Effects of Presowing Vernalization on Survival and Development of Several Grasses

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Improvement of plant establishment, especially on poor sites, is an important problem in seeding depleted watersheds and ranges in the Intermountain West. One approach to this problem lies in plant stimulation for more rapid development to enhance the young plant's chances for survival. The effectiveness of presowing vernalization of seeds in producing this kind of stimulation in some perennial grasses was investigated in experiments reported here. The procedure involves storing soaked seeds at near-freezing temperatures for several weeks as a conditioning process for subsequent phasic development and flowering, as distinguished from mere growth.

These studies are a follow-up of earlier work (Frischknecht, 1951) which suggested that in certain grasses fall planting stimulated faster growth and development than spring planting, aside from the fact that seedlings began growing earlier in the spring. Mountain rye *(Secale montanum)*, intermediate wheatgrass *(Agropyron intermedium)*, and a native strain of mountain brome *(Bromus carinatus)* were 3 of 16 grasses studied which merely stooled from spring plantings, but they flowered and produced seed the first year from late fall plantings (Figure 1) even though seedlings did not emerge until spring. Inasmuch as this behavior parallels reactions in so-called "winter" cereals, it was believed possible to "vernalize" seed of some perennial range grasses and thereby speed the development of spring plantings. It appeared that this could be done underneath snow, especially inasmuch as soil thermograph records had shown that winter temperatures near the ground surface under snow remained close to 32° F., which were similar to temperatures used in vernalizing cereals.

According to the symposium by Murneek and Whyte (1948) vernalization of seeds was considered to have great practical value in Russia at one time for hastening maturity in annual plants. Winter cereals had received most attention. However these authors concluded that vernalization would have little practical value in countries not experiencing extreme conditions of frost, drought, and floods especially when superior genotypes were found. An exception would be in the production of market garden crops where a few days' earliness would mean increased financial returns. Martin (1934) considered the process to have little practical value in the United States. McKinney (1940) noted that most investigators outside Russia attached little importance to the economic gain from vernalization of plants.

Investigations of vernalization of both plants and seeds have continued on a great variety of species, mostly in other countries. In this connection Wellensiek (1952) gave four illustrations of control of flowering: (1) plants that are insensitive to low temperatures and day length; (2) plants that require cold when they have reached a certain vegetative size (plant vernalization); (3) plants that react to seed vernalization; and (4) plants sensitive to plant vernalization but also to short day, provided it is followed by long days. Sechet (1953) grouped a number of plants into three categories with respect to their requirement of a period of vernalization as follows: (1) determinant, in which a cold period is valuable but not always indispensable for reproduction (winter cereals, tulips); (2) favorably responding to cold, in which flowering is precocious but occurs without a cold period (mustards, lupine, peas, etc.); (3) no favorable response to vernalization (onion, flax, bean, etc.).

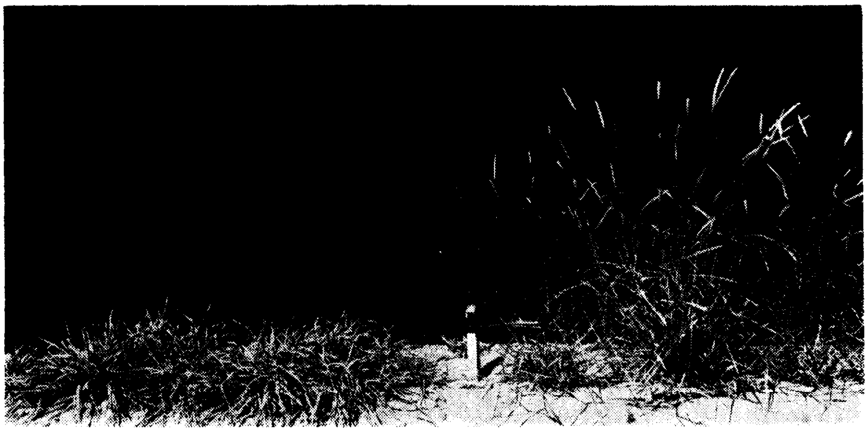

FIGURE 1. Mountain rye from spring planting *(left)* merely stooled during first growing season, whereas fall planting *(right)* stimulated flowering and seed production, although seedlings did not emerge until spring.

