Germination and root growth of 4 osmoconditioned cool-season grasses

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Abstract

Establishment of grass species used in range reseeding should improve if germination time can be decreased. Osmotically controlling the hydration of seed so that germination processes proceed other than radicle emergence (osmoconditioning) can decrease germination time of many plant species. Growth chamber experiments were conducted to evaluate effects of osmoconditioning at -1.5, -2.0, and -2.5 MPa for 4, 8, 12, 16, and 20 days on germination and root growth of 'Flintlock' western wheatgrass (*Pascopyrum smithii* (Rydb.) A. Löve) and 'Vinall' Russian wildrye (*Psathyrostachys juncea* (Fischer) Nevski) and at -2.0, -2.5, and -3.0 MPa for 4, 8, 12, 16, and 20 days on 'Nordan' Crested wheatgrass (*Agropyron desertorum* (L.) Gaertn.) and 'Tegmar' intermediate wheatgrass (*Agropyron intermedium* (Host) Beauv.). A second study looked at germination time of seed from the same species conditioned at osmotic potentials and durations producing the shortest time to 50% germination (optimum conditioning) and air dried for 0, 1, or 7 days. Conditioned seed of Russian wildrye and western wheatgrass germinated 2 to 4 days faster, respectively than untreated seed. Optimum conditioning of seed from all 4 species produced seedlings with roots 20 to 67% shorter 4 days after germination than seedlings from untreated seed. Conditioned western wheatgrass seed continued to germinate faster than untreated seed after being air dried for 7 days. Slow root growth from conditioned seed may negate any benefits derived from rapid germination.

Key Words: Russian wildrye, western wheatgrass, intermediate wheatgrass, crested wheatgrass

Many grass species have small seed that require a shallow planting depth (0.5-1.75 cm) for successful establishment (Cook et al. 1974). Successful establishment at shallow depths may be a problem because the top 2 cm of soil can dry from field capacity to wilting point within a single day (Army and Hudspeth 1960, Wiegand 1962, Reginato 1975) which is insufficient time for seed germination of many species. If seed germination and subsequent root extension occurs quickly, the probability of seedling survival is improved by the increased availability of soil moisture at greater depths (McGinnies 1973).

Mueller and Bowman (1989) reported that pregerminated seed

of cool-season grasses established deep roots more rapidly in a moist soil than untreated seed. Pregermination may reduce the number of days the soil surface would need to remain wet for successful seedling establishment.

Osmoconditioning is a method of controlled hydration that allows some germination processes to proceed but prevents radicle emergence (Bewley and Black 1985). Osmoconditioned seed have been shown to germinate and emerge faster than untreated seed (Brocklehurst and Dearman 1983; Hardegree and Emmerich 1992; Hardegree 1994). This process may improve establishment of small seeded range grass.

The purpose of this study was to evaluate the effects of osmoconditioning on germination and initial root growth of 4 cool-season grasses and to determine the effect of drying osmoconditioned seed on germination rate.

Materials and Methods

Experiment 1

Experiment 1 determined the effect of osmoconditioning on time to 50% germination, total germination percentage, and root growth of 4 cool-season grasses: 'Nordan' crested wheatgrass (*Agropyron desertorum* (L.) Gaertn.), 'Tegmar' intermediate wheatgrass (*Agropyron intermedium* (Host) Beauv.), 'Flintlock' Western wheatgrass (*Pascopyrum smithii* (Rydb.) A. Löve), and 'Vinall' Russian wildrye (*Psathyrostachys juncea* (Fischer) Nevski). The design of experiment 1 was a 3 × 5 factorial + 1 with 4 replications.

Russian wildrye and western wheatgrass seed were osmoconditioned with polyethylene glycol 8000 (PEG) at 3 osmotic potentials (-1.5, -2.0, and -2.5 MPa) for 5 durations (4, 8, 12, 16, or 20 days) plus a control. Polyethylene glycol is a nontoxic, high-molecular-weight compound that does not penetrate plant cell walls (Carpita et al. 1979, Tarkow et al. 1966). *Captan*¹ fungicide powder was added to the PEG solution (2.5g/liter) to help control fungus.

