

## LONG-TERM STORAGE OF GRASS SEEDS

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Seeds of crested wheatgrass, *Agropyron cristatum* (L.) Gaertn., intermediate wheatgrass, *A. intermedium* (Host.) Beauv., and smooth brome grass, *Bromus inermis* Leyss., were stored for 20 yr under various conditions. Temperature was a major factor affecting success with viability inversely related to storage temperature. At -7 and -18°C, viabilities of 80-90% were shown after 20 yr storage. Drying seed for 7.5 h at 60°C prior to storage gave little improvement over undried seed stored with 8% moisture. Plastic bags gave storage as good as glass jars with screw-top lids, although plastic bags were less effective in excluding moisture. It was concluded that adequate germination for reproduction of strains could be obtained after 25-30 yr of storage.

Key words: Storage, grass, seeds

[Longue conservation de semences de graminées.]

Titre abrégé: Longue conservation de semences de graminées.

Des semences d'agropyre à crête, *Agropyron cristatum* (L.) Gaertn., d'agropyre intermédiaire, *A. intermedium* (Host.) Beauv. et de brome inerme, *Bromus inermis* Leyss. ont été conservées pendant 20 ans dans diverses conditions ambiantales. La température est un facteur important de réussite puisque la viabilité des semences est inversement proportionnelle à la température de conservation. À -7 et -18°C, on obtient des viabilités de 80 à 90% au bout de 20 ans. Le séchage des semences pendant 7,5 heures à 60°C avant la conservation n'apporte que peu d'amélioration par rapport à l'entreposage des semences humides (8% d'humidité). Les sacs en plastique se prêtent aussi bien à la conservation que les bocaux en verre à couvercle vissé, encore que les premiers sont moins étanches à l'humidité. Les auteurs concluent qu'il est encore possible d'obtenir un bon taux de germination pour la reproduction des lignées au bout de 25 à 30 ans de conservation des semences.

Mots clés: Conservation, graminées, semences

Sub-freezing storage temperatures are proving very effective in prolonging the viability of grass seeds: Rincker and Maguire (1979) and Rincker (1981) found over 80% germination for several grasses stored for 14 yr at -15°C and 60% RH. In addition, there were no detrimental effects on the yields of crops grown from these stored seeds. Bass and Stanwood (1978) reported good viability of sorghum seed, *Sorghum bicolor* (L.) Moench, stored for 16 yr at -1 and -12°C, regardless of

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seed moisture content, and atmospheric environment of the storage medium. Knowles (1967) noted good viability of seeds of three grasses maintained for 10 yr at -18°C, with little advantage of sealed seed over unsealed seed at this temperature. Nakamura (1975) obtained 62-95% germination for various grass seeds after 13 yr of storage at 0°C using CaCl<sub>2</sub> as a desiccant. Evans (1957) found that reducing ryegrass seed moisture to 1.62% allowed good preservation of seed for 15 yr, with the first 8 yr at 15°C and the last 7 at

0°C. Burton (1979) emphasized seed storage in the preservation of germplasm and recommended drying seed to 5–7% moisture, sealing in glass containers, and storing at –20°C. The International Board for Plant Genetic Resources (1976) also recommended drying of seed to  $5 \pm 1\%$  moisture, sealing, and storing at –18°C or less.

This paper presents longevity data for seeds of three perennial grasses which were placed in storage in 1958, 1960, and 1962 using a variety of storage conditions.

## MATERIALS AND METHODS

Separate tests on seed storage were initiated in 1958, 1960, and 1962, and periodic determinations of germination were made until 1981. Following harvest, seeds were held indoors at room temperatures in cotton bags until the January–February period when tests were commenced.

### 1958 Test

This test, on which preliminary results were reported previously (Knowles 1967), involved seed of crested wheatgrass, smooth brome grass, and intermediate wheatgrass harvested in 1955 and 1957. Seeds were stored at 21, 1, and –18°C, using (a) glass bottles with screw-top lids, and (b) paper envelopes.

### 1960 Test

Seeds from the 1959 harvest of the same three grasses were dried at 50 and 60°C for 2.5, 5, and 7.5 h prior to storage. Storage was at 21°C and –7°C using (a) glass bottles, and (b) plastic bags of 1-, 2-, and 3-mil thickness. Moisture

determinations of seed were made immediately following drying in 1960 and again in 1961 and 1972.

### 1962 Test

Seeds harvested in 1959 and 1961 were placed in storage in February, 1962, at 21, 5, 1, –7, and –18°C. Seed was (a) dried at 60°C for 7 h and stored in glass jars, (b) not dried, and stored in glass jars, and (c) not dried, and left in paper bags. Between the date of harvest and storage, seed was kept in cotton bags at 21°C and approximately 23% RH.

