

# Germination response of hand-threshed Lehmann lovegrass seeds

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## Abstract

Germination of Lehmann lovegrass (*Eragrostis lehmanniana* Nees) was increased by seed after-ripening and by mechanical scarification of the seed coat. Hand-threshed seeds collected from 5 sites in southern Arizona were periodically germinated over the water potential range of 0 to  $-1.55$  MPa for 88 weeks after harvest. Nonscarified seeds exhibited very low germination at all water potentials for the entire length of the study. Total percent germination of scarified seeds peaked after 34 weeks. Seeds scarified before the after-ripening requirement was met germinated without further scarification at 46 weeks after harvest. Measurements of water uptake rates indicate that seed coat permeability to water contributes little to the increased germinability of scarified seeds.

**Key Words:** *Eragrostis lehmanniana* Nees, reduced water potential, mechanical scarification, after-ripening

Total percent germination and germination rate of Lehmann lovegrass (*Eragrostis lehmanniana* Nees.) have been shown to be highly variable among seed lots (Brauen 1967, Hardegree and Emmerich 1991). Two factors that may contribute to this variability are an after-ripening requirement and a strong positive response to any treatment that causes physical disruption of the seed coat (Brauen 1967, Haferkamp and Jordan 1977, Hardegree and Emmerich 1991). Variability among seed lots may, therefore, result from varying degrees of inherent dormancy or from mechanical scarification during harvest, threshing, and storage (Brauen 1967, Hardegree and Emmerich 1991). Haferkamp and Jordan (1977) have hypothesized that some of the germination enhancement of scarified seed results from increased permeability of the seed coat to water.

Brauen (1967) documented an after-ripening requirement for Lehmann lovegrass seeds but measured only total percent germination and did not record the effect of mechanical scarification over time. Hardegree and Emmerich (1991) measured germination variability among scarified and nonscarified seed lots of Lehmann lovegrass but had no control over seed lot age or harvest and storage conditions. Haferkamp et al. (1977) measured water uptake of scarified and nonscarified seeds but for only 1 seed lot during the first 18 hours of imbibition. The objectives of this study were to determine after-ripening requirements of hand-threshed Lehmann lovegrass seeds and the effects of mechanical scarification and reduced water potential on total percent germination, germination rate, and seed water uptake.

## Materials and Methods

Lehmann lovegrass seeds were collected from 5 sites in southern Arizona, USA, over a 3-week period in August and September, 1989. Seed collection sites were selected in a series of valleys across southern Arizona near the towns of Chiricahua (site 1, collected 19

September); Gleeson (site 2, collected 31 August); Sonoita (site 3, collected 18 September); Continental (site 4, collected 1 September); and Sasabe (site 5, collected 15 September). The sites were visited every few days after 1 August and the seed heads harvested by hand when the seeds could be removed easily by gentle hand rubbing. Harvested seed heads were allowed to air-dry in large paper bags prior to hand-threshing. The seeds were stored inside cloth bags at room temperature in the laboratory.

In the first experiment of this study, seed germination of both scarified and nonscarified seed were periodically monitored for 88 weeks after collection to determine after-ripening and scarification effects on germination response. Seeds were tested at a number of water potentials to broaden the range of conditions over which germination response could be evaluated.

Seeds were germinated inside vials designed for control of matric potential in the germination environment (Hardegree and Emmerich 1991, 1992). Seeds were placed on a cellulose membrane (Spectra/Por 3 dialysis membrane, Spectrum Medical Industries, Inc, Los Angeles, Calif.)<sup>1</sup> which was in contact with an osmotic solution of polyethylene glycol 8000 (Carbowax, Union Carbide, Danbury, Conn.). Matric potential on top of the membrane was determined by the osmotic potential of the PEG solution under the membrane. PEG was mixed with water to yield 7 solutions over the water potential range of 0 to  $-1.55$  MPa using equation 4 of Michel (1983) as suggested by Hardegree and Emmerich (1990). Seeds were germinated at 0,  $-0.09$ ,  $-0.31$ ,  $-0.63$ ,  $-0.94$ ,  $-1.24$ , and  $-1.55$  MPa following the procedure outlined by Hardegree and Emmerich (1991).

