Research Journal of Agronomy

MEDWELL PUBLICATIONS SINCE 2000

Yield, Biochemical Responses and Evaluation of Drought Tolerance of Two Barley Accessions "Ardhaoui" under Deficit Drip Irrigation using Saline Water in Southern Tunisia

[1, 2]Mohamed Bagues, [1]Ikbel Souli1, [1]Faiza Boussora and [1]Kamel Nagaz

[1]*Laboratory of Dryland and Oasis Cropping, Institute of Arid Regions of Medenine, ElFjè, Medenine 4119, Tunisia*

[2]*Faculty of Sciences of Sfax, 3000 Sfax, Tunisia*

Key words: Drought, proline, soluble sugars, minerals, yield, drought tolerance indices and barley

Corresponding Author:
Mohamed Bagues
Laboratory of Dryland and Oasis Cropping, Institute of Arid Regions of Medenine, ElFjè, Medenine 4119, Tunisia

Page No.: 17-23
Volume: 14, Issue 4, 2020
ISSN: 1815-9354
Research Journal of Agronomy

Abstract: In Southern Tunisia, two local barley accessions CV. Ardhaoui; "Bengardeni" and "Karkeni" were cultivated in field under deficit drip irrigation with saline water. Three treatments were used: control or full irrigation T0 (100% ETc) and stressed T1 (75% ETc), T2 (50% ETc). Proline and soluble sugars contents increases significantly under drought between accessions compared to control and varies between growth stages. Moreover, the increasing of Ca^{2+} concentration enhances the absorption of Na^+ ion, consequently K^+/Na^+ decrease significantly between accessions, these results suggest that a high tolerance of Bengardeni accession to drought stress. Therefore, drought tolerance indices (STI, SSI, MP, GMP, YSI and TOL) were used to identify high yielding and drought tolerant between accessions. MP explained the variation of GYi. GMP and STI explained the variation of GYs. The high values of MP, STI and GMP were associated with higher yielding accession. Higher TOL value is associated with significant grain yield reduction in stressed environment suggesting higher stress responses of accessions. Significant positive correlations between MP, STI and GMP and negative between YSI and SSI. MP, STI, GMP and YSI, TOL, SSI are not correlated with each other.

INTRODUCTION

In most African countries, food production was threatened by water scarcity for agricultural crops. Drought is the most serious abiotic stress at global scale[1] and its frequency is expected to increase as a consequence of climate changes[2]. To keep yield stability, it is important to grow plants having increased resistance to drought. Drought and salinity are the two major abiotic efforts limiting crop growth and productivity[3]. In nature, crops are subjected to the combination of different types of stress such as water stress and salinity. Also, soil salinity in large areas has been observed, especially in arid areas[4, 5]. Moreover, drought and salinity also weakened the metabolic mechanisms involved in the assimilation of CO^2 and the combined effects of stomatal closure and metabolic deficiency ultimately reduce the biomass accumulation[6]. Turgor phenomena in the plant cell are generally maintained during drought conditions by osmotic adjustment and this requires the synthesis and transport of compatible solutes and osmolytes, such as soluble sugars, polyols, proline and glycine betaine[7, 8].

Barley is one of the most important cereal crops in Asia, Europe, Middle East and in North and South Africa, even where eventually water scarcity and drought affect crops yielding[9]. As indicated[10], barley productivity depends essentially on water supplies, and it is threatened when the water stress is imposed at the pollination and flowering stages, rather than in the vegetative or seed filling stages[9]. In Tunisia, barley is usually grown in semi-arid and arid regions, this plant is widely cultivated in the region of production of cereals and pasture[11].