Drying was done in a Unitherm electric dryer with seed at 3 cm depth in screen trays. Germination tests were made using duplicate 100-seed samples. Only plump, well-filled seeds were used for germination tests. Seeds were placed between moistened paper towels at 21°C without prechilling. Final germination counts in 1981 were made at 14 days, though germination counts previous to this date were for 8–10 days. Moisture contents were reported on the basis of seed dry weight.

## RESULTS

### 1958 Test

All seed stored at room temperature (21°C) was dead in 1981. Seed stored at 1°C was also dead except for some 1957 seed stored in bottles (Table 1). Seed stored at –18°C germinated fairly satisfactorily when stored in glass jars, but less satisfactorily when stored in paper envelopes. This difference in germination was less evident in 1966 after 8 yr of storage (Knowles 1967). Seed from the 1955 harvest, which was not

Table 1. Percent germination of three grasses in 1958 and in 1981 after storage at 1°C and –18°C

Grass	Year of harvest	Initial germination 1958	Germination 1981			
			1°C storage		–18°C storage	
			Paper envelope	Glass jar	Paper envelope	Glass jar
Crested wheatgrass	1955	78	0	0	4	56
	1957	74	0	0	22	84
Smooth brome grass	1955	86	0	0	42	60
	1957	91	0	18	61	78
Intermediate wheatgrass	1955	97	0	0	61	90
	1957	94	0	12	81	92

stored at low temperatures until 1958, suffered more deterioration than the 1957 seed. This points to the desirability of cold storage commencing shortly after harvest. Intermediate wheatgrass, which had the largest seed of the three grasses, showed best seed viability in 1981. However, initial germination of intermediate wheatgrass was higher than for crested wheatgrass or bromegrass.

### 1960 Test

The effectiveness of drying treatments in reducing seed moisture content and of various containers in maintaining low mois-

ture content is shown in Table 2. The very low moisture content shown immediately following drying was not maintained, as shown by the 1972 moisture determinations. The moisture content of 3.3% for seed prior to drying seems unusually low. This may have resulted from a low relative humidity of 20% and indoor storage for several months prior to cold storage in 1960.

Seed stored at room temperature (21°C) showed poor germination in 1972 after 12 yr of storage (Table 3). There was little difference in germinability between dried and nondried seed or between types of stor-

Table 2. Average moisture content of 1959 seed of three grasses immediately after drying in 1960 and after storage to 1961 and 1972

Drying treatment 1960	Storage container	Moisture %				
		After drying 1960	21°C storage		-7°C storage	
			1961	1972	1961	1972
None	Glass jar	3.3	8.0	6.4	8.3	9.5
50°C, 2.5 h	Glass jar	1.8	6.1	6.1	5.9	7.4
50°C, 5 h	Glass jar	1.4	5.5	5.9	4.7	5.7
50°C, 7.5 h	Glass jar	1.2	4.8	5.9	4.0	4.6
60°C, 2.5 h	Glass jar	2.0	6.2	5.9	5.3	7.6
60°C, 5 h	Glass jar	0.9	5.1	5.7	3.9	6.3
60°C, 7.5 h	Glass jar	0.8	4.0	5.8	3.2	6.1
60°C, 7.5 h	Plastic 1 mil	0.8	6.5	5.4	3.6	9.7
60°C, 7.5 h	Plastic 2 mil	0.8	5.9	5.5	3.3	7.7
60°C, 7.5 h	Plastic 3 mil	0.8	5.5	5.8	3.4	6.8

Table 3. Average percent germination of 1959 seed of three grasses following various drying treatments 1960 and after storage to 1972 and 1981

Drying treatment 1960	Storage container	Storage condition				
		1960	21°C		-7°C	
			1972	1981	1972	1981
		Germination (%)				
None	Glass jar	93	10	0	96	96
50°C, 2.5 h	Glass jar	91	20	0	98	96
50°C, 5 h	Glass jar	90	11	0	97	97
50°C, 7.5 h	Glass jar	93	19	0	97	96
60°C, 2.5 h	Glass jar	89	9	0	98	95
60°C, 5 h	Glass jar	87	21	0	97	96
60°C, 7.5 h	Glass jar	88	26	0	96	95
60°C, 7.5 h	Plastic 1 mil	-	9	0	96	94
60°C, 7.5 h	Plastic 2 mil	-	15	0	97	89
60°C, 7.5 h	Plastic 3 mil	-	16	0	96	96

age container at 21°C. At -7°C, viabilities were well maintained to 1981, after 21 yr of storage. At this temperature there also was no benefit from drying seed and no benefit from bottling seed as against storing it in plastic containers. Drying seed at 50°C for as long as 7.5 h was not detrimental, but drying at 60°C caused a small reduction in germination. The low seed moisture contents prior to drying at these temperatures may have helped to prevent seed damage.

