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## Effect of high temperatures on seed germination of *Pinus sylvestris* and *Pinus halepensis*

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### Abstract

Fire is one of the most important ecological factors in the Mediterranean forest ecosystems. Most Mediterranean conifers are obligate seeders after fire. Seeds of two conifers species (*Pinus sylvestris* and *Pinus halepensis*) were heated to a range of temperatures similar to those registered on surface soil during natural fires (from 70° to 190°C) and a range of exposure times (from 1 to 5 min). Temperatures above 150°C have a negative effect on the germination of both species. With a temperature range from 70° to 130°C there is no difference between this treatment and the control. The increase in temperature and, of course, the longer exposure, generally decrease germination percentages in comparison with the control and this is much more marked in the case of *Pinus sylvestris*. © 2000 Elsevier Science B.V. All rights reserved.

**Keywords:** Fire; Heat treatment; *Pinus halepensis*; *Pinus sylvestris*; Seed germination

### 1. Introduction

Fire is an ecological factor which plays an important role in the distribution, organisation and evolution of Mediterranean ecosystems (Trabaud, 1980). Thus, fire can be considered a basic component of Mediterranean ecosystems. Intensity is one of the most significant characteristics of the disturbances and this is especially significant in the case of fire (Malanson, 1984; Sousa, 1984). Two factors characterise fire intensity: exposure time and temperature reached. Both factors have a considerable influence on plant community recovery capacity. Recovery of a burned area is accomplished by using two main strategies: (1)

vegetative reproduction from subterranean organs like bulbs or rhizomes, in species like *Quercus pyrenaica* (Calvo et al., 1991) or *Erica australis* (Calvo et al., 1998); or (2) sexual reproduction and seed liberation on the spot where the fire occurred in species of the *Pinus* and *Cistus* genera (Alonso et al., 1992; Tárrega et al., 1995; Valbuena et al., 1992).

Most Mediterranean conifers, excluding *Pinus canariensis* and several *Juniperus* taxa, are obligate seeders after a disturbance (Escudero et al., 1997). When a wildfire occurs, cones open and trees find an opportunity for natural regeneration (Walter, 1973). *Pinus sylvestris* (Toole, 1973) and *Pinus nigra* (Orlandini and Malcoste, 1972) seeds are normally stimulated by light. *Pinus halepensis* and *Pinus pinaster*, two common Iberian pines, have been characterised as typical pyrophytes, which regenerate well after a fire

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(Trabaud and Oustric, 1989a; Castro et al., 1990). However, several authors have recently noted that both pines are not real pyrophytes (Martínez-Sánchez et al., 1995), as they are not positively stimulated by high temperatures like other species.

Recovery in other pine species seems to be favoured by fire: *Pinus brutia* and *Pinus leucodermis* (Borghetti et al., 1989; Saracino and Leone, 1991; Thanos and Marcou, 1991; Trabaud and Campant, 1991). Pines are characterised by high seed production, dissemination by anemochory and heliophyllous germination. These characteristics favour pine germination and installation in areas opened up by fire.

Many Mediterranean shrub species need to be exposed to high temperatures for a certain period of time in order to germinate or, at least, their germination is stimulated under these conditions (Trabaud and Oustric, 1989b; Tárrega et al., 1992; Valbuena et al., 1992; Trabaud, 1995). Fire plays an important role in dormancy rupture in other seeds, mainly woody ones. In these cases, fire can act as a scarifying agent on the seed cover as in the case of *Pinus brutia* (Thanos et al., 1989). In addition, certain populations require periodic fires to maintain their position in the ecosystem and the role of fire has been recognized in maintaining species such as *Pinus longifolia* (Greswell, 1926), *Pinus palustris* (Chapman, 1946), *Pinus ponderosa* (Cooper, 1961) and *Pinus halepensis* (Trabaud, 1989).

Scots and Aleppo pines are some of the most important conifers in Spain, covering over 900 000 and  $1.1 \times 10^6$  ha, respectively. The surface covered by pine species is one of the most affected by wildfire in Castilla and León among different woodland areas (Del Hierro, 1998 personal communication). Increasing temperatures during wildfires can affect seed germination by pine species. This paper analyses the germination rate of pine seeds of *Pinus halepensis* and *Pinus sylvestris* seeds heated to different temperatures for different exposure times.