Besides, barely is among the most tolerant cereal crops to salinity. It has the ability to produceosmotic adjustments by toxic salt ions invacuoles which keeps a favorable K^+/Na^+ ratio in the cytoplasm at high leaf Na^+ concentrations, to causethe transport of Ca^{2+} ionsto the shoot; to distribute ions between mesophyll and epidermal cells and to trigger antioxidant metabolism[12-15]. However, at higher concentration than 8 dS m^{-1}, salt stress reduces barley growth causing changes to alterations in water and nutrient uptake rates and consequently decreases photosynthesis[16-18]. Nevertheless, despite much effort has been devoted to correlating these changes with the degree of salt adapter in plants, no agreement has been reached among researches[19, 20]. To maintain their growth, plants have developed several strategies when water availability is limited or unpredictable; an adaptation to the lack of water by improving their absorption, accumulation of compatible solutes and mineral nutrients during to maintain turgor, metabolic activity and absorption of water in case of fallsoil waterpotential[21]. Moreover, synthesis of osmo-regulators such as soluble sugars and proline in the event of severe stresses[22, 23].

To select adapted plant to Mediterranean-type climates, it is mandatory to study the phenological, morphological, physiological and agronomic parameters[24, 25] in order to improve plant adaptation to drought. Barley is one of suitable and profitable crops in these conditions[26]. Drought tolerance indices, basedon yield reduction under drought conditions in comparison to normal conditions were defined to provide a measure of drought constraint and to screen the most drought-tolerant genotypes[27].

The objectives of this work is to evaluate the biochemical and agronomic performances of two local barley accessions cultivar Ardhaoui subjected to deficit irrigation at different growth stagesin dryland, to assess the responses of this material to drought stress using drought tolerance indices as tool of selection and to determine the irrigation strategy to improve water productivity of barley in dryland.

MATERIALS AND METHODS

Experimental area: The experiment was conducted in the field at the Institute of Arid Regions is located 22 km southeast of Medenine (10°38'30.34"E, 33°29'53.23" Nalt 106 m). The climate is Mediterranean with hot, dry summers and mild winters an average annual rainfall of 125 mm. Minimum temperatures recorded during the months of November to May, respectively are included between 3.5 and 15.7°C while maximum temperatures are between 16 and 39.8°C in the same period.

Plant material: This study is performed on the cv. Ardhaoui (six rows) the local barley cultivar southern Tunisia known for its drought tolerance. Seeds were collected from two different accessions in Southern Tunisia ("karkeni" from Karkenah and "Bengardeni" from Bengardene) to compare their behavior when grown under conditions of water stress. Cultures were grown in field and irrigated with drip system.

Experimental device: The experimental plan in field consists of 3 blocks with an area of 352 or 117.3 m^2 block. Each block is divided into three equal plots of 39.1 m^2 T_0, T_1 and T_2 correspond, respectively to the irrigation treatments 100, 75 and 50% ETc, each treatment is divided into two sub-blocks contain the two accessions barley A1 and A2, each sub-block contains 6 rows spaced 40 cm (in total there will be 108 lines), the distance between the emitters is of 16 cm. Before sowing, 175 kg ha^{-1} N, 100 kg ha^{-1} P and 125 kg ha^{-1} K were equally distributed for all rows. The seeds are sown at a depth of 2-3 cm and a distance between them of the order of 2.5 cm, the planting density is approximately 230 plants per m^2. All blocks were drip irrigated with water from a will having an ECi of 10.8 dS m^{-1}. Each dripper had a 2 L h^{-1} flow rate. A control mini-valve in the lateral permits use or non-use of the dripper line.

Measurement

Soil salinity and humidity: The final average ECi values (0-60 cm soil depth) under different treatments are presented. Initial soil salinity determined at the time of planting was 11.23 dS m^{-1}. However, soil salinity decrease in deficit irrigation treatments (T_1 and T_2) than full irrigation (T_0). ECi values were in a decreasing order 50>7>100% ETc, also, soil salinity varies between growth stages due to the sourcing of rainfall (24, 48 and 49 mm) which causes increased soil moisture and consequently decreased salinity.

