Germination response to temperature of *Phyla canescens* (lippia)

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Summary *Phyla canescens* (Kunth) Greene (Verbenaceae) is an introduced invasive herb of floodplain pastures and wetlands in the Murray-Darling Basin. Its germination response to 81 constant and alternating temperatures was studied using a two-way thermogradient plate. The experiment was repeated for seed aged four months and 16 months, as well as seed collected from a population in another catchment. Germination prediction surfaces were generated using Geographical Information Systems-based techniques. All seed batches exhibited a ‘homothermophobic’ response, in which virtually no seed germinated at constant temperatures. The ecological significance of the homothermophobic germination response is discussed.

Keywords *Phyla canescens*, lippia, Murray-Darling Basin, germination, temperature, thermogradient plate, GIS, kriging.

INTRODUCTION

*Phyla canescens* is an invasive perennial herb native to South America. Introduced into Australia and elsewhere as a hardy ornamental groundcover, it continues to be sold in many areas. It is estimated to cover 5.3 million hectares of floodplain throughout the Murray-Darling Basin (including internationally significant wetlands), and costs the grazing industry $38 million in lost annual production (Earl 2003).

*Phyla canescens* has the potential to spread by seed and vegetative fragments but little is known about many fundamental aspects of the ecology of this species. It is thought to spread by floods and can develop substantial seedbank densities in Australia (2566 m⁻² derived from McCosker 1994) and in Argentina (3820 m⁻² derived from Alzugaray et al. 2003).

By better understanding the recruitment and seedbank processes, such as conditions required for germination, we anticipate being able to better target management interventions and reduce the impacts of this widespread and damaging species.

MATERIALS AND METHODS

Thermogradient plate  Multiple diel (daily) alternating temperature combinations can be simultaneously tested on a two-way thermogradient plate (Larson 1971).

For each experiment, 25 fruit (each consisting of two mericarps, hereafter called ‘seeds’) were pushed 1–2 mm into the surface of 1% agar gel set inside aluminium foil shells approximately 50 mm square and 20 mm deep. These were arranged in each of the 81 cells of the thermogradient plate (Figure 1). Thermocouples were also imbedded in the agar gel at 17 locations and temperatures logged at 1 h intervals. Within an hour of each alternation, temperatures had approximated the nominated temperature. Irradiance of 25 µmol m⁻² s⁻¹ (measured at the plate surface with

![Figure 1. Two-way thermogradient plate layout, showing 81 temperature combinations with constant temperature cells shaded (■), and location of 17 thermocouples (●).](image-url)
LI-COR model LI-250 light meter, John Morris Scientific) was provided for 12 h each day in phase with the day temperatures on the thermogradient plate. Agar was moistened as required to prevent dehydration. Each experiment lasted 28 days. Treatments were then transferred to an incubation cabinet set at alternating temperatures of 20:30°C (dark:light) with 12 h thermophotoperiod for another 28 days (irradiance = 20 μmol m\(^{-2}\) s\(^{-1}\)). The initial experiment was checked every 12 h and subsequent experiments daily. Seeds were considered germinated when cotyledons emerged from the seedcoat. For the initial experiment, seeds that had not germinated after 56 days were bisected and soaked in a 1% solution of tetrazolium chloride.

**Seed** Two seed batches were used. One was collected from ‘Kilmarnock’, Boggabri, adjacent to the Namoi River (30°42’S, 150°03’E) on 1 June 2004 and the germination experiments commenced four months (132 days) and 16 months (498 days) after harvest. The other was collected from ‘Old Dromana’ on the Gwydir River Watercourse, approximately 60 km west of Moree (29°19’S, 149°17’E) on 21 May 2005, and tested six months (178 days) after harvest. Seeds were stored in airtight containers in the dark at room temperatures.

**Data** Germination prediction surfaces were modelled using Geographical Information Systems (GIS)-based techniques (Tarasoff *et al.* 2005). Ordinary kriging was used with the ESRI® ArcGIS™ Geostatistical Analyst extension within ArcMap v9.0™ (ArcMap 2004). Four nearest neighbours in the search radius were used in the spherical variogram model.