Some investigations, such as the series by Gott, Gregory, and Purvis (1955), have been aimed at discovering the fundamental biological processes involved. Most of the recent investigations in the United States have centered around substances affecting plant development and flowering and control of the process by their application. Associated aspects of photoperiodism have received more attention than temperature — the main factor in vernalization. Little work has been done on seed vernalization in perennial grasses.

FIGURE 2. Two years after spring planting at Gunnison good stands were present only from snowbank-treated seeds of Russian wildrye *(right foreground)*, crested wheatgrass *(third row left)*, and fairway wheatgrass *(seventh row left)*. The other half of each row had been planted with untreated seeds.

Preliminary Attempt at Vernalization

The first of this series of studies, by the author, made in central Utah, involved soaking seeds of eight perennial grasses for 20 hours at room temperatures and then burying them in a snowbank at the ground surface for 50 days before planting. The grasses were mountain rye, intermediate wheatgrass (regular and Amur strains), pubescent wheatgrass *(A. trichophorum)*, tall wheatgrass *(A. elongatum)*, fairway wheatgrass *(A. cristatum)*, crested wheatgrass *(A. desertorum)*, and Russian wildrye *(Elymus junceus)*.

Duplicate samples of seeds thus treated and similar untreated samples were planted at comparable rates in early April 1952, at each of three locations in central Utah: (1) Benmore, big sagebrush *(Artemisia tridentata)* type, average annual precipitation 12.8 inches; (2) Tintic Valley, big sagebrush type, estimated average annual precipitation 10 inches; and (3) Gunnison, shadscale *(Atriplex confertifolia)* type, estimated average annual precipitation 8 inches. Seed samples were planted in separate 5-foot rows, spaced 20 inches apart. The soil was moist on all sites at the time of planting; this helped to prevent the moist snowbank-treated seeds from drying. A light rain fell at Gunnison on the day of planting, and moisture fell at the other two sites on the third day after planting.

Seed germination tests immediately following removal from the snowbank showed that viability was not impaired, except in mountain rye. Many seeds of mountain rye were decomposing after 50 days in the snowbank; similar damage was reported previously for dehulled seeds of this grass, tall oatgrass, and orchardgrass overwintering in cloth bags in the ground (Frischknecht, 1951). Apparently, if seed is dehulled, planting of these grasses should be done when there is reasonable chance for prompt germination.

Except for mountain rye, treated seeds produced good to excellent seedling stands on all plots at all three sites. None of the grasses produced seed the first year, but plants from all snowbank-treated seeds emerged a few days earlier than plants from untreated seeds, and they grew taller the first year. This was particularly striking in the two strains of intermediate wheatgrass: their seedlings from treated seeds tended toward culm elongation instead of stooling like seedlings from untreated seeds. A similar tendency was observed on the few plants of mountain rye that were present on each site from snowbank-treated seed. Such development would suggest that vernalization had occurred in these two species at least.

The generally good initial stands were maintained at Benmore. Rabbits greatly damaged the plantings at Tintic Valley by the end of the first season. The following spring only the two rows of Russian wildrye and one row of pubescent wheatgrass from snowbank-treated seed, plus one row of Russian wildrye from untreated seed showed good stands. High mortality occurred at the shadscale site, and by 1954 the only remaining good stands were from snowbank-treated seeds of Russian wildrye, crested wheatgrass, and fairway wheatgrass (Figure 2).

Snowbank versus Other Presowing Treatments

Other presowing treatments were compared with snowbank treatment in a subsequent test involving four selected species. Presowing treatments of seed included: (1) 20 hours' soaking at room temperature followed by snowbank storage for 48 days; (2) 20 hours' soaking followed by 48 days' storage in a locker at extremely cold temperatures (near 0° F.); (3) 36 hours' soaking followed by 3 days' locker storage; and (4) 36 hours' soaking followed by no cold treatment. Species used were intermediate wheatgrass, which is normally