Proline and soluble sugars determination: The technique used is that of Troll and Lindsley[28]. The principle is the quantification of the reaction by ninhydrin proline spectrophotometric measurement. Reactingis characterized by the appearance of the red color due to the proline-ninhydrin complex. Of each accession and each treatment, 100 mg of fresh material are removedfrom the leaves of the flourished 3rd row from the top. These samples were cut into small pieces and placed in test tubes containing screwed 2 mL Methanol (40%) and heated to boiling in a water bath at 85°C for 60 min. After cooling samples, 1 mL of the extract of each was removed and placed into new tubesto which was added 1 mL of acetic acid and 1 mL of a solution containing 120 mL H_2O, 300 mL acetic acid, 80 mL ortho-phosphoric acid "H_3 PO_4, density 1.7" and 25 mg of ninhydrin. The mixture is heated to boiling for 30 min. The solution turns to red. After cooling, was added 5 mL of toluene in each tube with stirring for two phases, then removed the upper phase in which a pinch of Na_2SO_4 was added toremoving water and the optical density was measured by a spectrophotometer thereafter (Bio-RadSmart SpecTM 3000) at the wavelength of 528 nm.

The content of soluble sugars, 100 mg of fresh material is taken to which is added 5 mL of 80% methanol and the samples were heated in a water bath at 70°C for 30 min. The soluble sugar content was determined by the phenol-sulfuric acid method. The 1 mL is removed from the extract which was added 1 mL of phenol 5% and 5 mL of concentrated sulfuric acid. After stirring and cooling, determine the absorbance in a spectrophotometer at 640 nm. The calibration is performed by glucose solutions with concentrations of 0.05-0.3 mg mL^{-1}.

$$\text{Concentration of soluble sugars}\left(\mu \text{mL mg}^{-}1\ \text{MF}\right) = \alpha * \text{Abs} * 1/\text{Re}$$

Where:

α = Slope of the calibration curve
Re = m/v = 0.1/5
Abs = Read

Yield and yield components: The measured yield componentsagronomic are:average height, dry matter, spike length, tillers number per m^2, spikes number per m^2, grain number per spike, 1000 grains weight and grain yield (q ha^{-1}).

Minerals analysis: Samples were totally dried at 100±5°C to constant weight. Then, 1g of each drysample was incinerated during 4 h at 550°C. Ashes were mixed with 4 mL of ultra pure water and 1 mL of concentrated HCl. The solution was heated until boiling, then filtered and adjusted to 100 mL with ultra pure water. This solution will be used for mineral analysis. All minerals contents were determined with an Atomic Absorption Spectrometry. The contents of sodium (Na), potassium (K) and Calcium (Ca) inthe dry matter plant were calculated as:

$$C\left(\text{mg/100g}\right) = c\ \left(\text{mg}^{-}l\right) * 10\text{-}3 * 50 * 100 * DF$$

where, C is the concentration of mineral (mg/100 g DM), "c" is the concentration of mineral (mg^{-1}), DF is the factor ofdilution and m = 100 is the mass of the extract (g).

Drought tolerance indices

Statistical analysis: All data presented were mean values of each treatments and were taken on three (sugars, proline and minerals) and seven (yield and yield components) replicates. Analyses of variance were carried out using the statistical package SPSS v 20. Relationships between yield and yield components were determined using Pearson's simple correlation test.

RESULTS AND DISCUSSION

Determination of proline content: Drought stress increase significantly proline content for both T_1 (75% ETc), T_2 (50% ETc) treatments compared to control T_0 (100% ETc, full irrigation) for the two barley accessions (Table 1). Furthermore, tailing stage present the high proline levels which increase from 0.085-0.1 mg g^{-1} FW for "Karkeni" accession and from 0.063-0.091 mg g^{-1} FW for"Bengardeni" accession, respectively for T_0 and T_2 treatments. Analysis of variance showed significant difference between the irrigation treatments at $p<0.01$.