Scarified and nonscarified seeds were evaluated at 3, 7, 11, 18, 34, 46, and 88 weeks after harvest. Seeds were germinated in a controlled-temperature room at  $25 \pm 1^\circ$  C under both fluorescent and incandescent light for 12 hour day<sup>-1</sup>. The mechanical scarification treatment followed that reported by Wright (1973) with a 0.5-ml seed sample and an 8-second scarification interval. The scarification treatment was replicated 6 times for each seed lot and 35 seeds from each scarification event were germinated at each water potential. Six sets of 35 nonscarified seeds from each seed source were also germinated at each water potential. Germination vials were randomly arranged within 6 blocks in the controlled-temperature room. Germinated seeds were counted and removed from the vials on days 1-5, 7, 9, 11, and 14 of a given test run. Germinated seeds were defined as those exhibiting  $\geq 2$  mm radicle extension. The cellulose membrane was treated with a 50  $\mu$ l suspension of fungicide (Daconil; 2.5g/100ml H<sub>2</sub>O) before the seeds were placed on the membrane surface.

Two germination indices were calculated for the seeds in each germination vial in the scarified treatments: total percent germina-

<sup>1</sup>Mention of a trademark name or proprietary product does not constitute endorsement by the USDA and does not imply its approval to the exclusion of other products that may also be suitable.

tion (G); and days to 50% of  $G_5(D_{50})$  as an index of germination rate. Cubic response surfaces were calculated relating total percent germination and germination rate to germination-solution water potential and post-harvest germination date for each seed source/scarification treatment following the procedure outlined by Evans et al. (1982). Regression equations were recalculated deleting first cubic then quadratic then linear terms that were not significant ( $P \leq 0.10$ ). Lower order terms that were not significant were left in the equation if a higher order term was significant. Germination index values were calculated from the regression equations and model confidence intervals ( $P \leq 0.05$ ) determined for each seed source-scarification treatment at each water potential.

Germination rate could not be calculated for treatments with zero germination. Treatments with very low germination exhibited large variability in germination rate because only a few seeds determined the germination characteristics of the entire treatment. Values for  $D_{50}$  were, therefore, included in the regression analysis only if at least 4 of the 6 treatment replicates exhibited germination. Nonscarified seed treatments exhibited such low total germination that germination rate was not calculated.

Treatments with near-zero germination were also excluded from the regression analysis of total percent germination. The relatively large numbers of treatments with zero or near-zero germination at low water potentials artificially reduced variability in the model.

The same criteria as for the rate index were used to determine inclusion of G values in the regression models.

A second experiment was undertaken to determine the effects of scarification on subsequent after-ripening. At 46 weeks after harvest, comparative seed germination response was determined for seeds that had been scarified at 3, 7, 18, 34, and 46 weeks after harvest. The same germination system was used but germination was evaluated only at 0 MPa water potential. Quadratic regression equations were calculated as before to relate germination index values for each seed lot to post-harvest time of scarification (Evans et al. 1982).

A third experiment was undertaken to determine seed-water uptake rates of scarified and nonscarified seeds. Seed water uptake rates were measured for seed lots 1, 4, and 5 shortly after the 88-week germination test, following the procedure of Hardegee and Emmerich (1992). These seed lots were chosen because they represented low, high, and intermediate germinability among the 5 original seed lots. PEG was mixed with water to yield 3 solutions of -0.09, -0.63, and -1.55 MPa water potential. Seeds with an approximate aggregate air-dry weight of 0.6 g were poured onto the membrane surface of a germination cup which was in contact with PEG solution in a germination vial. The time at which the seeds were loaded was recorded and the seeds allowed to equilibrate for up to 72 hours. Three replicate samples from each seed