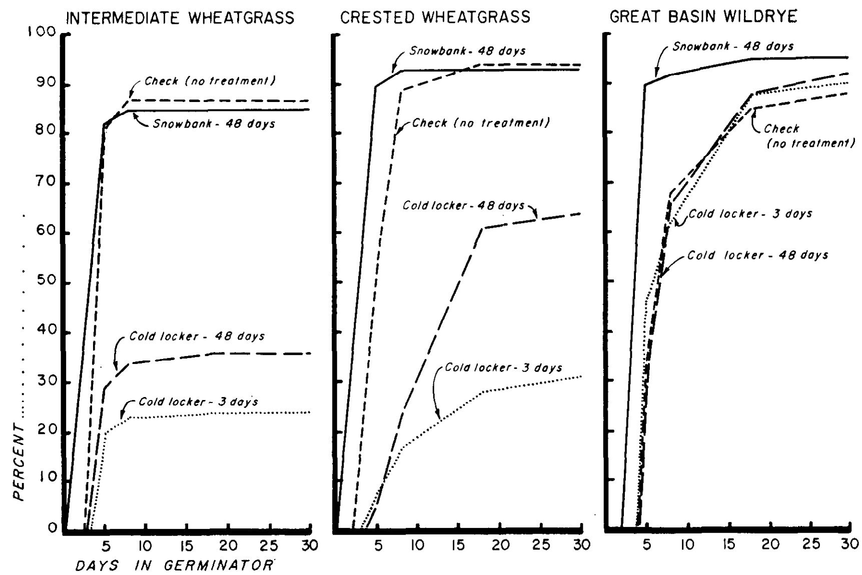

FIGURE 3. Seed germination in a germinator following different treatments.

quick to germinate; crested wheatgrass, almost equally fast to germinate; Great Basin wildrye *(E. cinereus),* considerably slower in germinating; and an extremely seed-dormant strain of Indian ricegrass *(Oryzopsis hymenoides).*

On April 2, 1953, triplicate 100-seed samples of each grass under each treatment, plus comparable samples of untreated seeds, were planted at Benmore in separate 5-foot rows, spaced 20 inches apart. Viability of all seed, except seed soaked for 36 hours followed by no cold, was tested in the germinator.

Response differences in the germinator (Figure 3) reflect inherent characteristics of the grasses. Indian ricegrass failed to germinate and thus showed no response to the treatments. On the other hand snowbank-treated seeds of the other grasses and untreated seeds of both wheatgrasses germinated readily and almost completely. Locker storage near 0° F. reduced germination of the wheatgrasses, but apparently did not affect Great Basin wildrye, as evidenced by the paralleled responses to untreated seed. Since the wildrye is normally the slowest of the three to germinate, it is likely that these seeds were metabolizing less rapidly than those of the wheatgrasses when placed in the cold storage locker and consequently were less susceptible to damage from cold temperature. Barton (1954) assumed that the more rapidly metabolizing seeds were more susceptible to injury. Lower germination of seeds from the 3-day locker treatment than the 48-day locker treatment in the wheatgrasses is believed due to the longer period of prior soaking rather than to length of locker storage; seeds soaked for 36 hours would have been nearer sprouting than those soaked for 20 hours, and therefore more susceptible to damage by extreme cold.

Indian ricegrass showed no emergence the first year from any of these treatments in field plantings at Benmore (Table 1). An average of 6.2 percent emergence a year following planting plus an additional 0.8 percent emergence 2 years following planting showed no effect of presowing cold treatment. The nonresponse in the germinator and delayed emergence in the field conform with results reported previously for this particular source of seed, and is associated with a hard seed coat and possibly a dormant embryo (Plummer and Frischknecht, 1952).

Emergence of snowbank-treated seeds of the other three species began about 21 days after planting at Benmore; these seeds also had the highest total emergence (Table 1). Although total emergence of intermediate wheatgrass from snowbank-storage and from the soaking treatment was about equal, the following year only intermediate wheatgrass from snowbank-treated seeds showed good stands; a very few plants of this species survived from other treatments. The only other survivors were a few plants of crested wheatgrass from the snowbank, soaked, and 3-day locker treatments.

Plantings similar to those at Benmore were made one day later near Ephraim at an elevation of 5,500 feet in the sagebrush zone, a site comparable to that at Benmore, (elevation 5,800 feet) and in Ephraim Canyon at an elevation of 7,200 feet in a moister, mountain-brush

Table 1. Seedling emergence of four grasses from spring seeding at Benmore following different presowing treatments.

Presowing treatment	Intermediate wheatgrass	Crested wheatgrass	Great Basin wildrye	Indian ricegrass*
	Percent	*Percent*	*Percent*	*Percent*
Snowbank 48 days	28.3	10.7	16.0	6.3
Cold locker 48 days	11.0	3.3	.0	3.7
Cold locker 3 days	7.3	1.3	.0	9.3
Soaked, no cold	26.3	4.3	.0	5.3
Check (no treatment)	12.0	4.0	.0	6.3

* Emergence of this species occurred one year following planting; for other species the data are for the year of planting.