## 2. Materials and methods

### 2.1. Seeds origin

The *Pinus sylvestris* and *Pinus halepensis* seeds used were obtained from the Centre for Forestry Improvement 'El Serranillo' (General Office for Nature Conservation, Ministry of the Environment). They

came from 'La Alcarria' in the case of *Pinus halepensis* ( $3^{\circ}15'W-2^{\circ}13'W$  longitude and  $41^{\circ}05'N-42^{\circ}40'N$  latitude) and 'Montaña Soriano-Burgalesa' ( $2^{\circ}20'W-3^{\circ}15'W$  longitude and  $41^{\circ}40'N-42^{\circ}40'N$  latitude) in the case of *Pinus sylvestris* and were harvested in 1994/1995. The seeds were stored in opaque paper bags at a constant temperature ( $20 \pm 2^{\circ}C$ ) until the experiment.

### 2.2. Experimental design

Seeds were exposed to high temperature for short periods to simulate the effects of fire (Trabaud and Casal, 1989; Tárrega et al., 1992).

Combinations of seven different temperatures ( $70^{\circ}$ ,  $90^{\circ}$ ,  $110^{\circ}$ ,  $130^{\circ}$ ,  $150^{\circ}$ ,  $170^{\circ}$  and  $190^{\circ}C$ ) and two exposure times (1 and 5 min) were studied together with a control (no treatment). Five replicates of 20 seeds each were used for each treatment and placed in Petri dishes. Germination was carried out in a germinator under constant conditions at  $21^{\circ}C$  in white light for 14 h and at  $17^{\circ}C$  in darkness for 10 h. They were kept like this for six weeks. Seeds were considered to have germinated when the radicle had grown 1 mm out of the tegument (Come, 1970).

The experimental design was completely randomised. The original data: the number of germinated seeds per sample was transformed so that distribution will be normal and the variances homogeneous. The transformation used was the arcsine of the square root of the proportion of germinated seeds per sample (Sabin and Stafford, 1990).

After carrying out an analysis of variance (ANOVA), the Duncan test was used to detect significant differences among the treatments with a probability of 95% ( $\alpha = 0.05$ ). Finally, orthogonal contrasts were used to detect significant differences between the exposure times and the control and between the temperatures that the Duncan test indicated as doubtful regarding their influence and the control. The temperatures thus studied were  $130^{\circ}$  and  $150^{\circ}C$  in the case of *Pinus halepensis* and  $110^{\circ}C$  in that of *Pinus sylvestris*.

## 3. Results

Fig. 1 shows that the maximum germination value for *Pinus halepensis* is obtained at a high temperature,