Preliminary trials showed that osmotic potentials of -1.5 MPa did not prevent germination of crested and intermediate wheatgrass seed, therefore seed of these species were conditioned at potentials of -2.0, -2.5, and -3.0 MPa. One-hundred seed were placed on blotter paper and *Kimpac*¹ germination paper soaked

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¹Mention of any trade name is for information only and does not imply endorsement.

with 100 ml of PEG solution inside a germination dish (32.5 × 33.8 × 7.5-cm). Germination dishes were then sealed with plastic tape and placed in growth chambers.

Growth chamber temperature and light settings during conditioning were: 16 hours dark at 20° C and 8 hours light at 30° C for crested wheatgrass, intermediate wheatgrass and Russian wildrye (Association of Official Seed Analysts 1981). Western wheatgrass was conditioned in the dark at temperatures of 15° C for 16 hours and 30° C for 8 hours each day. Osmoconditioning solutions were mixed to give stated osmotic potentials at 25° C (Michel and Kaufmann 1973). Growth chamber temperature changes caused a deviation in solution osmotic potentials of approximately ± 0.1 MPa. Osmotic potentials may have been slightly more negative than estimated because the equation used (Michel and Kaufmann 1973) to formulate the solution did not account for molecular exclusion of PEG from the germination paper substrate (Hardegree and Emmerich 1990).

Germination dishes were removed from the growth chamber after 4, 8, 12, 16, or 20 days. Seeds were then removed from the dishes, rinsed in distilled water, and placed in similar germination dishes on blotter and germination paper soaked with 100 ml of distilled water. Conditioned and untreated (control) seed were placed back in the growth chamber at a 36° angle (Jones and Cobb 1963) and germinated under the same temperature and light conditions used during osmoconditioning.

Germinated seed were counted daily the first week and alternate days thereafter for the study duration (21 days). Untreated seed radicle lengths were measured daily for the first 5 days after radicle emergence at which time the seedlings had to be taken out of the germination dish because of space limitations. Radicle lengths were measured daily for 5 days after conditioned seed were placed in the germination chamber. Thus, 4 day root growth analyses was limited to the optimum conditioning treatments in which seeds germinated within 1 day of being switched to distilled water. Seed were considered germinated if the radicle was showing. Since all germination did not occur the same day, root growth rate was calculated from the time of radicle emergence. Time to 50% germination of pure live seed (PLS) ($T_{50\%}$) was determined by extrapolating between the germination count (G_b) the day before (D_b) 50% germination occurred and the germination count (G_a) the day after (D_a) 50% germination occurred ($T_{50\%} = ((G_{50\%} - G_b) \times (D_a - D_b) / (G_a - G_b)) + D_b$). Fifty percent germination of pure live seed (PLS) equaled 0.5 × percent germination of untreated seed.

Experiment 2

Experiment 2 was conducted to determine effects of 0, 1, and 7 days of drying on germination of seeds that were osmotically conditioned using optimal conditioning treatments of those species evaluated in experiment 1 (-2.0 MPa and 8 days for crested and intermediate wheatgrass, -1.5 MPa and 20 days for western wheatgrass and Russian wildrye). The design was a single factor with 4 treatments (control and 3 drying periods) and 4 replications. Since there were no interactions main effect means were used to select optimal conditioning treatments. Conditioned seed were rinsed in distilled water, placed on blotter paper, and allowed to air dry at room temperature for either 0, 1, or 7 days before being put into the growth chamber. A set of untreated seed (control) was also included for germination testing. The same methodology used in the previous experiment to condition, germinate, and count the seed was used to evaluate treatments.

Data Analysis

Experiment 1 time to 50% germination and total germination percentage were analyzed using a 3 × 5 factorial analysis of variance excluding the control. Control means were compared with other means in the first experiment using error from analysis of variance on osmotic potential and treatment duration. Root growth in experiment 1 was analyzed using a single factor analyses of variance with 2 treatments and 4 replications. Experiment 2 was analyzed using a single factor analysis of variance with 4 treatments and 4 replications. Control means in the second experiment were part of the analysis of variance. Duncan's new multiple range test was used for mean separation. Significance was evaluated at $P \leq 0.05$.

