Percussion as an Alternative Scarification for New Mexico Locust and Black Locust Seeds[1]

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Hot water and sulfuric acid soaks are traditional treatments for seeds of many temperate woody legumes, including locusts. However, these scarification techniques often produce inconsistent germination. Percussion scarification, where seeds are repeatedly propelled against a hard surface, was compared with hot water scarification to evaluate treatment efficacy for New Mexico locust (*Robinia neomexicana*) and black locust (*R. pseudoacacia*) seeds. In the hot water treatment, seeds were placed in a 98^0C water bath, which was immediately removed from the heat source. For percussion scarification, seeds were placed in a soil sample tin and agitated in a paint shaker for 1, 2, 3, 4, 5 or 10 minutes. All treatments, including the control, were followed by 24-hour water soaks. Hot water baths resulted in 56% and 41% germination for New Mexico locust and black locust respectively. For both species, nearly all durations of percussion increased germination over the hot water treatment. Percussion durations of 4, 5 and 10 minutes for New Mexico locust and 3, 4 and 5 minutes for black locust resulted in at least 90% germination. Traditional scarification treatments randomly degrade the entire seed coat, which can lead to tissue damage during water uptake. Percussion scarification specifically weakens the strophiole, the natural source of water entry to the seed in papilionoid legumes. Following percussion, imbibition is controlled through the strophiole and underlying tissue is protected.

Keywords: New Mexico locust, black locust, scarification, percussion, germination

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Introduction

Locust trees (*Robinia* spp.) are aggressive pioneer species that quickly colonize disturbed land, fix atmospheric nitrogen in the soil, and stabilize slopes that are prone to erosion (Klemmedson 1994, Ashby et al. 1985) (Figures1,2). Recognition of these attributes has contributed to increased demand for locusts in reclamation projects. Locust seeds, however, exhibit physical dormancy, where a thick seed coat prevents the movement of water and gasses to the embryo (Leadem 1997).

Sulfuric acid or hot water soaks have traditionally been used to break seed dormancy in macro-propagation of locusts. Many growers are moving away from the use of sulfuric acid in treating locust seeds (Dreesen and Harrington 1997). In addition to being dangerous, acid soak durations must be specifically correlated to seed lot (Young and Young 1992). Collections of native seed can vary tremendously in seed size, seed weight and hard seededness, etc. This variability limits the utility of sulfuric acid soaks, as even within a given lot, some seeds may be over-treated and damaged, whereas others are under-treated and fail to imbibe (take up water). A survey of Southwestern container growers indicates that hot water baths are currently the most common scarification method for locust seeds (Hine et al. 1997). However, hot water baths can also produce inconsistent germination (Lin et al. 1996).

Percussion scarification, where seeds are repeatedly propelled against a hard surface, is an alternative dormancy-breaking method that has proven successful in legumes related to locusts (Hamly 1932, Barton 1947, Mayer and Poljakoff-Mayber 1982). We compared this alternative treatment to hot water scarification to evaluate treatment efficacy for New Mexico locust and black locust seeds.

Materials and Methods

Commercial New Mexico locust seeds (Western Native Seed, Coaldale, CO) were collected fall 2000 in Huerfano County, CO. Black locust seeds were collected September 2000 from Taos County, NM by harvesting ripe pods from several trees at a distance of up to 3 meters from ground level. Black locust seeds were allowed to air-dry for 3 weeks, and were threshed and separated from large chaff using a greenhouse fan. A Dakota Blower was used to remove fine chaff from both seed lots. Seeds were stored at 2-4^{0}C until the germination study was conducted in March 2001.

This study compared hot water scarification with percussion scarification using treatment and species as experimental factors. Along with a control, scarification treatments included immersion of seed in boiling water as well as percussion durations of 1, 2, 3, 4, 5 or 10 minutes. Four 100-seed samples were used to test each species by scarification treatment combination.

For the hot water treatment, test tubes were filled with 30mL water and placed in a water-filled beaker (Figure 3). The bath was raised to boiling (~98^{0}C at Las Cruces, NM elevation of 1300 meters). Each 100-seed replicate was placed in a test tube, and each test tube was immediately removed to cool at room temperature.

Percussion scarification was implemented using a pneumatic paint shaker (Central Pneumatic) (Figure 4). This allowed standardization of treatment intensity. Compressed air pressure was maintained at 80psi$\pm$5psi (530kPa$\pm$30kPa), resulting in approximately 350 oscillations per minute. Each 100-seed sample was placed in a 4oz soil tin for shaking. To maximize lateral movement of paint shaker, a paint can was used as a spacer to place soil tins at the end of the shaking arm (Figure 5). This allowed shaking to occur at the greatest distance from the pivot point (fulcrum). Following all treatments, including the control, seeds were soaked for 24 hours in distilled water prior to plating out.

Germination was evaluated on lab benches at room temperature. Each replicate of seeds was placed in a 10.0 cm petri dish on moistened filter paper, with humidity maintained by enclosing the petri dishes in ziplock bags. Germination was monitored daily for 14 days, in accordance with International Seed Testing Association standards for *Robinia* species (ISTA 1999). Germination was defined as protrusion of the radicle from the embryo by at least 1mm.

