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PERFORMANCE OF GRAIN YIELD, BIOCHEMICAL AND ANATOMICAL CHARACTERS UNDER WATER STRESS CONDITIONS IN SOME BREAD WHEAT GENOTYPES

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ABSTRACT: A field experiment was carried out during 2013/2014 and 2014/2015 winter growing seasons at the Experimental Farm, Kafr El-Hamam Agricultural Research Station, Zagazig, Sharkia Governorate, Egypt, to evaluate 20 wheat genotypes and cultivars under three irrigation regimes. Plants in the first regime were irrigated four times after planting irrigation (normal irrigation I_1). In the second regime plants were irrigated two times after planting irrigation (I_2). In the third regime plants were given one surface-irrigation 25 days after planting irrigation (I_3). A wide border (7m) was made to minimize the underground water permeability surrounded each experiment. The experiment was laid out in a split-plot design with three replications. Highly significant genotype differences were registered for grain yield and its components in both seasons and combined. The interactions between genotypes and water regime treatments for grain yield and its components were highly significant for all characters in both seasons and combined except for 1000 grain weight in first season. The obtained results showed that I_1 treatment gave the highest water consumptive use and applied irrigation water. Results indicated that values of water consumptive use in the first season were 520.1, 382.6 and 275.3 mm for irrigation regimes I_1 , I_2 and I_3 , respectively. However, in the second season the corresponding values of the water consumptive use were 494.7, 370.9 and 263.4 mm, respectively. Genotype No. 2 gave the least value of water consumptive use, while genotype No. 13 recorded the highest water consumptive use. Water utilization efficiency (kg grains/ m^3 applied water) revealed that I_3 gave the highest value, whereas I_1 was the lowest one. The highest water utilization efficiency was registered by Genotype 8, while genotype 3 recorded the lowest value for this measurement. Drought sensitivity index (DSI) reveal that genotypes No 1, 5, 8, 10, 12, 16 and 18 were tolerant to water stress. Moderate drought stress resulted in significant increase in total soluble sugars, proline and free amino acids in the studied genotypes. Under sever stress, the above mentioned characters significantly increased in genotypes No 1, 2 and 3 compared to other tested genotypes. Therefore wheat genotypes No.1, 2 and 3 could be classified as more tolerant to moderate drought stress. Anatomical features of 5 wheat genotypes were influenced by drought stress. It has been noticed that genotype No. 20 recorded the highest reduction in anatomical characters. On the other hand, the least reduction was detected in genotype No. 1 compared to other tested genotypes. Genotype No. 1 appeared to be more tolerant to drought stress as it exhibited DSI less than unity and gave increase in total soluble sugars, proline and free amino acids with lowest reduction in leaf anatomical characteristics. The study recommend genotype No. 1 as more tolerant to drought stress with good level of yield productivity.

Key words: Wheat, genotypes, drought stress, osmoprotactants, anatomy.

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INTRODUCTION

Wheat is the most important cereal crop all over the world and the main food crop in Egypt as in many other parts of the world. Egypt suffered a considerable gap between its national production and consumption. One of the strategies for narrowing the gap is growing wheat in the newly reclaimed areas. Irrigation water could be considered a limiting factor. So, reducing utilized amount of water will help to solve this problem and the breeders are always looking for germplasm more tolerant to drought tolerance.

The major abiotic stresses like drought, high salinity, cold, and heat negatively effect the survival, biomass production and yield of staple food crops up to 70% (Ahmad *et al.*, 2012). Water deficit is one of the most common environmental stresses that affects growth and development of plants (Bray, 1997). Drought, generally, limited water availability is the main factor limiting crop production (Seghatoleslami *et al.*, 2008).

The wheat growth period most sensitive to drought stress with respect to grain yield is from double ridge to anthesis stage due to the negative impact on number of spikelets and grains per spike (Sphiler and Bulm, 1991). El-Sayed (2003) reported that the irrigation level had a significant effect on the plant height, grain weight/spike and 1000- grain weight. Otherwise, Menshawy *et al.* (2006) found that wheat grain did not significantly decreased by reducing number of irrigation from five to two irrigations in clay soil at North Delta region. Water stress resulted in a shorter grain filling, smaller grains at maturity and an earlier loss of stem height (El-Banna *et al.*, 2002).

Drought resistance is the result of numerous morphological, anatomical and physiological characters, which interact with maintenance of growth and developmental processes under edaphically and climatic conditions (Steponkus *et al.*, 1980). In drought tolerance, plants are able to tolerate the conditions of water deficiency through manipulating the biochemical and physiological parameters and thus avoiding the injurious effects of drought.

One of the potentially important mechanisms of drought tolerance is osmotic adjustment, which can be achieved from the accumulation of compatible solutes (such as amino acids, sugars or sugar alcohols) in protoplasm (Bartels and Sunkar, 2005). The osmotic adjustment allows cell enlargement and plant growth during severe drought stress and allows stomata to remain partially open and CO₂ assimilation to continue during drought stress (Hare *et al.*, 1998). These help the cells to maintain their dehydrated state and the structural integrity of the membranes so as to provide resistance against drought and cellular dehydration (Ramanjulu and Bartels, 2002). The compatible solutes such as proline induced by water stress have been demonstrated to be involved in the sequestration of reactive oxygen species ROS, and hence in protection and/or repairing processes of some molecules and structures damaged by ROS toxicity (Moller *et al.*, 2007).

Anatomical changes induced by water deficits in higher plants are better observed indicators; they can be directly applied to agriculture and handled (Shao *et al.*, 2008). Plant tissues responses to water stress depend on the anatomical characteristics that regulate the transmission of the water stress effect to the cells (Olmos *et al.*, 2007). Tissues exposed to environments with low water availability have generally shown reduction in cell size and increase in vascular tissue and cell wall thickness (Guerfel *et al.*, 2009). Multiple characteristics of vascular structure have been investigated, such as modifications to the wall architecture and alteration of xylem/phloem ratio, which are thought to be involved in the resistance of the plant to environmental stresses (Child *et al.*, 2003).

Our objectives were to 1) compare the performance of agronomic traits of twenty spring wheat genotypes under normal and reduced irrigation, 2) identify genotypes with high yield potential under reduced irrigation, 3) determine the relative tolerance of bread wheat genotypes to drought stress and 4) study the importance of organic osmoprotectants and anatomical characters in relation to water stress.

MATERIALS AND METHODS

Field Experiments

A field experiment was carried out during 2013/2014 and 2014/2015 winter growing seasons at the Experimental Farm, Kafr El-Hamam Agricultural Research Station, Zagazig, Sharkia Governorate, Egypt,

The plant materials for this study comprised 20 bread wheat genotypes of them 16 promising lines and four commercial cultivars. Name and pedigree of these genotypes are shown in Table 1.

The entries were evaluated under three separate irrigation regime experiments. The first regime included plants irrigated four times after planting irrigation as normal irrigation (I_1). Plants in the second regime was irrigated two times after planting irrigation (I_2) and the third one was one surface-irrigation given 25 days after planting irrigation (I_3). A wide border (7m) to minimize the underground water permeability surrounded each treatment.

Entries were grown on 22nd November in both seasons using a split-plot design with three replications for each experiment. The three irrigation regimes were devoted in main plots, meanwhile the genotypes were allotted in sub-plots. The sub-plot consisted of six rows, 3m long and 20 cm apart, thus, the area harvest of each plot was 3.2 m². Seeds were drilled in rows with seeding rate of 350 seeds/ m². Nitrogen fertilizer was added in the form of urea (46% N) at a rate of 75kg N fad⁻¹. All rates of N was added before the first irrigation at tillering. All other culture practices were applied as recommended for wheat cultivation.

Data were collected for the following characters *i.e.*, days to heading, days to maturity, plant height, No. of spikes/m², No. of grains/spike, 1000 grain weight and grain yield (ardab/fad.). In addition to drought sensitivity index (DSI) which calculated according to Fisher and Wood (1979). Analysis of variance was done for each season and combined analysis was computed overall seasons according to Snedecor and Cochran (1990).

Particle size distribution and some soil-water constants of the experimental soil as determined

according to Klute (1986) are shown in Table 2. In addition, the prevailing weather conditions at the experimental site in winter seasons of 2013/2014 and 2014/2015 are listed in Table 3.