RESULTS

Experiment 1

Western wheatgrass time to 50% germination decreased as duration of osmoconditioning treatment increased. All treatment durations resulted in a lower time to 50% germination than the control (Table 1). Time to 50% germination for Russian wildrye

Table 1. Effect of osmoconditioning duration on time (days) to 50% germination of 4 cool season grasses.

Species	Control	Days in Solution				
		4	8	12	16	20
		(days)				
Crested wheatgrass	2.7a ¹	2.5a ²	2.3a	3.1a	2.6a	2.7a
Intermediate wheatgrass	2.0a	2.2a	1.9a	2.1a	2.5a	3.3b
Western wheatgrass	5.9d	4.4c	2.4b	2.3b	1.7a	1.6a
Russian wildrye	3.8b	4.3b	2.2a	2.3a	2.1a	1.7a

¹Control means compared with other means using error from osmoconditioning study.

²Means with same letter within a row are not significantly different ($P \leq 0.05$). Results are averaged across osmotic potentials.

was reduced after the seed had been in solution from 8 to 20 days. Time to 50% germination of crested wheatgrass was not affected by duration of osmoconditioning treatment. There was an increase in time to 50% germination of intermediate wheatgrass after seed had been in solution for 20 days.

Western wheatgrass time to 50% germination was lowered at the least negative osmotic potential treatments (Table 2). Time to 50% germination for Russian wildrye was less at -1.5 MPa than at lower osmotic potentials. Western wheatgrass and Russian wildrye seed conditioned at -1.5 MPa reached 50% germination

Table 2. Effect of osmoconditioning water potential (MPa) on time (days) to 50% germination of 4 cool season grasses.

Species	Control	Water Potential (MPa)			
		-1.5	-2.0	-2.5	3.0
		(days)			
Crested wheatgrass	2.7a ¹		2.3a	2.8a	2.8a
Intermediate wheatgrass	2.0ab		1.7a	2.7b	2.8b
Western wheatgrass	5.8d	1.6a	2.4b	3.4c	
Russian wildrye	3.7b	1.6a	3.2b	2.7b	

¹Control means compared with other means using error from osmoconditioning study.

²Means with same letter within row are not significantly different ($P \leq 0.05$). Results are averaged across treatment times.

in less than 2 days compared to approximately 6 and 4 days, respectively, for untreated seed. Osmotic potential had no effect on time to 50% germination of crested wheatgrass. Intermediate wheatgrass time to 50% germination was less for seed conditioned at -2.0 MPa than for seed conditioned at lower osmotic potentials.

Compared to the control, total germination percentage was reduced for crested wheatgrass after 8 days in solution, for intermediate wheatgrass after 12 days in solution, and for Russian wildrye after 16 days in solution (Table 3). Total germination of western wheatgrass was not affected by treatment duration.

Table 3. Effect of osmoconditioning duration on total germination (%) of 4 cool season grasses.

Species	Control	Days in solution				
		4	8	12	16	20
		----- (%) -----				
Crested wheatgrass	87b ¹	76ab ²	72a	67a	67a	65a
Intermediate wheatgrass	71b	66ab	67ab	60a	59a	60a
Western wheatgrass	85a	85a	86a	84a	86a	81a
Russian wildrye	76b	69ab	67ab	67ab	65a	66a

¹Control means compared with other means using error from osmoconditioning study.

²Means with same letter within a row are not significantly different ($P < 0.05$). Results are averaged across osmotic potentials.

An osmotic potential of -3.0 MPa lowered total germination of intermediate wheatgrass (Table 4). Total germination of crested wheatgrass and Russian wildrye was lower than the control at all osmotic potentials. Osmotic potential had no effect on total germination of western wheatgrass.

Root lengths of seed that were conditioned at optimum osmotic potential and treatment duration were less than the control 4 days after germination for western wheatgrass and 2, 3, and 4 days after germination for Russian wildrye (Fig. 1). Optimum conditioning of crested and intermediate wheatgrass seed produced root lengths that were lower than the control 3 and 4 days after germination.

Table 4. Effect of osmoconditioning water potential (MPa) on total germination of 4 cool season grasses.