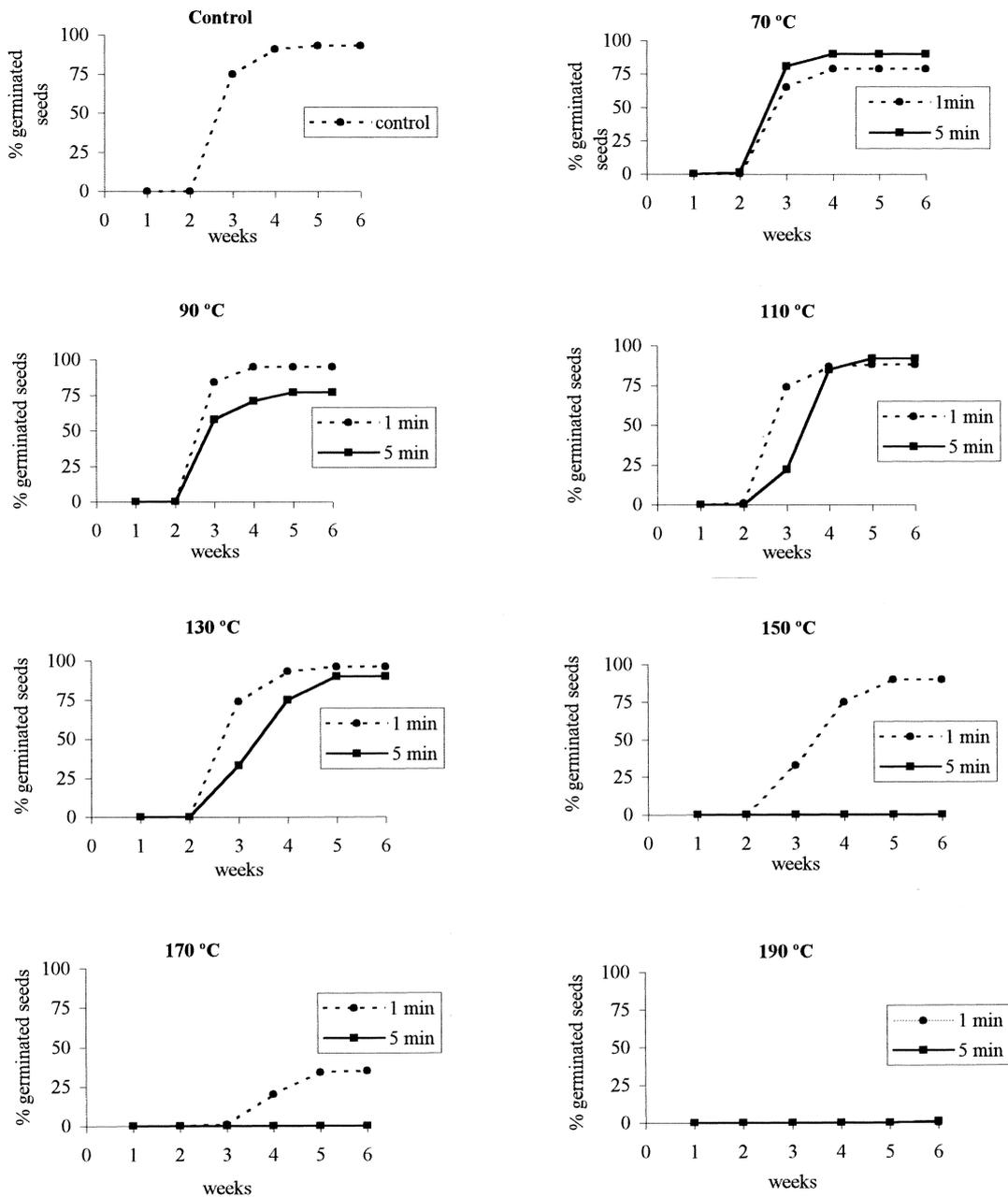


Fig. 1. Effects of temperature and exposure time on *Pinus halepensis* germination.

130°C, and short exposure time, 1 min. Only this treatment surpasses the control germination percentages but without significant differences. However, there are significant differences between control and temperature over 150°C. Increasing exposure time to

5 min produces a significant decrease in the number of seeds germinated. Germination is negatively affected from 130°C after 5 min to 170°C after 1 min and totally inhibited when subjected to 150°C after 5 min and 190°C after 1 min or longer.

