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Effect of seed vigour on stress tolerance of barley (*Hordeum vulgare*) seed at germination stage

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Summary

One of the prerequisites of efficient crop production is the use of high vigour seed that could guarantee a vigorous establishment. Seed production under organic and low input conditions is more difficult compared to conventional systems. Either nutrient or drought stress during seed development on parent plants can affect subsequent seed quality. The effects of both stresses on the subsequent germination of barley seeds were evaluated under drought and salt stress using either polyethylene glycol or NaCl at osmotic potentials of 0, -0.4, -0.8, -1 and -1.2 MPa. Barely seeds were produced under different irrigation (stressed and normal) and fertilizing systems (organic and chemical) in two cropping seasons (2007-2008).

Water stress in the field during seed development reduced 1000 seed weight and Mean germination time (MGT) in both years. The seeds produced by plants at severe or moderate drought stress treatments had a higher germination percentage and lower MGT in stressed conditions applied by PEG and NaCl compared to the control. Seeds produced at either organic or low input fertilizing systems, had a better germination percentage at different solution potentials compared to chemical fertilizing system. Drought stress (during grain filling) improved tolerance of the seeds to osmotic or salt stress during germination, imposed by PEG and NaCl.

Introduction

Barley (*Hordeum vulgare* L.) is an important cereal grain crop which is widely used as food, feed and for malt production. It ranks fifth among all crops in grain production in the world after maize, wheat, rice and soybean (FAO, 2008). It is largely grown in Asia and northern Africa along the dry marginal lands with 200-300 mm rainfall per year (Baik and Ullrich, 2008) where the water and salinity are the most important environmental stresses. Salinity and drought stresses could reduce seed germination by limiting seed water absorption (Bliss *et al*., 1986; Almansouri *et al*., 2001; Mer *et al*., 2000), changes in structural organization of seed (displacement of Ca by Na from critical cell wall binding sites which could disrupt cell wall synthesis and hence inhibit plant growth) (Dhanda *et*

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al., 2004; Othman *et al.,* 2006) or synthesis of proteins in embryos during germination process (Ueda *et al*., 2004; Hurkman and Tanka, 1996).

Ideally, seeds as propagation material should produce vigorous seedlings under a broad range of field conditions. This aspect of seeds is called "seed vigour". Seed vigour is a concept describing several characteristics which include the rate and uniformity of germination and growth, tolerance to environmental stresses after sowing and retention of performance after storage. Seeds which perform well in some or all of these aspects are termed high-vigour seeds (Black and Bewely, 2000). Thus to confirm a good germination and establishment of barley, high vigour seeds with reasonable tolerance to drought and salt stresses at germination stage are needed.

Barely seed vigour is controlled genetically as well as environmentally (Fenner, 2000). Drought stress, temperature and availability of nutrients during seed development can significantly affect seed vigour. Water and nutrient availability are limiting factors in sustainable barley seed production systems in Iran. These factors could affect tolerance of seeds to existing stresses during germination in the following season. Drought and high temperatures during seed filling result in a larger number of small seeds and poor seed vigour in soybean (*Glycine max* L.) (Dornbos and Mullen, 1991, Heartherly, 1993), pea (*Pisum sativum* L.) (Fougereux *et al*., 1997) and barley (*Hordeum vulgare* L.) (Szira *et al.,* 2008). Clua *et al.* (2006) founded that imposed drought stress during seed development of *Lotus glaber* which has hard seed, reduced germination percentage and increased germination rate.

Chlorophyll fluorescence, electrical conductivity, germination percentage, cold and drought tolerance, shoot length, root length and seedling dry weight have been used as various seed vigour parameters (Kim *et al.,* 1989; Verma *et al.,* 1998; Chloupek *et al.,* 2003; Jalink *et al.,* 1998; Groot *et al.,* 2006 and 2008). However, there is limited information about the effects of seed production conditions on subsequent salt and drought stress tolerance at the germination stage. This experiment was conducted to assess germination characteristics of barely seed produced under different drought or nutrient stresses.

Materials and methods

Plant material

Initial barley (cv. Turkman) seeds, used in the experiments for seed production, were provided by the Seed and Plant Breeding Research Institute, Karaj, Iran. The variety used is a spring malting barley type.