The present research trials aiming at evaluating the performance of 20 wheat genotypes exposed to soil moisture stress conditions induced due to different irrigation regimes comparing with the control.

Crop-Water Relationships under Study

Water consumptive use (CU)

Water consumptive use or actual evapotranspiration (ETc) values were calculated for each irrigation using the following formula (Israelson and Hansen, 1962).

$$WCU = \sum_{i=1}^{i=4} \frac{(\theta_2 - \theta_1)}{100} \times Bd \times D$$

Where:

WCU = seasonal water consumptive use (cm),

θ_2 = soil moisture content after irrigation (on mass basis, %),

θ_1 = soil moisture content before irrigation (on mass basis, %),

Bd = soil bulk density (g/cm³),

D = depth of soil layer (15cm each), and

I = number of soil layer.

Soil moisture content was gravimetrically determined in soil samples taken from consecutive depths of 15 cm down to 60 cm. Soil samples were collected just before each irrigation, 48 hours after irrigation and at harvest time.

Applied Irrigation water (AIW)

Submerged orifice with fixed dimensions was used to measure the amount of water applied according to the following equation (Michael, 1978).

$$Q = CA\sqrt{2gh}$$

Where:

Q = discharge through orifice, (cm³sec⁻¹).

C = coefficient of discharge, (0.61).

A = cross-sectional area of the orifice, cm².

Table 1. Pedigree of 20 genotypes of bread wheat

No. Genotype	Pedigree
1 SALE-6	ACHTAR*3//KANZ/KS85-8-4/3/ZEMAMRA-5 ICW01-00135-0AP-1AP-0AP-0AP-7AP-0AP- 0DZ/0AP-0DZ/0KUL/0SIN/0AP-0NJ/0AP-0ALK/0AP
2 HD2206/HORK"S"/3/2/ *NS732/HER//KAUZ"S"	HD2206/HORK"S"/3/2/*NS732/HER//KAUZ"S" ICW01-21075-2AP-12AP-0AP/0TS-0AP-6AP-0AP- 0DZ/0AP-0DZ/0KUL 0SIN/0AP-0NJ/0AP-0ALK/0AP
3 SANOBAR-4	SHA3/SERI//YANG87-142/3/2*TOWPE ICW00-0577-7AP-0AP-0AP-2AP-0AP-0DZ/0AP- 0DZ/0KUL/0SIN/0AP-0NJ/0AP-0ALK/0AP
4 REYNA-23	CHAM- 4/SHUHA 'S'/6/2*SAKER/5/RBS/ANZA/3/KVZ/HYS//YMH/TOB/ 4/BOW'S' ICW00-0634-6AP-0AP-0AP-35AP-0AP-0DZ/0AP- 0DZ/0KUL/0SIN/0AP-0NJ/0AP-0ALK/0AP
5 BOREJ-2	NWT/3/TAST/SPRW//TAW12399.75/4/ROOMY ICW98-0170-5AP-0APS-030AP-20AP-5AP-0AP-0DZ/0AP- 0DZ/0KUL/0SIN/0AP-0NJ/0AP-0ALK/0AP
6 NOUHA-1	NS732/HER//MILAN/SHA7 ICW99-0288-15AP-0AP-0AP-25AP-0AP-0DZ/0AP- 0DZ/0KUL/0SiN/I0AP-0NJ/0AP-0ALK/0AP
7 LOULOU-3	CBME4SA#4/FOW-2 ICW98-0047-1AP-0APS-030AP-1AP-3AP-6AP-0AP-0DZ/0AP- 0DZ/0KUL/0SIN/0AP-0NJ/0AP-0ALK/0AP
8 REYNA-16	CHAM- 4/SHUHA 'S'/6/2*SAKER/5/RBS/ANZA/3/KVZ/HYS//YMH/TOB/ 4/BOW'S' ICW00-0634-6AP-0AP-0AP-35AP-0AP-0DZ/0AP- 0DZ/0KUL/0SIN/0AP-0NJ/0AP-0ALK/0AP
9 DURRA-8	FOW'S'//NS732/HER/3/CHAM-6//GHURAB'S' ICW98-0035-5AP-0AP-S030AP-7AP-5AP-0AP-0DZ/0AP- 0DZ/0KUL/0SIN/0AP-0NJ/0AP-0ALK/0AP
10 NOUHA-3	NS732/HER//MILAN/SHA7 ICW02-00472-13AP/0TS-0AP-0AP-2AP-0AP
11 FIRDOUS-29	GIZA-164/YEBROUD-1//BOOMA-2 ICW02-00099-11AP/0TS-0AP-0AP-4AP-0AP
12 SOONOT-5	SAMAR-8//KAUZ'S'//CHAM-4/SHUHA'S' ICW02-00478-3AP/0TS-0AP-0AP-18AP-0AP
13 FANOOS-14	ANDALIEB-5//TEVEE-1/SHUHA-6 CMSS05B00137T-099TOPY-099M-099Y-099ZTM-11WGY-0B
14 REYNA-13	CHAM- 4/SHUHA 'S'/6/2*SAKER/5/RBS/ANZA/3/KVZ/HYS//YMH/TOB/ 4/BOW'S' CMSS05B00123T-099TOPY-099M-099NJ-6WGY-0B
15 SANOBAR	SHA3/SERI//YANG87-142/3/2*TOWPE CMSS05B00261T-099TOPY-099M-099NJ-099NJ-6WGY-0B
16 RUTH-1	F5 DERIVED Kenya (D.H) F2 CMSS05B00663S-099Y-099M-099Y-099TM-13WGY-0B
17 GEMMMIZA 11	
18 GIZA 168	MN / Bue // SERI
19 SIDS 12	BUC//7C/ALD/5/MAYA74/ON//1160.147/3/BB/GLL/4/CHAT"S"/6 /MAYA/VUL// CMH74A.630/4*SX.
20 SAKHA 93	Sakha 92/LTR 810328

Table 2. Particle size distribution (%) and some soil-water parameters and bulk density of the experimental site (mean of two seasons)

Parameter	Value						
Particle size distribution (%)							
Clay	52.1						
Silt	35.5						
Fine sand	11.4						
Coarse sand	1.0						
Textural class	Clayey						
Soil - water parameters and bulk density							
Soil depth, (cm)	Field capacity (FC)		Wilting Point (WP)		Available water (AW)		Bulk density (Mgm ⁻³)
	(%) (W/W)	(cm) depth	(%) (W/W)	(cm) depth	(%) (W/W)	(cm) depth	
0 - 15	44.2	7.29	21.8	3.60	22.4	3.70	1.10
15 - 30	40.8	7.34	20.3	3.65	20.5	3.69	1.20
30 - 45	36.9	6.92	19.1	3.58	17.8	3.34	1.25
45 - 60	34.7	6.87	18.8	3.72	15.9	3.15	1.32
		∑ 28.42		∑ 14.55		∑ 13.88	

Table 3. Meteorological data in 2013/2014 and 2014/2015 winter seasons*

Month	T.max.	T.min.	WS	RH	SS	SR	RF
	2013/2014						
November	28.2	15.2	3.6	53.2	10.5	326	5.3
December	20.1	8.5	3.8	54.7	10.1	268	21.1
January	20.9	8.5	3.0	58.9	11.0	280	9.8
February	22.5	8.2	3.7	57.5	11.1	354	17.4
March	25.6	10.4	4.0	45.1	11.7	441	13.3
April	30.7	13.6	3.8	40.5	12.0	519	5.7
May	33.8	17.6	4.1	37.1	13.5	585	5.5
	2014/2015						
November	25.4	13.3	3.5	57.5	8.4	432	0.6
December	22.7	10.3	3.2	55.2	9.5	514	21.4
January	18.9	7.1	4.3	53.6	10.5	572	37.3
February	18.3	7.0	3.7	54.8	11.7	354	13.1
March	25.5	10.7	3.8	49.1	11.8	441	1.8
April	29.1	12.0	4.3	44.2	12.8	519	5.5
May	34.1	16.8	3.9	41.7	13.6	585	0.0

* Data were obtained from the agro meteorological unit, Water Requirements and Field Irrigation Res. Dept., SWERI, ARC.