For all treatments and during the growth stages the difference between accessions is not significant. At Booting and Heading stages proline content decrease with a highly significant way ($p<0.001$).

Determination ofsoluble sugars content: Soluble sugars content increase significantly between the irrigation treatments for the tow barley accessions ($p<0.001$). Indeed in the tillering stage, the soluble sugars content increase in this order 4.19<4.97 mg g^{-1} FW and 3.76<4.65 mg g^{-1} FW respectively for "Karkeni" and "Bengardeni"for the treatments T_1 (75% ETc) and T_2 (50% ETc), compared with control T_0 (100% ETc). The difference between accessions is not significant (Table 1). Moreover, at booting and heading stages the difference between accessions is significantly at ($p<0.01$). However, at the booting stage, soluble sugars content reduce significantly ($p<0.01$) for the Karkeni accession compared to Bengardeni accession. Our results showed a highly significant ($p<0.001$) between growth stages.

Minerals compostion: Drought stress (T_1 and T_2) and growth stages caused a highly significant ($p<0.001$) increases in Na^+ and Ca^{2+} contents (from

Table 1: Drought tolerance indices

Index	Formula	Reference
Stress Tolerance Index	$STI = [(GYi)\times(GYp)/(G\bar{Y}i)^2]$	Fernandez[29]
Mean Productivity	$MP = (GYi+GYp)/2$	Rosielle and Hamblin[30]
Geometric Mean Productivity	$GMP = [(GYi)\times(GYp)]^{0.5}$	Fernández[29]
Stress Tolerance	$TOL = (GYi-GYp)$	Rosielle and Hamblin[30]
Stress Susceptibility Index	$SSI = [1-(Gyp)/(GYi)]/SI$	Fischer and Maurer[31]
Stress Intensity	$SI = \left[1-\left(G\bar{Y}p\right)/\left(G\bar{Y}p\right)/\left(G\bar{Y}i\right)\right]$	
Yield Stability Index	$YSI = GYp/GYi$	Bouslama and Schapaugh[32]

2182.65-2529.3 mg/100 g DW) for Karkeni accession and (from 1606.5-2075.4 mg/100 g DW) for Bengardeni accession respectively compared with full irrigation (T_0) and affected significantly K^+/Na^+ selectivity ($p<0.001$). The difference is significant between accessions at ($p<0.01$). For all growth stages and under drought stress, K^+ concentration decrease significantly at $p<0.001$ for the tow accessions compared with control (T_0). In fact, at Tillering stage, K^+ content decreased under drought from 2838.79-1606.5 mg^{-1}100 g DW in T_0 and from 2529.3-2075.4 in T_2, compared to full irrigation, respectively for T_0 and T_2 treatments. With drought stress T_1 and T_2 K^+/Na^+ ratio decreased (from 1.515-1.08 mg/100 g DW) in the same stage. At booting stage, the Bengardeni accession represented the high levels compared to Karkeni accession for the Na^+ contents ($p<0.01$).

Yield and yield components: To analyze the effect of full a nd deficit irrigation treatments on the final yield, eight criteria were retained: average height, tiller number, spike length, total dry matter, grain yield, spike number m^2, seed number/spike and 1000 grains weight.

Final average height was less significantly in the T_1 and T_2 than T_0 treatments also between accessions $p<0.001$. However, dry matter production was not significantly affected by irrigation treatment but we see a highly significant difference between the accessions $p<0.001$. Therefore, increasing water stress caused a significantly reduction in the tiller number and a subsequently reduction on the number of spike per plant. Spike length, seed number per spike and 1000 grains weight decreased significantly between the two accessions in the T_1 and T_2 treatments compared to full treatment T_0. Consequently, barley grain yield shows a highly significant between all irrigation treatments $p<0.001$. Furthermore, we noticed that grain yield was positively correlated to spike length (r = 0.784**), seed number per spike (r = 0.836**) and 1000 grains weight (r = 0.881**) at $p<0.01$.