Seed production conditions

Field studies were conducted at the Experimental Farm of the Department of Agronomy and Plant Breeding, University of Tehran, Iran (35°56'N and 50°58'E with an altitude of 1312 m) during the cropping seasons of 2006-2007 and 2007-2008. The soil texture of the experimental site was clay loam. Experimental design was a split plot arrangement based on a randomized complete block design with four replications. The barley seeds were sown on 17th of March 2007 (for the 2007 harvest) and 5th of December 2007 (for

the 2008 harvest) at a density of 300 seed/m^2. The treatments consisted of three irrigation regimes (main plots) and six soil fertilizing systems (sub-plots). Three different irrigation treatments were applied related to the phenological stages of barley according to Zadoks scale (1974) (table 1). Normal irrigation was performed at weekly intervals when soil moisture reached 50% available soil water at root growth zone. There was no effective rainfall during seed development and air temperature and relative humidity measurements indicated that 2008 was a drier year than 2007 (table 2). Six different fertilizing systems were employed (table 3). Fertilizer application rate was based on soil analyses.

Table 1. Irrigation regimes employed on the plots during seed production.

NS	Non-stressed: Normal irrigation until the end of the plant physiological maturity;
MS	Medium stress: Ceased irrigation from the beginning of flowering (Zadoks 65) to the initiation of seed filling stage (Zadoks 70);
SS	Severe stress: Ceased irrigation from the initiation of flowering stage to the end of the physiological maturity).

Table 2. Total monthly precipitation, average monthly temperature, average monthly relative humidity and maximum air temperature (T_{max}) for the period 1 April to 31 July in 2007 and 2008, Karaj, Iran.

Month	Precipitation (mm)		Temperature (°C)		Relative humidity (%)		T_{max} (°C)	
	2007	2008	2007	2008	2007	2008	2007	2008
April	3.3	0.1	14.4	17.7	52	34	23.6	33
May	0.4	0	20.3	22	45	34	22.4	35
June	0.4	0	24.3	24.6	38	36	38	37
July	0.2	0	27	27.8	37	34	38.4	39.8

Table 3. Different fertilization regimes on the sub plots employed during seed production.

NF	No fertilization
NB	Nitrogen and phosphorous biofertilizers (Biofertilizer is a complex of different free living nitrogen fixing and phosphorus solubilizing bacteria)
CF	100% Chemical fertilizer (NPK) (based on soil chemical analysis)
VC	5t/ha of Vermicompost (or V-compost), is a heterogeneous mixture of decomposing vegetable or food waste, bedding materials, and pure vermicast (the residue of earth worms) produced during the course of normal vermiculture operations, it is a nutrient-rich organic fertilizer and soil conditioner (Kelly and Knutzen, 2008)
CV	50% chemical fertilizer (NPK) + 50% vermicompost (2.5 t/ha)
CB	50% chemical fertilizer (NPK) + 50% biofertilizer

Seed quality tests

Standard germination test

Laboratory germination tests were performed at Plant Research International, Wageningen, Netherlands on harvested seeds from the productions in both experimental years (2007 and 2008) according to the methods of the International Seed Testing Association (ISTA, 1999). Seeds were considered germinated when radicles emerged at least 2mm. Seedling quality was evaluated after 7 days in respect to the number of normal and abnormal seedlings (ISTA, 2006). Parameters related to germination, such as maximum germination (G_{max}) and mean germination time (MGT, calculated by integration of the fitted germination curve) were calculated using the software package SeedCalculator V3.0 (Plant Research International, Wageningen, The Netherlands).

Drought and salt stress germination tests

To evaluate drought stress and salt tolerance during germination, 25 seeds of each treatment were placed in 9 cm sterile Petri dishes containing two Whatman No.1 filter papers moistened either with 13 ml of sterile demineralised water or the same solution added with polyethylene glycol (PEG 6000) or NaCl. These osmotic agents were used in four osmotic potentials (-0.4, -0.8, -1.0 and -1.2 MPa) created using either PEG-6000 suggested by Michael and Kaufman (1973) or dissolving 9.4, 18.9, 23.6 and 28.3 gr/kg NaCl salt in demineralised water, to create the corresponding osmotic pressures. Three replications were used for each treatment. Experimental design was a randomized complete block with split-split plot arrangements. Samples from the drought stress treatments in the field were assigned to the main plots, while those from the fertilizing systems were considered as sub plots. The water or salt concentration levels during the germination tests were allocated to the sub-sub plots. To limit water loss, the petri dishes were packed in two plastic bags. During the daily evaluations demineralised water was added to the Petri dishes if needed to maintain water and salt concentrations near target levels. The imbibed seeds were placed in an incubator at approx. 20°C in the dark. Radicle protrusion was scored daily for seven days to evaluate the germination rate and the final germination percent after this period was recorded for the other germination characteristics. The final number of germinated seeds after the germination period (7 days) were used in analysis of variance by transformation by function of $y = \arcsin\sqrt{p/100}$, were p is the germination percentage.