**Table 1. Calculated values for Total Percent Germination (G) as a function of time and water potential for scarified seed collected from 5 locations in southern Arizona. Values in parentheses represent one-half of the width of calculated confidence intervals ( $P \leq 0.05$ ).**

Seed lot	Post-harvest time (weeks)	Water Potential (MPa)					
		0	-0.09	-0.31	-0.63	-0.94	-1.24
		----- (%) -----					
1	3	—	—	—	—	—	—
	7	1(6)	—	—	—	—	—
	11	16(5)	14(4)	10(6)	6(7)	—	—
	18	36(5)	30(4)	19(5)	9(5)	—	—
	34	54(5)	42(4)	19(5)	3(8)	—	—
	46	50(6)	35(5)	9(6)	—	—	—
2	88	23(7)	12(6)	7(8)	—	—	—
	3	8(6)	—	—	—	—	—
	7	31(5)	22(4)	11(5)	3(6)	—	—
	11	49(4)	40(3)	26(4)	16(5)	4(7)	—
	18	70(5)	59(4)	42(4)	29(5)	14(7)	—
	34	78(5)	65(4)	42(4)	23(6)	—	—
3	46	63(6)	49(5)	24(5)	3(7)	—	—
	88	57(8)	43(6)	19(7)	6(8)	3(9)	—
	3	1(7)	—	—	—	—	—
	7	24(5)	21(4)	14(5)	3(6)	—	—
	11	43(4)	39(3)	30(4)	17(4)	5(7)	—
	18	65(5)	60(4)	48(4)	31(4)	15(7)	—
4	34	80(5)	73(4)	56(4)	31(6)	—	—
	46	70(6)	62(5)	42(5)	14(7)	—	—
	88	69(7)	60(6)	40(5)	11(9)	—	—
	3	13(6)	12(5)	10(4)	2(5)	—	—
	7	35(4)	34(3)	28(3)	17(4)	3(6)	—
	11	53(4)	50(3)	42(3)	28(4)	13(4)	—
5	18	74(4)	69(4)	56(4)	39(4)	23(4)	8(8)
	34	83(5)	74(4)	53(4)	29(4)	14(5)	—
	46	69(6)	57(5)	32(5)	7(5)	-5(7)	—
	88	63(8)	47(6)	20(7)	7(9)	—	—
	3	—	—	—	—	—	—
	7	19(5)	11(4)	0(5)	—	—	—
6	11	37(4)	28(3)	15(4)	11(6)	—	—
	18	58(4)	47(4)	31(4)	23(5)	11(9)	—
	34	67(5)	54(4)	33(4)	17(5)	—	—
	46	53(6)	39(5)	17(5)	-3(7)	—	—
	88	52(7)	40(5)	22(5)	9(8)	—	—

<sup>1</sup>Low germination treatments with more than 2 replicate samples exhibiting zero germination were not included in the regression analysis.

lot/water potential treatment were removed for water content determination after approximately 2, 4, 6, 8, 10, 12, 24, 36, 48, and 72 hours. Seeds were weighed immediately after removal from the germination cup, dried at 105° C for 24 hours, and reweighed. The initial air-dry water content was determined for 6 samples of each seed lot for both scarified and nonscarified seeds.

## Results

The effects of scarification, time after harvest, and water potential on total percent germination varied by seed lot but followed a general pattern. All seed lots exhibited low initial germination regardless of the scarification or water potential treatment (Table 1). Total percent germination for scarified seeds in the high water potential treatments peaked at about 34 weeks, followed by a slow decline in germinability except for seeds from site 1 (Table 1). Seeds from site 1 had a lower germination peak and a more rapid subsequent decline in G (Table 1). Total percent germination was lower but peaked sooner in some of the reduced water potential treatments (Table 1). Total percent germination of nonscarified seeds slowly increased over the experimental period but averaged less than 6% at 46 weeks and less than 9% at 88 weeks in the 0 MPa water potential treatment.

