

## **Study of respond seed germination of barley (*Hordeum vulgare* L.) to different primings**

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**ABSTRACT:** In order to evaluate the effect of different seed priming techniques on germination and morphological characters of barley an experiment was conducted in 2011\_ in a factorial experiment based on the complete randomized block design with 2 factors in Iran. Seeds were primed for 20, 40 and 60 hours in seven priming media (PEG 5%, PEG 10%, KNO<sub>3</sub> 1%, KNO<sub>3</sub> 2%, KCl 2%, KCl 4% and distilled water as control). Maximum seed germination percentage was observed when seed primed by KNO<sub>3</sub> 2%. The most seedling length and radical length were obtained for seeds with KCl 2% for 60 h and KCl 4% respectively. Rate of germination was improved when the seed soaked KNO<sub>3</sub> 2% compared with PEG, KCL and water. Increasing of seed soaking duration improved some parameters such as seedlings length, radicle length, stem dry weight and rate of germination. There was interaction between seed priming media × priming duration showed the beneficial effects on number of germination and seedling length.

**Keywords:** Germination, PEG, priming and seed

### **INTRODUCTION**

Barley is mainly grown for grain, green fodder and straw for small ruminants during winter (Khan et al., 1999). It is grazed continuously by the livestock in the driest seasons with no grain harvest. It is mostly grown on irrigated land while in rainfed areas it occupies one third of land. Among all crops the salt tolerance of barley is the highest (Mass & Hoffman, 1977) and differences in salinity tolerance in barley and wheat species (Mano & Takeda, 1998; Forster et al., 2000; Shafi et al., 2010) have been reported. Improved plant growth in saline areas lead to generate salt tolerance in crops. Salt tolerance not only varies among species but also amongst different genotypes within species (Maas & Hoffman, 1977; Storey & Wyn Jones, 1978; Marschner et al., 1981; Shafi et al., 2011). One of the most important Abiotic factors limiting plant germination and early seedling stages is water stress

brought about by drought and salinity (Almansouri, et al. 2001), which are widespread problems around the world (Soltani, et al. 2006). Salinity and drought affect the plants in a similar way. Reduced water potential is a common consequence of both salinity and drought. Water stress acts by decreasing the percentage and rate of germination and seedling growth. Germination of seeds, one of the most critical phases of plant life, is greatly influenced by salinity. Polyethylene glycol (PEG) compounds have been used to simulate osmotic stress effects in Petri dish (In vitro) for plants to maintain uniform water potential throughout the experimental period (Kulkarni, et al. 2007). The adverse effect of water shortage on germination and seedling growth has been well reported in different crops such as corn (Mohammadkhani and Heidari, 2008). Solutions of high molecular weight, polyethylene glycol, are often used to control water potential in seed germination studies

(Hardegree and Emmerich, 1990). The polyethylene glycol (PEG)-induced inhibition of germination has been attributed to osmotic stress (Dodd and Donovan, 1999; Sidari et al., 2008).

The main objective of this study was to evaluate the effects of different priming media and duration treatments on seed germination behavior of barley.

## MATERIAL AND METHODS

This experiment was conducted in Islamic Azad University, Boroujerd branch, Iran in March 2011 to determine seed priming effects on germination, Rate of germination and morphological characters of barley. The study involved laboratory. Seeds was fully immersed in priming media at a temperature of 25°C for durations of 20, 40 and 60 h. All seed was then rinsed thoroughly with distilled water and lightly hand dried using blotting paper. Laboratory research measured the rate of germination using a two factor factorial complete randomized block design with 21 treatment combinations replicated three times. The two treatment factors were three duration time (20, 40 and 60 h) and seven priming media. The seven priming media were: water; 5% PEG; 10% PEG; 2% KNO<sub>3</sub>; 1% KNO<sub>3</sub>; 2% KCl; 4% KCl. The PEG6000 concentration is chosen in such a way that it dose not allow seed to absorb enough water to germinate. All priming media were prepared in distilled water. 100 seeds from each of the treatments were placed on 90 mm-diameter Whatman No.2 filter paper that was moistured with 10 ml distilled water in each Petri-dish. Seeds were kept at 25°C in air temperature under normal light. 7 days after treatment, length of radicle and stem, dry weight (stem, radical), length of seedling and number of germination were measured and percentage of germination and rate of germination were calculated. Germination Percent (GP) was calculated based on following equation.