Statistical analysis

Data were statistically analyzed separately for each production year by analysis of variance (ANOVA) using MSTATC and SAS programs (Michigan State Univ., East Lansing, MS, USA). Homogeneity of error variances was tested using Bartlett's Chi-square. Since value of χ^2 was not significant, so combined analysis of data was performed for two years data. Probability of significant differences among treatments and interactions by Duncan test ($p < 0.05$) were used to compare means within and among treatments.

Results

Germination in a standard germination test

In both years the 1000 seed weight was reduced by water stress and this effect was greater in the relative dryer production year of 2008 (table 4). Seeds produced under severe stress during development geminated faster, expressed as a shorter mean germination time (MGT), than seeds produced under full irrigation. Seeds produced under mild water stress showed an intermediate rate of germination. Total germination was not influenced by the irrigation regime.

Seed weight was hardly influenced by the fertilizing systems applied (table 4). Only when the results over the two production years were combined, it became apparent that seeds produced with nitrogen fixing and phosphorus solubilising bacteria (NB) were heavier than those produced with 100% chemical fertilizer (CF).

Seeds produced with 50% chemical fertilizer plus 50% biofertiliser (CB) fertilization germinated significantly faster, while absence of fertilizer (NF) or fertilization with vermicompost (VC) resulted in significantly slower seed germination (table 4). Total germination was only influenced in the 2008 production year, but on average there was no significant influence of the fertilization regimes on those parameters.

Application of nitrogen fixing and phosphorus solubilising bacteria alone or along with chemical fertilizer reduced MGT in 2007 but application of vermicompost along with chemical fertilizer reduced MGT in 2008 (table 4). Results of combined analysis of variances for two years showed a significant interaction from the effect of irrigation and production year on 1000 seed weight. In both years 1000 seed weight decreased as drought severity increased (7.16% (MS) and 16.27% (SS) reduction in 2007 and 32% (MS) and 37.45% (SS) in 2008). The detrimental effect of increasing water stress on 1000 seed weight was higher in 2008 because of higher temperature and lower relative humidity during grain filling in this year. But 1000 seed weight under normal irrigation in 2008 was higher than 2007 which could be related to the growth period that was longer in 2008 because of earlier sowing date.

Germination under drought and salt stress

The results showed that germination was suppressed by -1.2 MPa potential induced by PEG and NaCl treatments. Germination increased as water potential decreased to -0.4 MPa (table 5). The seeds subjected to severe stress during seed development, had a better germination percentage under stressed conditions applied by either PEG or NaCl, compared to those produced under moderate and normal conditions (figure 1).

Mean germination time was significantly less in 2008 compared to 2007 for both PEG and NaCl tests (table 5). The seeds developed under drought stress during grain filling were slightly more tolerant to osmotic stress during the germination phase because their MGT was less than the normal irrigation treatment (table 5). Also the germination percentage in these seeds was significantly higher under PEG and NaCl stresses compared to normal seeds (seeds produced under normal irrigation treatment) (table 5). Fertilizing system had a slight significant effect on germination in NaCl solutions. The seeds produced at application of chemical fertilizer along with vermicompost (CV) treatment germinated

Table 4. Effects of irrigation and fertilizing treatments on 1000 seed weight, mean germination time, and final germination percentage of c.v Turkaman barley seeds produced in 2007 and 2008.

		1000 seed weight	MGT	Germination (%)
2007				
Irrigation system	NS	45.67 a	1.88 a	84.49 a
	MS	42.4 b	1.74 ab	84.68 a
	SS	38.24 c	1.64 b	82.85 a
Fertilizing system	NF	42.95 a	1.91 a	85.81 a
	NB	42.46 a	1.71 bc	85 a
	VC	41.66 a	1.82 ab	81.34 a
	CV	42.74 a	1.77 ab	82.53 a
	CB	42.90 a	1.61 c	83.54 a
	CF	39.86 a	1.71 bc	85.81 a
2008				
Irrigation system	NS	47.69 a	2.05 a	86.73 a
	MS	32.42 b	1.89 ab	82.66 a
	SS	29.83 c	1.73 b	77.75 a
Fertilizing system	NF	35.70 a	1.92 ab	76.87 c
	NB	39.97 a	1.95 ab	83.17 abc
	VC	37.73 a	2.01 a	85.00 ab
	CV	34.45 a	1.76 c	77.24 bc
	CB	35.89 a	1.84 bc	85.81 a
	CF	36.13 a	1.88 bc	86.18 a
2007 and 2008 combined				
Year	2007	42.10 a	1.89 a	84.00 a
	2008	36.65 b	1.75 b	82.38 a
Irrigation system	NS	46.67 a	1.97 a	85.61 a
	MS	37.41 b	1.82 b	83.67 a
	SS	34.03 c	1.69 c	80.301 a
Fertilizing system	NF	39.32 ab	1.91 a	81.34 a
	NB	41.22 a	1.83 ab	84.08 a
	VC	39.70 ab	1.92 a	83.17 a
	CV	38.60 ab	1.77 bc	79.88 a
	CB	39.40 ab	1.72 c	84.68 a
	CF	38.00 b	1.79 bc	86.00 a