Germination rate response was similar to total germination response (Table 2). Germination rate increased ( $D_{50}$  decreased) for

about 34 weeks and then either stabilized or decreased (Table 2). The decrease in germination rate after 34 weeks was relatively greater at reduced water potential (Table 2).

Seeds scarified at 3, 7, 11, 18, 34, and 46 weeks after harvest that were tested for germination at 46 weeks after harvest all exhibited high total percent germination (Table 3). Three of the seed sources in this test exhibited maximum germination for seeds scarified at the time of the first experimental run (Table 3). Germination rate was uniform and apparently unaffected by date of scarification when germinated at 46 weeks (Table 3).

Scarified seeds took up more water than nonscarified seeds in the -0.09 MPa treatment but not necessarily at a greater rate (Fig. 1). Differences in water uptake were small in the -0.63 and -1.55 MPa water potential treatments (Fig. 1). Seed water uptake patterns for seed sources 1 and 4 (data not shown) were similar to that shown for seed source 5 (Fig. 1).

All of the regression models in Tables 1 and 2 were significant at the  $P \leq 0.01$  level. Confidence limits were included in Tables 1-3 to provide an estimate of model variability.

The regression models were unconstrained with respect to absolute possible minimum levels for total percent germination as this would have masked some of the variability in the data for low germination treatments. The unconstrained models provided the best fit for the data but resulted in 2 seemingly erroneous predic-

**Table 2.** Calculated values for Days to 50% of G ( $D_{50}$ ) as a function of time and water potential for scarified seed collected from 5 locations in southern Arizona. Values in parentheses represent one-half of the width of calculated confidence intervals ( $P \leq 0.05$ ).

Seed lot	Post-harvest time (weeks)	Water Potential (MPa)					
		0	-0.09	-0.31	-0.63	-0.94	-1.24
----- (days) -----							
1	3	---	---	---	---	---	---
	7	4.6(1.8)	---	---	---	---	---
	11	3.6(1.2)	3.5(1.1)	3.3(1.6)	3.1(2.1)	---	---
	18	2.4(1.2)	2.2(1.0)	2.2(1.3)	2.8(1.5)	---	---
	34	1.4(1.2)	1.3(1.0)	1.6(1.1)	3.9(2.2)	---	---
	46	1.8(1.3)	1.6(1.2)	2.2(1.4)	---	---	---
2	88	1.7(1.9)	1.4(1.4)	2.9(2.1)	---	---	---
	3	2.4(0.7)	---	---	---	---	---
	7	2.2(0.6)	2.2(0.5)	2.3(0.7)	2.8(0.7)	---	---
	11	2.1(0.6)	2.1(0.4)	2.2(0.6)	2.8(0.6)	3.8(1.1)	---
	18	1.9(0.5)	1.8(0.4)	2.0(0.5)	2.6(0.5)	3.9(1.0)	---
	34	1.5(0.6)	1.5(0.5)	1.6(0.5)	2.5(0.5)	---	---
3	46	1.4(0.7)	1.3(0.5)	1.5(0.6)	2.6(0.6)	---	---
	88	1.8(1.0)	1.7(0.8)	2.0(1.0)	3.6(1.1)	6.5(1.6)	---
	3	3.2(1.1)	---	---	---	---	---
	7	2.7(0.8)	2.9(0.6)	3.4(0.8)	4.7(1.1)	---	---
	11	2.3(0.7)	2.4(0.5)	2.9(0.6)	4.1(0.8)	5.8(1.3)	---
	18	1.8(0.8)	1.9(0.7)	2.3(0.7)	3.3(0.7)	4.8(1.2)	---
4	34	1.5(0.8)	1.5(0.6)	1.7(0.6)	2.5(0.9)	---	---
	46	1.7(1.0)	1.7(0.8)	1.9(0.8)	2.6(1.1)	---	---
	88	1.6(1.2)	1.8(1.0)	2.3(0.9)	3.5(1.6)	---	---
	3	2.5(0.6)	2.5(0.6)	2.8(0.5)	3.8(0.6)	---	---
	7	2.2(0.5)	2.2(0.4)	2.4(0.4)	3.4(0.5)	5.1(0.7)	---
	11	2.0(0.4)	1.9(0.4)	2.1(0.4)	3.0(0.4)	4.7(0.5)	---
5	18	1.7(0.5)	1.6(0.4)	1.8(0.4)	2.6(0.5)	4.2(0.5)	6.4(0.9)
	34	1.5(0.5)	1.5(0.4)	1.6(0.4)	2.3(0.5)	3.8(0.6)	---
	46	1.7(0.7)	1.6(0.6)	1.8(0.5)	2.6(0.6)	4.2(0.8)	---
	88	1.6(0.8)	1.8(0.7)	2.5(0.6)	4.2(1.2)	---	---
	3	---	---	---	---	---	---
	7	2.7(0.5)	2.8(0.4)	3.0(0.5)	---	---	---
6	11	2.3(0.4)	2.4(0.3)	2.7(0.4)	3.1(0.5)	---	---
	18	1.8(0.4)	1.9(0.4)	2.2(0.4)	2.7(0.4)	3.3(0.8)	---
	34	1.4(0.4)	1.5(0.3)	1.8(0.4)	2.4(0.4)	---	---
	46	1.5(0.5)	1.6(0.4)	2.0(0.4)	2.6(0.5)	---	---
	88	1.7(0.6)	1.9(0.5)	2.4(0.5)	3.2(0.8)	---	---

