Gürkan 2005). However, according to Ferrandis et al. (1999), this does not mean that these species are exclusively dependent on fire. Under natural conditions, in the absence of fire, seed coat of *Cistus* species could be weakened by different ecological processes, such as microbial degradation or changes in soil humidity and temperature (Baskin and Baskin 2014). Nevertheless, interesting differences were found in the germination response to fire of *C. ladanifer* and *C. salvifolius* seeds. Apart from slight differences in their optimal heat shock treatments, important variations were found in their germination kinetics, which can play a determinant role in plant regeneration after fire. Although lower FGP was achieved in *C. ladanifer* compared to *C. salvifolius*, germination initiated faster and occurred at a higher speed. These features, along with the high number of seeds produced per plant (Chamorro et al. 2016), make *C. ladanifer* a very competitive species for occupying burnt areas, thus explaining its consideration as a typical pyrophyte species.

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