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# Study on Osmotic Stress Tolerance in Promising Durum Wheat Genotypes Using Drought Stress Indices

Ali Akbar Moayedi, Amru Nasrulhaq Boyce, Syed Shahar Barakbah

Institute of Biological Sciences, University of Malaya, 50603, Kuala Lumpur, Malaysia

**Abstract:** One way to overcome the negative effects of water stress on crop production is the development of drought tolerant cultivars. In the present study we have attempted to quantify the drought tolerance of several durum wheat genotypes using stress indices. The study was laid out in factorial experiments based on a completely randomized design (CRD) with three replications and two factors. Twenty promising durum wheat genotypes were germinated under four (0.0, -0.3, -0.6 3 and -0.9 MPa) osmotic stresses conditions produced using different concentrations of Polyethylene glycol (PEG 6000) at 20 ˚C. The results showed that the effect of osmotic stress on the germination stress index (GSI) was highly significant ($P < 0.01$) and increasing osmotic stress significantly decreased the GSI values. In terms of the germination stress tolerance index (GSTI), a comparison of the different genotype responses to osmotic stress based on root length, root dry weight and seedling dry weight, showed that genotype number 4 (RASCON_39/TILO_1) was most tolerant under low osmotic stress (-0.3MPa) while G10 (RASCON_37/BEJAH_7) exhibited the highest GSTI under severe osmotic stress (-0.9 MPa) conditions. On the other hand genotypes G17 (GARAVITO_3/RASCON_37//GREEN_8) and G7 (HAI-OU_17/GREEN_38) showed the lowest GSTI under osmotic stress conditions. These results indicate that genotypes number 10 and 4 may be suitable for planting in arid and semi-arid areas that are subjected to severe or mild drought stresses.

**Key words:** Germination, seedlings, osmotic stress, durum wheat, drought tolerance

## INTRODUCTION

One of the important challenges facing crop physiologists and agronomists is understanding and overcoming the major abiotic stresses in agriculture which reduces crop productivity and yield. One of these, particularly predominant in arid and semi-arid regions is drought stress, which brings about a decrease in plant growth and development and as a result crop yield. Begg & Turner [4] and Ashraf *et. al.* [3] have suggested that development of drought tolerant varieties can be a useful approach to increase crop production and yield under water stress conditions. As such the release of drought tolerance genotypes, including desirable traits associated with water limitation has become an established applied method for developing cultivars under dry conditions, Izanloo *et. al.*[12].

Varietals and genotype differences in drought tolerance have been reported previously in wheat and several other crops, Kulshrestha and Jain [14], Steiner *et. al.*[22]. Recently the results of Radhouane [18] showed that genotypes with longer root length under water stress conditions are able to access deeper water in the soil. He suggested that the increase in root length was an adaptive response. Several researchers have reported the relationships between water stress with drought tolerance using drought indices in different cereals such as durum wheat, Fernandez [8], Arzani [2], Golabadi *et. al.*[10, 11], bread wheat, Ghodsi [9] and triticale, Nazeri [16].

The objective if this study is to identify drought tolerant genotypes under different levels of osmotic stress conditions using drought stress indices. In addition to this the germination sensitivity thresholds of the promising durum wheat genotypes were also determined. As seed germination is considered to be the most critical growth stage especially, under water stress conditions for the successful stand establishment of crop plants it was used and determined in this study.

## MATERIALS AND METHODS

The experiments were carried out in the Institute of Biological Sciences, Faculty of Science, and University of Malaya. It was laid out in factorial experiments based on a completely randomized design

**Corresponding Author:** Ali Akbar Moayedi, Institute of Biological Sciences, University of Malaya, 50603, Kuala Lumpur, Malaysia
E-mail: moayediali@yahoo.com
phone: +60 17-233-6782

(CRD) with three replications and two factors. The first factor studied was osmotic stress at four levels, i.e. 0.0 MPa (D1, distilled water, control), -0.3MPa (D2), -0.6 MPa (D3) and -0.9 MPa (D4). The second factor was the promising durum wheat genotypes. The seeds (Table 1) of the various durum wheat genotypes were obtained from the elite durum yield trial (EDYT) of 2006-2007, carried out in the Seed and Plant Improvement Institute, Iran.