T. max, T. min = maximum and minimum temperatures °C, WS = wind speed (m sec⁻¹), RH = relative humidity (%), SS = actual sunshine duration (h day⁻¹), SR = solar radiation (Cal cm⁻² day⁻¹) and RF = rainfall (mm month⁻¹).

g = acceleration due to gravity, cm/sec.^2 (981 cm/sec.^2).

h = pressure head, causing discharge through the orifice, cm.

Water Productivity

Water productivity is an efficiency term calculated as a ratio of product output over water input. The output could be biological goods such as crop grain, fodder....*etc.* So, water productivity, in the present study, is expressed as kilogram of wheat seed obtained per the unit of applied irrigation water. The water productivity values (kilograms of wheat grains m^{-3} applied water) were calculated as follows:

$\text{WP (kg m}^{-3}\text{)} = \text{grain yield (kg fad}^{-1}\text{)}/\text{applied water (m}^3 \text{fad}^{-1}\text{)}$ (FAO, 2003).

Estimation of total soluble sugars (TSS)

Total soluble sugars were measured in an ethanolic extract of the studied 20 wheat genotypes leaves during the second growing season (2014/2015), using phenol-sulfuric according to the method of Dubois *et al.* (1956).

Estimation of proline

Proline content in wheat genotypes leaves during the second growing season (2014/2015), was determined using the method of Bates *et al.* (1973).

Estimation of total free amino acids (FAA)

Total free amino acids in wheat genotypes leaves during the second growing season (2014/2015), were determined using ninhydrin reagent according to Moore and Stein (1954).

Anatomical Investigation

Anatomical characters were made on samples of five representative wheat genotypes, received four irrigations after planting irrigation (I_1) or received only one irrigation after planting irrigation (I_3). These five wheat genotypes were selected based on their great differences in biochemical and yield characters. Samples were collected from the blades of flag leaves at booting stage through the second growing season (2014/2015). These specimens were cut into pieces of 1.0 cm length, then killed and fixed for 24 hours at least in plant fixative which is known as FAA (formalin acetic alcohol) represented by the following formula: 10 ml. formaldehyde (37- 40%), 5ml. glacial acetic acid, 50 ml. ethyl alcohol (95%) and 35 ml.

distilled water. Then the specimens were washed and dehydrated in ascending concentrations of ethyl alcohol series, then cleared in transferring concentrations of xylene and absolute alcohol. Specimens were embedded in pure paraffin wax of melting point $52\text{-}54^\circ\text{C}$. Sections were prepared using EPMA a rotary microtome at 14 microns. Paraffin ribbons were mounted on slides and sections were stained in safranin and light green. Sections were mounted in Canada balsam (Willey 1971). Selected sections were examined to detect histological manifestations of the chosen treatments using light microscope (Olympus) with digital camera (Canon power shot S80) connected to computer; the photographs were taken by Zoom Browser Ex Program. The dimensions of leaf blade sections were measured by using Corel Draw program ver.11.

RESULTS AND DISCUSSION

Effect of irrigation treatments on earliness characters and plant height are presented in Table 4 which shows that exposing wheat genotypes to water stress caused early heading and maturity in both growing seasons and combined. The earliest heading and maturity were found under I_3 (98.4 and 142.4 days) and I_2 treatments (99.1 and 144.9 days) rather than I_1 treatment which recorded (100.6 and 146.2 days) for days to heading and to maturity in the combined. A decrease in plant height was observed due to reducing the amount of irrigation water, so that the shortest plants were observed in I_3 during the two seasons valued (104.1 and 110.4 cm), respectively. Similar results were recorded by Saleem (2003).

Highly significant differences which recorded among wheat genotypes for the three characters might reflect, partially their different genetic backgrounds. Genotype No. 10 was the earliest for days to heading, while genotype No. 5 found to be the latest one. Moreover, the earliest and latest genotypes for days to maturity were genotypes No. 8 and No.13, respectively. The observed significant variation among the genotypes might reflect partially their different genetic structure. These results are in harmony with those reported by Menshawy (2007) and Gab-Allah (2007). For plant height Genotype No. 18 was the shortest while, genotype No.7 was the tallest one.

Table 4. Effect of irrigation regimes (I₁, I₂ and I₃), genotypes and their interaction on days to heading, days to maturity and plant height for 2013/2014 and 2014/2015 seasons and the combined

Character	Days to heading			Days to maturity			Plant height		
	2013/ 2014	2014/ 2015	Comb.	2013/ 2014	2014/ 2015	Comb.	2013/ 2014	2014/ 2015	Comb.
Irrigation									
I ₁	99.6	101.7	100.6	143.2	149.2	146.2	110.2	124.3	117.2
I ₂	97.9	100.2	99.1	142.2	147.7	144.9	107.8	117.4	112.6
I ₃	97.5	99.3	98.4	138.1	146.6	142.4	104.1	110.4	107.2
F-test	**	**	**	**	**	**	**	**	**
LSD	0.952	0.672	0.484	2.031	0.207	0.248	4.465	3.039	2.243
Genotype									
1	98.4	100.6	99.5	142.6	147.4	145.0	116.7	130.6	123.6
2	97.9	100.4	99.2	142.6	147.1	144.8	119.4	120.6	120.0
3	98.7	100.6	99.6	142.1	148.6	145.3	105.0	121.1	113.1
4	98.9	100.4	99.7	141.1	148.7	144.9	106.7	120.0	113.3
5	100.7	103.8	102.2	139.9	150.2	145.1	106.1	118.3	112.3
6	98.1	100.2	99.2	139.8	147.8	143.8	126.7	111.7	119.2
7	98.2	101.1	99.7	138.8	147.9	143.3	117.2	133.9	125.6
8	98.6	100.8	99.7	140.1	146.3	143.2	106.7	133.3	120.0
9	97.6	100.9	99.2	141.8	147.9	144.8	108.3	117.8	113.1
10	96.7	98.8	97.7	139.9	147.0	143.4	110.6	117.2	113.9
11	98.8	100.1	99.4	140.3	147.0	143.7	106.7	110.6	108.6
12	98.7	99.9	99.3	140.8	148.4	144.6	102.2	122.2	112.2
13	98.8	101.0	99.9	143.1	148.0	145.6	99.4	106.7	103.1
14	99.3	100.2	99.8	142.9	147.3	145.1	105.0	110.0	107.5
15	97.7	98.8	98.2	140.2	147.0	143.6	102.2	107.8	105.0
16	98.3	99.4	98.9	140.9	149.4	145.2	108.9	115.0	111.9
17	98.1	99.6	98.8	141.0	148.2	144.6	106.7	113.3	110.0
18	99.1	101.4	100.3	142.2	146.1	144.2	100.0	105.0	102.5
19	97.0	100.3	98.7	141.7	146.1	143.9	94.4	113.3	103.9
20	96.4	100.1	98.2	141.3	147.9	144.6	98.3	118.9	108.6
F-test	**	**	**	**	**	**	**	**	**
LSD	0.96	1.368	0.827	1.248	1.022	0.298	8.022	5.808	4.902
Irrigation									
I × G	**	NS	**	**	**	**	NS	**	**

I₁ = 4 irrigations (control).I₂ = 2 irrigations.I₃ = 1 irrigation.

Significant interaction between $I \times G$ was registered in all cases, except for days to heading in 2014/2015 season and plant height in 2013/2014 season which was insignificant.

Grain Yield and its Components

Statistical analysis revealed that irrigation treatments had a highly significant effect on grain yield and its components in both seasons and combined (Table 5). The results illustrated significant increase in No. of spikes/m², No. of grains/spike, 1000 grain weight and grain yield (ardab/fad.) by increasing number of irrigation. I₁ treatment recorded higher No. of spikes /m², No. of grains spike⁻¹, 1000 grain weight and grain yield rather than I₂ and I₃ treatments.

Highly significant genotype differences were occurred for grain yield and its components in both seasons and combined (Table 5). These variations among genotypes might be due to their different genetic back grounds. Genotype No. 6 recorded the lowest value for No. of grains spike⁻¹, while genotype No. 15 was the highest one among the studied wheat genotypes.