<sup>1</sup>Low germination treatments with more than 2 replicate samples exhibiting zero germination were not included in the regression analysis.

Table 3. Calculated values for G and D<sub>50</sub> as a function of initial scarification time when germinated at 0 MPa, 46 weeks after harvest. Values in parentheses represent one-half of the width of calculated confidence intervals ( $P \leq 0.05$ ).

Germ Index	Seed Lot	Post harvest time					
		3	7	11	18	34	46
G(%)	1 <sup>1</sup>	55(5)	55(5)	55(5)	55(5)	55(5)	55(5)
	2	80(8)	81(5)	82(5)	81(6)	71(7)	56(9)
	3	82(7)	81(6)	80(5)	78(5)	74(6)	70(9)
	4	85(5)	83(4)	82(4)	79(3)	73(5)	69(7)
	5	83(6)	80(5)	78(5)	73(4)	63(5)	55(8)
D <sub>50</sub> (days)	1 <sup>1</sup>	1.6(<0.1)	1.6(<0.1)	1.6(<0.1)	1.6(<0.1)	1.6(<0.1)	1.6(<0.1)
	2 <sup>1</sup>	1.5(<0.1)	1.5(<0.1)	1.5(<0.1)	1.5(<0.1)	1.5(<0.1)	1.5(<0.1)
	3	1.5(<0.1)	1.6(<0.1)	1.6(<0.1)	1.6(<0.1)	1.6(<0.1)	1.5(<0.1)
	4	1.5(<0.1)	1.5(<0.1)	1.5(<0.1)	1.5(<0.1)	1.5(<0.1)	1.5(<0.1)
	5 <sup>1</sup>	1.6(<0.1)	1.6(<0.1)	1.6(<0.1)	1.6(<0.1)	1.6(<0.1)	1.6(<0.1)

<sup>1</sup>The regression model for this seed source and germination index had no significant regression coefficients. Predicted germination index values for this seed source and germination index, therefore, represent the mean value across all treatment.

tions of total germination less than 0% (Table 1). In both cases, however, the model confidence interval of these values overlapped the minimum possible value for G.