Osmotic potentials (-0.3, -0.6, and -0.9 MPa) were produced using different concentrations of polyethylene glycol 6000 (PEG) at 20 ˚C according to the method of Michel and Kaufmann[15]. The seeds were germinated using the paper method, in 9 cm diameter Petri dishes on the top of filter papers. Twenty healthy and equal-sized seeds of each genotype were selected and then sterilized with 5% sodium hypochlorite solution for three seconds before the seeds were put in covered sterilized Petri dishes containing germination paper moistened with 8 ml of the different solutions of PEG-6000. The Petri dishes were kept in an incubator for 8 days at 20 ± 0.5 ˚C (Rehman *et. al.* [21] and Ghodsi [9]. Data were recorded daily for 8 days. For germination purposes, only those seeds that presented approximately 2mm of root length were considered to have germinated and were used for germination percentage and rate calculations, Sapra *et. al.* [21] and Afzal *et. al.*[1]. The numbers of seeds germinated were counted daily and the germination percentage and rate were estimated. Mean germination time (MGT) was calculated to assess the germination rate (GR) according to results of Ellis and Roberts [7] and Sapra *et. al.*[21]. At the end of eighth day, 5 seedlings were randomly selected and the coleoptiles root, shoot and also seedling length measured. Additionally, root, shoot and seedling dry weight were measured after drying samples at 76 ˚C for 48 hours in an oven. As according to the Bouslama and Schapaugh [5] formula, the germination stress index (GSI) was calculated as follows:

$$GSI = (PISS / PICS) \times 100$$

In this formula, PISS is the promptness index of stressed seed while the PICS is the promptness index of control seed. The promptness index (PI) was calculated as:

$$PI = nd2\ (1.00) + nd4\ (0.80) + nd6\ (0.60) + nd8\ (0.40)$$

Where, nd2, nd4, nd6 and nd8 are germination percentages on the second, fourth, sixth and eighth day, consecutively. Stress tolerance index (STI) was calculated during the germination stage using the Fernandez [8] formula as follows:

$$\mathbf{GSTI = (Yp \times Ys) / (\bar{Y}^2_p)}$$

In this formula, yield potential (Yp) and yield stress (Ys) shows the value of each genotype under normal and the stress conditions. $\bar{Y}^2 p$ is the mean square of the considerate trait for all genotypes under normal and stress conditions. The data were statistically analyzed by MSTAT-C software package and comparative analyses of the means were performed by Duncan's Multiple Range Test ($P < 0.05$ and $P < 0.01$).

## RESULTS AND DISCUSSION

**Germination Stress Index (GSI):** The germination stress index (GSI) shows the germination rate under osmotic stress to normal conditions ratio. The results indicated that with increasing osmotic stress, the germination stress index (GSI) gradually decreased from the D2 to D4 treatments. Thus the reduction of the GSI value from 92.97 in the D2 treatment to 31.41 in the D4 treatment was related to a similar decrease in germination percentage and rate, under the osmotic stress conditions (Figs.1 and 2). It has been suggested that the germination stress index also indicates the sensitivity threshold of the cultivars and genotypes to drought stress during the germination stage Nazeri [16] and Ghodsi, [9]. The results of the germination percentage and rate showed that there were no significant difference between the D1 (distilled water) and the D2 (-0.3MPa) treatments. While, with increasing osmotic stress the germination percentage, germination rate and germination stress index significantly decreased in the D3 and D4 treatments (Figs.1 and 2). From this we can conclude that the -0.6 MPa treatment (D3) can be the germination sensitivity threshold in these durum wheat genotypes studied. Similarly Nazeri [16] and Ghodsi [9] in separate experiments, reported that -0.6 MPa and -0.9 MPa osmotic stress levels are the germination sensitivity threshold for triticale and bread wheat cultivars, respectively.

**Germination Stress Tolerance Index (GSTI):** The stress tolerance index, at the germination stage, has also been used to investigate drought stress tolerance in durum and bread wheat genotypes, Fernandez [8]. With regard to this, Dhanda *et. al.* [6] and Nazeri [16] suggested that root length, root dry weight and seedling dry weight are the major traits to select for studying tolerant genotypes under water stress conditions. As shown in table 2, a comparison of the genotype responses to germination stress tolerance index, based on root length, root dry weight and seedling dry weight, showed that with increasing osmotic stress GSTI decreased. However, it depended on genotypic difference under osmotic stress conditions. In addition, the G6 and G10 genotypes had the highest GSTI value based on all of the calculated traits under mild (-06MPa) and severe (-0.9MPa) osmotic stress conditions. However the highest GSTI value under lower stress conditions (-0.3MPa) belonged to different genotypes (G4, G10 and G16). Many researchers, such as Sapra [21], Khan *et. al.* [13], Nazeri [16], Dhanda *et. al.* [6], Ghodsi [9], Okçu *et. al.* [17], Rauf *et. al.*[19], Yamur and Kaydan [23] have studied and reported the important and significant relationship between root and seedling dry weight with germination percentage, germination rate, root length, and shoot dry weight in response to the drought tolerance as is the case in the present study. The higher values observed in some of the genotypes (Table 2) can be related to the root to shoot length ratio, where the genotype G10 showed the highest root to shoot length ratio among all the genotypes under severe osmotic stress condition. It supports the results of Radhouane [7] that genotypes exhibiting longer root length under water limitation show an adaptive reaction to increase water uptake ability by the seeds.