Genotype No. 13 found to be the least in 1000 grain weight in the two seasons and the combined, while genotype No. 11 was the heaviest for this character.

Also, genotype No. 11 produced the highest value for No. of spikes/m² while genotype No. 4 recorded the lowest value for this character. Genotype No.8 recorded the highest grain yield 22.2 ardab/fad., while genotype No.3 gave the lowest grain yield 16.7 ardab/fad. Also it was reported that grain yield was significantly decreased by decreasing number of irrigations (Table 5). These results are in agreement with those obtained by Menshawy *et al.* (2006) and Gab-Allah (2007).

The interaction between genotype and water treatments for grain yield and its components was significant for all characters in both seasons except for 1000 grain weight in the first one. These results revealed that the genotypes responded differently to water regimes and reflect the possibility of selection the most tolerant genotypes under water stress environments.

Screening drought tolerant genotypes

Water stress consistently lowered the yield of wheat genotypes rather than non-stress conditions. Based on drought sensitivity index (DSI) for grain yield (Table 6) for 2nd and 3rd treatments relative to I₁, genotypes No. 1, 5, 8, 10, 12, 16 and 18 appeared to be more tolerant to drought as they exhibited DSI less than unity. Otherwise, genotypes No. 3, 6, 9, 11 and 15 were sensitive to water stress (DSI >1), furthermore the remaining wheat genotypes were moderate. Similar results were recorded by Richards *et al.* (2014).

Water Relation Parameters

Applied irrigation water (AIW)

As shown in Table 7, the average amounts of irrigation water for the first and second seasons and combined were 2184, 2078 and 2131m³/fad., for (I₁) respectively, 1605, 1556 and 1580 m³/fad., for I₂, respectively as well as 1156, 1106 and 1131m³/fad., for I₃, respectively. Results revealed that irrigation treatment I₁ consumed the highest amount of irrigation water followed by I₂ and then I₃. Similar results has been recorded by Eisa *et al.* (2002).

Seasonal actual water consumptive use (Evapotranspiration, ETa)

Seasonal actual water consumptive use (ETa) values as affected by irrigation treatments and wheat genotypes and their interactions are recorded in Fig. 1. The main effect of the irrigation treatments show that the highest irrigation regime (I₁) gave the highest consumptive use followed by the I₂ and then I₃. The values of water consumptive use in 2013/2014 were 520.1, 382.6 and 275.3 mm for I₁, I₂ and I₃, respectively. The same respective orders in 2014/2015 were 494.7, 370.9 and 263.4 mm. Differences between results of the two seasons may be due to high temperature especially in March, April and May and to the relatively lower humidity in the first season. These results indicate that ETa value was increased for the treatment of irrigating wheat plants without withholding irrigation, while, subjecting wheat plants to water deficit or withholding irrigation caused decrease in ETa values. So, subjecting plants to water stress in the late stage would affect the absorption of water from the soil and

Table 5. Effect of irrigation regimes (I₁, I₂ and I₃), genotypes and their interaction on No. of spikes/m², No. of grains/spike, 1000 grain weight (g) and grain yield (ardab^{*}/fad.^{}) for 2013/2014 and 2014/ 2015 seasons and their combined**

Character	No. of spikes/m ²			No. of grains/spike			1000 grain weight (g)			Grain yield (ardab/fad.)		
	2013/ 2014	2014/ 2015	Comb.	2013/ 2014	2014/ 2015	Comb.	2013/ 2014	2014/ 2015	Comb.	2013/ 2014	2014/ 2015	Comb.
Irrigation												
I ₁	460	453	546	54.4	55.5	53.5	48.8	47.4	48.1	22.8	26.0	24.4
I ₂	408	391	399	49.2	49.6	48.9	44.9	41.7	43.3	18.6	17.9	18.2
I ₃	353	345	349	45.8	45.0	46.7	41.0	36.8	38.9	15.6	14.4	15.0
F-test	**	**	**	**	**	**	**	**	**	**	**	**
LSD	12.729	16.796	8.753	1.354	1.945	0.984	0.852	0.675	0.451	0.455	0.379	0.246
Genotypes												
1	365	361	363	46.9	53.9	50.4	49.4	40.5	45.0	18.8	17.8	18.3
2	430	424	427	47.0	53.4	50.2	42.0	45.9	44.0	17.4	16.3	16.9
3	440	390	415	41.7	49.1	45.4	41.0	43.7	42.3	15.0	18.4	16.7
4	317	347	332	51.9	46.9	49.4	52.8	42.5	47.6	18.9	18.3	18.6
5	376	369	373	55.4	44.3	49.4	38.5	40.7	39.6	20.7	18.7	19.7
6	411	408	409	44.2	46.3	45.3	50.0	39.1	44.5	21.6	21.4	21.5
7	423	371	397	51.6	43.4	47.5	41.8	40.8	41.3	16.5	19.8	18.2
8	423	459	441	45.9	48.3	47.1	43.0	39.8	41.4	24.3	20.1	22.2
9	394	393	394	48.7	46.3	47.5	41.9	45.2	43.6	18.1	20.1	19.1
10	451	379	415	44.6	51.6	48.1	49.4	42.1	45.7	21.0	20.0	20.5
11	448	445	447	45.9	47.0	46.4	51.1	44.4	47.8	17.1	20.1	18.6
12	338	386	387	53.1	55.9	54.5	50.6	43.4	47.0	21.4	17.6	19.5
13	479	414	447	52.4	50.9	51.7	37.0	37.7	37.4	18.3	20.2	19.3
14	398	416	407	49.2	53.4	51.3	44.3	43.9	44.1	18.4	21.0	19.7
15	398	355	376	56.8	55.3	56.1	46.0	38.5	42.3	16.4	20.1	18.2
16	389	388	389	59.8	49.0	54.4	44.2	43.8	44.0	18.6	18.1	18.3
17	394	383	389	54.1	51.9	53.0	49.9	42.6	46.3	18.0	20.6	19.3
18	446	420	433	48.0	55.9	51.9	41.8	42.8	42.3	20.0	19.1	19.5
19	388	434	411	52.1	49.6	50.8	43.0	39.5	41.2	19.8	21.5	20.7
20	370	391	381	44.1	47.8	45.9	40.6	42.6	41.6	19.0	19.0	19.0
F-test	**	**	**	**	**	**	**	**	**	**	**	**
LSD	16.513	22.91	13.977	2.8	3.3	2.144	2.012	2.099	1.439	0.726	0.948	0.590
Irrigation												
I × G	**	**	**	**	**	**	**	**	**	**	**	**

I₁ = 4 irrigations (control). I₂ = 2 irrigations. I₃ = 1 irrigation. ardab^{*} = 150 kg, fad.^{**} = faddan = 4200 m²

Table 6. Drought sensitivity index (DSI) for grain yield of twenty wheat genotypes based on combined data

Drought sensitivity index for grain yield (ardab/fad.)		
Genotype	I ₁ - I ₂ DSI	I ₁ - I ₃ DSI
1	0.26	0.63
2	1.09	1.08
3	1.29	1.31
4	1.14	0.99
5	0.79	0.65
6	1.20	1.23
7	0.85	1.01
8	0.71	0.89
9	1.28	1.18
10	0.73	0.65
11	1.61	1.26
12	0.71	0.84
13	1.33	0.97
14	1.13	1.03
15	1.59	1.33
16	0.91	0.89
17	1.15	1.09
18	0.64	0.82
19	0.84	1.04
20	0.44	0.92

I₁ = 4 irrigations. I₂ = 2 irrigations. I₃ = 1 irrigation.

Table 7. Amount of irrigation water applied (m³/fad.) for irrigation regimes (I₁, I₂ and I₃) to wheat genotypes for 2013/2014 and 2014/2015 seasons and combined.

