

# Dormancy in Seed of Dwarf Spikerush *Eleocharis coloradoensis* (Britt.) Gilly)<sup>1</sup>

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## ABSTRACT

Dormant seed (achenes) of dwarf spikerush (*Eleocharis coloradoensis* (Britt.) Gilly) stored for 12 months at 4 C in water that was exchanged once a month with fresh water germinated 67%. Dormant seed stored at a high volume ratio of 6 parts water to 1 part seed and a low volume ratio of 1 part water to 6 parts seed germinated 60 and 41%, respectively. Seed stored 3 months with the basal ends of the pericarps removed germinated 62%, whereas, seed stored

similarly with uncut pericarps germinated 4%. Germination of non-dormant seed incubated in a diluted extract of pulverized dormant seed was 11%; in a non-dormant seed extract it was 30%. The germination of non-dormant seed incubated in the leachate obtained from dormant seed was 8%, compared to 54% germination with the same seed incubated in distilled water.

*Key words:* Seed-dormancy, inhibitor, germination, leachate, incubation, extract, allelopathy.

## INTRODUCTION

Dwarf spikerush is a low-growing perennial that can

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displace certain unwanted rooted submersed aquatic weeds (2, 4). Because the plant is short and does not restrict water-flow, boating, or other aquatic activities like the tall weedy species that grow to the surface, it is beneficial to grow the plant in weedy aquatic sites. One method used to establish stands of dwarf spikerush in lakes, ponds, and canals is to sow seed. However, newly harvested seed are dormant. Several methods for breaking dormancy and enhancing the germination of dwarf spikerush seed were investigated (3, 5). Procedures for germinating dwarf spikerush seed showed that germination could be significantly enhanced by scarifying them in 5.25% sodium hypochlorite or chilling seed wet at 4 C for 24 months.

Mechanical methods for enhancing germination were only partially successful. Scoring of the pericarp with sand, cutting the pericarp at random and rupturing the pericarps by applying pressure, all resulted in low percentages of germination. The pericarp of dwarf spikerush is composed of several layers of annulated cells filled with a waxlike substance (1). The waxlike material may have helped resist mechanical methods of inducing germination.

The ratio of water to seed used for storing seed wet was observed by the senior author to be important in retaining viability.<sup>2</sup> During the period from 1972 to 1976, quantities of approximately 1 L of seed were stored at 4 C in 8 L containers filled with water. These seeds had an average percentage germination of 40% after one year of storage. With the addition of a seed nursery and additional field harvests, larger quantities of seed (a volume of approximately 6 L) were stored in 1 L of water to save space. When these seeds were washed and incubated in distilled water after being stored wet at 4 C for one year, only 0 to 3% germinated.

Conditions for incubating dwarf spikerush were studied (10). The seed germinated well at 20 to 22 C in subdued light. Incubating seed in subdued light was preferable because algae occasionally formed in the incubating dishes when placed in full sunlight. Photoperiod did not affect the total percentage germination, but the rate of germination was accelerated when the seeds were incubated in continuous light.

The objective of this study was to determine if a water-soluble inhibitor was present in the seed of dwarf spikerush. The following tests were made: (1) Cutting the bases of the pericarps, (2) storing dormant seed in different volumes of water or in water replaced each month with fresh water, (3) incubating non-dormant seed in different concentrations of extracts made from pulverized dormant and non-dormant seed, and (4) incubating non-dormant seed in leachate obtained from dormant seed that had been stored in water.

#### MATERIALS AND METHODS

*Incubation.* Seed treated before or during incubation were placed in 35 x 10 mm plastic Petri dishes. Distilled water was added to bring the incubating medium to a total volume of 10 ml in the large dishes and to 4 ml in the small dishes. Each treatment was replicated 3 times and each replicate consisted of 100 or 50 seed per dish. The dishes containing the seeds were set in subdued sunlight

at a temperature range of 20 to 22 C. The average light intensity in the subdued sunlight at midday during the tests (November through February) varied from 600 to 800 lux.

