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Delayed Germination of California Oatgrass, Danthonia californica[1]

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CALIFORNIA oatgrass, *Danthonia californica* Boland, is a long-lived perennial bunch grass found throughout range lands in the western United States. The species has almost disappeared from heavily grazed areas and efforts are now being made to encourage its re-establishment (Jones 1948). Reported instances of difficulty in reseeding the grass and our own failures to get stands in experimental work led to this investigation in this species.

Germination within the genus has been studied by several investigators. The poor germination of *Danthonia spicata* was improved by sulfuric acid treatment and by prechilling the seed according to Toole (1927) who considered restriction of gas exchange through the seed coat as the primary cause of difficulty. Trumble (1937) related low germination of Danthonias to the "obstructiveness of the flowering glume", and reported that treatment with concentrated sulfuric acid markedly stimulated germination under both field and laboratory conditions (Trumble 1927).

Cockayne (1916) suggested that poor germination of Danthonia in New Zealand may result from the seed being harvested before it is sufficiently mature. He considered that good lines should germinate of 80%, but that an average germination of 50% is satisfactory. The slow establishment and slow seedling growth of Danthonia was stressed by Cashmore (1932). After studying numerous samples of six species this investigator concluded that the viability and rate of germination of the seed is largely a function of the strain used. Moore (1946) reported difficulty in obtaining a stand from dehulled seed of *Danthonia semi-annularis* when sown with a drill. He considered that dehulled seed is likely to be planted at too great a depth, and concluded that best results are obtained when the seed is planted not deeper than ¼ inch and at a time when soil is likely to remain moist for 2 to 3 weeks.

In preliminary trials ad Davis in 1946 with two lots of California oatgrass seed planted in greenhouse soil, seedling emergence of less than 3% was obtained with untreated controls. Treatment with concentrated sulfuric acid for 15 minutes resulted in emergence ranging from 20% to 40% within 21 days. It appeared feasible, therefore, to study the nature of germination in this species more extensively.

Materials and Methods

Mature seed was collected in 1947 from 16 selected native stands of *Danthonia californica*[3] located in California from Santa Cruz County in the south to Humboldt County in the north (Beetle 1947). This covers a north-south distance of approximately 275 miles. Table I presents the collections made. Seed was stripped from the terminal inflorescence in all stands, and the axillary cleistogenes (Chase 1918) were collected from six of them.[4] The results in this paper are for terminal inflorescence seed unless otherwise indicated. All seed was stored in the laboratory until used.

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[3] Danthonia californica as used in this paper includes both *Danthonia californica* Boland, and *Danthonia californica* var. *americana* (Scribn.) Hitchc. Both the species and variety were found to occur together at most of the collection sites and behaved in like manner to the treatments employed.

[4] The cleistogenes used were those of the multi-floret spikelets occurring along the upper regions of the culm, not the uni-floret spikelets with enlarged caryopsis found at the basal nodes of the culm.

The large number of unfilled florets found among those stripped from the terminal inflorescence necessitated care in detecting them from full florets. To insure that only florets containing caryopses were employed in the test, the seed was candled over a frosted glass. This made the shadow of the caryopsis visible through the enclosing lemma and palea and permitted separation of filled from empty florets. However, the indurated lemma of the cleistogenes rendered them to opaque to allow this technique, so full florets of cleistogenes were determined by hand.

Germination tests in the laboratory were carried out in a Minnesota seed germinator at constant temperature of 21°C. Seed was germinated in Petri dishes on paper toweling moistened with tap water. A seed was considered germinated when both the plumule and radicle had broken through the pericarp.

Greenhouse tests were run in unsterilized soil in benches or wooden flats. Field plantings were made in a weed-free plot receiving only natural rainfall. Planting depth in both cases was ¼ inch. In greenhouse and field soil only living seedlings were counted so the emergence and survival figures vary somewhat with the data of the reading as new seedlings emerged or old ones died.