Under irrigation treatments, Duncan test is made each accession apart (groups A, B and C for Karkeni accession and groups a, b and c for Bengardeni accession). A significant positive correlation between GYi and SSI, STI, MP, GMP and TOL, YSI showed high negative correlation. Similar results were observed with GYs against STI, GMP, YSI and MP but SSI and TOL were highly negative correlated to GYs. MP was the strongly correlated index to GYi and highly correlated to GYs; whilst STI and GMP were the highest correlated to GYs and highly correlated to Gyi.

In our study in addition to water deficit, the experiment was conducted in field and in saline conditions (initial soil salinity ECi = 11.23 dS m^{-1}, salinity of irrigation water ECi = 10.8 dS m^{-1}), so the plants that grow on this type of soils are often subjected to salinity[33, 34]. Indeed, our results show that the water deficit responses triggers biochemical and agronomic changes on barley plantsand at the same time promotes adaptations mechanisms to ensure the survival and productivity of plants in serious conditions[35]. However, ECi values increases under drought (T_1 and T_2) than control T0 in all growth stages in a order 50>75> 100% ETc, this increasing it may be attributed to little leaching of the soil expected under drought[36]. Accumulation of osmo-regulators such as proline and soluble sugars facilate osmo-regulation under drought and salinity has been observed in plants[37]. In present study, proline and soluble sugars increases under irrigation treatments and varies between accessions during the growth stages studied, this augmentation enable plants to tolerate drought[38]. Proline and soluble sugars contents presents the higher levels at Tillering than booting and heading stages, this variation can be explained by rainfall and by physiological mechanisms of plants[39].

Drought effect increase significantly Ca^{2+} concentration between barley accessions and irrigation treatments, it may be assumed that maintaining higher translocation of Ca^{2+} is an important way to reduce drought stress or beneficial to improve plant tolerance to drought. Increase in Na^+ion and decrease in K^+ ion concentration uptake interrupted ionic balance as observed in the tow accessions under all treatments[40, 41]. The accumulation of Na^+ ion was the expense of the K^+ absorption (r = -0.013), K^+/Na^+ ratio decreased significantly ($p<0.01$) between barley accessions, it is more important in Bengardeni accession than Karkeni under drought treatments. The high K^+/Na^+ ratio indicate that the Bengardeni accession is less susceptible than Karkeni accession to drought under saline conditions. These results are similar find that by Wu *et al*.[42] working on Tibetan wild and cultivated barley.

In addition, drought stress showed a highly significant difference between barley accessions in yield and yield components ($p<0.001$). In fact, a highly significant decrease on plant height, dry matter, grains per spike and 1000 grains weight in the two barley accessions. A significant positive correlation observed between tiller number ($r = 0.561^{**}$), number of spikes per plant ($r = 0.960^{**}$), spike length ($r = 0.784^{**}$), number of seed per spike ($r = 0.881^{**}$) and grain yield. For both accessions, drought stress affected high significantly ($p<0.001$) grain yield.

Drought tolerance indices such as MP, STI and GMP to be the best predicates for both conditions. SSI and TOL showed disparity against GYi and Gyp indicating the population segregated for genes conditioning yield potential and drought resistance. Mardeh *et al*.[43] suggested that these traits can contribute to increase yield under stress and reduce stress susceptibility. Golabadi *et al*.[44] reported that selection for TOL will be worthwhile only when the target environment is no-drought stressed. STI and GMP were not correlated to YSI, SSI and TOL. Low correlation between STI, GMP and MP against YSI, TOL and SSI suggest that each index may be a potential indicator of different biological response to drought[45]. Golabadi *et al*.[44] leading to similar results, stated that combination is biologically attainable in wheat, thereby combining traits that associate with each index. Thus, accession that have high STI, MP and GMP and low SSI were suited for both irrigated and stressed environments. This is the case of Karkeni accession.