Germination Percentage = (total seed germination after 14 days / total seeds) × 100

Experimental data were analyzed by using SAS (Statistical software, SAS institute, 2002) and treatment means were compared using Duncan's multiple range tests at 5% level of probability.

## RESULTS AND DISCUSSION

Seed priming(A) had significantly positive effect on characters such as seedling length, Length of R/S, radicle dry weight, germination percentage and rate of germination. Radicle dry weight was significantly affected by seed priming treatment in  $p < 0.05$ . Priming duration(B) had significantly effect on characters such as seedling length, number of germination, germination percentage and rate of germination. Priming media ×

during time(A\*B) interaction had significant effect on seedling length and number of germination (Table 1).

The maximum and minimum seedlings length were obtained from seeds primed in KCl 4% (18.1mm) and control (9.2mm). The maximum and minimum radicle length were obtained from seeds primed in KCl 4% (18.2mm) and PEG 10% (10mm). However, the maximum and minimum length of R/S was attained from application PEG 10% (1.45mm) and KNO<sub>3</sub> 1% (0.89mm). Also the maximum and minimum stem dry weight was attained from application KNO<sub>3</sub> 2% (1.33g) and PEG 10% (0.9g). The maximum radicle weight was attained from PEG 5% (0.31cm). Maximum number of germination, germination percentage and rate of germination were KNO<sub>3</sub> 2% (48.11), (94%) and (38.9%) respectively (Table 2).

With increasing of priming time from 20h until 60h seedlings length, radicle length, stem dry weight and rate of germination were increased but the number of germination and germination percentage were decreased (Table 3).

Maximum seedling length and number of germination were obtained from seed prim × duration time in KCl 2% for 60h and KNO<sub>3</sub> 1% for 60h respectively (Table 4). Priming the seeds for 60h had better effect on stem and radicle length and stem and radicle dry weight compared with other during temperature. In present study PEG caused the maximum radicle dry weight and GP and number of germination and rate of germination. In conclusion, wheat seed priming with PEG 10% increased stem and radicle length, stem and radical dry weight, germination percentage and rate of germination.

Pre-sowing seed treatments have shown to enhance stand establishment in non-saline areas (Khan, 1992; Bakht et al., 2010; Bakht et al., 2011) and have potential in saline areas as well (Ashraf & Ruaf, 2001). Priming is a pre-sowing soaking of seeds in different solutions, which enhances germination and seedling emergence uniformity under adverse environmental conditions (Bakht et al., 2010). Salt priming could be used as an adaptation method to improve the salt tolerance of seeds (Bakht et al., 2011). Our results show that barley germination characteristics and seedling growth may be affected by different priming and duration of them. There were differences in final germination percentage of prim duration depending on stress intensity. Willenborg et al. (2005) also reported that germination characteristics were affected by moisture stress in six western Canadian oat genotypes. However, greater efficiency of osmohardening with CaCl<sub>2</sub> and KCl is possibly related to the osmotic advantage that both K<sup>+</sup> and Ca<sup>2+</sup> have in improving cell water saturation, and that they act as co-factors in the activities of numerous enzymes. There was a little reduction in germination in solutions of KCl. In contrast, a more significant reduction in germination was observed in solutions of PEG. Barley differed significantly in its response to KNO<sub>3</sub>, KCl and osmotic stress (PEG). Such responses have been reported

by many workers over a wide range of halophytic and glycophytic plant species (Hampson & Simpson, 1990a, b; Falleri, 1994; Huang & Redmann, 1995; Katembe et al., 1998; Raza et al., 2006; 2007). The general view is that a decrease in water potential gradient between seeds and their surrounding media adversely affects seed germination and subsequent growth processes. The physical process of water uptake leads to the activation of metabolic processes as the dormancy of the seed is broken following hydration (Katembe et al., 1998). Elevated KCL and PEG concentrations slowed down water uptake by seeds, thereby inhibiting the germination process. In this research, it was found that PEG to be more inhibitory to germination. These results agree with those of Hampson & Simpson (1990a, b) for wheat, as well as Huang & Redmann (1995) for barley and Brassica, Gulzar & Khan (2001) for *Aeluropus lagopoides*. However, such findings