The percentage germination was obtained by counting and removing the newly-germinated seed that had elongated cotyledonary sheaths. Germinated seed were removed every few days to avoid the difficulty of sorting ungerminated seed from the fibrous roots. The germination period lasted 28 days. The data were analyzed by comparing the replicate means of the percentages using Duncan's multiple range test at the 5% level.

*Effect of cutting pericarps.* The effect of exposing the seed of dwarf spikerush directly to water by cutting the pericarp at the basal end of the achene was studied. Dormant seed harvested in 1979 at the Aquatic Plant Research Laboratory at Davis, California were used in this study. They were stored dry at 4 C until the experiment was initiated. Each treatment consisted of 3 lots of 50 seed each that had the following: (1) Seed uncut, stored dry or in water at 4 C for 3 months; (2) bases of pericarps cut, stored dry or in water at 4 C for 3 months; (3) intact seed stored in water at 4 C for 3 months and then the bases of the pericarps cut. Seed selected for the study were black or dark brown, unbroken, and sank in water when thoroughly wetted.

The bases of the pericarps were removed by carefully cutting across the base without damaging the embryo using a new razor blade. At the end of each storage period or treatment following the storage period, the seed were incubated and the percentage of germination determined.

*Effect of storing seed in different amounts of storage water on dormancy.* Two studies were conducted on the influence of storing seed wet in different volumes of storage water on the percentage germination. The study was conducted twice. Test 1 was initiated in August 1979 and Test 2 in March 1980. Seed harvested in June 1979 were used in both tests. In March 1980, dormant seed were weighed, placed in 35 ml plastic containers, and treated as follows: (1) 16 g of seed stored dry at 4 C; (2) 16 g of seed stored dry at 21 C; (3) 16 g of seed stored wet at 4 C in a low volume of water (ratio of 6 parts of seed to 1 part water); (4) 2 g of seed stored wet at 4 C in a large volume of water (ratio of 1 part seed to 6 parts water); and (5) 9 g of seed stored wet at 4 C at a volume ratio of 1 part seed to 1 part water. There were approximately 119,000 seed in 16 g. In treatment 5, the storage water was exchanged monthly with fresh distilled water, each final exchange was preceded by five complete rinses. Each rinse was made by filling the containers with fresh distilled water, agitating, and then as soon as the seed settled the water was decanted.

At the beginning of each month, for 12 months, a sample of seed was removed from each of the stored containers and rinsed with fresh distilled water. Three lots (replications) of 100 seed were counted and incubated.

Test 2 employed the same techniques as Test 1. This study was continued for 10 months. The data for the 5th month was inadvertently not taken.

*Effect of an extract from pulverized seed on the percentage germination of viable seed.* A study was made to determine if viable seed incubated in an extract of pulver-

<sup>2</sup>Unpublished data. R. R. Yeo. 1979.

ized dormant seed would reduce germination. Four grams of dormant seed (harvested in 1979) that had been stored dry at 4 C for 12 months and had a percentage germination of 3% and 4 g of non-dormant seed that had been stored wet at 4 C for 12 months and had a percentage germination of 38%, were each placed in a polytron and ground for 8 minutes. After pulverizing, they were each put into a 200-ml Erhlenmeyer flask and distilled water added to make stock solutions of 50 ml. The flasks were placed on a shaker table and agitated for 18 hr at room temperature. The extractions were then filtered through an 80-mesh screen. Quantities of 1, 2.5, 5, and 10 ml of each extract were then added to three 60 x 15 disposable plastic Petri dishes. To obtain the necessary final dilutions of the potentially inhibitive material, distilled water was added to each dish to make a total volume of 10 ml. Only distilled water was added to the control treatments. Conductivities of comparable solutions were 390 and 410  $\mu$ mho. Viable dwarf spike-rush seed that had been harvested 1975 and stored wet at 4 C for 16 months to break dormancy were added to each dish. The seed were then incubated to determine the effect of each concentration of the extracts on the percentage germination of viable seed.