TABLE 1. –*Seed collections of Danthonia californica made in 1947 from native stands in California.*

Collection No.	County in which collected	Date collected	Full florets, %
1*	Santa Cruz	June 14	51
2*	Sonoma	June 16	45
3	Sonoma	June 18	54
4	Marin	June 15	52
5	Marin	June 15	82
6	Sonoma	June 17	77
7	Sonoma	June 19	49
8*	Humboldt	June 26	63
9*	Humboldt	June 18	78
10	Mendocino	June 16	66
11	Mendocino	June 11	70
12	Mendocino	June 13	57
13	Humboldt	June 27	72
14	Humboldt	June 28	71
15*	Lake	July 15	42
16*	Nevada	July 22	60

* Cleistogenes also collected from the stands.

The sulfuric acid treatment employed concentrated acid (sp. gr. 1.84). Dry seed was covered with an excess of acid in beakers for the desired time, stirred as necessary, then washed thoroughly in tap water and dried before planting.

Untreated seed was hulled by rolling the caropses out of the lemma and palea between soft rubber mats. This did not visibly scratch or break the seed coats.[55] Those nicked with a scalpel were cut over the endosperm on the dorsal side of the seed.

Results and Discussion

Duplicate 100-seed samples of all collections were given a uniform 15-minute sulfuric acid treatment and planted in greenhouse soil along with untreated controls. Rows were randomized within blocks. The planting date was approximately 4 months after the collection of seed. The soil was kept moist during the entire text period. Day temperatures averaged 75 ° F and night

[5] The individual roles of pericarp and integuments were not determined. These tissues are considered together as seed coats in this paper.

temperatures averaged 60° F in the greenhouse. Weekly counts of living seedlings were made during the 4 month following planting.

Greater emergence was recorded for the acid-treated samples compared to the controls for the same collections in all cases. An analysis of variance made on the data for the fourth week after planting indicated this greater emergence to be statistically significant in 14 of the 16 collections. Differences between collections were highly significant.

Table 2 presents typical results of this trial on terminal inflorescence seed for collections 1, 8, and 15, which gave significant response to acid treatment, and for collection 10, one of the two in which differences were not statistically significant. The gradually increased emergence of the untreated lots during the 16-week period illustrates the delayed germination behavior of *Danthonia californica*. The more rapid and greater emergence of the treated samples is apparent.

TABLE 2.—*Percentage of Danthonia californica seedlings emerged and surviving in greenhouse soil, each figure the mean of duplicate 100-seed samples of terminal inflorescence seed.*

Collection No.	Sulfuric acid treatment	Weeks after planting						
		2	4	6	8	10	12	16
1	None	0.5	2.0	2.5	3.0	4.0	4.0	5.5
	15 min.	12.5	15.5	17.5	19.5	19.5	18.5	19.0
8	None	0.5	2.5	7.0	10.0	13.0	14.5	14.5
	15 min.	13.0	19.5	24.0	24.5	25.5	26.5	27.0
10	None	0.0	0.5	1.0	1.5	2.0	2.5	3.5
	15 min.	3.0	4.5	5.0	5.5	6.5	6.0	7.0
15	None	1.0	2.5	3.0	4.5	5.5	5.5	9.5
	15 min.	18.5	26.5	27.0	27.5	27.5	27.5	24.0

The 15-minute acid treatment seemed too severe for the auxillary cleistogenes. Treated cleistogenes appeared to have been injured and emergence results substantiated this view.

The best duration of concentrated sulfuric acid treatment was determined by studying the effect of a series of treatment times on terminal inflorescence seed of collections 8 and 10, and cleistogenes of collection 15. Treatments of 5, 10, 20, 25, 30, and 45 minutes were used. Each treatment sampled was divided into three parts. One part was germinated in Petri dishes in the seed germinator, one planted in greenhouse soil, and one planted in the field.

Results are presented in Table 3. Collections 8 and 10 responded in much the same manner although 8 yielded somewhat higher germination and emergence. The maxima of 85% for collection 8 and 76% for collection 10 in the germinator indicate good viability with suitable treatment. Much higher germination was obtained in Petri dishes than emergence in soil, and field emergence was reduced compared to that in the greenhouse. The great interval of days required for appreciable field emergence was due in large measure to a period of 6 weeks of dry weather following planting.