Treatments within a column followed by the same letter are not significantly different with the Duncan test at 0.05 level. NS = normal irrigation until the end of the plant physiological maturity; MS = ceased irrigation from the beginning of flowering (Zadoks, 65) to the initiation of seed filling stage (Zadoks, 70); SS = ceased irrigation from the initiation of flowering stage; NF = no fertilizing, NB = phosphatic and nitrogenous biofertilizer, VC = vermicompost, CV = 50% chemical fertilizer including NPK+50% vermicompost, CB = 50% chemical fertilizer including NPK + 50% biofertilizer and CF = 100% chemical fertilizer

Table 5. Effects of year, irrigation and fertilizing treatments, PEG and NaCl on mean germination time, and final germination percentage of Turkaman cultivar in 2007 and 2008.

		PEG		NaCl	
		MGT (day)	Germination (%)	MGT (day)	Germination (%)
Year	2007	2.80 a	71.86 a	2.76 a	72.41 a
	2008	1.80 b	70.07 a	1.89 b	69.97 a
Irrigation system	NS	2.36 a	68.31 b	2.32 a	67.96 b
	MS	2.18 b	71.17 ab	2.25 b	71.85 a
	SS	2.15 b	72.88 a	2.14 c	73.02 a
Fertilizing system	NF	2.34 a	73.18 a	2.27 a	72.57 a
	NB	2.19 a	72.16 a	2.24 a	71.26 a
	VC	2.14 a	71.49 a	2.21 a	70.87 a
	CV	2.14 a	70.77 a	2.24 a	70.52 a
	CB	2.27 a	69.06 a	2.24 a	72.62 a
	CF	2.30 a	68.06 a	2.22 a	67.81 b
Osmotic potential	0.0	0.79 d	80.81 a	0.78 e	81.24 a
	-0.4	1.24 c	82.45 a	1.18 d	79.53 a
	-0.8	2.79 b	65.51 b	2.06 c	74.93 b
	-1.0	4.09 a	54.38 c	2.87 b	67.58 c
	-1.2	0.00 e	0.00 a	4.30 a	51.43 d

Treatments within a column followed by the same letter are not significantly different with the Duncan test at 0.05 level. NS = normal irrigation until the end of the plant physiological maturity; MS = ceased irrigation from the beginning of flowering (Zadoks, 65) to the initiation of seed filling stage (Zadoks, 70); SS = ceased irrigation from the initiation of flowering stage; NF = no fertilizing, NB = phosphatic and nitrogenous biofertilizer, VC = vermicompost, CV = 50% chemical fertilizer including NPK+50% vermicompost, CB = 50% chemical fertilizer including NPK + 50% biofertilizer and CF = 100% chemical fertilizer

less well in NaCl solutions compared to other treatments (table 5). Germination was delayed by moderate osmotic potential (-0.4 MPa for both PEG and NaCl solutions). However, germination percentage was not affected by drought stress and was equal to the control (demineralised water) treatment. Higher osmotic potential significantly reduced germination percentage (table 5).

There was no significant interaction between fertilizing system and osmotic potential on MGT or germination percentage under drought stress induced by PEG application. However, in combined analysis of data, a significant interaction between irrigation system and osmotic potential was observed on germination percentage under drought stress imposed by PEG (figure 2). Stress in the field affected seed germination in different PEG solutions. This showed that drought stress during field production could induce drought stress tolerance in subsequent produced seeds. This could explain the greater germination percentage in PEG potentials (-0.4, -0.8 and -1.0 MPa) by stressed seeds especially in SS treatment at -1.0 MPa osmotic potential (figure 2).

