Effect of Potassium Nitrate Priming on Seed Germination of Seashore Paspalum

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Abstract. Germination of seashore paspalum (Paspalum vaginatum Swartz) is a critical factor influencing seedling establishment when seeded directly in fields. In this research, pregermination priming with osmotic solution was used to improve the germination percentage of seashore paspalum. The goal of this study was to develop techniques that improve the germination of a seashore paspalum cultivar (SeaSpray). Seeds were subjected to priming in petri dishes with solutions of KNO₃ at 25 °C for 24, 48, and 72 h in growth chambers. Germination percentage differed by priming duration as well as concentration of priming media. Based on the germination percentage 14 days after imbibition, the most promising priming condition was the treatment with 0.2% or 0.5% KNO3 for 72 h at a constant temperature of 30 °C and 0.2% or 0.5% KNO3 for 48 to 72 h at alternating temperature of 25/35 °C. Both concentrations showed reasonable germination percentage greater than 85% at alternating temperature condition. Priming with KNO₃ solution for 48 to 72 h improved not only germination percentage, but also uniformity. The increased duration of priming with KNO3 was positively correlated with an improved germination percentage. The effect of increasing concentration was the most apparent at a constant temperature (30 °C) regime with the treatment of 0.2% KNO₃ priming. Germination percentage was increased from 34.3% to 68.0% 2 weeks after imbibition (WAI) as the priming duration was increased from 24 to 72 h. Priming with KNO3 for 3 days also had a modest effect on germination percentage (greater than 74.7%) at 1 WAI. Therefore, priming with 0.2% or 0.5% solution of KNO3 for 72 h is a recommended method that can be practically applied for increasing germination of paspalum under an alternating temperature (25/35 °C) condition. The regression analysis between odds of germination percentage and germination time showed that priming treatment increased internal activities during the second stage of seed germination.

Seashore paspalum (*Paspalum vaginatum* O. Swartz, 2n = 2x = 20) is a promising warm-season turfgrass species resulting from its high tolerance to salt stress (Carrow and

Duncan, 1998; Duncan, 1999; Lee et al., 2005), drought (Huang et al., 1997), and flooding (Malcolm, 1969) and adaptation to a broad soil pH range (Duncan, 1994). In addition, the species adapts well to low irradiance and weak shade conditions (Jiang et al., 2004). Although the species has not been as popular as other turfgrasses, the potential for vigorous proliferation by its fast-growing characteristics has been noted by many independent researchers and seed companies (Duncan, 1994, 1999; Jiang et al., 2004). Recently, the demands for a turfgrass

tolerance to adverse conditions have increased and the commercial development of seashore paspalum cultivars has accelerated.

Seed germination and seedling establishment are important stages in the field performance of crop plants. In the field, seed germination is controlled by many environmental factors and innate conditions, which subsequently affect seedling establishment. Seeds are often placed to specific conditions unfavorable for germination. Therefore, seed priming is commonly used to improve seed germination under unfavorable conditions. Although seed priming is a practical method to improve rates and uniformity of germination (Parera and Cantliffe, 1994), priming methods differ depending on crop species and seed and germination conditions (Bradford, 1986; Bush et al., 2000; Heydecker et al., 1973; Khan, 1992; McDonald, 2000). The effect of priming has been attributed to metabolic repair and activation of seed during water imbibition (Basra et al., 1988, 2005; Bray et al., 1989).

Many treatment techniques have been developed to improve the germination of perennial turfgrass seed with each species having a different pattern of germination (Bush et al., 2000; Frett and Pill, 1995; Hacisalihoglu, 2007). Thus, there is no universal technique for improving seed germination. Of the known germination methods, chemical pretreatment is the most promising because of ease of application, scale of economies, and labor-saving attributes compared with methods in which the environment must be controlled for prolonged periods of time.

A seeded seashore paspalum cultivar has been used for establishment of golf courses and sports fields. Therefore, the knowledge of the appropriate chemical pretreatment that can be applied easily for bulk treatment is necessary before releasing any new breed of seashore paspalum cultivars. The objective of this study was to determine effects of seed priming with KNO₃ solution on the subsequent germination of seashore paspalum.

Materials and Methods

Seeds of seashore paspalum (cv. Sea-Spray) produced in a pilot production field were provided by Turf-Seed, Inc. of Hubbard, OR. The seeds were air-dried and stored at 4 °C until use. Seeds were classified into viable or nonviable based on the results of a tetrazolium test (Grabe, 1970).