Thirty-minute acid treatment for collection 8 and 45-minute treatment for collection 10 resulted in the highest germination in Petri dishes. In greenhouse soil both collections gave the best emergence following the 20-minute acid treatment. In the field both were superior in response to 15-minute acid. The importance of recommending field treatments on the basis of

field results is apparent. The best germinator treatments were poor in the greenhouse and of little to no benefit in the field.

The cleistogenes of collection 15 followed the same trends but in response to shorter treatment times. Best germinator results were obtained with the 10- and 15-minute treatments while both greenhouse and field plantings responded best when treated only 5 to 10 minutes.

The 20-minute sulfuric acid treatment was employed uniformly on terminal inflorescence seed of all 16 collections for a greenhouse test approximately 9 months after the seed had been collected. Duplicate 100-seed samples both treated and untreated were planted in randomized blocks on greenhouse benches. Seedling emergence and survival counts were made for 16 weeks following planting. Results are presented in Table 4.

TABLE 3.—*Sulfuric acid treatments of Danthonia californica compared in seed germinator, in greenhouse soil, and in the field; figures the mean of duplicate 50-seed samples*

Sulfuric acid treatment	Germinator					Greenhouse					Field				
	Germination %					Emergence and survival, %					Emergence and survival, %				
	Days in germinator					Days after planting					Days after planting				
	5	10	13	16	21	14	21	35	49	56	73	84	98	115	146
Seed Collection No. 8															
None	2	3	4	4	5	0	1	0	2	2	3	5	5	5	5
5 min	4	10	10	10	11	4	5	5	8	7	2	6	6	6	6
10 min	3	23	25	26	26	9	12	13	13	14	6	12	13	11	11
15 min	10	34	43	44	47	15	17	18	17	17	16	24	23	21	20
20 min	22	58	64	67	71	26	28	28	27	29	7	11	10	9	7
25 min	38	62	69	72	75	23	26	26	25	25	4	8	7	8	6
30 min	42	79	84	84	85	22	21	20	20	20	6	8	6	6	5
45 min	43	65	66	66	67	1	2	1	1	1	1	0	2	1	0
Seed Collection No. 10															
None	0	1	1	1	1	0	0	2	4	4	0	1	0	0	1
5 min	0	4	6	9	11	1	2	2	4	4	1	1	1	1	1
10 min	2	16	18	21	21	5	6	7	10	12	1	2	3	3	3
15 min	4	23	24	25	29	5	11	12	13	14	3	9	10	9	8
20 min	5	32	42	45	47	12	17	15	19	18	0	5	4	2	2
25 min	10	51	56	59	60	9	10	10	10	11	3	7	4	4	4
30 min	22	58	65	69	71	8	9	8	8	9	1	4	3	2	2
45 min	22	64	70	76	76	4	6	5	5	5	1	2	0	0	0
Seed collection No. 15 (Cleistogenes)															
None	0	0	0	0	--	0	1	2	3	3	0	0	0	0	0
5 min	1	46	56	59	--	5	11	10	10	10	0	2	6	5	4
10 min	14	76	78	81	--	7	7	7	7	7	2	4	6	5	4
15 min	13	62	70	71	--	0	0	0	0	0	1	1	1	1	1
20 min	2	30	31	31	--	0	0	0	0	0	0	0	0	0	0
25 min	4	36	37	37	--	0	0	0	0	0	0	0	0	0	0
30 min	5	29	32	33	--	0	0	0	0	0	0	0	0	0	0
45 min	0	15	17	17	--	0	0	0	0	0	0	0	0	0	0

The marked delayed germination of all untreated samples is evident, as is the hastening of emergence and increased number of seedlings in the treated lots. In no case did untreated samples exceed the treated in percentage emergence. All acid treatments at 4 weeks resulted in significantly increased emergence compared with the controls for the same collection. After 16 weeks the acid-treated samples still exhibited significant increases in all but four collections. Variations in response among collections were statistically significant, and suggest the possibility of selecting strains for improved germination.

Seed remaining ungerminated in moist soil for 16 weeks was recovered and found to be sound and plump. Occasional seeds were still commencing to germinate but had not yet emerged.

Some indications of the nature of the delayed germination was obtained through study of the water uptake of acid-treated and untreated seed and study of the effect of hulling, and of breaking the seed coat by nicking with a sharp scalpel.