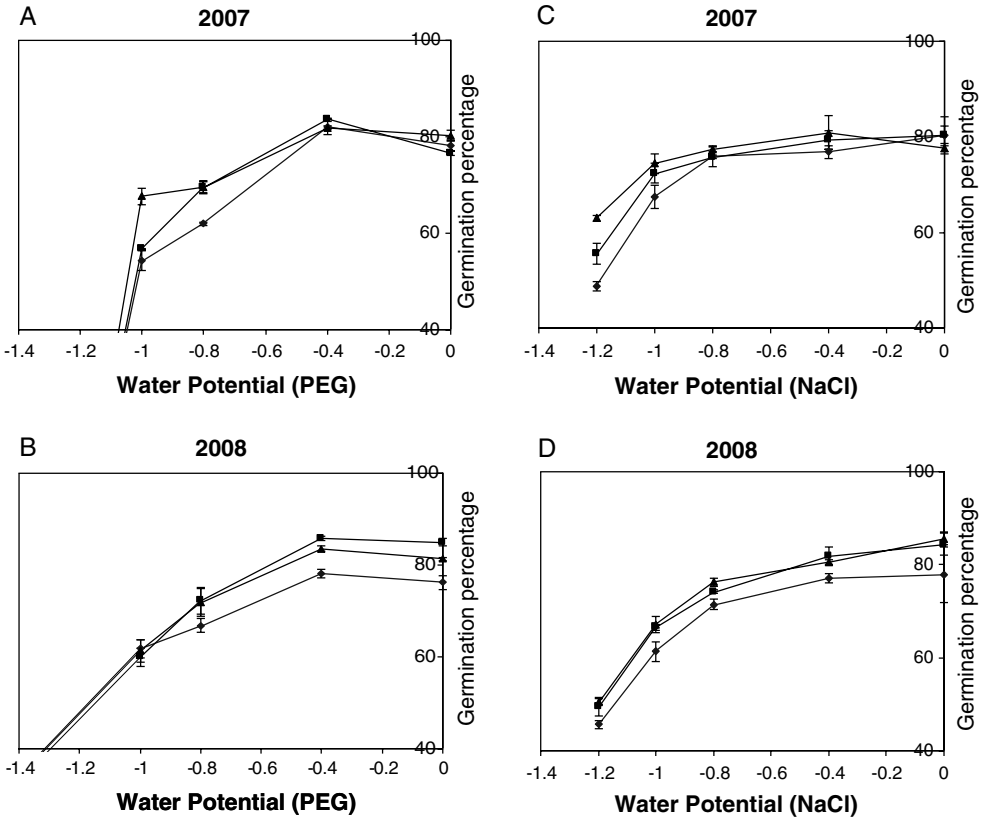

Figure 1. Effects of PEG (A and B) and NaCl (C and D) treatments and irrigation regimes on germination of barley seeds in 2007 and 2008. Bars indicate ± SD of means. -◆- NS: Non-Stress (normal irrigation until the end of the plant physiological maturity); -■- MS: Medium Stress (creased irrigation from the beginning of flowing (Zadoks, 65) to the initiation of seed filling stage (Zadocks, 70)); -▲- SS: Severe Stress (ceased irrigation from the initation of flowing stage (Zadocks, 65) to the end of the physiological maturity).

There was a significant interaction between irrigation and fertilizing system across all water potential levels induced by NaCl on both MGT and germination percentage (figure 3 and 4). However, there was only a significant interaction of irrigation and osmotic potential by NaCl on MGT (figure 5). Seeds produced in all fertilizing treatments at SS irrigation system had less MGT compared to seeds produced under the NS irrigation system. The same trend was also observed for all seeds produced in MS irrigation system except for the VC treatment (figure 3). Irrigation and fertilizing systems affected seed germination in different NaCl solutions. The seeds produced in the SS treatment had a greater germination percentage compared to NS and it was significant in seeds produced in treatments containing chemical fertilizer (CV, CB and CF) (figure 4).

Drought stress on parent plants during grain filling significantly affected produced seeds and reduced MGT in all NaCl potentials but this reduction was significant only in NaCl potentials higher than -0.4 MPa (figure 5).

