2.1 = Warmer temperatures determine the germination window of *Platanus orientalis* L.



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Recent anthropogenic climate change is disrupting natural environmental patterns, which is affecting species' distributions and life history events, putting natural populations at risk and creating challenges for biodiversity conservation (1). The most threatened species are those with restricted ecology. This might be the case of wetland species in the Mediterranean bioregion.

Platanus orientalis L. is a long-lived tree distributed from the central Mediterranean to the Himalaya (2; Fig. 1) that occupies lowland riparian forests, a geographically restricted ecological niche within its distribution (Habitat Natura 2000 code 92C0). P. orientalis is currently in decline in Europe, where it is considered vulnerable due to habitat loss, fungal infection by Ceratocystis fimbriata Ellis & Halsted f. sp. platani (Walter) and ongoing climate change (3).

The aim of our work was to better understand seed germination behavior of Sicilian populations from the Iblei and Peloritani Mountains (Fig. 2) at the western range edge of P. orientalis in Europe. We recently observed that the number of individuals of P. orientalis in natural populations has significantly declined in the last few decades, and seedling recruitment is very low in almost all monitored locations. Here, we conducted a series of experiments to explore the effects of temperature and light on seed germination of *P. orientalis*.



Figure 1 - Distribution map of *Platanus orientalis* (from Wazen & Fady, 2016)

Figure 2 - a) *Platanus orientalis* in its natural habitat; b) fruits dispersed by water

Experiments started two weeks after seed collection and tested germination performance at constant temperature (5°C increments from 5-35°C), either with 12/12h light/dark or full darkness photoperiod.

The results allowed us to exclude the presence of primary dormancy. Seeds are dispersed between late winter and early spring and are immediately germinable, but require temperatures above 10°C, with almost complete thermo-inhibition of germination below 15°C, either in light and dark conditions. In the light, final germination percentage reached 90% in the optimal temperature range (20 to 30°C) and remained high at temperatures either side of the optimal range (around 80% at both 15 and 35°C). At temperature above 20°C, mean germination time (MGT) was very short (3 days), and increased with decreasing temperature (up to 10 days at 15°C). The presence of light improved germination at 15 and 20°C (near zero germination in full darkness), but had no effect at higher temperatures (Fig. 3).

				light/dark (12/12 h) full darkness	light/dark (12/12 h)
Source of variation	df	Mean Square	F	MSD 0.05 = 10.96 MSD 0.01 = 14.78 MSD 0.05 = 5.08 MSD 0.01 = 6.91	15
Seed provenance	1	11.580	0.271 NS		
Temperature	6	15292.910	357.912 ***	90	12
Light regime	1	13248.321	310.061 ***	ಷ್ಟಿ 80 Ґ	
Seed provenance x Temperature	6	281.864	6.597 ***		
Seed provenance x Light regime	1	369.299	8.643 **		
Temperature x Light regime	6	3449.592	80.734 ***		
Seed provenance x Temperature x Light regime	6	43.096	1.009 NS		
Error Three-way ANOVA of temperature, light regin	84 the o ne ar	42.728 effects of seed ad their interac	provenance, tions on the		3

germination of P. orientalis. (NS= not significant; **=p<0.01; ***=p<0.001)



Iblei Peloritani

Figure 3

The laboratory experiments supported the likelihood of a late-spring germination window (April to May), when the temperature increases and water availability in the soil is assured. In agreement with this, we observed seedlings of *P. orientalis* in the field in early May (Fig. 4). Seed germination characteristics in *P. orientalis* were quite similar to those reported for many temperate wetland species that require high temperature, temperature fluctuation and light for ideal germination conditions. For example, seeds of *Salix* spp. are also non-dormant at the time of dispersal and can germinate in a wide range of temperatures (4). This combination of characteristics is known as the 'Typical Wetland Germination Requirement (TWGR)' (5,6). Considering the thermal germination needs observed in our experiments, the long-term population survival of *P. orientalis*, particularly at the western edge of its distribution, could be furtherly threatened by reduced spring precipitation, increased temperatures and more frequent extreme weather events (7). There may be a second window of suitable germination conditions between late summer and early autumn, so our future research will focus on testing the natural germination seasons of *P. orientalis*, and how they may shift between spring and autumn. Indeed, understanding how germination strategies will respond to ongoing climate change is important for implementing active conservation measures.



Figure 4 - Seedlings of *P. orientalis* in the field in early May

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