Genotype	The applied water (m ³ /fad.)									Grand mean
	2013/ 2014	2014/ 2015	Mean	2013/ 2014	2014/ 2015	Mean	2013/ 2014	2014/ 2015	Mean	
	I ₁			I ₂			I ₃			
1	2111	2031	2071	1583	1525	1554	1080	1028	1054	1560
2	2081	2018	2049	1561	1514	1537	1075	1024	1050	1545
3	2209	2109	2159	1612	1570	1591	1198	1146	1172	1641
4	2102	2059	2081	1579	1522	1551	1150	1097	1124	1585
5	2255	2187	2221	1607	1647	1627	1232	1179	1206	1685
6	2128	1963	2046	1562	1530	1546	1077	1026	1052	1548
7	2312	2113	2212	1605	1644	1624	1219	1164	1192	1676
8	2258	2198	2228	1681	1586	1634	1248	1193	1220	1694
9	2270	2189	2230	1652	1616	1634	1253	1202	1228	1697
10	2213	2151	2182	1671	1532	1601	1202	1149	1175	1653
11	2216	2169	2192	1622	1587	1604	1218	1163	1191	1662
12	2008	1943	1975	1537	1477	1507	1032	979	1006	1496
13	2331	2282	2306	1708	1655	1681	1291	1236	1263	1750
14	2072	1906	1989	1581	1487	1534	1024	1024	1024	1516
15	2229	2058	2143	1612	1575	1594	1197	1144	1170	1636
16	2222	2061	2142	1590	1537	1564	1191	1140	1165	1624
17	2009	1897	1953	1512	1478	1495	977	925	951	1466
18	2317	2114	2216	1675	1591	1633	1228	1173	1201	1683
19	2119	2071	2095	1563	1511	1537	1057	1005	1031	1554
20	2226	2039	2133	1584	1531	1557	1178	1125	1151	1614
Mean (I)	2184	2078	2131	1605	1556	1580	1156	1106	1131	1614

I₁ = 4 irrigations. I₂ = 2 irrigations. I₃ = 1 irrigation.

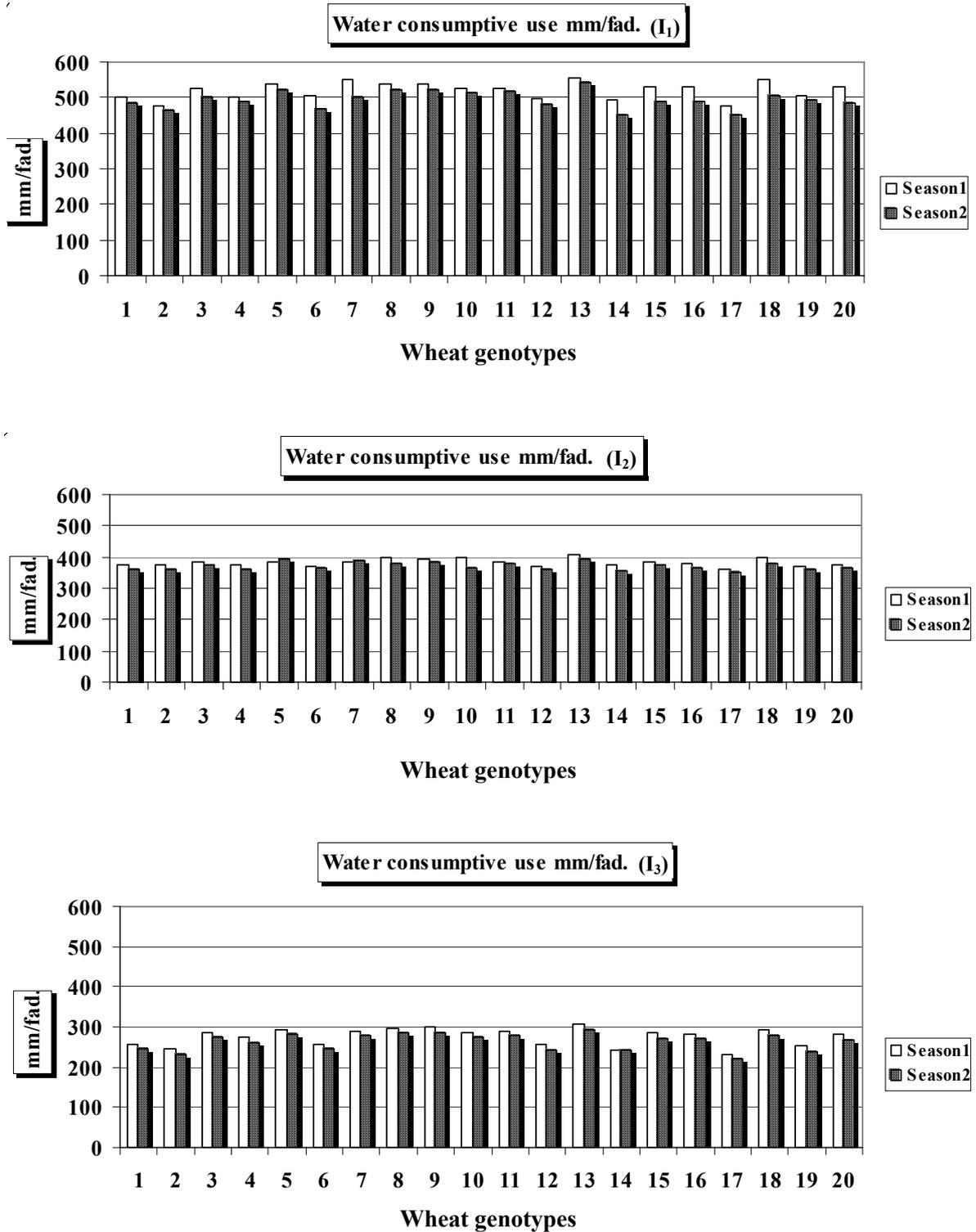


Fig. 1. Water consumptive use of wheat genotypes under irrigation regimes (I₁, I₂ and I₃) in 2013/2014 and 2014/2015 growing seasons

movement of minerals and solvents to the plant. These results indicate the importance of adequate soil moisture during growth stages.

Data presented in Fig.1 also indicate that the genotypes were differed in water consumptive use during the two seasons. These variations among genotypes might reflect, partially their different genetic backgrounds. Genotype 2 recorded the lowest value of water consumptive use while genotype 13 exhibited the highest value for this character.

These results are in full agreement with those obtained by Eisa *et al.* (2002) who found that the highest reduction of applied water was resulted from withholding irrigation 3 times and ranged from 33.7 to 43.9% less than the regular irrigation.

Water utilization efficiency (WUE)

Water utilization efficiency (WUE) is represented here as the amount of yield produced by one cubic meter of irrigation water used by crop. The main effect of irrigation treatments shows that average values of WUE in 2013/2014 were 1.57, 1.74 and 2.03 kg grains/m³ of the applied water for I₁, I₂ and I₃, respectively. Values in 2014/2015 were 1.85, 1.76 and 1.97 kg grains/m³ of the applied water for the same respective treatments as shown in Fig. 2. It is clear that wheat plants which given one surface-irrigation 25 days after planting irrigation (I₃), resulted in higher water use efficiency compared to the other irrigation treatments. This may be due to that withholding irrigation from vegetative growth stage to harvest could save water by about 47% and 28% with acceptable grains yield reduction of about 38% and 18% compared to I₁ and I₂, respectively.

With respect to wheat genotypes, WUE values in 2013/2014 ranging from 1.31 to 2.19 kg grains/m³ of the applied water and in 2014/2015, the corresponding values were 1.65 to 2.22 kg grains/m³ of the applied water Fig. 2. The highest (WUE) was recorded by genotype No. 6 followed by genotype No.12, while genotype

No. 3 recorded the lowest value for water utilization efficiency. The interaction between WUE and wheat genotypes indicate that genotype No. 1 gave the lowest value of water utilization efficiency under I₁, while genotype No. 8 recorded the highest value under I₂. These results are in agreement with the observations mentioned by Shah *et al.* (2006) and Abd El-Hay (2008). Furthermore, Eisa *et al.* (2002) found that withholding irrigation at any stage of growing season resulted in higher water use efficiency values compared to the adequate irrigation.