Terminal inflorescence seed treated with 20-minute concentrated sulfuric acid and untreated hulled seed having no detectable seed coat injury were soaked in tap water for 24 or 48 hours and the per cent water uptake on a dry weight basis determined. (The acid-treated seed was hulled by the acid treatment.) The average water uptake of hulled seed after 24 hours of soaking was 32.58% of the original dry weight of the seed, while that of acid-treated seed was 32.64%. After 48 hours soaking the hulled seed averaged 44.07% uptake and the acid-treated averaged 43.47%. Striking similarity in water absorption is indicated in acid-treated and untreated hulled seed.

TABLE 4.—*Percentage of Danthonia californica seedlings emerged and surviving in greenhouse soil, each figure the mean of duplicate 100-seed tests of terminal inflorescence seeds*

Collection No.	Sulfuric acid treatment	Weeks after planting							
		1.5	4	6	8	10	12	14	16
1	None	0.5	4.5	5.5	6.5	8.5	10.5	10.5	10.5
	20 min.	25.5	33.5	33.5	33.0	32.5	33.0	34.5	34.5
2	None	0.0	1.5	2.0	3.0	3.0	3.0	3.0	3.0
	20 min.	20.0	20.0	20.5	20.5	20.5	20.0	21.0	20.5
3	None	0.0	1.5	1.5	3.0	3.5	3.5	5.0	5.5
	20 min.	20.5	30.0	31.5	32.0	33.5	33.0	34.0	32.5
4	None	3.0	7.0	8.0	8.5	11.0	11.5	14.0	14.0
	20 min.	18.0	22.0	21.5	22.0	21.5	22.5	24.0	23.0
5	None	1.0	3.0	4.5	6.0	7.0	9.5	13.0	11.0
	20 min.	20.0	25.5	29.0	28.0	31.0	31.0	30.0	30.0
6	None	2.5	5.0	7.0	8.5	8.0	11.0	13.5	16.0
	20 min.	19.0	31.0	28.0	29.0	30.5	32.0	31.5	32.5
7	None	0.0	2.5	4.0	4.5	5.5	6.5	7.5	10.0
	20 min.	11.5	13.5	13.5	13.5	13.5	13.5	13.5	13.0
8	None	1.0	2.5	4.0	4.0	4.5	6.0	6.0	6.5
	20 min.	13.0	23.5	21.5	23.0	24.0	24.0	25.0	24.5
9	None	0.0	0.5	2.0	1.5	1.5	3.0	3.0	3.5
	20 min.	17.5	29.5	29.5	29.5	28.0	28.5	29.5	28.5
10	None	0.0	1.5	1.5	1.0	2.0	2.0	3.0	3.0
	20 min.	5.0	10.5	12.0	13.0	16.0	17.0	17.5	17.5
11	None	0.5	2.5	5.5	6.0	7.0	7.5	9.5	9.0
	20 min.	4.5	11.5	12.0	12.5	13.0	13.0	15.0	15.5
12	None	0.5	2.0	3.5	3.5	3.5	4.0	5.0	5.5
	20 min.	7.0	10.5	11.0	12.5	14.0	16.0	17.0	19.0
13	None	1.0	2.0	3.0	3.5	4.5	6.0	8.5	9.0
	20 min.	9.5	21.5	22.0	23.5	23.5	22.5	25.0	26.0
14	None	0.0	2.0	3.5	4.0	5.5	6.5	8.5	11.5
	20 min.	4.5	9.0	10.5	10.5	12.0	13.0	14.5	15.5
15	None	0.5	0.0	3.5	4.5	5.5	7.0	7.5	8.5
	20 min.	17.0	25.5	25.5	25.5	26.0	26.5	29.0	28.5
16	None	0.0	3.5	4.5	5.5	6.0	6.0	6.5	8.5
	20 min.	6.5	9.0	8.0	8.0	8.0	8.0	8.5	8.5
L. S. D. 5 % level.....		---	4.8	---	---	---	---	---	8.0
L. S. D. 1 % level.....		---	6.5	---	---	---	---	---	10.8

Table 5 presents germination and emergence results following hulling, and mechanically nicking the seed coat with a scalpel. The effect of hulling alone suggest but slight improvement in germination. Only the greenhouse planting of collection 8 yielded a marked increase. Hulled

seed nicked with a scalpel to break the seed coats, however, resulted in substantial increases in germination and emergence in all cases.