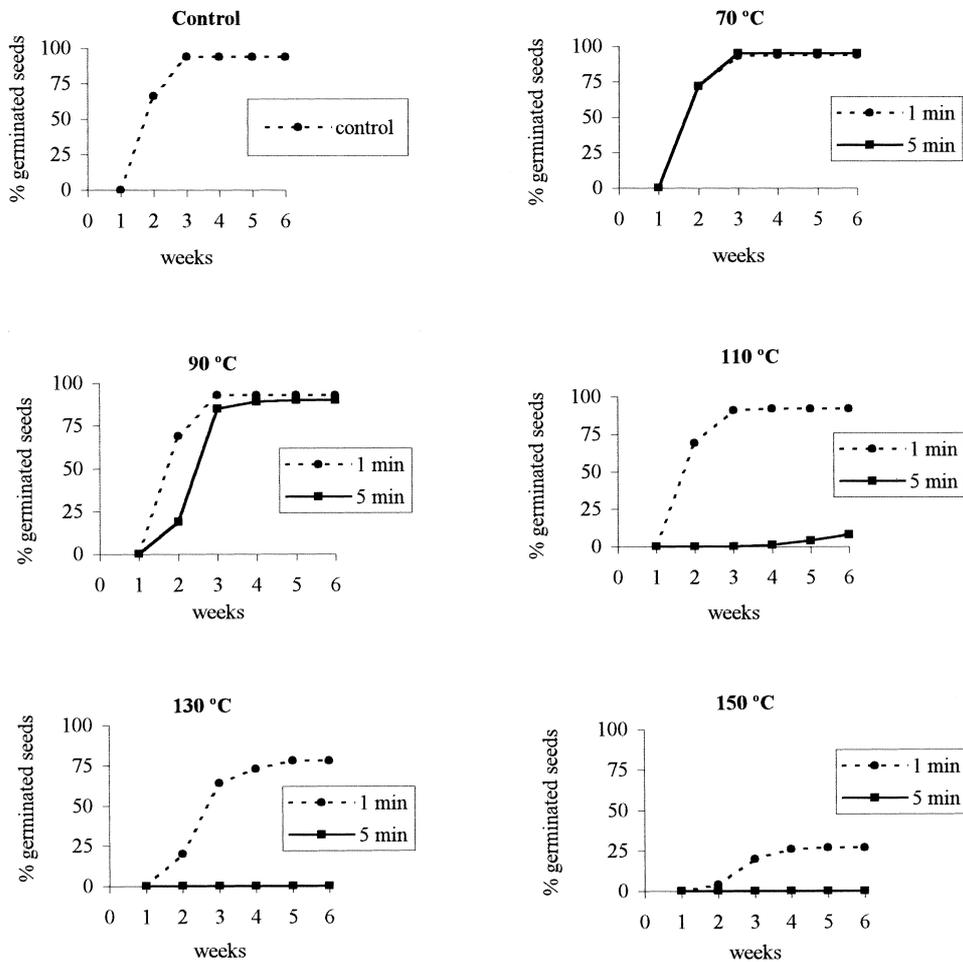


Fig. 2. Effects of temperature and exposure time on *Pinus sylvestris* germination.

At temperatures of 150°C and over *Pinus halepensis* seeds are more sensitive to exposure time than temperature; germination is nil for the three highest temperatures and 5 min exposure, while germination decreases as temperature increases with 1 min exposures. However, when the temperature is below 150°C, germination behaviour depends on temperature as much as exposure time. Germination decreases at 90° and 130°C when exposure time increases. It increases at 70°C when exposure is increased. Exposure time has no influence at 110°C.

In the case of *Pinus sylvestris* (Fig. 2) maximum germination is reached at 70°C and 5 min exposure, with percentages similar to those for the control (no

significant differences were found). As in the case of Aleppo pine high germination values are obtained at temperatures below 130°C after 1 min.

Exposure times have a significant influence on germination of Scots pine. This species is more sensitive to high temperatures than *Pinus halepensis*. Germination is nil at temperatures of 130° and 150°C if exposure time is equal to 5 min and at over 170°C independently of exposure time. Exposure time has no influence at 70° and 90°C.

The analysis of variance (Tables 1 and 2) shows that there are significant differences in germination between the different thermal treatments in the case of both *Pinus halepensis* and *Pinus sylvestris*. A

Table 1  
Analysis of variance of the *Pinus halepensis* germination results<sup>a</sup>

Source	g.l.	Sum of squares	Probability > F
Model	14	22.69849	0.0001
Error	60	1.29686	
Total	74	23.99536	
<i>Contrast</i>			
1 min vs. control	1	1.16462616	0.0001
1 min vs. 5 min	1	0.02191049	0.3181
5 min vs. control	1	1.32984584	0.0001
130°C vs. control	1	0.00009479	0.9474
150°C vs. control	1	3.76835888	0.0001

<sup>a</sup> The design is completely aleatory and the variable has been transformed (see text).

decrease in germination was observed in both species at high temperatures (150° and 110°C, respectively) and 5 min exposure.

Table 3 shows the groups separated by the Duncan test. The orthogonal contrasts show that no significant differences exist between the 1 and 5 min thermal treatments in the two species, although the probability levels are different (0.3181 for *Pinus halepensis* and 0.0116 for *Pinus sylvestris*). No significant differences were detected between the control and the thermal treatments equal to or below 130°C in the tests with *Pinus halepensis*, but they were between the control

Table 2  
Analysis of variance of the *Pinus sylvestris* germination results<sup>a</sup>

Source	g.l.	Sum of squares	Probability > F
Model	14	28.69766	0.0001
Error	60	0.89309	
Total	74	29.59076	
<i>Contrast</i>			
1 min vs. control	1	2.19251163	0.0001
1 min vs. 5 min	1	0.10088209	0.0116
5 min vs. control	1	2.68803541	0.0001
110°C vs. control	1	1.50367387	0.0001