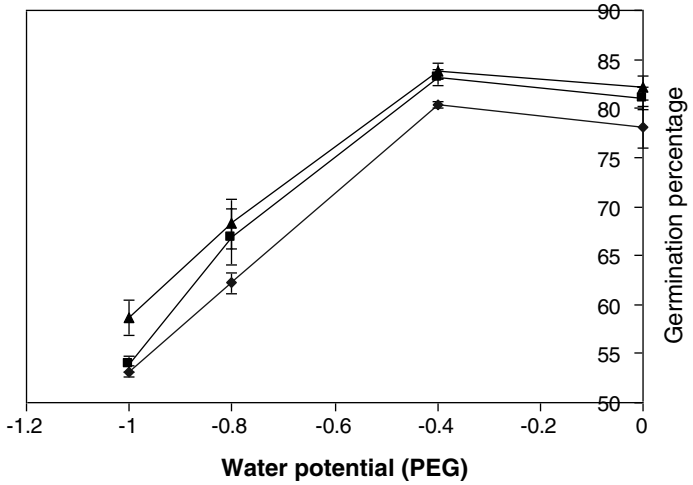

Figure 2. Effects of PEG treatment and irrigation regimes on germination of barley seeds over two years. Bars indicate ± SD of means. -◆- NS: Non-Stress (normal irrigation until the end of the plant physiological maturity); -■- MS: Medium Stress (creased irrigation from the beginning of flowing (Zadoks, 65) to the initiation of seed filling stage (Zadocks, 70)); -▲- SS: Severe Stress (ceased irrigation from the initation of flowing stage (Zadocks, 65) to the end of the physiological maturity).

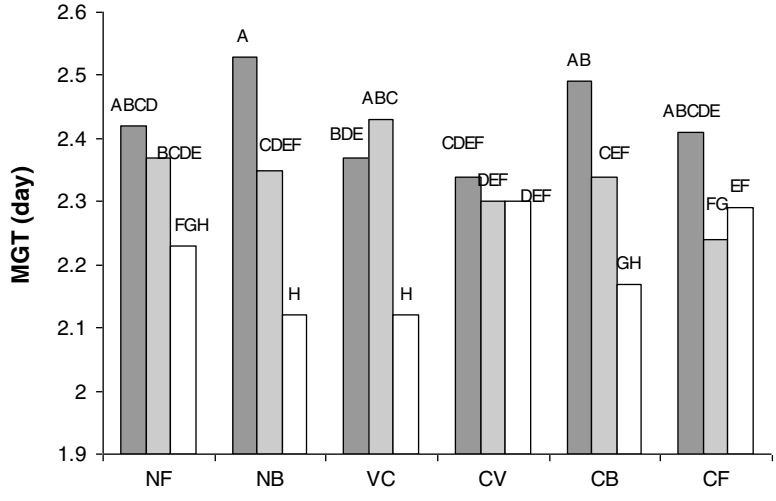

Figure 3. Effect of fertilizer and irrigation regimes across all water potential levels induced by NaCl on mean germination time of barley seeds. Means with the same letter(s) are not significantly different at $P < 0.05$ level. NF= no fertilizing, NB= phosphate and nitrogenous biofertilizer, VC=vermicompost, CV=50% chemical fertilizer including NPK+50% vermicompost, CB= 50% chemical fertilizer including NPK + 50% biofertilizer and CF= 100% chemical fertilizer. -▩- NS: Non-Stress (normal irrigation until the end of the plant physiological maturity); -▨- MS: Medium Stress (creased irrigation from the beginning of flowing (Zadoks, 65) to the initiation of seed filling stage (Zadocks, 70)); -□- SS: Severe Stress (ceased irrigation from the initation of flowing stage (Zadocks, 65) to the end of the physiological maturity).