Organic Osmoprotectants

Figs. 3, 4 and 5 show that organic osmoprotectants *i.e.* total soluble sugars, proline and free amino acids (FAA) are significantly increased in leaves of the 20 wheat genotypes under drought stress. Plants which received two irrigations (I₂) have the highest concentrations of TSS, proline and FAA than those received four (I₁) or one (I₃) irrigations. The three wheat genotypes No. 1, 2 and 3 recorded the highest concentrations of TSS, proline and FAA under moderate stress compared to other genotypes where, these organic compounds approximately increased in stressed plants more than 2 folds than in unstressed plants. In the same trend, Loutfy *et al.* (2012) and Khoshro *et al.* (2013) found that drought stress caused a rapid increase in soluble sugars, proline and amino acids contents in wheat genotypes.

The accumulation of soluble carbohydrates in plants has been widely reported as a response to drought (Zhang *et al.*, 2009). Carbohydrates seem to play a key role in the integration of plant growth and appear to be part of a wider mechanism for balancing carbon acquisition and allocation within and between organs (Farrar *et al.*, 2000). Under water stress, soluble sugars can function in two ways which are difficult to separate, namely osmotic agents and osmoprotectors (Yong *et al.*, 2006). As osmotic agents, soluble sugars facilitate osmotic adjustment, as osmoprotectors they stabilize

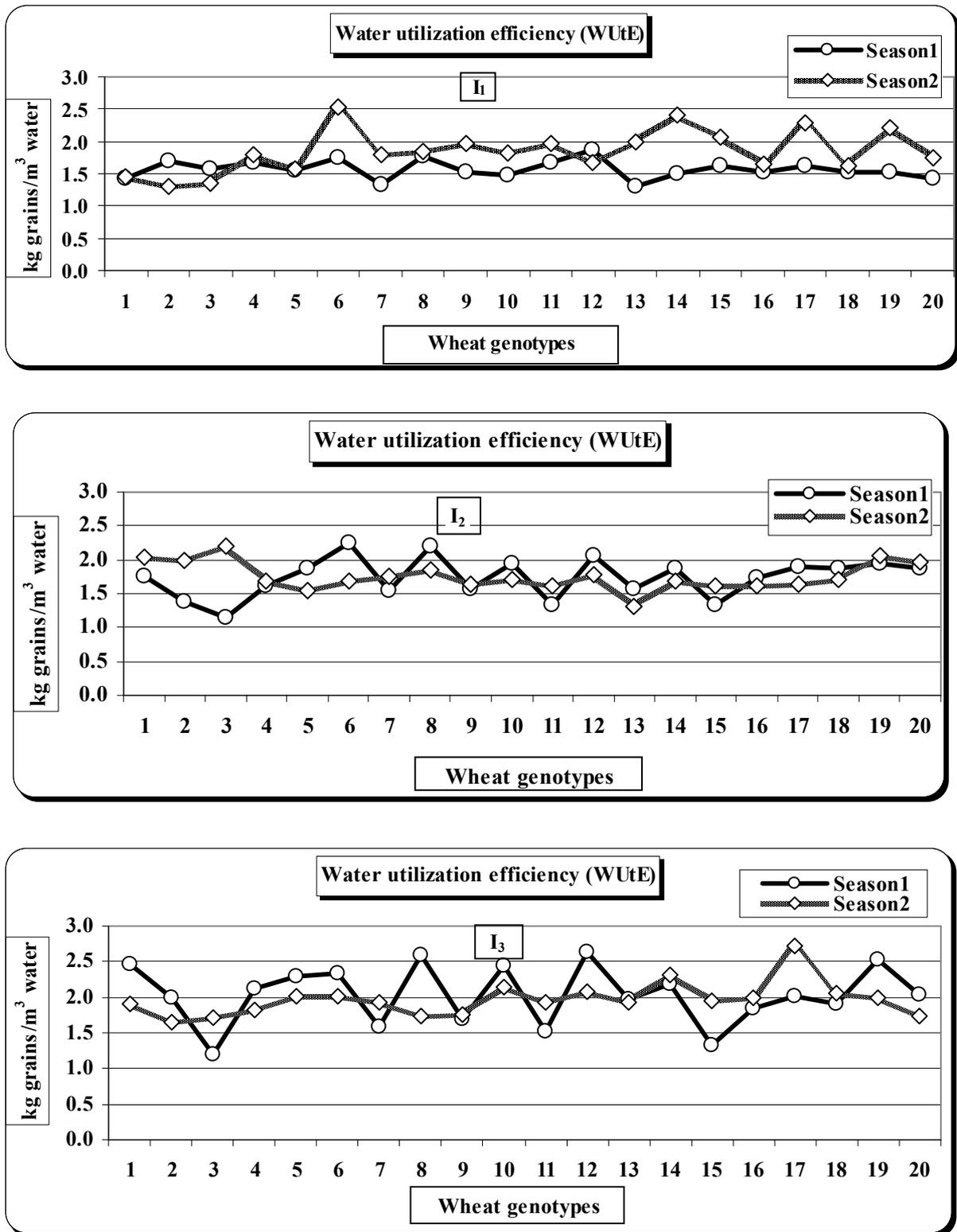


Fig. 2. Water utilization efficiency (WUE) of wheat genotypes under irrigation regimes (I₁, I₂ and I₃) in 2013/2014 and 2014/2015 growing seasons

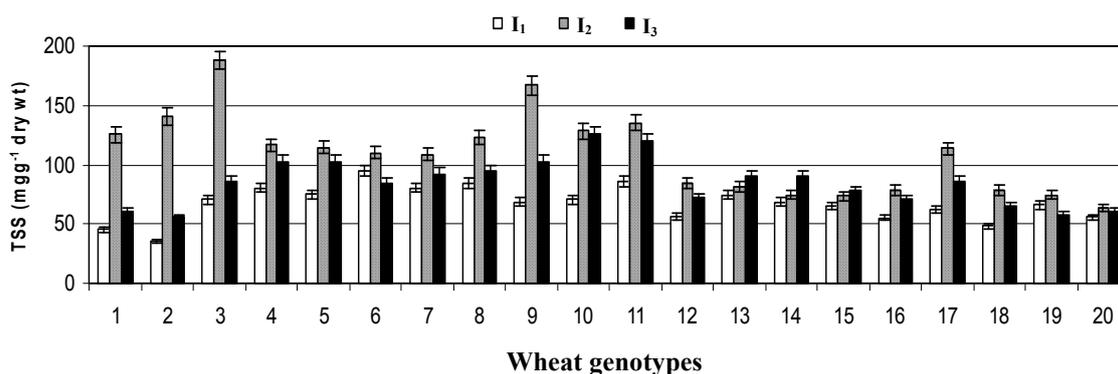


Fig. 3. Total soluble sugars (TSS) of 20 wheat genotypes subjected to three levels of water stress. Where, I_1 - plants received four irrigations, I_2 - plants received 2 irrigations and I_3 - plants received 1 irrigation after planting during the second growing season (2014/2015)

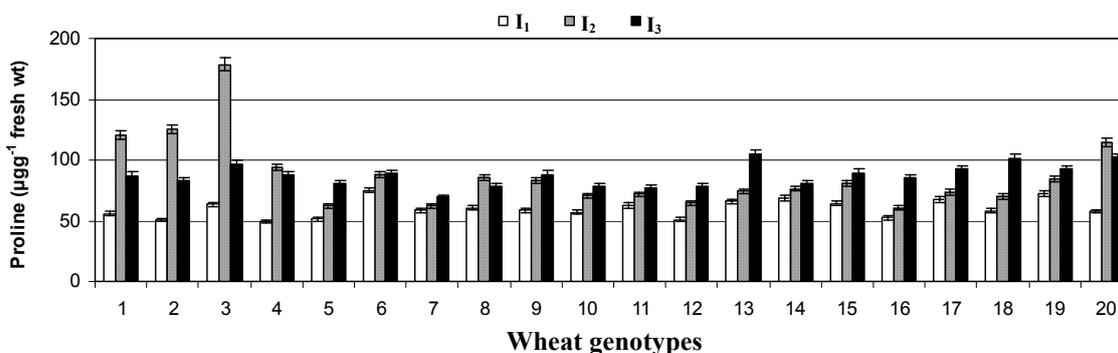


Fig. 4. Proline content of 20 wheat genotypes subjected to three levels of water stress. Where, I_1 - plants received four irrigations, I_2 - plants received 2 irrigations and I_3 - plants received 1 irrigation after planting during the second growing season (2014/2015)