Categorical Analysis of Variance using Proc Catmod (SAS Institute 1989) was used to determine treatment differences in germination percentages for each source. This procedure is an extension of a Chi-square test of homogeneity using the natural log of the ratio of germinated to non-germinated seeds for each treatment. P-values less than 0.05 were considered significant. A limited set of pairwise-comparisons was conducted to compare treatment means using a conservative alpha value of 0.05 divided by the number of comparisons.

Results

Percussion maximized germination for both New Mexico locust and black locust. Figure 6 shows three germination response levels for New Mexico locust. First, there was a low response in the control, indicating a dormant seed lot. The hot water treatment and one-minute duration of percussion share a second, intermediate response. Germination was highest at percussion levels from 2 to 10 minutes.

The representative percussion treatment produced the steepest germination curve, indicating the fastest germination rate (Figure 7). The hot water treatment produced an intermediate curve, or relatively slower germination rate. The control curve was very flat, indicating the slowest germination rate for New Mexico locust.

Control seeds germinated poorly for black locust as well (Figure 8). Hot water scarification improved germination over the control, but all levels of percussion improved germination over hot water. The optimal durations of percussion treatment were bracketed, with a significant increase from the 1 to 2 minute percussion level, a high germination response from 2 to 5 minutes of percussion, and finally a significant decrease at the 10-minute level. This drop-off suggests that 10 minutes of percussion over-treats seeds from this lot of black locust.

The representative percussion treatment also had the fastest germination rate for black locust (Figure 9). The hot water curve is somewhat flatter, indicating a slower germination rate. The control curve is very flat, indicating a slow germination rate.

A qualitative difference between hot water and percussion-treated seeds was also observed. Along with healthy germinants, the hot water treatment produced over-treated seeds (Figure 10). Examples of damaged germinants include seeds where the radicle did not elongate (Fig 10.2), seeds where the radicle elongated but was delayed in freeing itself from the seed coat (Figure 10.3) and seeds where the radicle broke off completely from the embryo (Figure 10.4). In addition to these examples of over-treatment, there were numerous small, dark seeds that failed to imbibe water. For percussion treatment, nearly all seeds took up water (Fig 11.1). Germination was rapid and uniform, with little damage evident (Fig 11.2).

To summarize, both New Mexico locust and black locust seed lots were dormant. The standard hot water scarification treatment improved germination, but only to 56% and 41% for New Mexico locust and black locust respectively. Nearly all percussion durations improved total germination as well as germination rate over the hot water treatment. Germination was very high, over 90%, for percussion durations of 4, 5 and 10 minutes for New Mexico locust and 3, 4 and 5 minutes for black locust.

Discussion

Papilionoid seeds have a specific anatomical feature known as the strophiole, or lens—the natural site of water entry to the seed (Hamly 1932). The strophiole is located on the cotyledonary lobe of a locust seed (Fig 12). When a papilionoid legume seed is percussed for the appropriate amount of time, repeated hits on the integrated seed coat loosen the constrained cells of the strophiolar region, without excessively damaging the rest of the seed coat (Ballard 1976).

When a percussed seed is soaked, water enters exclusively through the strophiole in a controlled manner (Kelly and Staden 1987). This regulated entry of water to the embryo is associated with even pressure on underlying seed tissues. This contrasts with seeds that have been hot water or acid scarified, treatments that can randomly degrade the seed coat. Unlocalized cracks in the seed coat can promote irregular water uptake associated with uneven pressure on underlying seed tissues and subsequent seed damage.

Papilionoids are the largest subfamily of legumes, covering almost all legumes occurring in temperate climates (Baskin and Baskin 1998). As with New Mexico locust and black locust, these legumes often play an integral role in the revegetation of disturbed lands. Developing superior scarification methods to the standard hot water and acid treatments should facilitate the use of these legumes in restoration projects.

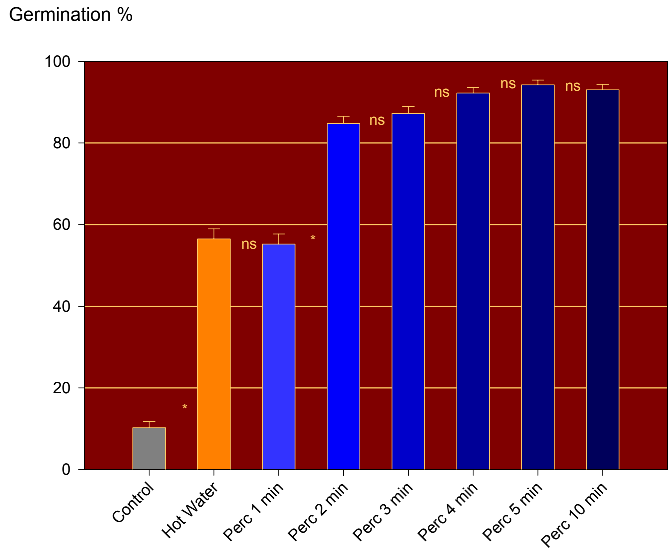

Figure 6. Effect of scarification treatments on germination percentage for New Mexico locust. ns = no significant difference between adjacent treatment bars. * = significant difference at α = 0.0045 (0.05/11).