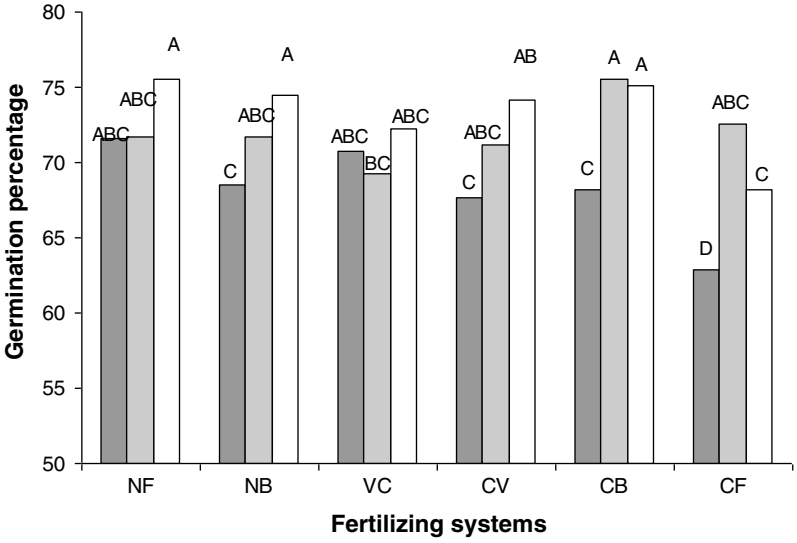

Figure 4. Effect of fertilizer and irrigation regimes across all water potential levels induced by NaCl on germination percentage of barley seeds. Means with the same letter(s) are not significantly different at $P < 0.05$ level. NF= no fertilizing, NB= phosphate and nitrogenous biofertilizer, VC=vermicompost,CV=50% chemical fertilizer including NPK+50% vermicompost, CB= 50% chemical fertilizer including NPK + 50% biofertilizer and CF= 100% chemical fertilizer. -▩- NS: Non-Stress (normal irrigation until the end of the plant physiological maturity); -▨- MS: Medium Stress (creased irrigation from the beginning of flowing (Zadoks, 65) to the initiation of seed filling stage (Zadocks, 70)); -□- SS: Severe Stress (ceased irrigation from the initation of flowing stage (Zadocks, 65) to the end of the physiological maturity).

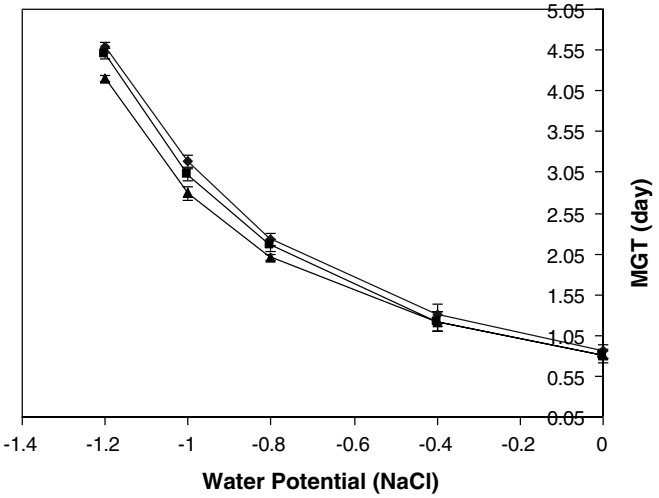

Figure 5. Effects of NaCl treatments and irrigation regimes on mean germination time of barley seeds over two years. Bars indicate ± SD of means.-◆- NS: Non-Stress (normal irrigation until the end of the plant physiological maturity); -■- MS: Medium Stress (creased irrigation from the beginning of flowing (Zadoks, 65) to the initiation of seed filling stage (Zadocks, 70)); -▲- SS: Severe Stress (ceased irrigation from the initation of flowing stage (Zadocks, 65) to the end of the physiological maturity).

Discussions

Stress during grain filling significantly reduced 1000 seed weight because of reduction in post–anthesis photosynthesis and in the amount of remobilisable assimilates (Kobata *et al.,* 1992). The reduction was pronounced by higher temperature (Dornbos and Mullen, 1991) and drier air in 2008 that tended to lower seed weight. The earlier sowing date in 2008 provided a longer growing season for plants leading to better vegetative growth which caused more assimilate production and heavier 1000 seed weight. The results of the standard germination test showed that barley germination was only affected by nutrient availability during the growing season in 2008. Geus *et al.,* (2008) found different results with organic and conventional farming systems on seed germination in two consequent years. They suggested that crop condition during the growing season, soil fertility and nutrient availability during the crucial phases of seed development and maturation may have influenced seed quality. Rate of germination (MGT) is an important factor leading to a quick and good establishment in the field and malting industry. Irrigation regimes significantly affected this parameter in both years. Benech Arnold *et al.,* (1992) in their experiment with sorghum concluded that water stress during seed development reduces dormancy of *Sorghum halepense* seeds through modification in properties of the glumes that apparently result in enhancement of their permeability to oxygen diffusion. On the other hand Ellis and Marshal (1998) found that in barley grain size affected germination time, though small seeds germinated faster than big ones.

MGT was affected by application of nitrogen fixing and phosphorus solubilising bacteria alone or along with chemical fertilizer in 2007 and by application of chemical fertilizer along with vermicompost (CV) in 2008. Nitrogen fixing and phosphorus solubilising bacteria are able to supply phosphorus from mineral sources. This will provide more available phosphorus for plants leading to greater phosphorus content in seeds produced under these fertilizer treatments. This phenomenon could help the seeds to germinate faster. This reaction was probably more pronounced in the CV fertilizer treatment in 2008. Fertilizer systems containing chemical fertilizer showed less MGT in both years. It seems that in these treatments mineral nutrients are more readily available than other treatments which could help the seeds to germinate faster. Nutrient availability also increased germination in CF and CB fertilizing treatments in 2008.

