

Study of Some Physiological Responses of Drought Stress in Hexaploid and Tetraploid Wheat Genotypes in Iran

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Abstract: Drought stress is the most significant factor restricting plant production on majority of agricultural fields of the world. Wheat is usually grown on arid-agricultural fields and drought stress was assessed in terms of relative water content (RWC), chlorophyll (Chl) and carotenoids (Car) contents and membrane stability index (MSI) physiological responses were studied under water stress by keeping water supply during parenthesis phase at 50, 60 and 70 days after sowing (DAS) and samples were collected at 60, 70 and 80 DAS. Irrigated and fully turgid plants were used as a control. The plant materials used in this experiment were Hexaploids (*Triticum aestivum*) Bezostaja (drought susceptible) and Cascogne (drought tolerance) and 2 Tetraploids (*Triticum durum*) Boeuffi and Leucurum significant reduction were observed in RWC, MSI and Chl contents of all the cultivars under water stress. Of these, Bezostaja showed the highest RWC, Chl and Car under irrigation condition and Leucurum showed the lowest. Compared with tetraploid wheat's, hexaploid wheat's were found to possess the highest total Chl content at all the stages under irrigation condition. While the cultivars studied showed no significant difference at any stage in their Car contents under irrigation condition, Boeuffi showed the lowest Car content under stress condition.

Key words: Carotenoids content • Chlorophyll content • Membrane stability • Water content

INTRODUCTION

Dryness of the most important factor limiting production of crops including wheat in the world and Iran. This topic is more important in dry and semi-arid regions of the world [1]. Plants are exposed to numerous stress factors during their life, which have a significant effect on the growth of plants. Biotic (pathogen, competition with other organisms) and abiotic (drought, salinity, radiation, high temperature or freezing etc.) stress caused changes in normal physiological function of all plants, including economically important cereals as well. All these stresses reduced biosynthetic capacity of plants and might cause some destructive damages on plants [2]. Drought stress has the largest percentage (26%) when the usable areas on the earth are classified in view of stress factors. It is followed by mineral stress with 20% part, cold and freezing stress with 15% part. While the other stress gets 29% part whereas only 10% area is one exposed to any stress factor [3]. Therefore, drought stress is the most widespread environmental stress, which affects growing and productivity, it induces many physiological, biochemical and molecular responses on plants, so that

plants are able to develop tolerance mechanisms which will provide to be adapted to limited environmental conditions [4].

Wheat is a widely adapted crop, providing one-third of the world population with more than half of their calories and nearly half of the protein. Wheat is mainly grown on rainfed land and about 35% of the area of developing countries consists of semiarid environments in which available moisture constitutes a primary constraint on wheat production. Climatic variability in these marginal environments causes large annual fluctuations in yield. Selection of wheat genotypes with better adaptation to drought stress should increase the productivity of rainfed wheat [5]. Improvement of wheat productivity for this abiotic stress is therefore an important objective of research. Because of their better adaptation under hot and arid regions, *Triticum durum* wheat is usually regarded as more tolerant to stress conditions than hexaploid wheat [6]. In addition among crop plants, durum wheat, which is often grown in water limited conditions, is an attractive study system due to the natural genetic variation in traits related to drought tolerance.

However, the physiological basis of their stress tolerance is not well understood. An understanding of how plants respond to water deficits and in certain instances are able to tolerate them should lead us eventually to ways of optimizing plant productivity in marginal environments [7]. According to the previous studies, there is a link between various physiological responses of crop plants to drought and their tolerance mechanisms such as high relative water content and water potential [8, 9] membrane stability [10, 11] and pigment content stability under stress [11, 12]. On the other hand, reports concerning variation in these physiological parameters on genotypic basis or owing to of ploidy levels are very rare. Zaeifyzadeh and goliov[12] studied the relationship between genotype and environmental conditions (dry and normal) on the amount of chlorophyll content and the amount of super oxide Dismutase reported that drought-resistant cultivars increase Dismutase Super oxide stress increases but in susceptible cultivars decrease chlorophyll Super oxide Dismutase showed. Also a good variety between the native masses of durum wheat in North-West Iran and Azerbaijan in terms of drought resistance and SRAP (Sequence related amplified polymorphism) but did not found any significant relationship between coefficient of drought tolerance and SRAP. The present study was undertaken to assess the selection criteria for identifying drought tolerance in durum wheat genotypes, so that suitable genotypes can be recommended for cultivation in the drought prone area of Iran and Ardabil.