Seed priming was conducted in H_2O and 0.2% and 0.5% (w/v) KNO₃ for 1, 2, and 3 d at 25 °C. Preweighed seeds (5 g) were placed on two blotter papers in 9-cm diameter petri dishes saturated with appropriate osmotic solutions and the dishes were covered with aluminum foil. After the required priming period, the seeds were quickly rinsed using tap water and air-dried at room temperature until seed moisture content became similar to that of untreated seeds. The seeds were transferred to paper bags and stored at 10 °C until testing.

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All germination tests were conducted in transparent $11 \times 11 \times 3.5$ -cm polycarbonate boxes with friction fitting lids (Hoffman Manufacturing, Inc., Albany, OR) and with two layers of steel blue blotter paper (Anchor Paper, St. Paul, MN). Each experimental unit consisted of 50 seeds placed onto germination paper soaked in 17 mL of H₂O and positioned in a Conviron Model E15 growth chamber (Controlled Environments Limited, Winnipeg, Manitoba, Canada) under a 12-h light period.

The germination boxes were rearranged daily to minimize effects of potential temperature differences within the chambers. When the emerging radicle could be observed by the naked eye, the seed was considered germinated. Germination was checked daily and calculated as a percentage (germination percentage) and as a weighted germination percentage. The weighted germination percentage (WGP) was calculated as follows:

WGP =
$$\sum [(T - t_i + 1)n_i] \times 100/(T \times S)$$

where t_i = the ith day, n_i = number of germinated seeds at t_i , T = total germination period in day, and S = total number of seed per germination box.

Mean germination time (MGT) was calculated to enable knowledge of the velocity of germination and calculated as follows:

$$MGT = \sum (t_i n_i) / N$$

where t_i = the ith day, n_i = number of seed germinated at t_i , and N = number of germinated seed ($N = \sum n_i$).

The germination percentage was transformed as odds. Regression analysis between odds and germination time was performed to know regression coefficient during the second stage of germination. The period showing a linearity of relation between odds and time was considered as the second stage of germination. Odds were calculated as follows:

$$Odds = (germination percentage)/$$

(100 - germination percentage)

The effects of priming on germination were compared by means of germination percentage, weighted germination percentage, and mean germination time. Germination experiment was a completely randomized block design with three replications. All data were statistically analyzed using a SAS program (Version 8.0; SAS Institute, Cary, NC). Analysis of variance was used to analyze the data with three replications. When the analysis of variance indicated significance (P < 0.05), means were compared using Duncan's multiple range test. Regression analysis was performed using the PROC REG procedure.

Results and Discussion

Viability of seashore paspalum seeds used in the experiment was 91% that was enough to conduct the germination test for evaluating the effect of priming treatments. Germination percentage and germination speed were improved by priming with H_2O and KNO_3 (Table 1). Germination percentage of primed seed was higher at alternating temperature 25/35 °C than 30 °C regardless of the priming treatment. A previous study (Shin et al., 2006) reported that the alternating temperature was more effective for germination of seashore paspalum and that light was required to obtain an appropriate germination percentage.

Potassium nitrate solution has long been known as a suitable chemical approach for promoting germination in various plant species and generally as a priming agent or germination media (Argerich and Bradford, 1989; Bush et al., 2000; Madakadze

Table 1. Effect of priming media with different imbibition times on the germination percentage of seashore paspalum at an alternating (25/35 °C) and constant (30 °C) temperature.

	Alternating	(25/35 °C) Constan		nt (30 °C)
	1 WAI ^z	2 WAI	1 WAI	2 WAI
Priming media		%-		
H ₂ O 24 h	45.0 g ^y	65.0 f	22.7 d	25.3 e
H ₂ O 48 h	56.7 e	78.0 c	28.7 cd	30.0 de
H ₂ O 72 h	55.7 e	76.0 cd	26.7 cd	30.0 de
KNO3 0.2% 24 h	49.3 f	71.3 de	29.7 cd	34.3 d
KNO3 0.2% 48 h	69.7 c	87.7 ab	58.0 a	61.7 b
KNO3 0.2% 72 h	74.7 a	88.0 a	63.0 a	68.0 a
KNO3 0.5% 24 h	60.7 d	81.7 bc	43.7 b	52.7 c
KNO ₃ 0.5% 48 h	72.3 bc	90.0 a	60.3 a	66.7 ab
KNO ₃ 0.5% 72 h	79.0 a	89.7 a	62.3 a	68.0 a
Control ^x H ₂ O	3.7 i	42.7 g	4.3 f	12.7 f
Control ^x KNO ₃	10.3 h	69.7 ef	10.7 e	55.0 c

^zWeek after imbibition.