**Conclusions:** The overall results of the present study showed that with increasing osmotic stress, the germination stress index decreased significantly. Hence, the highest and lowest value for GSTI was observed in low (-0.3MPa) and severe osmotic stress treatments (-0.9 MPa). With regard to germination rate and the germination stress index, treatment with -0.6 MPa can be the germination sensitivity threshold in the durum and bread wheat genotypes studied. On the other hand, the comparison between the GSI and GSTI values for selection of the tolerant genotypes revealed that the results obtained were considerably similar for both of the studied indices. For this purpose, the genotypes G4 (RASCON_39/TILO_1) and G10 (RASCON_37/BEJAH_7) were the most tolerant genotypes under low and severe osmotic stress. Whereas genotypes G17 (GARAVITO_3/RASCON_37//GREEN_8) and G7 (HAI-OU_17/GREEN_38) exhibited the lowest GSTI value under osmotic stress conditions.

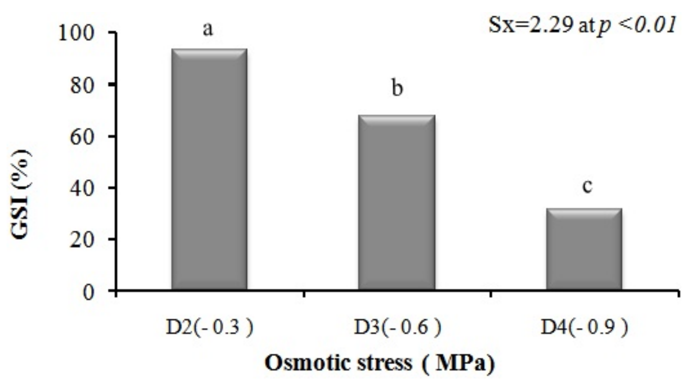


**Fig.1:** The effect of osmotic stress on germination stress index

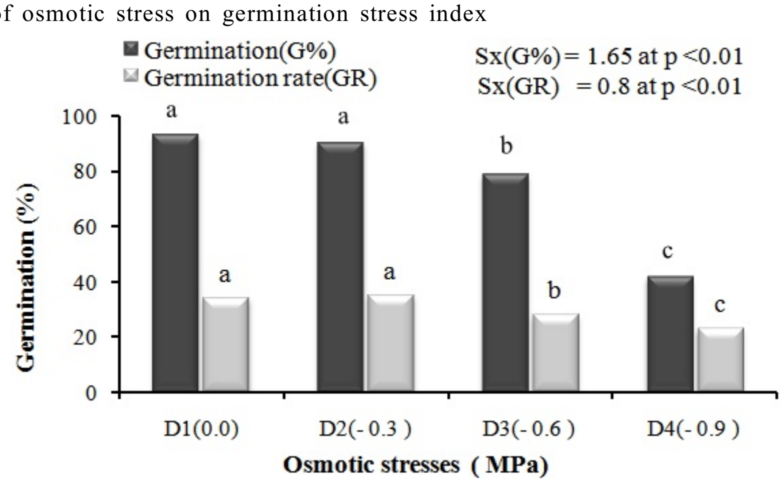


**Fig. 2:** The effect of osmotic stress on germination percentage and rate

**Table1:** List of durum wheat genotypes used in study

| Genotype | Pedigree | Genotype | Pedigree |
|---|---|---|---|
| G1 | ARIA | G11 | GREEN_2/HIMAN_12 |
| G2 | PISHTAZ | G12 | HUI/YAV79//RASCON_37 |
| G3 | STOT//ALTAR 84/ALD | G13 | LIRO_3/LOTAIL_6 |
| G4 | RASCON_39/TILO_1 | G14 | MUSK_1//ACO89/FNFOOT_2 |
| G5 | E90040/MFOWL_13//LOTAIL_6 | G15 | CADO/BOOMER_33 |
| G6 | BRAK_2/AJAIA_2//SOLGA_8 | G16 | PLATA_3//CREX/ALLA/3/LOTAIL_6 |
| G7 | HAI-OU_17/GREEN_38 | G17 | GARAVITO_3/RASCON_37//GREEN_8 |
| G8 | SN TURK MI83-84 375/NIGRIS_5//TANTLO_1 | G18 | BOOMER_18/KITTI_1//LUND_4 |
| G9 | RAFI97/RASCON_37//BEJAH_7 | G19 | CPAN.6018/2*RAJ1555//2*PORRON_4 |
| G10 | RASCON_37/BEJAH_7 | G20 | YDRANASSA30/SILVER_5//SILVER_3/RISSA |