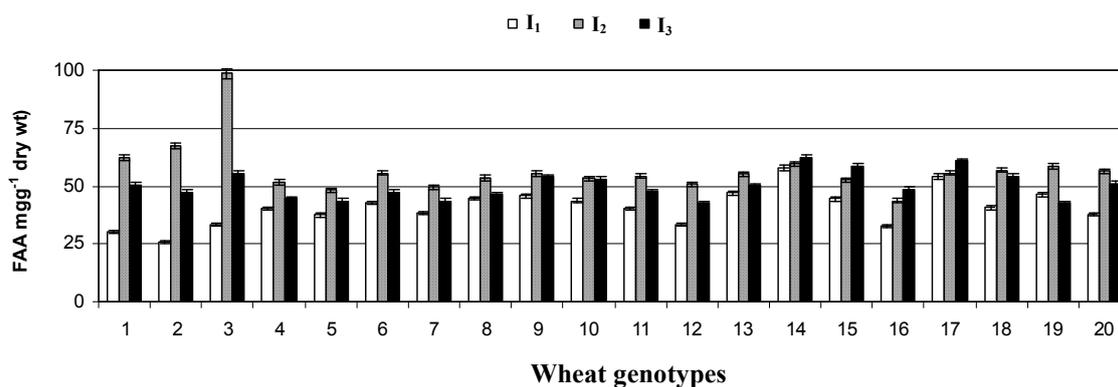


Fig. 5. Free amino acids (FAA) content of 20 wheat genotypes subjected to three levels of water stress. Where, I_1 - plants received four irrigations, I_2 - plants received 2 irrigations and I_3 - plants received 1 irrigation after planting during the second growing season (2014/2015)

proteins and membranes, most likely substituting water in the formation of hydrogen bonds with polypeptide polar residues and phospholipids phosphate groups.

Proline, which is widely found in higher plants, accumulates in stressed plants in larger amounts than other amino acids (Ghaderi and Siosemardeh, 2011). Proline regulates the accumulation of useable nitrogen, is osmotically active and contributes to membrane stability (Bandurska *et al.*, 2008; Javadi *et al.*, 2008). It may also act as a signaling regulatory molecule able to activate multiple responses that are components of the adaptation process (Maggio *et al.*, 2002).

Anatomical Features

Effect of irrigation treatments on anatomical features of flag leaves blades for representative wheat genotypes are shown in Fig. 6 and tabulated in Table 8. Results indicated that, anatomical features of wheat genotypes influenced by water stress. Generally, the measured dimensions of the studied wheat genotypes leaves *i.e.* midrib thick., midvein bundle length, midvein bundle width, phloem thick., average diameter of meta xylem vessel, lamina thick., mesophyll thick., upper epidermis thickness and lower epidermis thick were reduced by giving wheat plants one irrigation only (I_3) compared with (I_1) which received four irrigations in all genotypes with high differences among genotypes.

It has been noticed that genotype No. 20 recorded the highest reduction in the aforementioned measurements with reduction percentage of 68.40, 68.58, 37.80, 58.80, 47.52, 65.40, 66.72, 54.15 and 69.68%, respectively. On the other hand, the lowest reduction was registered in genotype No. 1 which reached 7.80, 11.43, 10.74, 15.06, 9.06, 9.70, 7.75, 16.92 and 13.98%. Therefore, genotype No. 1 could be considered as more tolerant to water stress.

Water stress decreased most of leaf anatomical characters has been registered by Ghanem (2008) and Hameed *et al.* (2002).

Moreover Adhikary *et al.* (2007) reported that, the drought tolerance and sensitive genotypes revealed differentiating parameters in leaf anatomy.

Thick cutical is the character feature of xeric conditions and this may be and adaptations of xeric grasses (Ubeda, 1993), as well as Ramon and Chang (1982) also reported that thick cuticle is the most reliable traits for drought resistance of four clones of tea.

The decrease in mesophyll tissue, xylem and phloem leads to a slow rate on the translocation of photo assimilates towards the developing grains through the peduncle and spike rachilla. Furthermore, the decrease in the diameter of metaxylem vessels in the leaf blade results in lowering the accumulation of necessary water required for photosynthesis. The lowest reduction in leaf anatomical characteristics has been observed in genotype No.1 led to drought tolerance ($DSI < 1$) and enhance wheat grain yield (18.3 ardab/fad.).

Conclusion

Water stress caused a significant decrease in yield of wheat genotypes rather than non – stress conditions based on drought sensitivity index (DSI) of grain yield. Genotypes No. 1, 5, 8, 10, 12, 16 and 18 appeared to be more tolerant to drought as they exhibited DSI less than unity. Otherwise, genotypes No.3, 6, 9, 11 and 15 were sensitive to water stress ($DSI > 1$), furthermore the remaining wheat genotypes were moderate tolerant to water stress resulted in increase of osmoprotactants (total soluble sugars, proline and free amino acids) in the studied genotypes. Under sever water stress, the osmoprotactants increased in three genotypes No.1, 2 and 3 compared to other tested genotypes. Noticeably, genotypes No.1, 2 and 3 recorded the highest osmoprotactants under moderate water stress. The present study indicated that genotype No. 1 appeared to be more tolerant to drought stress as they exhibited DSI less than unity, gave increase in osmoprotactants and lowest reduction in flag leaf blade anatomical characteristics has been observed. The study recommended by sowing this genotype for saving water and increasing grain yield.