^yMeans within columns with the same letters are not significantly different at 5% according to Duncan's multiple range test.

*Seeds were germinated without priming.

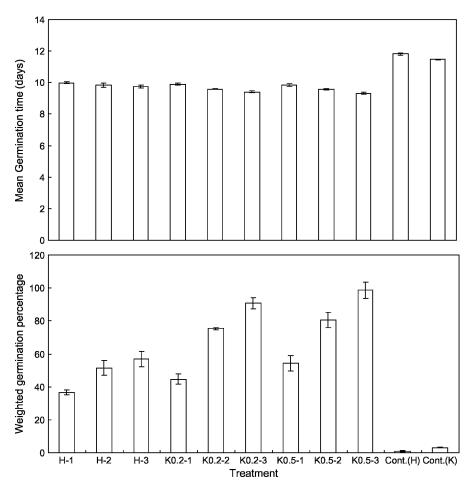


Fig. 1. Effects of priming on days to germination and weighted germination percentages of seashore paspalum at germination temperature of 25/35 °C. Seeds were primed with H₂O (H), 0.2% KNO₃ (K0.2), or 0.5% KNO₃ (K0.5) for 1, 2, or 3 d. Nonprimed seeds (control) were imbibed and germinated in H₂O or 0.2% KNO₃.

et al., 1993; McDonald, 2000). The duration of priming with KNO_3 influenced germination percentage differentially, i.e., prolonged priming had a more positive effect on germination.

Treatment with KNO₃ solution for 3 d showed practically acceptable germination percentage (over 74.7%) at 1 week after imbibition (WAI). Therefore, priming with KNO₃ solution of a minimum concentration of 0.2% or 0.5% solution was deemed appropriate for increasing germination percentages under alternating temperature (25/35 °C). In practice, priming with KNO₃ solution would be a simple procedure that can be performed in bulk scale.

Even seeds without a priming treatment showed more rapid germination (69.7% at 2 WAI) in 0.2% KNO3 germination media as compared with the H₂O control (42.7% at 2 WAI). As noted previously (Shin et al., 2006), an alternating temperature regimen was effective in promoting germination of seashore paspalum. The effectiveness was especially observed in the priming experiments using KNO₃ solutions. Both the 0.2% and 0.5% KNO3 solutions increased germination; however, the final germination percentages at 2 WAI implied that pretreated seeds germinated at a constant 30 °C did not attain a level adequate for practical use. The lower germination percentage of treated seeds might be the result of the constant temperature being less favorable to germination.

Mean germination times and weighted germination percentages reflect germination speed that cannot be measured by germination percentage. There were minor differences in MGT among priming treatments, although all priming treatments examined in this experiment reduced MGT compared with the control (Fig. 1). Therefore, it was concluded that the priming of seashore paspalum, in general, resulted in a shortened MGT. The MGT is also dependent on the duration of imbibition and/or internal metabolic activities after imbibition (the second stage of germination). Priming activates internal metabolism required for furthering the germination process (Basra et al., 2005). In this experiment, the swelling pattern of seeds was similar among treatments (data not shown); therefore, the differences of MGT were likely the result of the different level of metabolic activity between treated and control seeds in the second stage of germination.

The WGP values indicate clear differences between treatments and treatment durations. The tendency of priming effect on WGP was similar in KNO₃ treatments regardless of treatment durations. Germination data at 2 WAI showed a little difference between a KNO₃ priming duration of 2 and 3 d (Table 1). However, WGP revealed significant differences between priming durations. This result is considered to be the result of the higher germination percentage until 1 WAI. Hence, we assume that KNO₃ priming longer than 2 d increases germination velocity without an increase in final germination percentage.