In conclusion we aimed to investigate drought resistance of two hexaploid wheat commonly grown in Iran(Ardabil) and the drought resistance of which was only determined by agronomic observation and two tetraploid wheat which should be of the ability of resisting drought on account of its genetic structure according to above mentioned characters.

MATERIALS AND METHODS

The present study was conducted during the winter season of 2008-2009 under pot-culture conditionals. The experiment was laid out in randomized complete (RCD). physiological responses were studied under water stress by keeping water supply during parenthesis phase at 50, 60 and 70 days after sowing (DAS) and samples were collected at 60, 70 and 80 DAS. Irrigated and fully turgid plants were used as a control. The plant materials used in this experiment were Hexaploids (*Triticum aestivum*) Cascogne (drought susceptible) and Bezostaja (drought tolerance) and 2 Tetraploids (*Triticum durum*) Leucurum

and Boeuffi. Earthen pots 40x40 cm in size was filled with clay-loam soil and farm yard manure in 6:1 ratio. Each pot was fertilized equivalent to 120, 60 and 60 kg ha⁻¹ of N, P and K, respectively. 6 seedlings were formed for each kind in every kind whereas totally 180 pots in investigation period and trial randomized plots was construed according to trial leaf samples were randomized taken from these pots. It was determined that while moisture content in irrigated stage of pot soil ranged from 36 to 39.2% moisture content to which drought stress was applied was 11.2 to 13.2% Elmetron PWT 101 was used in the evaluation of soil moisture content. Plants were collected in quadruplicate from control and stressed plants between 9.00 am to 11.00 am from the first fully expanded leaf. The response of drought stress was assessed in term of relative water content (RWS), chlorophyll (Chl) and carotenoid (Cart) content and membrane stability (MSI).

RWS was calculated according to the method of Weatherly [14]. Leaf samples (0.5 g) were saturated in 100 ml of water for 4 h and their turgid weights were recorded. Then, they were rolled in a dried butter paper at oven dried at 65.c for 48 h and their dry weights were recorded RWC was calculated as:

$$RWC = \left[\frac{(\text{fresh weight} - \text{dry weight})}{(\text{turgid weight} - \text{dry weight})} \right] \times 100$$

Leaf membrane stability index (MSI) was determined according to the method of Premachandra *et al.* [15] as modified by Sairam [16]. Leaf stripes (0.2 g) of uniform size were taken in test tubes containing 10 ml of double distilled water in two tests. Test tubes in one set were kept at 40.c in water bath for 30 min and electrical conductivity of the water containing the sample was measured (C₁) using a conductivity bridge these tubes in the other set incubated at 100°C in the boiling water bath for 15 min and their electrical conductivity was measured as above (C₂). MSI was calculated using the formulae given below:

$$MSI = [1 - C_1/C_2] \times 100$$

Chlorophyll and carotenoids were extracted by the non-maceration method of Hiscox and Israelstam [17]. leaf samples (0.05 g) were incubated in 5 ml of dimethyl sulfoxide (DMSO) at 65°C for 4 h. Absorbance were recorded at 645, 663 and 470 nm and chlorophyll a, b and total chlorophyll were estimated according to Arnon [18] and carotenoids content according to Lichtenthaler and Wellburn [19].