CONCLUSION

The main conclusions of this research is in the one hand, to understand the biochemical and agronomic behaviors of two local barley accessions under deficit drip irrigation using saline water and in the other hand to compare their performance against the water stress in order to select the best adaptive accession to drought. However, Karkeni accession is less susceptible to water stress than Bengardeni as shown by their relatively high proline and soluble sugars contents. Moreover, drought tolerance of Karkeni accession was found to be associated with a lower selectivity K^+/Na^+ ratio. Furthermore, drought tolerance indices in grain yield showed a genotypic variation between accessions: Karkeni accession is the less susceptible to drought and the most productive than Bengardeni under three water regimes, also, it can be concluded that the full irrigation (100-ETc) strategies offer significant advantage for yield and reduce the build-up of salinity compared to the (50-ETc) irrigation practices in barley production under arid conditions. As a result of this research, full irrigation 100-ETc is recommended for irrigation of barley crop under the carried climate of southern Tunisia.

ACKNOWLEDGEMENT

We are very grateful for all staff of “Laboratory of Dryland an Oasis Cropping, Arid land Institue, Medenine, Tunisia”, especially Tebra TRIKI and Belgacem LACHIHEB two laboratory technicians for all help during experiments.

REFERENCES

01. Pennisi, E., 2008. Plant genetics: The blue revolution, drop-by-drop, gene by gene. Science, 320: 171-173.
02. Ceccarelli, S., S. Grando, M. Maatougui, M. Michael and M. Slash *et al*., 2010. Plant breeding and climate changes. J. Agric. Sci., 148: 627-637.
03. Wang, W., B. Vinocur and A. Altman, 2003. Plant responses to drought, salinity and extreme temperatures: Towards genetic engineering for stress tolerance. Planta, 218: 1-14.
04. Rabhi, M., Z. Barhoumi, R. Ksouri, C. Abdelly and M. Gharsalli, 2007. Interactive effects of salinity and iron deficiency in *Medicago ciliaris*. Comptes Rendus Biol., 330: 779-788.
05. Yousfi, S., H. Mahmoudi, C. Abdelly and M. Gharsalli, 2007. Effect of salt on physiological responses of barley to iron deficiency. Plant Physiol. Biochem., 45: 309-314.
06. Flexas, J., J. Bota, F. Loreto, G. Cornic and T.D. Sharkey, 2004. Diffusive and metabolic limitations to photosynthesis under drought and salinity in C_3 plants. Plant Biol., 6: 269-279.
07. Foyer, C.H., M.H. Valadier, A. Migge and T.W. Becker, 1998. Drought-induced effects on nitrate reductase activity and mRNA and on the coordination of nitrogen and carbon metabolism in maize leaves. Plant Phys., 117: 283-292.
08. Morison, J.I., N.R. Baker, P.M. Mullineaux and W.J Davies, 2008. Improving water use in crop production. Philo. Trans. R. Soc. London B Biol. Sci., 12: 639-658.
09. Ceccarelli, S., S. Grando and M. Baum, 2007. Participatory plant breeding in water-limited environments. Exp. Agric., 43: 411-435.
10. Shakhatreh, Y., O. Kafawin, S. Ceccarelli and H. Saoub, 2001. Selection of barley lines for drought tolerance in low-rainfall areas. J. Agron. Crop Sci., 186: 186-187.
11. Oueslati, O., M. Ben Hammouda, M.H. Ghorbal, M. Guezzah and R.J. Kremer, 2005. Barley autotoxicity as influenced by varietal and seasonal variation. J. Agron. Crop Sci., 191: 249-254.