### 1962 Test

This test was designed to test a wide range of temperatures on seed viability as well as the effects of drying and type of seed container. Prior to drying in February 1962, seed was maintained in inside storage and showed an average of 6.8% moisture for the three grasses, and average germination of 97%. Following drying at 60°C for 7 h the average moisture content was reduced to 3.2% with germination still at 97%. Moisture contents increased substantially by 1969 and again by 1981 (Table 4). The high moisture contents for seed stored in bottles appears to indicate leakage or the accumulation of moisture when bottles were opened periodically for germination tests. Some moisture may also have resulted from respiration of the seed.

Excellent retention of viability was shown at -18°C, regardless of type of storage or drying treatment (Table 5). At -7°C, good viabilities were shown when seed was stored, dried or undried, in glass

jars but not when placed in envelopes. At 1°C there was a clear advantage for storage in glass jars compared to envelopes, as shown by 1969 germinations. By 1981 only the combination of jar storage with drying of seed gave appreciable viability at 1°C. Temperatures of 5°C showed little advantage over 21°C in maintenance of viability. Here again, sealing of seed in bottles along with drying was desirable.

Higher germination percentages were shown in 1969 and 1981 than immediately after drying in 1962. This appears to reflect variation in germination techniques between germination dates, especially a shorter germination period in 1962.

Although results for 1959 seed lots were not included in Table 5, they were in keeping with results of the 1961 seed. In most instances, the viability of 1959 seed was below that of 1961 seed, showing that cold storage should commence within a few months of seed harvest.

### DISCUSSION

The value of low temperatures for the preservation of viability of grass seeds is evident from this and other studies. The limits of longevity under the most favorable conditions must await additional years of storage. However, it would appear that 25-30 yr of storage is quite possible, using temperatures in the range of -7 to -18°C. Our present cold storage facilities became available in 1958, and 18 seed lots of these three grasses outside the present tests and representing 1951 to 1956 harvests were

Table 4. Moisture content of 1961 seed following various storage treatments, 1962 test, averages for three grasses

Storage temperature (°C)	Envelope not dried		Glass jar not dried		Glass jar dried	
	1969	1981	1969	1981	1969	1981
	<i>Moisture (%)</i>					
21	6.2	6.9	5.5	7.2	4.7	6.9
5	16.3	13.3	12.3	14.5	9.8	14.6
1	16.7	14.5	11.1	15.2	7.3	13.8
-7	15.0	13.1	8.6	12.7	6.4	11.8
-18	14.9	13.3	7.8	11.1	5.1	8.7

Table 5. Percent germination of 1961 seed of three grasses placed in different types of storage in 1962 and tested in 1969 and 1981

Storage temperature (°C)	Germination (%)					
	1969			1981		
	Envelope	Glass jar	Glass jar dried	Envelope	Glass jar	Glass jar dried
	<i>Crested wheatgrass</i>					
21	7	31	65	0	0	0
5	0	49	87	0	0	0
1	0	79	84	0	0	25
-7	66	92	95	1	90	90
-18	92	83	95	90	88	93
	<i>Smooth bromegrass</i>					
21	7	32	64	0	0	0
5	0	78	88	0	0	0
1	22	90	93	0	0	82
-7	70	96	90	1	91	94
-18	88	93	95	95	92	96
	<i>Intermediate wheatgrass</i>					
21	12	57	75	0	0	0
5	0	80	92	0	0	0
1	0	97	94	0	0	58
-7	94	95	97	26	92	90
-18	93	93	97	95	98	97

stored at  $-18^{\circ}\text{C}$  in glass bottles. All these lots showed sufficient germination in 1981 to allow reproduction of strains.

Longevities of the three grasses were fairly similar, although intermediate wheatgrass which had the largest seed showed somewhat better longevity than crested wheatgrass which had the smallest seed. There was little indication that bromegrass seeds had less longevity than other grasses as reported by Rincker (1981). A few samples of timothy, *Phleum pratense* L., reed canarygrass, *Phalaris arundinacea* L., and hard fescue, *Festuca ovina* var. *duriuscula* (L.) Koch, stored at  $-7^{\circ}\text{C}$  in plastic bags had good viability after 20 yr. This suggests that the present results apply generally to grass seeds.

The choice of seed container for maximum longevity was not clear from these studies. At the lowest temperature of  $-18^{\circ}\text{C}$  there was good retention of viability in both paper containers and glass bottles (Table 5). However, at  $-7^{\circ}\text{C}$  the use

of bottles was preferable. Light polyethylene bags had limited value in excluding moisture as was pointed out by Justice and Bass (1978). They recommended 5- to 10-mil polyethylene bags as giving better protection. Plastic bags are convenient where large and variable amounts of seed are stored and have been adopted for general use at this Station.

Drying of seed in these studies did not greatly prolong seed viabilities. Since seeds prior to storage were in the 3-8% moisture range, it is likely that benefits from artificial drying were minimal. Storing seed indoors at  $20^{\circ}\text{C}$  and at 20% RH in mid-winter appears to be an effective method of drying seed preparatory to storage.

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