Dealing with Physical Dormancy in Tasmanian *Pomaderris* (Rhamnaceae) collections: Heat shock, seed size and mould issues

JAMES ALEXANDER WOOD

Royal Tasmanian Botanical Gardens.
Corresponding author: James.Wood@rtbg.tas.gov.au

The presence of physical dormancy (PY) in the seeds of Rhamnaceae is widely recognised by seed practitioners and has had some study (Hanley and Lamont 2000, Turner *et al.* 2005, Haines *et al.* 2010 Ooi *et al.* 2014). The need to develop an efficient technique to alleviate PY arose from a larger study into seed dormancy in Tasmanian populations of *Pomaderris* species. A dry heat shock treatment was selected for investigation, as hot water treatments were considered cumbersome when processing large quantities of small seed in a short amount of time. Initial investigations were conducted on a collection of *Pomaderris pilifera*. Heat shock (HS) treatments were applied with using a laboratory, fan assisted oven (Binder FD115) and two 6 cm deep, stainless steel steam pans filled with 1.5 cm depth of dry sand (Figure 1a). To apply HS the steam pans were placed in the oven and brought to temperature over two hours. Seed samples were placed in a mono layer within folded aluminium foil envelopes. The envelopes are then placed flat on the sand surface of one tray (Figure 1b) and the second tray is then placed on top to sandwich the envelopes between the two bodies of heated sand and returned to the oven (Figure 1c). Envelopes were retrieved after the allotted time (Figure 1d). A control treatment (i.e., seeds neither scarified nor heated) and a manual scarification trial were also conducted to gauge the effectiveness of the HS treatments. Germination trials consisted of 3 x 40 seeds from each treatment sown on 9 cm Petri dishes of 1% agar. Remaining seeds were cut test at the end of the test. Treatments and results are presented in Table 1.

The control test confirmed the presence of PY in the *Pomaderris* collection. Surprisingly the HS trials proved to be far more effective than the manual scarification trial, partly due to seed succumbing to mould. Short durations at 90°C proved to be ineffective at alleviating PY and on cut test most seeds were found to have not imbibed. Treatments at 105-120°C were found to effective at alleviating PY with little differences in final percentage result or germination rate (Table 1). The amount of fine mould on seeds and elaiosome did differ however with decreasing amounts of mould observed as treatments became hotter and longer. At 120°C for 10 minutes almost no mould growth was observed.

Table 1. Heat shock trial results for *P. pilifera*.

Treatments	mean %	s.e	t50 (d)
15°C (control)	0.8	0.8	14.0
Chip-> 15°C	30.8	5.5	22.5
HS (90°C/1m)-> 15°C	34.7	1.4	30.3
HS (90°C/2m)-> 15°C	41.5	2.3	25.7
HS (90°C/5m)-> 15°C	62.7	7.5	27.7
HS (90°C/10m)-> 15°C	78.2	5.4	29.4
HS (105°C/1m)-> 15°C	84.6	3.9	26.3
HS (105°C/2m)-> 15°C	89.2	4.4	25.7
HS (105°C/4m)-> 15°C	80.7	4.3	25.7
HS (105°C/10m)-> 15°C	88.3	4.1	25.7
HS (120°C/1m)-> 15°C	87.3	0.1	24.7
HS (120°C/4m)-> 15°C	89.2	5.8	24.1
HS (120°C/10m)-> 15°C	94.1	3.1	24.6

(mean % = mean of final germination result; s.e. = standard error of final germination results; t50 = mean time (in days) to achieve 50% of final germination result.)

Further testing was conducted with a collection of *Pomaderris elliptica* to assess the upper tolerance level for HS treatments. Heat shock temperatures of 100, 120, 140 and 160°C were applied for 5-, 10- and 15-minute intervals. The results (not shown) confirmed that 120°C for 10 minutes is within a safe optimal range for breaking physical dormancy with good results at 5, 10 and 15 minutes. 140°C for 5 minutes killed nearly all seeds and durations and temperatures above that were completely lethal. These findings are in line with other reports of HS treatments in Rhamnaceae (Hanley and Lamont 2000, Ooi *et al.* 2014). Hanley *et al.* (2003) and Williams *et al.* (2003), studying a range of Australian legumes, reported that smaller seeded species have higher temperature tolerances. The seed mass of *Pomaderris* sits right at the lower end of the seed weights for the legumes in those studies, so it looks as though the correlation extends beyond Legumes. Overbeck *et al.* (2006) observe the same seed size/heat correlation for Brazilian grassland herbs, but Gashaw and Michelson (2002) report the reverse correlation for Ethiopian savannah species with larger seeds surviving higher temperatures.

Figure 1. Sand pan heat shock application. Photos: J. Wood

Routine testing of *Pomaderris* collections within the Tasmania Seed Conservation Centre (TSCC) has identified a small number of collections (9 out of 55) succumbing to mould very rapidly and reducing total germination. As all 55 collections had been handled identically the reason for this is unclear but the pathology is most likely imbibition shock. Higher germination results by slowly imbibing the seeds over water (after PY alleviation but before sowing on agar) appears to confirm this. It was also found that higher levels of germination were scored if these collections were sown into pots of compost rather than onto plates of agar. This suggests that issues of imbibition shock are in part a product of the testing medium and therefore the phenomenon would not carry over to field sowing. It may be worth considering whether boiling water treatments can also result in imbibition shock, if poor results are achieved using that technique.

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