TABLE 5.—*Effect of hulling and of cutting the seed coats of Danthonia californica compared in seed germinator, in greenhouse soil, and in the field; figures based on duplicate 50-seed samples.*

Treatment	Germinator					Greenhouse					Field				
	Germination, %					Emergence and survival, %					Emergence and survival, %				
	Days in germinator					Days after planting					Days after planting				
	5	10	13	16	21	14	21	35	49	56	73	84	98	115	146
Seed Collection No. 8															
None	0	1	1	2	2	1	1	1	3	4	0	0	0	0	0
Hulled	2	5	6	7	9	3	4	12	19	22	4	6	6	5	5
Hulled, nicked with scalpel	33	76	81	84	84	33	36	32	31	32	19	20	16	15	12
Seed Collection No. 10															
None	0	0	0	1	3	0	0	1	1	1	0	1	3	1	2
Hulled	1	4	5	5	5	1	2	3	5	5	0	2	2	2	1
Hulled, nicked with scalpel	9	56	71	78	81	19	18	16	17	17	13	16	13	10	10
Seed Collection No. 15 (Cleistogenes)															
None	0	0	0	0	--	0	0	0	1	2	0	1	2	2	1
Hulled	0	1	1	1	--	0	0	4	7	9	0	3	2	2	2
Hulled, nicked with scalpel	0	5	20	29	--	15	21	24	23	22	9	18	20	18	15

Examination of acid-treated terminal seed under a dissecting microscope revealed very definite etched areas on the seed coats where 15 minutes or more of acid treatment had been employed. It would appear that weakening of the seed coats is essential in overcoming the delayed germination of *Danthonia californica*. The seed coats may hinder germination through mechanical restraint or restriction of gas exchange or both. They do not prevent water absorption.

Mechanical scarification of California oatgrass seed to weaken the seed coats does not appear feasible as the protruding embryo is located in such an exposed position that embryo injury results. Sandpaper scarification of hulled seed did not increase emergence in soil in preliminary tests.

General Discussion

The faster seedling emergence and the greater total emergence following treatment of *Danthonia californica* seed with sulfuric acid are desirable fro the standpoint of competitive ability and stand establishment. Although the different collections studied varied in the degree of response to acid treatment, all benefited. The experience of some ranchers in this state has been to note new seedlings of Danthonia near animal droppings. Many factors may be involved in such seedling establishment, but the possibility of natural chemical treatment while in the digestive tract of the animal is worthy of consideration.

There is evidence to suggest that the intensity of the delayed germination in seed of *Danthonia californica* from a given stand varies from one seed crop to the next. A study is in progress to verify this and to give a clearer insight into environmental factors which possibly may influence delayed germination. Seed from selected native stands is being collected annually

and tested in the hope that germination results when compared with climatic data will shed light on this relationship. Germination following different periods of seed storage is also receiving attention.

Summary

Delayed germination of *Danthonia californica* was studied in 16 seed collections obtained from native stands in California in 1947.

Seedling emergence 16 weeks after planting ranged from only 2.5 % to 16.0 % when untreated terminal inflorescence seed was planted in unsterilized greenhouse soil. Concentrated sulfuric acid treatment of seed hastened emergence and increased the number of seedlings.

Response to treatment was compared in the seed germinator, greenhouse, and field. Field plantings benefited most from acid treatments of considerably shorter duration than those yielding the best results in the generator.

In field tests the 15-minute sulfuric acid treatment yielded the greatest seedling emergence from terminal inflorescence seed. Axillary cleistogenes were found to require less treatment and exhibited the best field response to a 5- to 10-minute acid treatment.

Weakening of the seed coats appears essential in overcoming the delayed germination of Danthonia californica. In these experiments this was accomplished by sulfuric acid treatment or by cutting the seed coats. Hulling without seed coat injury was of little benefit.

The seed coats appear to delay germination through mechanical restraint or restriction of gas exchange or both.; They do not prevent water absorption.

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