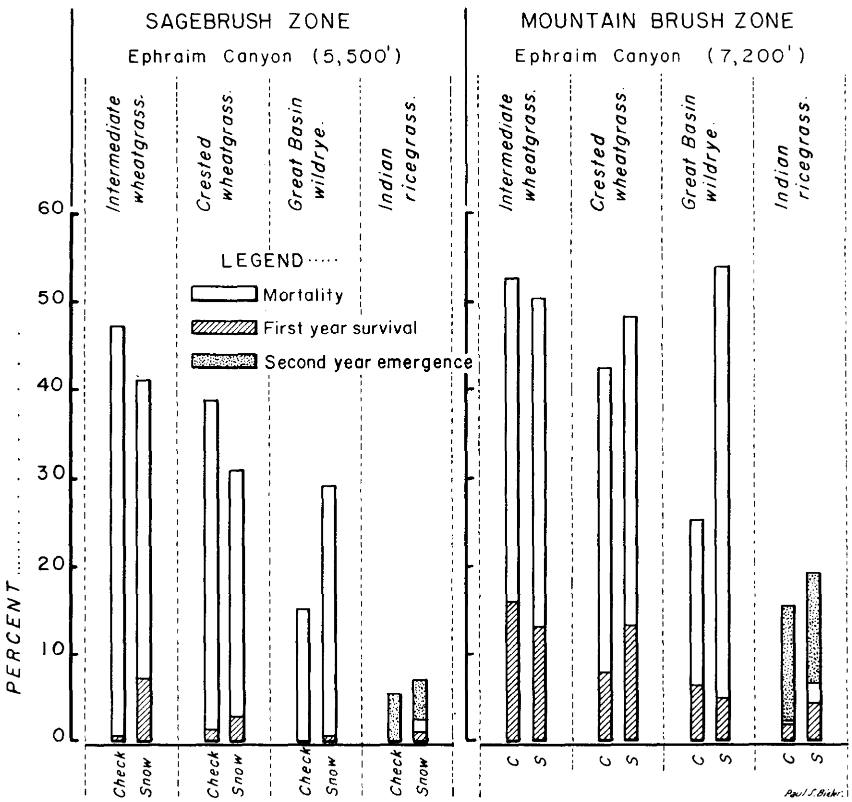

FIGURE 4. Emergence, mortality, and survival of plants from snowbank-treated and untreated seeds (check) at two sites near Ephraim.

site having more fertile soil. Only seedlings from snowbank-treated and untreated seeds were counted, but general observations were made on the other plantings. Figure 4 shows that survival of all species in the sagebrush zone was somewhat better from snowbank-treated seeds than from untreated seeds, but differences were less consistent in the mountain-brush zone. At both sites first-year emergence and survival of Indian ricegrass were best from snowbank-treated seeds; again, the somewhat higher second-year emergence of this grass showed no advantage for or against treatment.

Although there was little difference in total emergence or in first-year survival of intermediate wheatgrass at the mountain-brush site, plants from the snowbank-treated seeds flowered and produced seed in the first growing season—conclusive evidence of vernalization (Figure 5). These effects were apparently carried over into the second year because on two of the three replications, plants from vernalized seeds produced more than half again as much herbage as the untreated and about one-third more flower stalks. On the other replication, plants from vernalized and untreated seeds were about equal in herbage yield and flower stalk production. Cepikova (1935) reported that effects of presowing vernalization were carried over into the second year on *Phleum pratense, Alopecurus pratense*, and *Trifolium pratense*.

One plant of intermediate wheatgrass from the 48-day locker treatment bolted and flowered the first year; otherwise there was no evidence of vernalization in this species from any but the snowbank treatment. Although a greater percentage of plants of the other three species flowered the first year from seed that had the snowbank treatment than from untreated seeds, some plants flowered under all treatments. This and other observations indicate that vernalization treatment is unnecessary for first-year flowering when these three grasses are grown under favorable conditions. Apparently they could be classified with the second group of plants described by Sechet (1953) in which flowering was more precocious as re-

FIGURE 5. Vernalization of intermediate wheatgrass seeds prior to planting stimulated flowering and seed production *(rear row)*, whereas plants from untreated seeds *(foreground)* only stooled in first growing season (mountain-brush site, Ephraim Canyon).

sult of vernalization. On the other hand, intermediate wheatgrass would fall in his first class, where vernalization was "determinant" for first-year flowering. It is not known whether the earlier emergence from the treated seed contributed to the increased flowering in the three species: likely, it had little or no effect on intermediate wheatgrass.