Species	Control	Water Potential (MPa)			
		-1.5	-2.0	-2.5	-3.0
Crested wheatgrass	87b ¹		70a	73a	66a
Intermediate wheatgrass	71b		60ab	72ab	56a
Western wheatgrass	85a	86a	84a	84a	
Russian wildrye	76b	67a	64a	69a	

¹Control means compared with other means using error from osmoconditioning study.

²Means with same letter within a row are not significantly different ($P \leq 0.05$). Results are averaged across treatment times.

Experiment 2

Intermediate wheatgrass and Russian wildrye seed that were not dried or had been air dried at room temperature for 1 day after optimum conditioning germinated more rapidly than untreated seed (Table 5). Conditioned western wheatgrass seed germinated more rapidly than untreated seed after 0, 1, or 7 days of air dry-

Fig. 1. Mean root length (mm) of western wheatgrass and Russian wildrye grown from seed osmoconditioned at -1.5 MPa for 20 days and crested and intermediate wheatgrass osmoconditioned at -2.0 MPa for 8 days.

Table 5. Effect of drying on time (days) to 50% germination of 4 cool season grasses grown from osmoconditioned seed.

Species	Treatment	Control	Days dried		
			0	1	7
			(days)		
Crested wheatgrass	-2.0 MPa, 8 days	3.5a	2.6a ¹	2.1a	2.3a
Intermediate wheatgrass	-2.0 MPa, 8 days	2.8b	1.5a	1.0a	2.6b
Western wheatgrass	-1.5 MPa, 20 days	7.3d	0.9a	1.6b	2.5c
Russian wildrye	-1.5 MPa, 20 days	4.1c	0.9a	2.8b	3.5bc

¹Means with same letter within a row are not significantly different ($P \leq 0.05$)

ing. Crested wheatgrass seed did not exhibit faster germination after optimum conditioning than untreated seed at 0, 1, or 7 days of drying.

Discussion

Results from experiment 1 generally agree with previous studies showing enhanced germination response from osmoconditioned seed (Heydecker and Coolbear 1977), and detrimental conditioning effects at low water potentials and long duration treatments (Hardegree and Emmerich 1992, Gray et al. 1990, Ely and Heydecker 1981). Exceptions were the beneficial effects of longer treatment times on time to 50% germination of Russian wildrye and western wheatgrass. If treatment times had been longer than 20 days results may have been different. Loss of seedling vigor was beginning to show in both species as indicated by slower root growth from osmoconditioned seed than from the untreated seed 2 to 4 days after germination.

Conditioning seed to minimize time to 50% germination slowed root growth in all 4 species. Slower root growth brought about by conditioning could negate any benefits in establishment obtained by more rapid germination, especially for species that germinate quickly. Russian wildrye root growth from untreated seed would be expected to exceed root growth from conditioned seed within 4 days after the conditioned seed had germinated, even though the untreated seed germinated 2 days later than the conditioned seed. Research on perennial ryegrass (*Lolium perenne* L.) and bell pepper (*Capsicum annum* L.) indicates that the initial advantage in rapid root growth from conditioned seed may not persist. Perennial ryegrass roots from osmoconditioned seed ceased to be longer than those from untreated seed 118 hours after being placed on germination blotters soaked with distilled water (Danneberger et al. 1992). Bell pepper seedlings from untreated seeds had higher root weights 14 days after seeding than seedlings from osmoconditioned seed (Stoffella et al. 1992).

Conditioned seed that are dried but still retain rapid germination are more practical and convenient for sowing and storage. Western wheatgrass was the only species to retain rapid germination beyond 1 day of air drying. Drying the seed slowly by controlling humidity during the process may prolong germination rates achieved through conditioning. Rapid drying of imbibed seed has been shown to decrease the rate of germination in perennial ryegrass (Debaene-Gill et al. 1994). However, drying the seed slowly would entail additional equipment to control humidity.

Even with osmoconditioning, slow root growth may negate any benefits derived from rapid germination. Field studies need to be conducted to evaluate the combined effects of slower root growth and rapid germination before conditioned seed can be recommended for revegetation of grasses under dryland conditions.

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