are not universal. For example, Katembe et al., (1998) observed the opposite in wild rye, wheat grass and *Atriplex* species. However, tolerance to high salt stress may also be due to the tolerance of these

plant species (halophytic) to high salt-induced osmotic stress. Unlike PEG, KCL may readily cross the cell membrane into the cytoplasm of the cell unless an active metabolic pump prevents accumulation of the ions (Katembe et al., 1998). However, germination was more sensitive to PEG-induced stress. It has been reported that PEG-induced osmotic stress can cause hydrolysis of storage compounds that further lower the internal osmotic potentials of the seed (Hampson & Simpson, 1990a). In conclusion, barley is more sensitive to osmotic stress at germination stage. However, at early growth stage both salt-induced osmotic stress and CL toxicity reduced growth.

Table 1. Analysis of variance for different traits

	df	Seedling length	Radicle length	Length R/S	Stem dry weight	Radicle Dry weight	dry weight R/S	Number of germination	Germination percentage	Rate of germination
Block	2	0.77	1.25	0.0145	0.0022	0.0019	0.44	3.25	28	3.258
A	6	25.1**	4.1	0.5**	0.0044	0.0078*	4.1**	14.22*	101.33*	198.3**
B	2	10.1*	22.5	0.066	0.002	0.0031	0.398	49.36**	299.36**	211.36**
A*B	12	2.74**	4.22	0.049	0.0011	0.002	0.311	7.22**	40	99
Error	40	0.77	4.1	0.029	0.00048	0.003	0.25	4.1	28.98	52
CV%		8.8	12.3	12	8.88	14.1	19.3	9.66	7.24	21.3

\*, \*\*, Significant at p=0.05 and 0.01 level, based on a F-test

Table 2. Means of the estimated traits under experiment conditions for priming media

A	Seedling Length (mm)	Radicle length (mm)	Length R/S (mm)	Stem dry Weight (g)	Radicle Dry Weight (g)	Dry weight R/S (g)	Number of germination	Germination percentage	Rate of Germination %
PEG 5%	9.11 <sup>b</sup>	14.2 <sup>ab</sup>	1.45 <sup>a</sup>	1.1 <sup>b</sup>	0.31 <sup>a</sup>	2.11 <sup>b</sup>	35.2 <sup>b</sup>	82.8 <sup>b</sup>	25 <sup>bc</sup>
PEG 10%	11 <sup>ab</sup>	10 <sup>b</sup>	1.29 <sup>a</sup>	0.9 <sup>c</sup>	0.22 <sup>bc</sup>	3.3 <sup>a</sup>	40.2 <sup>b</sup>	79.2 <sup>a</sup>	21.5 <sup>c</sup>
KNO3 1%	15.1 <sup>a</sup>	12 <sup>ab</sup>	0.89 <sup>b</sup>	0.99 <sup>c</sup>	0.29 <sup>ab</sup>	1.2 <sup>c</sup>	42.2 <sup>b</sup>	92 <sup>a</sup>	19.2 <sup>c</sup>
KNO3 2%	10.33 <sup>ab</sup>	11.2 <sup>b</sup>	1.1 <sup>b</sup>	1.33 <sup>a</sup>	0.19 <sup>c</sup>	1.9 <sup>c</sup>	48.11 <sup>a</sup>	94 <sup>a</sup>	38.9 <sup>a</sup>
KCl 2%	10.2 <sup>ab</sup>	14.15 <sup>ab</sup>	1.02 <sup>b</sup>	1.1 <sup>b</sup>	0.122 <sup>c</sup>	1.8 <sup>c</sup>	34.2 <sup>c</sup>	81 <sup>b</sup>	24 <sup>bc</sup>
KCl 4%	18.1 <sup>a</sup>	18.2 <sup>a</sup>	1.03 <sup>b</sup>	0.99 <sup>c</sup>	0.122 <sup>c</sup>	2.2 <sup>b</sup>	33.1 <sup>c</sup>	84 <sup>b</sup>	20.2 <sup>c</sup>
control	9.2 <sup>b</sup>	12.2 <sup>ab</sup>	1 <sup>b</sup>	1.29 <sup>ab</sup>	0.29 <sup>ab</sup>	1.4 <sup>c</sup>	40.1 <sup>b</sup>	92 <sup>a</sup>	26.6 <sup>bc</sup>