Fall Planting versus Presowing Treatments and Spring Planting

Further trials were conducted to see how snowbank storage followed by spring planting compared with fall planting; also, to check refrigerator storage at near-freezing temperatures *vs.* snowbank storage. Fall plantings of the four grasses were made into moist soil in late November 1953, at the three locations and in the manner previously described. Then, early in April 1954, similar plantings were made of: (1) untreated seed, (2) soaked seed that had been in snowbank storage for 60 days, and (3) soaked seed that had been in refrigerator storage in plastic bags at 31° to 34° F. for 60 days.

Previous work (Frischknecht, 1951) had shown that seedling mortality was not serious underneath snow but that the period during and immediately following snowmelt was critical for seedlings. In the present study, seedlings from fall plantings did not emerge until after snowmelt, but it is possible that they were adversely affected during this period while in the pre-emergence stage. Emergence from fall plantings was first observed at the two sites near Ephraim on March 15, and at Benmore on March 24. Continuous snow cover had remained on fall plantings near Ephraim from December 1 to March 8 at the mountain-brush site, and from December 1 to February 22 at the sagebrush site. The snow cover was thinner and sometimes intermittent at Benmore during this period. Subsequent storms left the ground covered with snow for rather brief periods. Emergence of spring plantings began about April 25 — approximately 2 weeks after planting at all three sites.

Figure 6 shows varied results, some favoring fall planting and some favoring snowbank or refrigerator treatment followed by spring planting. All three produced germination generally superior to that from untreated