<sup>a</sup> The design is completely aleatory and the variable has been transformed (see text).

and the 150°C treatments, due to the strong inhibitory effect this temperature has on germination. These results confirm a greater resistance to fire on the part of *Pinus halepensis* seeds. Significant differences are only detected between the control and thermal treatments over 110°C in the case of *Pinus sylvestris*. Due to the fact that there is a marked decrease in germination as a result of the inhibitory effect of high temperatures or long exposure times, it can be stated that there are no significant differences in comparison with the control in the case of the thermal treatments with a high germination value. It can be seen that the

Table 3  
Groups defined by the Duncan test for *Pinus halepensis* and *Pinus sylvestris*

Treatment	<i>Pinus halepensis</i> <sup>a</sup>				<i>Pinus sylvestris</i> <sup>a</sup>				
	A	B	C	D	A	B	C	D	E
Control	*	*			*				
70°C		*			*				
5 min	*	*			*				
90°C					*				
5 min		*			*				
110°C		*			*				
5 min	*	*						*	
130°C	*	*				*			
5 min			*						*
150°C	*	*					*		
5 min				*					*
170°C			*						*
5 min				*					*
190°C				*					*
5 min				*					*

<sup>a</sup> The groups with the same letter are not significantly different (group A has the highest number of germinated seed mean and group E the lowest).