**RESULTS**

**Germination** Maximum germination varied between experiments; 54% (Figure 2a), 50% (Figure 2b) and 62% (Figure 2c). Germination was significantly lower for seed subjected to hot (maximum = 45°C), cold (15°C or below for ‘Kilmarnock’ and 20°C or below for ‘Old Dromana’ seed) or constant (diel fluctuation ≤5°C) temperature treatments than for seed from fluctuating (diel fluctuation ≥10°C) temperature treatments (Figure 3). This difference was not significant once seeds from cold and constant treatments were subjected to fluctuating temperatures. Germination and viability of seed from hot treatments remained significantly different from fluctuating regimes, even after 28 days of incubation and the tetrazolium test.

**DISCUSSION**

The germination response of *P. canescens* to temperature is characterised by reduced germination at

Figure 2. Germination prediction surfaces for ‘Kilmarnock’ seed four months (a), 16 months (b) and ‘Old Dromana’ seed, six months (c) after harvest, showing low germination along the constant temperature diagonal.
We have termed this response ‘homothermophobia’. We consider this not to be a form of dormancy, but quiescence, equivalent to the ‘enforced dormancy’ of Harper (1977). We regard diurnally fluctuating temperatures to simply be a ‘normal physical environmental factor’, without which this species will not germinate (Baskin and Baskin 2004). This is supported by the subsequent germination of seeds after being moved from constant temperature to fluctuating temperature conditions. These seeds showed the same pattern as those moved from positions apparently too cold for germination (Figure 3).

Homothermophobia has been recorded to varying degrees in many species. In some cases there is still substantial, though reduced, germination at constant temperatures. There are also some species, which seem to exhibit almost absolute homothermophobia. Examples include; *Cynodon dactylon* (L.) Pers., Poaceae; *Typha latifolia* L., Typhaceae (Morinaga 1926); *Lycopus europaeus* L., Lamiaceae (Thompson 1969); *Fimbristylis littoralis* Gaudich., *Scirpus juncoides* Roxb., both Cyperaceae (Pons and Schröder 1986) and *Phalaris arundinacea* L., Poaceae (Leck 2004). These obligate homothermophobes, although from a wide range of families, all occupy aquatic, semi-aquatic or floodplain habitats. *P. canescens* appears to be a classic homothermophobe. This generalisation is consistent across geographically disjunct populations, as well as with seed age.

The ecological and adaptive significance of this phenomenon has been the subject of speculation. Explanations include soil depth-sensing, ecological-gap-sensing and inundation-recession-sensing (Thompson and Grime 1983). Greater germination of *Sorghum halepense* (L.) Pers. has been recorded at shallower depths of burial and attributed to homothermophobia (Gersha et al. 1992). Likewise, larger soil temperature fluctuations induced by a reduction in leaf canopy has been implicated in greater germination of the same species (Benech Arnold et al. 1988). Many wetland species require alternating temperatures to germinate (Thompson and Grime 1983). This was related to an increase in temperature fluctuations due to the diminution of the ‘insulating effect’ of water as wetlands dry out. The seeds are therefore able to delay germination until there is bare, exposed ground.
How these competing detection hypotheses interact depends upon other aspects of the biology of the seed and the habitat under study. Depth-sensing may be redundant in species which also require light for germination. This may be particularly true for small seeds because temperature fluctuations of sufficient amplitude for germination may penetrate deeper than the ability of the seed to emerge (Thompson and Grime 1983). Light would then be a more sensitive ‘depth-sensor’, particularly in fine-textured soils. As P. canescens also appears to require light to germinate (Macdonald unpublished data), we suggest that inundation-recession-detection is the main adaptive driver of homothermophobia in this species.

Approximately half of the seed collected from both populations is apparently not viable. This suggests the potential for increased fecundity. One possible cause is a lack of genetic diversity in the populations studied. The introduction of new genetic material through continued sale may release P. canescens from this low viability. This reinforces the importance of understanding the breeding system and genetic variation in this species.

The majority of viable, freshly collected seed appears non-dormant. Under the conditions of this study, temperature appears not to induce dormancy. The significance of the 4% of seed, which remained ungerminated across all initial treatment categories, but stained positive for tetrazolium is unknown. If it is actually viable, it may make an important contribution to the seedbank. The reduced overall viability of seeds from hot initial regimes indicates some mortality under these conditions.

Homothermophobia may be pre-adaptive for general weediness (Thompson and Grime 1983). For P. canescens, it appears to be an important mechanism driving the ability to develop persistent seedbanks, and recruit successfully after flooding.

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References