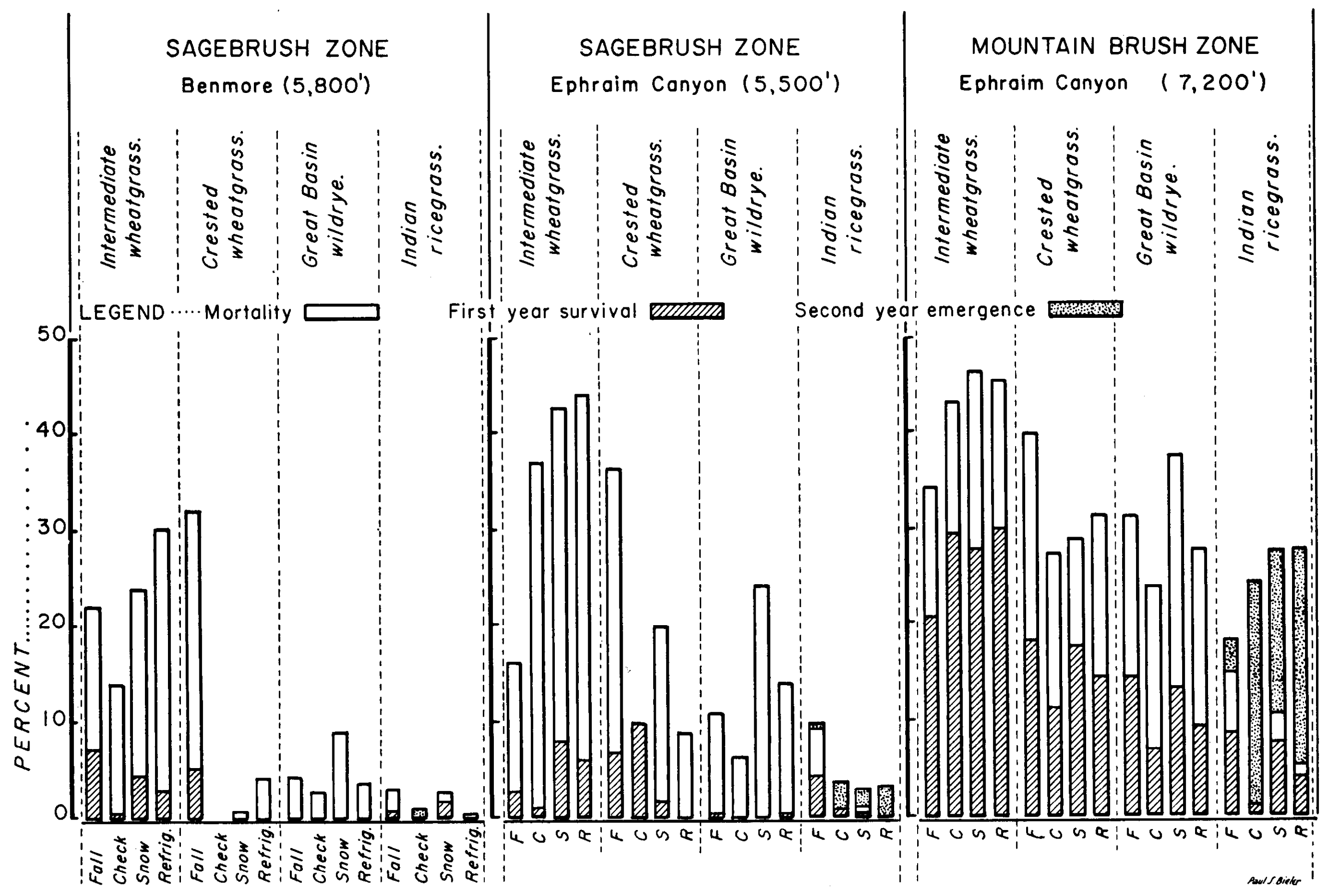

FIGURE 6. Percentage of emergence, mortality, and survival of four grasses at three sites in relation to planting season and vernalizing seed treatments: *F*, fall-planted, untreated; *C*, (check) spring-planted, untreated; *S*, spring-planted, snowbank-treated; *R*, spring-planted, refrigerator-treated.

spring plantings. Differences between snowbank and refrigerator storage appeared to favor the former, but some differences were inconsequential. It is to be expected that fall plantings will produce superior stands in some years and vernalized spring plantings in others, depending upon weather and soil conditions during and shortly after emergence. In the present test, crested wheatgrass showed relatively high emergence from fall planting, whereas highest emergence of Great Basin wildrye resulted from snowbank storage and spring planting. As in previous tests, mortality was high on all plantings, especially in the sagebrush zone.

Survival of intermediate wheatgrass, in particular, was enhanced on the two dry sites by vernalization, either from fall planting or snowbank or refrigerator storage prior to spring planting. Although growing conditions were not conducive to maximum development as in the mountain-brush zone, the increased survival in the sagebrush zone was probably more important. Seemingly small differences become quite important when it is considered that a 5-percent survival in these tests means an average of one plant per linear foot of row.

Indian ricegrass showed generally higher first-year emergence from fall planting than from other treatments, but snowbank-treated seeds gave much better response than the untreated seeds. Survival from snowbank treatment compared very favorably with that from fall planting. Second-year emergence of this grass was relatively high at the mountain-brush site, but many of these seedlings failed to become established in the rows where plants originated the first year. Even in rows without year-old plants it appeared that competition from the adjacent rows was severe, and the slowly developing new plants were at a distinct disadvantage (Figure 7). However, second-year emergence, which in this case was particularly high from spring plantings, should aid in securing a stand where initial establishment is poor.

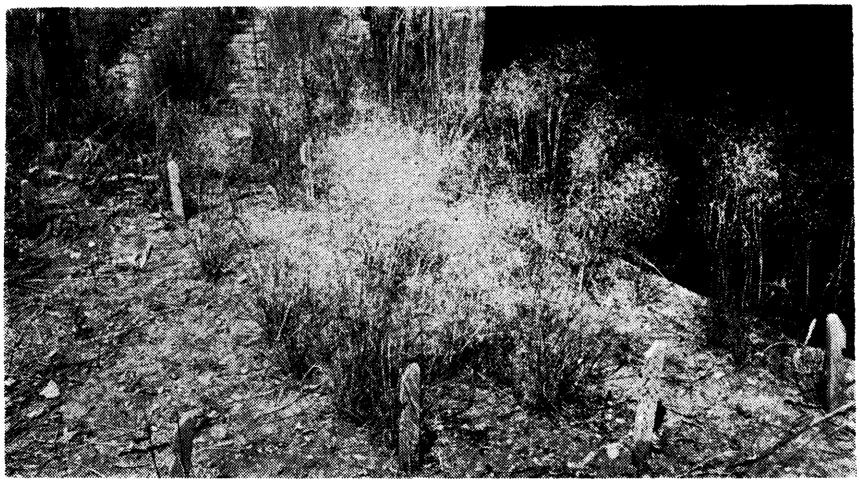

FIGURE 7. Young plants of Indian ricegrass in left row, which originated 1 year after planting, are at a disadvantage with plants originating a year earlier from: *(left to right)* fall-planted, untreated seed; spring-planted, refrigerator-treated seed; and spring-planted, snowbank-treated seed.