Table1: Effect of water stress on plant height and total dry matter production at harvest in wheat genotypes

Cultivar	Plant height (cm)			Total dry matter (g plant ⁻¹)		
	Irrigated	Stress	Percent decrease	Irrigated	Stress	Percent decrease
Bezostaja-1	84.4	74.5	12.7	67.4	48.4	25
Cascogne	90.3	82.7	9.04	51.9	36.9	24.9
Boeuffi	88	80.1	6.43	43.47	31.5	23.1
Leucurum	86.2	77.3	9.09	47.9	36.7	19.8
Cv	3.7			4.27		
Stress	2.65			4.01		

Water stress was applied to 4 pots of each genotype at parenthesis stage (50 DAS) for 10 days and after final stress period, the pots were irrigated and kept separately for yield analysis. After maturation the plants were harvested from stressed and irrigated pots. Some growth parameters such as plant height and total plant biomass were determined for comparative study and effect of moisture stress on such characteristics (Table 1).

Data of the all parameters were statically analyzed for Spss-16 and differences with $P < 0.05$ were considered significant.

RESULTS

Under irrigated conditions the highest RWC was obtained by hexaploid wheat Bezostaja-1 and Cascogne, whereas under stress conditions RWC in Tetraploids was noted to be higher than that of Hexaploids. RWC in Boeuffi was the highest at 70 and 80 DAS (Fig. 1).

Tetraploid Leucurum and Hexaploid wheat exhibited similar membrane stability index (MSI) values under irrigated conditions but Boeuffi showed a higher percent decline especially at 60 and 70 DAS. MSI values of hexaploid Bezostaja were observed to be the highest under irrigated conditions, while the lowest under stress conditions, followed by Boeuffi. MSI values were similarly high in both hexaploid Cascogne and Leucurum (Fig. 2).

Tetraploid Leucurum and Hexaploids exhibited the highest chlorophyll contents (Chl) under both irrigated and water stress conditions, while among the Tetraploids both Leucurum and Boeuffi had similar values especially at 80 DAS. In this study Bezostaja was found to have the highest Chl content (Fig. 3).

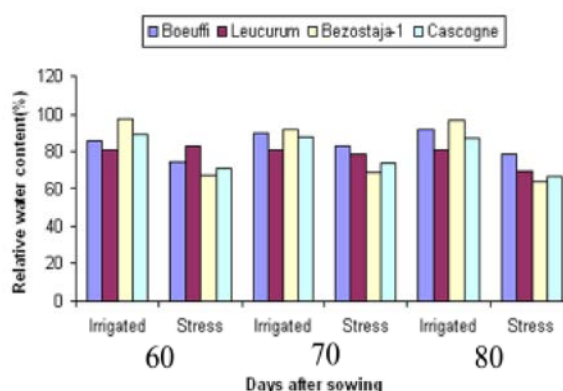


Fig. 1: Effect of water stress on relative water content

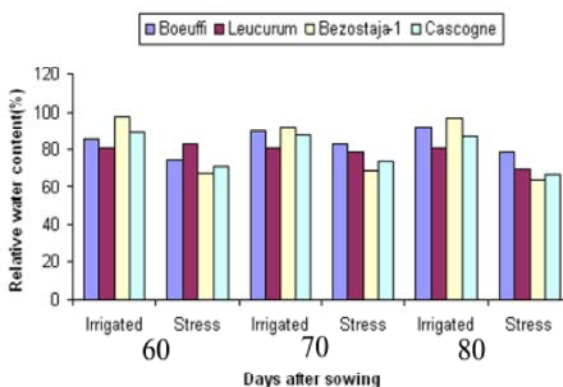


Fig. 2: Membrane stability index in wheat genotypes

The CAR contents of Cascogne at 60 and 70 DAS were observed to be highest under irrigated conditions, while the lowest under water stress conditions. No significant differences were observed between CAR contents of Bezostaja-1 and Cascogne especially 60 and 70 DAS under irrigated conditions. The highest CAR contents at all stages were exhibited by Tetraploid Leucurum under stress conditions (Fig. 4). Tas and Tas [20] reported in their study on Hexaploid and tetraploid wheat cultivars that were reported Hexaploid wheats had the lowest value. The result of this study was similar.