In each column, any two means having a common letter are not significantly at p=0.05 based on Duncan's multiple range test

Table 3. Means of the estimated traits under during time for priming

B	Seedling Length (mm)	Radicle length (mm)	Length R/S (mm)	Stem dry Weight (g)	Radicle Dry Weight (g)	Dry weight R/S (g)	Number of germination	Germination percentage	Rate of Germination %
20h	9.2 <sup>c</sup>	10.22 <sup>b</sup>	1.2	1.1 <sup>b</sup>	0.22	3.11	49.25 <sup>a</sup>	92.25 <sup>a</sup>	28.2 <sup>b</sup>
40h	12.22 <sup>b</sup>	14.2 <sup>a</sup>	1.11	1.65 <sup>ab</sup>	0.25	3.22	45.25 <sup>a</sup>	88.3 <sup>a</sup>	26.3 <sup>b</sup>
60h	15.2 <sup>a</sup>	15.22 <sup>a</sup>	1.19	2.1 <sup>a</sup>	0.23	3.25	38.2 <sup>b</sup>	79.25 <sup>b</sup>	36.25 <sup>a</sup>

In each column, any two means having a common letter are not significantly at p=0.05 based on Duncan's multiple range test

Table 4. Means of the estimated traits under experiment conditions for priming media  $\times$  during time

A	B	Seedling Length (mm)	Number of germination
PEG 5%	20h	8.12 <sup>c</sup>	36.2 <sup>b</sup>
PEG 5%	40h	8.86 <sup>c</sup>	38 <sup>b</sup>
PEG 5%	60h	9.22 <sup>c</sup>	41.2 <sup>ab</sup>
PEG 10%	20h	6.9 <sup>c</sup>	32.1 <sup>bc</sup>
PEG 10%	40h	8.2 <sup>c</sup>	33 <sup>bc</sup>
PEG 10%	60h	8.8 <sup>c</sup>	36 <sup>b</sup>
KNO3 1%	20h	10.51 <sup>b</sup>	42.1 <sup>a</sup>
KNO3 1%	40h	11.2 <sup>b</sup>	41.1 <sup>a</sup>
KNO3 1%	60h	11.33 <sup>b</sup>	44.2 <sup>a</sup>
KNO3 2%	20h	8.1 <sup>c</sup>	36.9 <sup>ab</sup>
KNO3 2%	40h	10.1 <sup>b</sup>	35.2 <sup>b</sup>
KNO3 2%	60h	13.3 <sup>ab</sup>	35.26 <sup>b</sup>
KCl 2%	20h	13.3 <sup>b</sup>	33.2 <sup>bc</sup>
KCl 2%	40h	12.99 <sup>b</sup>	36.2 <sup>b</sup>
KCl 2%	60h	15.22 <sup>a</sup>	38.2 <sup>ab</sup>
KCl 4%	20h	11.22 <sup>b</sup>	29.8 <sup>c</sup>
KCl 4%	40h	14.2 <sup>a</sup>	31.2 <sup>bc</sup>
KCl 4%	60h	13.1 <sup>ab</sup>	33.1 <sup>bc</sup>
control	20h	7.11 <sup>c</sup>	37.2 <sup>b</sup>
control	40h	8.1 <sup>c</sup>	40.2 <sup>a</sup>
control	60h	8.81 <sup>c</sup>	39.2 <sup>a</sup>

In each column, any two means having a common letter are not significantly at  $p=0.05$  based on Duncan's multiple range test

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