The results of the drought and salt stress tests at germination indicated that the inhibitory effect of PEG and NaCl is because of the osmotic stress they impose during the early phases of germination. Germination was suppressed by the osmotic stress probably because of inhibition in seed hydration which was greater in higher osmotic potentials. Moderate drought stress (-0.4 MPa) at germination delayed germination because of restricted hydration. However, with increased osmotic water potential, germination was suppressed more severely by PEG rather than NaCl. These results corresponded with findings of Heikal and Shaddad (1982), Dodd and Donovan (1991), Huang and Redman (1995), Almansouri *et al.* (2001) and Kaya *et al.* (2005), who suggested that increasing internal osmotic potential by ion uptake (Na^+ and Cl^-) in seeds lead to more water absorption and higher germination in high osmotic potential. However, this phenomenon does not occur in drought induced by PEG treatments because it is not a penetrating

molecule. Barley could stand NaCl up to -0.4 MPa without any decrease in germination but higher water potentials induced by NaCl significantly diminished germination which is in agreement with findings of Othman *et al.* (2006). Na^+ increase inside plants has a toxic effect on seed germination mainly by affecting the plant water relations or through displacement of Ca by Na from critical cell wall binding sites which could disrupt cell wall synthesis and hence inhibit plant growth (Othman *et al.,* 2006).

Drought stress and its duration during the grain filling period in parent plants induced more salinity and drought tolerance in seeds. This phenomenon was more pronounced when the drought stress period was longer during the grain filling period of parental plants. Any increase in osmotic potential prolonged germination time (decreased germination rate) due to inhibition of water uptake, but this reduction in germination rate was less pronounced in MS (ceased irrigation from the beginning of flowering to the initiation of seed filling stage) and SS treatments (ceased irrigation from the beginning of flowering to the end of the physiological maturity) compared to the control. Tolerance to drought stress is adjusted by osmoprotectants and regulation of water permeability (Ueda *et al.,* 2004, Dhanda *et al.,* 2004). Thus differences in drought tolerance among SS and MS treatments compared to NS (control) could be better explained by some changes in protein synthesis and membrane integrity during grain filling. Fenner (2000) described that drought during seed maturation may affect seed germinability by changing the properties of maternal tissues surrounding the seed which also could cause seed to switch from the seed-developing to germination stage. This switch involves changes in protein patterns and messenger RNA.

There is evidence that ABA starts to increase during seed filling which regulates senescence in plants (Tasda *et al.,* 1999). Yange and Zhange (2006) observed increases in ABA content in root and stem after soil drying treatments. Also Skriver and Mundy (1990) described that ABA levels increase in tissues subjected to osmotic stress by desiccation, salt, or cold as well as oxidative stress (Chizhova *et al.,* 2005). It promotes tolerance of the plants to desiccation, enabling them to survive under adverse conditions or to colonize areas with scarce water availability and regulates seed germination (Ton *et al.*, 2009).

Thus, it could be concluded that in our experiments the ABA content could have increased in drought stress treatments. Galli and Levi (1982) observed that pretreatment of seed by ABA caused better stress tolerance and improved germination percentage. Therefore, increased ABA content in these stressed plants during grain filling could well explain the reduction in the MGT and the better germination rate under water and salinity stress conditions.

Significant interactions between irrigation and fertilizing systems on MGT (figure 3) showed that stress in the field decreased MGT in all fertilizing systems, but in organic fertilizers (NB and VC) MGT decreased significantly more than the other fertilizers. It shows the importance of these organic fertilizers in arid environments. Significant differences in the effects of irrigation and fertilizing systems on seed germination (figure 4) demonstrates the beneficial effect of drought stress during grain filling on germinability of produced seeds. However, under chemical fertilizing treatment (CF) the severe drought stress (SS) may cause toxicity in seeds (because of high concentration of minerals) which lead to less germination compared to corresponding treatment in other fertilizing systems.

From the results of the two years of this experiment, it could be concluded that germination percentage derived from the standard germination test is not a reliable criteria to assess barley seed vigour which has been produced under stress conditions. Germination tests under stressed conditions could be a more reliable assay in this regard. According to our experiments, barley is a drought and salinity tolerant crop and can produce seeds with higher vigour in severe conditions compared to normal environments.

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