Discussion

Although both snowbank and refrigerator storage were effective in vernalizing seeds, the snowbank treatment had some practical advantages in that it required no special equipment and seeds did not dry out, making it unnecessary to add water during the process. Ten pounds of intermediate wheatgrass seed that had been soaked in a tub of water for 20 hours weighed 17 pounds after removal and drainage for 2 hours, a weight increase of 70 percent. After storage in a snowbank for 45 days, the weight remained exactly the same. Lojkin (1936) reported that the effective moisture content for vernalizing winter wheat was 50 to 70 percent on the basis of dry weight before treatment; and where the initial moisture content was less than 60 percent and subsequent drying occurred, it was necessary to add water during vernalization.

Refrigerator storage in plastic bags was superior to storage in cloth bags. Seeds in cloth bags tended to dry; this made necessary the periodic adding of water from an adjacent covered jar, kept at the same temperature as seeds. This was unnecessary for the seeds in plastic bags, but the bags were turned over occasionally because moisture from the seeds tended to condense on the upper inside surfaces. Also, sprouting of the wheatgrasses began in cloth bags after 29 days in the refrigerator, but there was no sign of sprouting in plastic bags until after 46 days.

Snowbank storage of 10 pounds, and later 20 pounds, of intermediate wheatgrass seed was simply accomplished by burying burlap sacks containing the soaked seeds underneath snow on a north-facing hillside in February. Packing snow over the bags caused these mounds to remain after other snow had melted in the spring. During the course of study, seeds were stored in snow for 38, 45, 50, 60, and 67 days—periods similar to those reported by others for vernalizing cereals. Every treatment period stimulated culm production in intermediate wheatgrass, but excessive sprouting usually began after about 45 days in the snowbank.

It was not difficult to drill vernalized seed of intermediate wheatgrass if sprouting had not occurred, and it appeared that other species could be drilled as well. Although the swollen seeds were moist when removed from the snowbank, they dried rapidly on the outer surface when exposed to air and slid through the drill box fairly easily. However, an agitator in the drill would have been beneficial.

It is doubtful whether a few hours' drying to facilitate ease of drilling would have been detrimental, but drying for 7 days in a basement following snowbank storage resulted in about 50 percent lowered emergence when seeds were planted in the field. However, inasmuch as some plants flowered the first year, devernalization did not occur completely, if at all, as has been reported for some other plants (Lojkin, 1936; Purvis and Gregory, 1952). In fact, supplementary observations showed that some seeds of intermediate wheatgrass remained vernalized for at least 1 year when dried and stored in a basement after removal from the snowbank. The extremely low rate (5 percent) of germination of this seed may have been influenced by the fact that much sprouting occurred before these seeds were dried. Seeds of crested wheatgrass, Great Basin wildrye, and Indian ricegrass which had been similarly stored for 1 year would not germinate in the germinator.

Summary and Conclusions

Fall planting or storage of soaked seed in a snowbank or refrigerator prior to spring planting accomplishes vernalization of some perennial grasses, notably intermediate wheatgrass and mountain rye. Other grasses used in this study, with the possible exception of Russian wildrye, are capable of first-year flowering from spring plantings without cold treatment, but it appeared that a higher percentage of plants flowered when seeds had been treated prior to spring planting; to what extent a few days' earlier emergence from treated seeds was a contributing factor is not known. The other species investigated were four wheatgrasses—pubescent, tall, crested, and fairway—and Indian ricegrass and Great Basin wildrye.

Whereas both snowbank and refrigerator storage effectively vernalized seeds, snowbank storage was advantageous in that it required no special equipment and seeds did not dry during the process. Use of plastic bags instead of cloth bags in the refrigerator lessened the problem of seeds' drying.

These cold treatments hastened emergence from spring planting and aided survival on dry sites. With such treatment spring plantings compared favorably with fall plantings.

This study suggests that use of vernalized seed in spring planting of some range grasses may help obtain successful stands when fall planting has not been feasible. Also, this procedure might produce a crop of seed from spring plantings a year earlier than otherwise or increase seed yield the following year, at least in certain species.

Although practical possibilities of vernalizing perennial grasses or other species have not been fully tested, it is apparent that plant stimulation, either by this process or other means, offers a fruitful approach for research in wild-land revegetation.

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