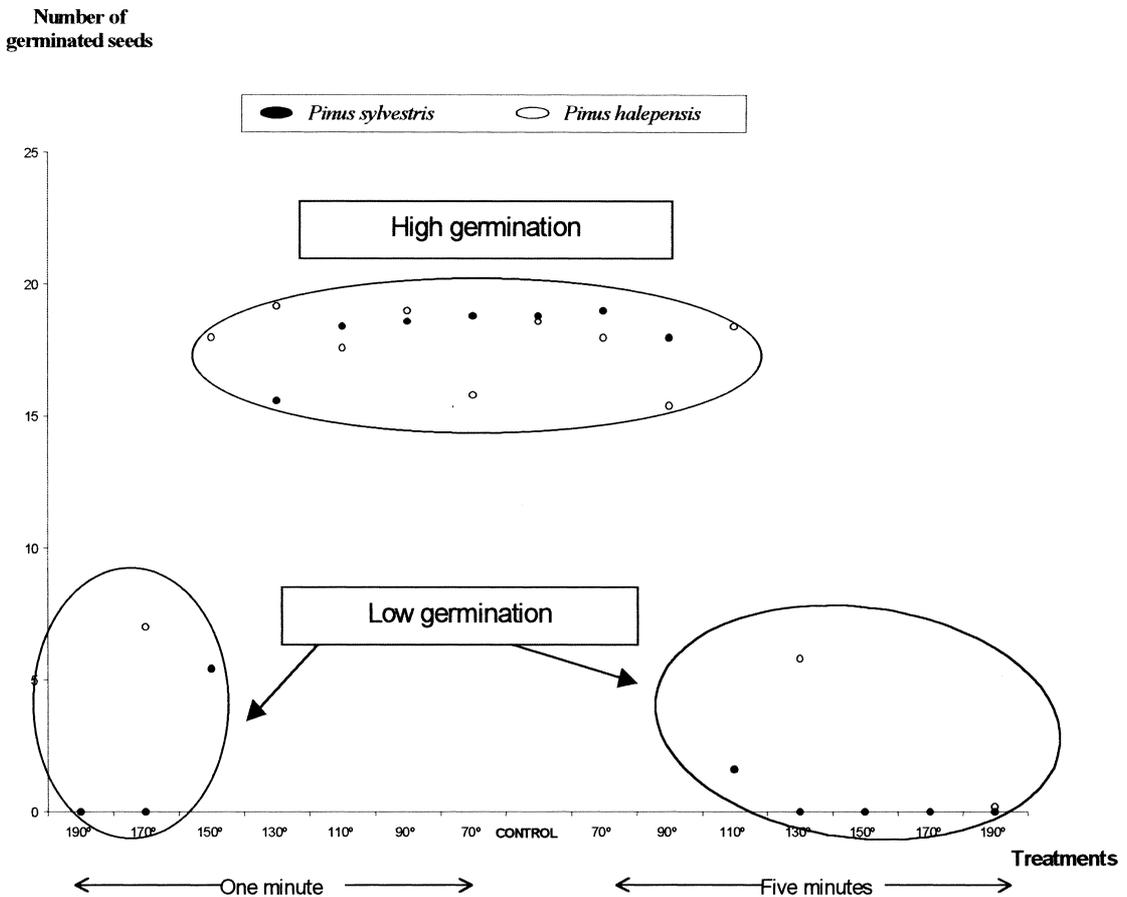


Fig. 3. Seeds germinated according to treatments and species. The Y-axis represents the number of germinated seeds and the X-axis the treatments. In the centre of this axis we can see the control, on the left of the control there the different temperatures when time exposure is equal to 1 min and on the right of the control the different temperatures when time exposure is equal to 5 min.

decrease in germinated seeds in the case of *Pinus sylvestris* occurs at lower temperatures than in the case of *Pinus halepensis*, which indicates that the former is more sensitive to temperatures above 130°C.

Fig. 3 shows the influence of time exposure and temperature on the number of germinated seeds. The Y-axis represents the number of germinated seeds and the X-axis the treatments. In the central of this axis we can see the control, on the left of the control are the different temperatures when time exposure is equal to 1 min and on the right of the control the different temperatures when time exposure is equal to 5 min). Exposure time reinforces the effect of temperature on germination. So the decrease in germination rates occurs at lower temperatures when exposure is

5 min than when it is 1 min. As can be seen the ellipse on the right is larger than the one on the left.

#### 4. Discussion and conclusion

In Spain, Scots pines grow in mountainous areas, normally between 1000 and 2000 m asl. In these areas the climate is continental with a short period of aridity and a great difference between average temperatures in summer (20°C) and winter (7°C). Mean rainfall in summer is over 200 mm. In addition, soils are both siliceous and calcareous. Aleppo pines grow on a wide range of soil conditions, frequently on calcareous ones, from sea level to 1600 m. The climate is arid

with severe droughts in summer. Average minimum temperature is  $-3^{\circ}\text{C}$  and annual rainfall are between 400 and 1000 mm.

Dissemination of the mature seeds of *Pinus sylvestris* coincides with the end of spring and lasts throughout the summer (Vega, 1977). Natural dissemination is after a cone maturation period of two years. An increase in environmental temperature is absolutely necessary for cones to open (Francelet, 1970). However, the availability of the seed for germination is not the same in all the species, either in time or in space.

*Pinus halepensis* can keep the seed in its pinecone for several seasons (Vega, 1977) as occurs with *Pinus radiata* (Reyes and Casal, 1995), *Pinus banksiana* (Chandler et al., 1983) and *Pinus brutia* (Lotan, 1975), which only open after a fire and thus ensure regeneration (Reyes and Casal, 1995).

The increase in temperature and, of course, the longer exposure generally decrease germination percentages in comparison with the control and this is much more marked in the case of *Pinus sylvestris*. Some authors studying different species found similar behaviour to that recorded in this study in certain cases. Using *Pinus halepensis* seeds, Trabaud and Oustric (1989a) observed that high temperatures lowered germination with respect to the control and the same occurred with *Pinus contorta* (Knapp and Anderson, 1980), *Rosmarinus officinalis* (Trabaud and Casal, 1989) and *Cytisus multiflorus* (Añorbe, 1988). However, there is a large group of species whose germination is favourably influenced by high temperatures, especially in Mediterranean areas: *Cistus* sp (Trabaud and Oustric, 1989b; Valbuena et al., 1992), *Genista florida*, *Cytisus scoparius* (Tárrega et al., 1992), *Ulex europaeus* (Pereiras, 1984).

The results of this study suggest that high temperatures do not increase the germination of *P. halepensis* and *P. sylvestris*. So the seedlings found in Scots and Aleppo pine stands after fire do not come from a positive influence of forest fire on germination. After a forest fires the high germination of pines is due to two factors: opening of the pinecones or the preparation of an appropriate seedbed. These results coincide with the conclusions arrived by other authors (Trabaud, 1987; Reyes and Casal, 1995; Martínez-Sánchez et al., 1995). *P. sylvestris* seeds are more sensitive to external factors and, in the case of a moderately severe fire, lose

their germinative capacity more rapidly than *P. halepensis*.

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