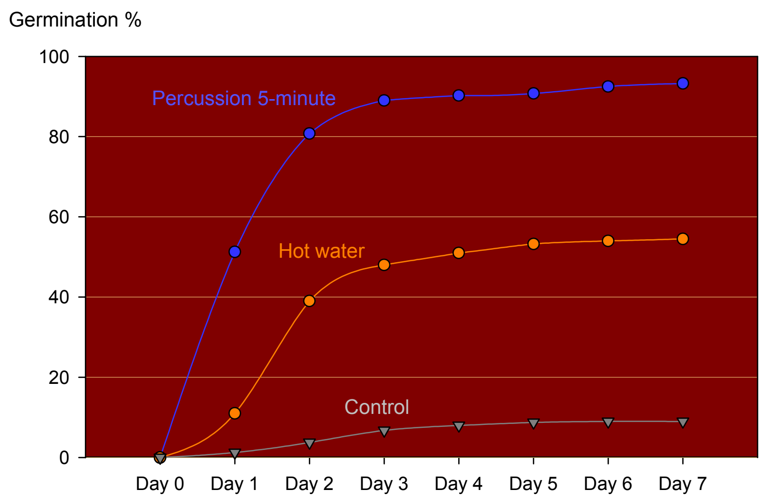

Figure 7. Effect of scarification on germination rate for New Mexico locust. Only 7 days of the 14 day study are shown. 5-minute percussion curve representative of 2 to10-minute treatment durations. Germination rate defined by slope of the curve—the increase in germination with increase in time.

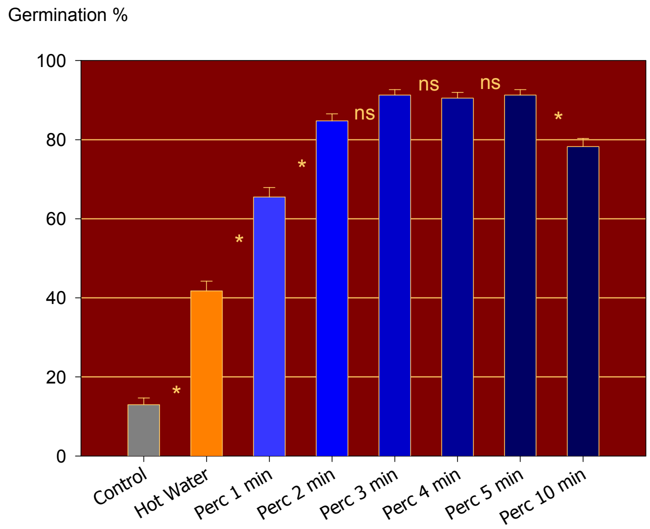

Figure 8. Effect of scarification treatments on germination percentage for black locust. ns = no significant difference between adjacent treatment bars. * = significant difference at α = 0.0045 (0.05/11).

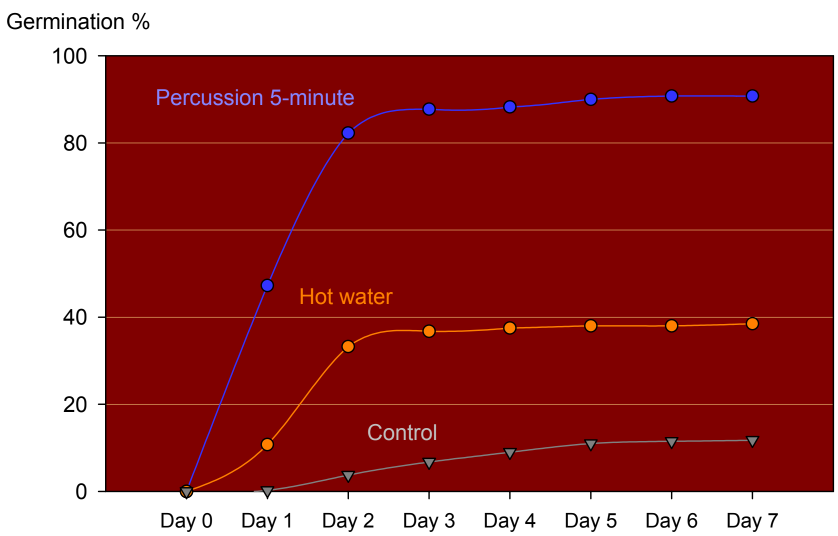

Figure 9. Effect of scarification on germination rate for black locust. Only 7 days of the 14-day study are shown. 5-minute percussion curve representative of 2 to 5-minute percussion durations. Germination rate defined by slope of the curve—the increase in germination with increase in time.

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