*Effect of the leachate from dormant stored seed on the germination of non-dormant seed.* Leachate from dormant stored seed was examined for its inhibitive characteristics on non-dormant viable seed. Seed harvested in May 1977 and stored dry at 4 C until November 1977 were placed in wet storage at 4 C for 6 months to obtain storage water containing leachate from dormant seed. A volume ratio of approximately 6 parts seed to 1 part distilled water were placed in a 2-L jar and stored in a refrigerator at 4 C for 6 months. In May 1978, aliquots of the leachate were placed in 35 x 10 mm plastic Petri dishes in quantities of 0, 1, 2, and 4 ml. Distilled water was added to bring the amount of solution in each dish to 4 ml, resulting in concentrations of 0, 25, 50, and 100% of the leachate. The seed were allowed to incubate for 4 days, then the number of germinated seed were counted. The lengths of the cotyledonary sheaths of the germinated seed were also measured

to determine if the leachate contained a substance that would inhibit elongation.

## RESULTS AND DISCUSSION

*Effect of cutting pericarps.* Cutting and removing the basal ends of the pericarps and submersing the dormant seed in water for 4 months at 4 C, significantly increased the percentage germination to 62%, compared to 28% with seed that were uncut (Table 1). Removing the bases of the pericarps after storage in water for 3 months, did not increase germination more than that of intact seed identically treated. This suggested that an inhibitive substance may have leached more rapidly from the seed with cut pericarps than with seed with uncut pericarps. Apparently an intact pericarp does not allow seed to have sufficient contact with water to permit rapid leaching, but the seed does imbibe some water so the leaching process can proceed slowly.

*Effect of storing seed in different amounts of storage water on dormancy.* Seed that were stored wet at 4 C and had the storage water exchanged once a month for 12 and 10 months gave the largest percentage germination, 67 and 69%, respectively (Table 2). The inhibitive substance appeared to be sufficiently soluble to have most of it removed after 6 monthly changes of storage water. Exchanging the storage water after the 6th month did not increase the percentage germination.

TABLE 1. EFFECT OF CUTTING THE PERICARPS ON THE GERMINATION OF DWARF SPIKERUSH SEED.<sup>1</sup>

Treatment	Percentage germination
Uncut, stored dry at 4 C for 3 mo	4 a
Cut, stored dry at 4 C for 3 mo	4 a
Uncut, stored wet at 4 C for 3 mo	28 b
Stored wet at 4 C for 3 mo, then cut	30 b
Cut, stored wet at 4 C for 3 mo	62 c

<sup>1</sup>Means in a column followed by a common letter are not significantly different at the 5% probability level, according to Duncan's multiple range test.

TABLE 2. EFFECT OF DIFFERENT STORAGE METHODS ON THE PERCENTAGE GERMINATION OF DWARF SPIKERUSH SEED. THE VALUES IN THE TABLE ARE THE PERCENTAGE GERMINATION AFTER VARIOUS MONTHS OF SEED STORAGE IN THE TREATMENTS.<sup>1</sup>

Storage treatment	Months											
	1	2	3	4	5	6	7	8	9	10	11	12
	Test 1											
Dry 4 C	3 a	3 a	3 a	1 a	2 a	4 a	3 a	8 a	5 a	6 a	9 a	7 a
Dry 21 C	2 a	4 a	4 a	4 b	7 b	6 a	11 b	9 a	9 a	11 a	17 a	20 b
Wet 4 C, 6:1 ratio <sup>2</sup>	3 a	8 b	13 b	26 b	27 c	31 b	24 b	19 b	39 b	42 b	43 b	41 c
Wet 4 C, 1:6 ratio <sup>3</sup>	4 a	8 b	15 b	30 c	32 c	39 b	38 d	46 c	52 bc	56 bc	59 bc	60 d
Wet 4 C, rinsed <sup>4</sup>	6 b	21 c	27 c	38 d	45 d	64 c	67 e	64 d	67 c	67 c	64 c	67 e
	Test 2											
Wet 4 C, 6:1 ratio <sup>2</sup>	3 a	4 a	6 a	9 a	—	17 a	22 a	31 a	38 a	48 a		
Wet 4 C, 1:6 ratio <sup>3</sup>	3 a	4 a	9 a	25 a	—	36 b	44 b	55 b	57 b	58 b		
Wet 4 C, rinsed <sup>4</sup>	10 b	25 b	34 b	52 b	—	69 c	71 c	68 c	69 b	69 b		