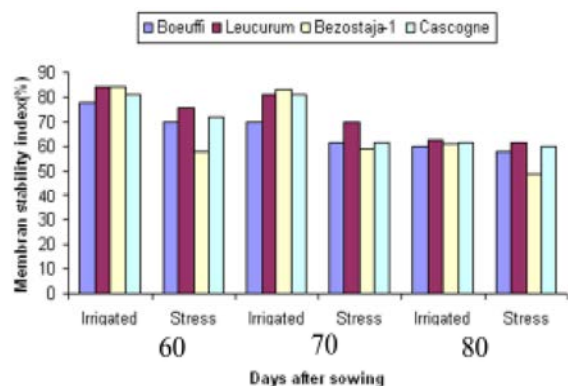


Fig. 3: Chlorophyll content in wheat genotypes

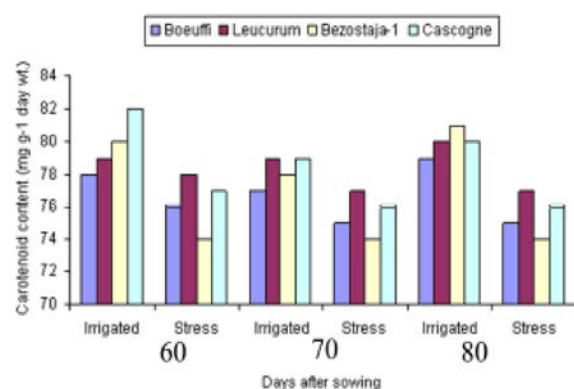


Fig. 4: Carotenoid content in wheat genotypes

DISCUSSION

Considering the results and past results are determined by the resistance of tetraploid wheat is more significant differences in RWC/water potential in tolerant and susceptible genotypes of wheat [12]. The Tetraploid genotypes showed the highest RWC as well as a lower percent decline under stress at all growth stages. The results are in agreement with the findings of Al-Hakimi and Mannoveux [20] who reported that leaf water potential and RWC under drought of Hexaploid was higher than Tetraploids Bezostaja-1 under water stress.

The plasma membrane is generally protected from desiccation-induced damage by the presence of membrane-compatible solutes, such as sugars and amino acids. Therefore, a link may exist between the capacity for osmotic adjustment and the degree of membrane protection from the effect of dehydration [20]. Maintenance of membrane integrity and function under a given level of dehydration stress has been used as a measure of drought tolerance by Premachandra *et al* [15] and Tas and Tas [20], they indicated that the underground part of the plant plays an important role

under drought stress conditions. Decrease in MSI reflects the extent of lipid per oxidation caused by active oxygen species [4]. The best performance of Tetraploid genotypes under water stress in terms of MSI points to their better adaptation under adverse conditions. However, in our study Bezostaja hexaploid wheat had the best result, which is not exactly in consistent with what the researchers found.

Chlorophyll maintenance is essential for photosynthesis under drought stress in tolerant genotype of wheat have also been reported by Sairam *et al* [22] and Kraus *et al* [12]. The results suggested that drought tolerant genotypes Hexaploid Cascogne, Tetraploid Leucurum and Hexaploid Bezostaja showed lower reduction than Tetraploid Boeuffi.

Higher levels of Car under drought tolerant genotypes have also been reported by Sairam *et al* [22] and Kraus *et al* [12]. Carotenoids also have critical role as photoprotective compounds by quenching triplet chlorophyll and singlet oxygen derived from excess light energy, thus limiting membrane damage. This area of carotenoid function has been reviewed extensively elsewhere [20]. As well as their function in photosynthetic tissue, carotenoids play an important role in plant reproduction, thought their role in attracting pollinators and in seed dispersal and are essential components of human's diets. It is thus apparent that drought tolerance of a given genotypes is not limited to a particular physiological character. Carotenoids are responsible for the scavenging of singlet oxygen [20], thus comparatively higher Car levels in Leucurum, Bezostaja and Boeuffi demonstrates their tolerance capacity.

It was determined that of the Tetraploid wheat Leucurum and Hexaploid wheat Bezostaja showed a better performance in view of resistance to drought stress. However if this study is realized on field conditions, it will increase the reliability of the above mentioned results.

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