**Table 2:** Response of the durum wheat genotypes to germination stress tolerance index (GSTI) based on different germination traits

| Genotype | GSTI based on root dry weight | | | GSTI based on seedling dry weight | | | GSTI based on root length | | |
|---|---|---|---|---|---|---|---|---|---|
| | D2(-0.3) MPa | D3(-0.6) MPa | D4(-0.9) MPa | D2(-0.3) MPa | D3(-0.6) MPa | D4(-0.9) MPa | D2(-0.3) MPa | D3(-0.6) MPa | D4(-0.9) Mpa |
| G1 | 1.04 | 0.53 | 0.19 | 0.76 | 0.28 | 0.08 | 0.59 | 0.29 | 0.11 |
| G2 | 0.85 | 0.33 | 0.02 | 0.66 | 0.16 | 0.01 | 0.66 | 0.18 | 0.05 |
| G3 | 1.18 | 0.22 | 0.04 | 0.78 | 0.10 | 0.02 | 0.72 | 0.24 | 0.05 |
| G4 | 1.93 | 0.68 | 0.09 | 1.18 | 0.30 | 0.04 | 0.52 | 0.18 | 0.12 |
| G5 | 0.93 | 0.72 | 0.05 | 0.65 | 0.44 | 0.02 | 0.47 | 0.31 | 0.08 |
| G6 | 1.42 | 0.90 | 0.12 | 0.91 | 0.58 | 0.05 | 0.57 | 0.36 | 0.11 |
| G7 | 0.80 | 0.48 | 0.05 | 0.55 | 0.24 | 0.01 | 0.87 | 0.31 | 0.13 |
| G8 | 1.53 | 0.33 | 0.06 | 0.92 | 0.13 | 0.02 | 0.47 | 0.10 | 0.09 |
| G9 | 1.18 | 0.38 | 0.08 | 0.61 | 0.14 | 0.02 | 0.54 | 0.21 | 0.14 |
| G10 | 1.75 | 0.73 | 0.27 | 1.21 | 0.30 | 0.1 | 0.55 | 0.18 | 0.24 |
| G11 | 1.53 | 0.55 | 0.06 | 0.99 | 0.25 | 0.03 | 0.75 | 0.32 | 0.13 |
| G12 | 1.27 | 0.16 | 0.10 | 0.86 | 0.07 | 0.04 | 0.73 | 0.15 | 0.12 |
| G13 | 1.39 | 0.30 | 0.05 | 0.77 | 0.11 | 0.02 | 0.55 | 0.10 | 0.11 |
| G14 | 1.39 | 0.67 | 0.06 | 0.85 | 0.29 | 0.02 | 0.53 | 0.31 | 0.05 |
| G15 | 1.59 | 0.43 | 0.03 | 0.98 | 0.16 | 0.01 | 0.59 | 0.17 | 0.03 |
| G16 | 1.22 | 0.40 | 0.02 | 0.83 | 0.16 | 0.01 | 1.11 | 0.35 | 0.02 |
| G17 | 1.23 | 0.38 | 0.01 | 0.78 | 0.16 | 0.00 | 0.59 | 0.21 | 0.01 |
| G18 | 1.34 | 0.36 | 0.04 | 0.91 | 0.15 | 0.02 | 0.74 | 0.21 | 0.06 |
| G19 | 1.56 | 0.46 | 0.05 | 1.06 | 0.22 | 0.02 | 0.96 | 0.33 | 0.08 |
| G20 | 1.56 | 0.46 | 0.02 | 1.14 | 0.21 | 0.01 | 0.82 | 0.18 | 0.05 |

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