<sup>1</sup>Means in a column followed by a common letter are not significantly different at the 5% probability level, according to Duncan's multiple range test.

<sup>2</sup>Low volume of storage water.

<sup>3</sup>High volume of storage water.

<sup>4</sup>Storage water changed monthly.

Storing small volumes of seed in large volumes of water (1:6 ratio) resulted in percentages of germination of 60% in Test 1 and 58% in Test 2 after 12 and 10 months, respectively. Large volumes of seed stored in small volumes of water (6:1 ratio) germinated 41% in Test 1 and 48% in Test 2 after 12 and 10 months, respectively. When the storage water was unchanged in both tests, the percentages of germination generally showed gradual increases with each additional month of treatment. These monthly increases in germination suggested that an inhibitive substance may have been slowly released from the seed. The substance leached slowest from seed stored in small quantities of storage water. After the percentages of germination reached the highest levels in both tests, small fluctuations in the percentages occurred, apparently the result of random sampling of the seed.

Seed stored dry at 4 and 21 C for 12 months germinated 7 and 20%, respectively. Comparatively little is known about the removal of dormancy during dry storage.

*Effect of an extract from pulverized dormant seed on the percentage germination of viable seed.* The extract made from dormant seed was significantly more inhibitive to the germination of viable test seed than the extract made from non-dormant seed (Table 3). Test seed did not respond to small differences in the concentrations of the extracts. In-

TABLE 3. PERCENTAGE GERMINATION OF VIABLE SEED OF DWARF SPIKERUSH INCUBATED IN DIFFERENT AMOUNTS OF EXTRACTS MADE FROM DORMANT AND NON-DORMANT DWARF SPIKERUSH SEED.<sup>1</sup>

Amount of extract (ml)	Germination in Different seed extracts	
	Dormant (%)	Non-dormant %
10.0	2 a	7 b
5.0	2 a	9 b
2.5	6 ab	10 b
1.0	11 b	30 c
0.02	38 d	

<sup>1</sup>Means in both columns followed by a common letter are not significantly different at the 5% probability level, according to Duncan's multiple range test.

<sup>2</sup>Test seed germinated in distilled water.

TABLE 4. PERCENTAGE GERMINATION AND ELONGATION OF COTYLEDONARY SHEATHS OF NON-DORMANT DWARF SPIKERUSH SEED INCUBATED IN THE LEACHATE OF WET-STORED SEED.<sup>1</sup>

Concentration of leachate (%)	Germination (%)	Average length of cotyledonary sheath (mm)
100	8 a	0.2 a
50	33 b	1.6 b
25	33 b	1.8 b
0	54 c	2.6 c

<sup>1</sup>Means followed by a common letter are not significantly different at the 50% probability level, according to Duncan's multiple range test.

incubation of seed in large dilutions of each extract (1 ml extract in 9 ml distilled water) caused a significant difference in percentage germination when compared to incubation in undiluted extract (10 ml of extract only).

*Effect of the leachate from dormant stored seed on the germination of non-dormant seed.* When non-dormant seed were incubated in undiluted leachate, the percentage germination was 8% as compared to 54% for controls (Table 4). The growth of the cotyledonary sheath was significantly inhibited when test seed were incubated in the leachate, regardless of concentration. The cotyledonary sheaths of seedlings germinated in the leachate were thin and white, compared to the green cotyledonary sheaths on seedlings germinated in distilled water.

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