12. Fricke, W., R.A. Leigh and A.D. Tomos, 1996. The intercellular distribution of vacuolar solutes in the epidermis and mesophyll of barley leaves changes in response to NaCl. J. Exp. Bot., 47: 1413-1426.
13. Munns, R., R.A. James and A. Lauchli, 2006. Approaches to increasing the salt tolerance of wheat and other cereals. J. Exp. Bot., 57: 1025-1043.
14. Perez Lopez, U., A. Robredo, M. Lacuesta, C. Sgherri, A. Mena Petite, F. Navari Izzo and A. Munoz Rueda, 2010. Lipoic acid and redox status in barley plants subjected to salinity and elevated CO_2. Physiol. Plant., 139: 256-268.
15. Perez Lopez, U., A. Robredo, M. Lacuesta, C. Sgherri, A. Munoz Rueda, F. Navari Izzo and A. Mena Petite, 2009. The oxidative stress caused by salinity in two barley cultivars is mitigated by elevated CO_2. Physiol. Plant., 135: 29-42.
16. Perez-Lopez, U., A. Robredo, J. Miranda-Apodaca, M. Lacuesta, A. Munoz-Rueda and A. Mena-Petite, 2013a. Carbon dioxide enrichment moderates salinity-induced effects on nitrogen acquisition and assimilation and their impact on growth in barley plants. Environ. Exp. Bot., 87: 148-158.
17. Perez-Lopez, U., A. Robredo, M. Lacuesta, A. Mena-Petite and A. Munoz-Rueda, 2012. Elevated CO_2 reduces stomatal and metabolic limitations on photosynthesis caused by salinity in *Hordeum vulgare*. Photosyn. Res., 111: 269-283.
18. Perez-Lopez, U., J. Miranda-Apodaca, A. Mena-Petite and A. Munoz-Rueda, 2013b. Barley growth and its underlying components are affected by elevated CO_2 and salt concentration. J. Plant Growth Regul., 32: 732-744.
19. Chen, Z., M. Zhou, I. Newman, N. Mendham, G. Zhang and S. Shabala, 2007. Potassium and sodium relations in salinised barley tissues as a basis of differential salt tolerance. Funct. Plant Biol., 34: 150-162.
20. Genc, Y., G.K. McDonald and M. Tester, 2007. Reassessment of tissue Na^+ concentration as a criterion for salinity tolerance in bread wheat. Plant Cell Environ., 30: 1486-1498.
21. Hummel, I., F. Pantin, R. Sulpice, M. Piques and G. Rolland *et al*., 2010. Arabidopsis plants acclimate to water deficit at low cost through changes of carbon usage: An integrated perspective using growth, metabolite, enzyme and gene expression analysis. Plant Physiol., 154: 357-372.
22. Ashraf, M. and M.R. Foolad, 2007. Roles of glycine betaine and proline in improving plant abiotic stress resistance. Environ. Exp. Bot., 59: 206-216.
23. Delauney, A.J. and D.P.S. Verma, 1993. Proline biosynthesis and osmoregulation in plants. Plant J., 4: 215-223.
24. Gonzalez, A. and L. Ayerbe, 2010. Effect of terminal water stress on leaf epicuticular wax load, residual transpiration and grain yield in barley. Euphytica, 172: 341-349.
25. Gonzalez, A., I. Martin and L. Ayerbe, 1999. Barley yield in water stress conditions. The influence of precocity, osmotic adjustment and stomatal conductance. Field Crop Res., 62: 23-34.
26. Gonzalez, A., I. Martin and L. Ayerbe, 2007. Response of barley genotypes to terminal soil moisture stress: Phenology, growth and yield. Aust. J. Agric. Res., 58: 29-37.
27. Mitra, J., 2001. Genetics and genetic improvement of drought resistance in crop plants. Curr. Sci., 80: 758-763.
28. Troll, W. and J. Lindsley, 1955. A photometric method for the determination of proline. J. Biol. Chem., 215: 655-660.
29. Fernandez, G.C.J., 1992. Effective Selection Criteria for Assessing Stress Tolerance. In: Proceedings of the International Symposium Adaptation of Vegetables and Other Food Crops in Temperature and Water Stress, Kuo, C.G. (Ed.)., Food Crops in Temperature and Water Stress Publication, Tainan, Taiwan.
30. Rosielle, A.A. and J. Hamblin, 1981. Theoretical aspects of selection for yield in stress and non-stress environment. Crop Sci., 21: 943-946.
31. Fischer, R.A. and R. Maurer, 1978. Drought resistance in spring wheat cultivars. I. Grain yield responses. Aust. J. Agric. Res., 29: 897-912.
32. Bouslama, M. and W.T. Schapaugh, 1984. Stress tolerance in soybeans. I. Evaluation of three screening techniques for heat and drought tolerance. Crop Sci., 24: 933-937.
33. Abbas, G., M. Saqib, J. Akhtar and M.A.U. Haq, 2015. Interactive effects of salinity and iron deficiency on different rice genotypes. J. Plant Nutr. Soil Sc., 178: 306-311.
34. Biswas, A. and A. Biswas, 2014. Comprehensive approaches in rehabilitating salt affected soils: A review on Indian perspective. Open Trans. Geosci., 1: 13-24.
35. Bartels, D. and R. Sunkar, 2005. Drought and salt tolerance in plants. Crit. Rev. Plant Sci., 24: 23-58.
36. Schoups, G., J.W. Hopmans, C.A. Young, J.A. Vrugt, W.W. Wallender, K.K. Tanji and S. Panday, 2005. Sustainability of irrigated agriculture in the *San joaquin* valley, California. Proc. Natl. Acad Sci. USA., 102: 15352-15356.
37. Trovato, M., R. Mattioli and P. Costantino, 2008. Multiple roles of proline in plant stress tolerance and development. Rend. Lincei, 19: 325-346.