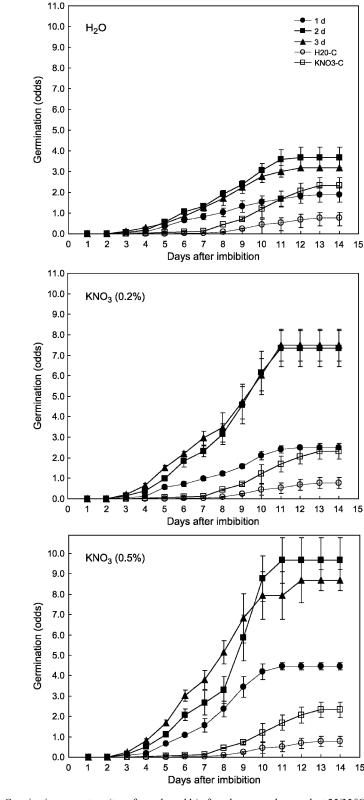


Fig. 2. Germination percentage (transformed as odds) of seashore paspalum seeds at 25/35 °C after priming with H_2O and KNO_3 for 1, 2, and 3 d. Nonprimed controls were imbibed and germinated in H_2O or 0.2% KNO_3 . Odds were calculated using the equation: Odds = (germination percentage)/(100 – germination percentage).

Priming with KNO₃ solutions showed a germination-promoting effect. Considering the higher tolerance of seashore paspalum to salt conditions (Carrow and Duncan, 1998), priming with KNO₃ might show no harmful effects on the germination of sea-

shore paspalum. The difference between the KNO_3 concentrations used in this study, however, was not significantly different as priming time was increased (greater than 48 h). Nevertheless, the duration of priming indicated differences, especially between

Table 2. Regression analysis for the comparison between odds of germination percentage and germination time during the second germination stage (5 DAI to 10 DAI).

Treatment	Duration (days)	Regression equation	R^2
H ₂ O	1	y = 0.235x - 0.826	0.997** ^z
H ₂ O	2	y = 0.462x - 1.753	0.990**
H ₂ O	3	y = 0.416x - 1.547	0.983**
0.2% KNO3	1	y = 0.302x - 1.074	0.978**
0.2% KNO3	2	y = 0.838x - 3.229	0.970**
0.2% KNO ₃	3	y = 0.789x - 2.536	0.990**
0.5% KNO3	1	y = 0.669x - 2.775	0.969**
0.5% KNO3	2	y = 1.201x - 5.064	0.891**
0.5% KNO3	3	y = 1.187x - 4.182	0.993**
Controly (H ₂ O)	_	y = 0.039x - 0.200	0.836**
Control (KNO ₃)		v = 0.115x - 0.578	0.870**

^zSignificant at 1% level.

^ySeeds were germinated on H_2O or 0.2% KNO₃ as germination media without pretreatment. DAI = days after imbibition.

1 d and 2 to 3 d. To better understand the differences, the germination percentage data were transformed into odds as described in "Materials and Methods". The transformed data resulted in two interesting findings: 1) there was no difference between 2 and 3 d of priming; and 2) there was linearity from 5 d to 10 d after imbibition (Fig. 2). Based on the regression between odds of germination percentage and germination time, we calculated the slope using the regression equation (Table 2). We assumed the duration from 5 to 10 d after imbibition involved the second stage of germination during which internal reserve materials are degraded to supply raw materials and energy required for metabolism. Because the priming treatment acts during this second (lag) stage of germination by changing metabolic activities (Basra et al., 1988, 2005), the change in slope might be a response to the internal changes accelerated by exposure to KNO₃. The slopes indicate that a 0.5% KNO3 priming solution enhanced germination the most. The accelerated germination rate then might be the result of the increased internal activity during the second germination stage for any subsequent germination process. Because the slope was greater in seeds treated for 2 d compared with those treated for 3 d, an optimal priming duration of 2 d was accepted for both 0.2% and 0.5% KNO₃ solutions. These results reflect the notion that a prolonged priming duration (greater than 2 d) could reduce the germination-promoting effect of KNO₃. Indeed, this tendency was similar for all priming solutions (H₂O, 0.2% KNO₃, and 0.5% KNO₃).

In conclusion, seed germination of seashore paspalum can be improved by priming in 0.5% KNO₃ solution for 2 d. Although MGT was not influenced differently by priming duration and priming solutions, nonetheless, the priming shortened MGT. The germination of primed seeds was promoted at an alternating temperature of 25/35 °C compared with a constant temperature. In practice, it is recommended that seashore paspalum seeds can be treated in 0.2% to 0.5% KNO3 solution for 2 d and germinated at the alternating temperature of 25/35 °C to obtain an appropriate seed germination speed and rate for favorable seedling establishment in the field.

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