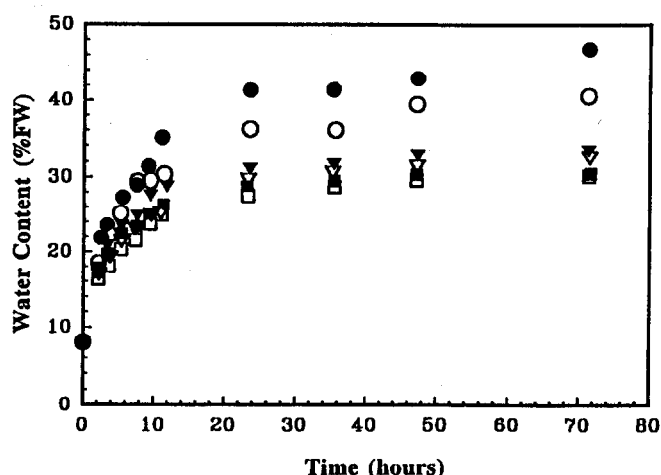


Fig. 1. Seed water content of seed source 5 as a function of time, for scarified (closed symbols) and non-scarified (open symbols) seeds at -0.09 (●, ○), -0.63 (▼, ▽), and -1.55 (■, □) Mpa water potentials.

### Discussion

Our understanding of the germination response of Lehmann lovegrass is complicated by high variability among seed sources (Hardegree and Emmerich 1991). In the current study, hand-threshed but non-scarified seeds averaged less than 9% germination 88 weeks after harvest. This suggests that seed lots previously tested for scarification effects may have already been partially scarified during harvest, threshing, or storage (Hardegree and Emmerich 1991). Differences among seed lots in this study may have been caused by preharvest environmental conditions, but our study was not set up to evaluate within-seedlot variability or to assign significance to environmental conditions at the collection sites. All of the seedlots used in this study, however, shared similar patterns of after-ripening response and scarification effects. Differences among seed lots in this study were relatively small compared to those found in previous studies (Hardegree and Emmerich 1991).

The trend toward lower germinability with time is inconsistent with the data from some previous studies. Hardegree and Emmerich (1991), found high germinability over an extended water potential range for seeds tested much later than 88 weeks after harvest. The slow decline in germinability after 34 weeks may have

been due to the storage conditions particular to this study, but there was no way to test this with our data. Data from the second experiment, however, indicate that there is an interaction between scarification and after-ripening. Seeds generally exhibited higher total germination 46 weeks after harvest if they were scarified before the after-ripening requirement was met (Table 3). Perhaps the relatively high total germination of older seed lots from previous studies was partially due to inadvertent scarification before the seeds were fully ripe.

The difference in total water uptake between scarified and non-scarified seeds is not proportional to the relatively large effect of scarification on total percent germination. Scarified seeds took up more water than non-scarified seeds at a water potential of -0.09 MPa but treatment differences were negligible at -0.63 and -1.55 MPa (Fig. 1). Hardegree and Emmerich (1991) showed that scarification can result in a large increase in total percent germination at water potentials lower than -0.63 MPa but did not measure water uptake patterns.

In the current study, there was so little germination of non-scarified seeds that a comparison of scarification effects on germination rate could not be made. Hardegree and Emmerich (1991), however, found that scarification could advance mean germination time by several days. It is unlikely that the negligible change in imbibition rates found here and in other studies (Haferkamp et al. 1977) could be solely responsible for a several day increase in mean germination time.

After-ripening was an important factor for these seed lots only in the first 9 months after harvest. Low initial germinability is an advantage for Lehmann lovegrass since it produces seeds relatively late in the summer rainy season in southern Arizona. Seedbed conditions are suitable for germination at this time but late summer seedlings would be vulnerable to dry conditions later in the fall.

Germination of Lehmann lovegrass is affected by a multitude of environmental variables that will be expressed to a different degree at every point in the seed bank (Cox and Martin 1984, Frasier 1989, Haferkamp and Jordan 1977, Hardegree and Emmerich 1991, Martin and Cox 1984). Natural chemical and physical scarification events in the field may insure that some seeds are always viable when seed bed conditions become favorable for establishment (Brauen 1967, Hardegree and Emmerich 1991). As in previous studies, our results show that germination rate of Lehmann lovegrass can be relatively high for scarified and after-ripened seeds (Hardegree and Emmerich 1991).

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