38. Kocsy, G., R. Laurie, G. Szalai, V. Szilagyi, L. Simon-Sarkadi, G. Galiba and J.A. de Ronde, 2005. Genetic manipulation of proline levels affects antioxidants in soybean subjected to simultaneous drought and heat stresses. Physiol. Plant., 124: 227-235.
39. Szira, F., A.F. Balint, A. Borner and G. Galiba, 2008. Evaluation of drought-related traits and screening methods at different developmental stages in spring barley. J. Agron. Crop Sci., 194: 334-342.
40. Ahmed, I.M., H.X. Dai, W. Zheng, F.B. Cao, G.P. Zhang, D.F. Sun and F.B. Wu, 2013. Genotypic differences in physiological characteristics in the tolerance to drought and salinity combined stress between Tibetan wild and cultivated barley. Plant Physiol. Biochem., 63: 49-60.
41. Qiu, L., D. Wu, S. Ali, S. Cai and F. Dai *et al*., 2011. Evaluation of salinity tolerance and analysis of allelic function of HvHKT1 and HvHKT2 in Tibetan wild barley. Theor. Applied Genet., 122: 695-703.
42. Wu, D., L. Qiu, L. Xu, L. Ye and M. Chen *et al*., 2011. Genetic variation of HvCBF genes and their association with salinity tolerance in Tibetan annual wild barley. PLoS One, Vol. 6, No. 7. 10.1371/journal.pone.0022938
43. Mardeh, A.S.S., A. Ahmadi, K. Poustini and V. Mohammadi, 2006. Evaluation of drought resistance indices under various environmental conditions. Field Crop Res., 98: 222-229.
44. Golabadi, M., A. Arzani and S.A.M.M. Maibody, 2006. Assessment of drought tolerance in segregating populations in durum wheat. Afr. Agric. J. Res., 1: 162-171.
45. Hassanzadeh, M., A. Asghari, S. Jamaati-e-Somarin, M. Saeidi, R. Zabihi-e-Mahmoodabad and S. Hokmalipour, 2009. Effects of water deficit on drought tolerance indices of sesame (*Sesamum indicum* L.) genotypes in moghan region. Res. J. Environ. Sci., 3: 116-121.