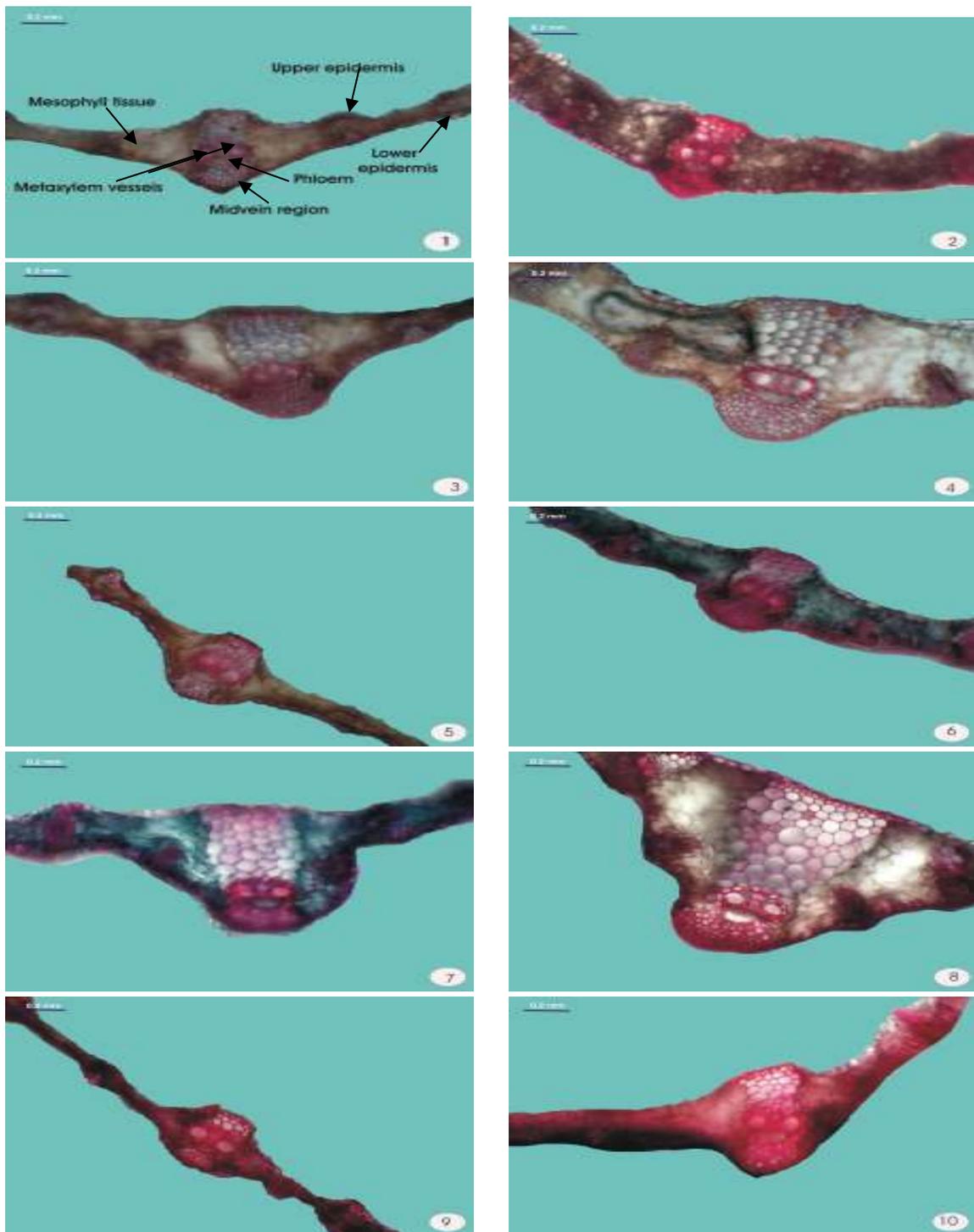


Fig. 6. Changes in transverse sections in the flag leaf blade of five wheat genotypes (1, 6, 9, 18 and 20) received four irrigations or received only one irrigation after planting during the second growing season 2014/2015 (The bar for all plates = 0.2mm)

- 1) Genotype 1 received one irrigation.
- 3) Genotype 6 received one irrigation.
- 5) Genotype 9 received one irrigation.
- 7) Genotype 18 received one irrigation.
- 9) Genotype 20 received one irrigation.

- 2) Genotype 1 received four irrigations.
- 4) Genotype 6 received four irrigations.
- 6) Genotype 9 received four irrigations.
- 8) Genotype 18 received four irrigations.
- 10) Genotype 20 received four irrigations.

Table 8. Anatomical features of flag leaf blade of representative wheat genotypes, received four irrigations (I₁) or received only one irrigation (I₃) after planting irrigation during the second growing season (2014/2015)

Genotype	Genotype 1			Genotype 6			Genotype 9			Genotype 18			Genotype 20			
	Treatments	I ₁	I ₃	Reduction (%)	I ₁	I ₃	Reduction (%)	I ₁	I ₃	Reduction (%)	I ₁	I ₃	Reduction (%)	I ₁	I ₃	Reduction (%)
Dimensions of the midvein bundle	Midrib thick. (μ)	598.58	551.90	7.80	975.61	845.38	13.35	596.88	314.33	47.34	1186.41	712.08	39.98	743.65	435.02	68.40
	Length (μ)	224.54	198.87	11.43	297.53	259.03	12.94	250.85	145.41	42.03	296.33	180.32	39.15	265.78	83.52	68.58
	Width (μ)	359.27	320.69	10.74	372.38	332.20	10.79	311.33	196.08	37.02	359.55	224.00	37.70	303.06	188.51	37.80
	Phloem thick. (μ)	71.39	60.64	15.06	90.97	74.29	18.34	80.06	57.80	27.80	98.70	51.30	48.02	86.19	35.51	58.80
	Average diameter of meta xylem vessel (μ)	67.96	61.80	9.06	91.54	82.33	10.06	87.67	64.21	26.76	101.69	66.16	34.94	87.73	46.04	47.52
Dimensions of the lamina	Lamina thick. (μ)	311.23	281.03	9.70	412.83	359.83	12.84	262.70	154.48	41.20	441.81	236.19	46.54	302.75	104.74	65.40
	Mesophyll thick. (μ)	237.55	219.13	7.75	321.07	286.82	10.67	196.18	89.53	54.36	353.70	183.57	48.10	226.99	75.54	66.72
	Upper epidermis thick. (μ)	38.19	31.73	16.92	50.20	39.15	22.01	36.40	36.31	0.25	44.64	27.19	39.09	40.11	18.39	54.15
	Lower epidermis thick. (μ)	35.49	30.53	13.98	41.56	33.86	18.53	30.11	28.64	4.88	43.47	25.43	41.50	35.65	10.81	69.68

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سلوك محصول الحبوب والصفات الكيموحيوية والتشريحية تحت ظروف الإجهاد المائي لبعض التراكيب الوراثية لقمح الخبز

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أجرى هذا البحث بمحطة بحوث كفر الحمام محافظة الشرقية - مصر خلال موسمي الزراعة ٢٠١٣/٢٠١٤ و ٢٠١٤/٢٠١٥ حيث تم تقييم ٢٠ سلالة وصنف من قمح الخبز تحت ثلاث معاملات ري: معاملة الكنترول (I_1) أعطيت ٤ ريات بعد رية الزراعة و (I_2) أعطيت ريتان بعد رية الزراعة (I_3) أعطيت رية واحدة بعد رية الزراعة وتم ترك مسافة ٧ متر بين كل معاملة ري والأخرى، تم استخدام تصميم القطع المنشقة مرة واحدة وكانت معاملات الري في القطع الرئيسية والأصناف في القطع المنشقة الأولى وتم توزيع الأصناف بطريقة عشوائية في ثلاث مكررات وكانت أهم النتائج كالتالي: أظهرت النتائج فروق عالية المعنوية في محصول الحبوب ومكوناته للتراكيب الوراثية في كلا الموسمين والتحليل التجميحي كما أظهرت النتائج تفاعل عالي المعنوية بين التراكيب الوراثية ومعاملات الري في كلا الموسمين والتحليل التجميحي في كل الصفات عدا وزن الألف حبة في الموسم الأول، أظهرت معاملة الري I_1 (الكنترول) أعلى قيمة للاستهلاك المائي وكمية المياه المضافة، وكانت قيم الاستهلاك المائي في الموسم الأول ١، ٥٢٠، ٦، ٣٨٢، ٣ و ٢٧٥، ٣ ملم / فدان لمعاملات الري I_1 و I_2 و I_3 ، على التوالي، وفي الموسم الثاني كانت قيم الاستهلاك المائي ٧، ٤٩٤، ٩، ٣٧٠، ٤ و ٢٦٣، ٤ ملم / فدان على التوالي، وسجل التركيب الوراثي رقم ٢ أقل قيمة للاستهلاك المائي، في حين سجل التركيب الوراثي رقم ١٣ أعلى قيمة للاستهلاك المائي، أعطت المعاملة I_3 أعلى كفاءة للاستفادة من المياه (كجم حبوب/ ٣ م ماء مضاف) في حين سجلت I_1 أقل قيمة وسجل التركيب الوراثي رقم ٨ أعلى كفاءة لاستخدام المياه ، بينما سجل التركيب الوراثي رقم ٣ أقل قيمة لهذا المقياس، دليل الحساسية للجفاف أظهر أن التراكيب الوراثية أرقام ١، ٥، ٨، ١٠، ١٢، ١٦ و ١٨ كانت متحملة للإجهاد المائي، أدى الإجهاد المائي المتوسط إلى زيادة جوهرية في كل من السكريات الكلية الذائبة والبرولين والأحماض الأمينية الحرة في الأصناف تحت الدراسة، وتحت ظروف الإجهاد الشديد سجلت زيادة جوهرية للصفات السابقة في التراكيب الوراثية رقم ١، ٢، ٣ مقارنة بباقي التراكيب الوراثية، تأثرت الصفات التشريحية لأوراق السلالات الخمسة الممثلة للتراكيب الوراثية بواسطة الإجهاد المائي، وسجل التركيب الوراثي رقم ٢٠ أعلى نقص في الصفات التشريحية بينما سجل التركيب الوراثي رقم ١ أقل نقص مقارنة بباقي الأصناف المدروسة، أظهر التركيب الوراثي رقم ١ مقاومة عالية للإجهاد المائي الذي سجل دليل حساسية للجفاف (الأقل من الواحد) وأعطى زيادة معنوية في كل من السكريات الكلية الذائبة والبرولين والأحماض الأمينية الحرة وأظهر أقل تأثر في الصفات التشريحية مقارنة بباقي التراكيب الوراثية، لذا توصي الدراسة بزراعة التركيب الوراثي رقم ١ لانه متحمل للإجهاد المائي ويعطى مستوى جيد من المحصول.

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