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GROWTH, YIELD AND ANTIOXIDANTS STATUS OF WHEAT (*Triticum aestivum* L.) CULTIVARS UNDER WATER DEFICIT CONDITIONS

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Water scarcity is one of the major impacts of climate change leading to great reduction in wheat crop yields all over the world. Therefore, identification of drought tolerant or sensitive wheat cultivars has become an essential approach to enhance the food production on sustainable basis. A study was planned to evaluate the potential of nine approved wheat cultivars [Fareed-2006, Millat-2011, Miraj-2008, AARI-2011, Lassani-2006, AAS-2011, Shafaq-2006, Sahar-2006 and Punjab-2011] against water deficit conditions on the basis of their antioxidants status. The cultivars were grown under water deficit and well-watered conditions (50% and 100% field capacity), respectively. The cultivars showed differential but statistically similar response during germination and seedling establishment under applied water treatments. Drought (50% field capacity) caused significant reduction in growth and yield contributing parameters i.e. plant height, number of grains per spike, 100 grain weight and grain yield per plant in all the wheat cultivars studied. However, performance of AARI-11 ensured the maximum yield and yield contributing components under deficit and normal watered conditions with more activities of enzymatic antioxidants [superoxide dismutase (SOD), peroxidase (POD) catalase (CAT)], higher contents of non-enzymatic antioxidants [ascorbic acid (AsA) and total phenolic contents (TPC)], also greater contents of leaf K^+ and photosynthetic pigments (chlorophyll "*a"* and "*b"*). Nevertheless, antioxidants status was significantly higher at reproductive stages (booting and heading) as compared to vegetative stage (leaf initiation) in all the cultivars, under water deficit and well-watered conditions. Moreover, AARI-11 showed the major grain protein profiling determined through sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-page). In the present study, the cultivar AARI-11 proved itself a drought tolerant cultivar with maximum productivity.

Keywords: Drought tolerance, enzymatic and non-enzymatic antioxidants, SDS-page, water stress.

INTRODUCTION

Agriculture is leading source of income for more than half of the population of Pakistan where wheat (*Triticum aestivum* L.) is staple food for more than 40-60% of the world's population (Nawaz *et al*., 2013). Its grain is the main source of protein and carbohydrates. It is cultivated in Rabi season (October–November to March–April) which is usually prone to short term drought (Kamran *et al*., 2009) as a result of climate change, seasonal variation, uneven spatiotemporal distribution of rainfall and decreasing capacity of water reservoirs. For the reason, a dire need to explore ways and means to achieve yield goals under reduced water availability (Hussain *et al*., 2014; Nawaz *et al*., 2015).

Plants show complex responses to water deficit involving many biochemical and physiological processes such as the production of antioxidants (enzymatic and non-enzymatic), proteins and mineral elements (Zhu, 2002). The impacts of drought on crop plant varies depending upon growth stages, duration, frequency and intensity of drought (Pospisilova *et al*., 2000). Seed germination is the first stage of plant growth which is highly sensitive to water deficit condition (Ali *et al*., 2007) whereas in semiarid to arid climates of Pakistan, low moisture availability at critical growth stages is the main growth limiting factor. However, seedling establishment under these stressful conditions is a good indicator for determining crop development and maturity (Rauf *et al*., 2012). The reduction in soil moisture or osmotic potential significantly decreases the mass flow and diffusion of mineral nutrients, ultimately reducing the availability of nutrients to plant roots which diminishes the yield and yield components (Arora *et al*., 2002). The oxidative stress resulting from water deficit disrupts photosynthetic system and triggers the generation of reactive oxygen species (ROS) in crop plants so inhibiting the normal growth of plants (Farooq *et al*., 2009). To ameliorate the damaging effects of reactive oxygen species (ROS), plants produce antioxidants (enzymes and non-enzymes) which convert H_2O_2, O^- and OH^- into nontoxic forms (Banowetz, 1998). Drought tolerance is involved in the production or inhibition of various proteins in plants under stress. The profiling of these proteins illustrates the taxonomic and evolutionary aspects of various crop species due to abiotic stresses (Khan *et al*., 2007; Iqbal *et al*., 2005). The plants have intrinsic ability to cope adverse environmental conditions through avoidance/tolerance/adaptation. Therefore, identification of

drought tolerant/ resistant wheat cultivars is necessary to feed the ever increasing population under reduced water conditions (Baligar *et al*., 2001; Ahmad *et al*., 2014).

Following the literature and water scarcity situation in Pakistan, the present study was planned to screen drought tolerant wheat cultivars on the basis of antioxidant defense potential, growth, yield parameters and grain protein profiling under water deficit conditions.

MATERIALS AND METHODS

***Experimental layout*:** The experiment was conducted in pots during Rabi season 2012-13 at Experimental Farm, Faculty of Agricultural Sciences and Technology, Bahauddin Zakariya University, Multan, Pakistan. The climate of the Multan region is semi-arid and subtropical. The meteorological data for Rabi season 2012-13 is presented in Fig. 1.

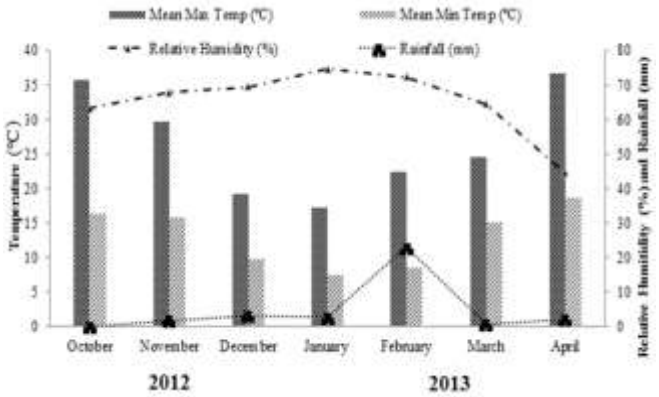


Figure 1. Monthly averages of meteorological data for growing period of wheat crop during 2012-2013.

The experimental design was completely randomized design (CRD) in factorial arrangement with three replications. There are two factors including nine approved wheat cultivars widely grown in the drought prone areas of Southern Punjab i.e. Fareed-2006 (06), Millat-2011 (11), Miraj-2008 (08), AARI-2011 (11), Lassani-2006 (06), AAS-2011 (11), Shafaq-2006 (06), Sahar-2006 (06) and Punjab-2011 (11) and two moisture regimes based on field capacity (FC 100% and FC 50%). The pot depth x width was 15 x 9 inches filled with 10 kg soil in a mixture of 1:1:1 (weight basis) sand, silt and clay. The maximum water holding capacity/ field capacity of the soil was 15% on weight basis. Moisture level was established in pots prior to sowing and maintained up to harvesting on the basis of soil water holding capacity through moisture meter (Model: Irrometer Tensiometer "R", Irrometer Company, Riverside, California, USA). Fifteen seeds were sown in each pot on 1st November, 2012. Fifteen days after sowing, thinning was done maintaining three seedlings per pot. The recommended practices were adopted throughout the growing season of the crop. The observations recorded during course of study for various growth, yield and biochemical aspects were as following.

***Germination (%)*:** The number of seeds germinated were counted daily for 10 days up to completion of germination. Seeds were considered germinated, when the radical obtained 2 mm length. Germination (%) was calculated using the following formula;

Germination (%) = (Germinated seed/ total seed) ×100

***Germination index (GI)*:** The germination index (GI) was calculated as described by the Association of Official Seeds Analyst (Anonymous, 1983) by using the following formula;

$$GI = \frac{\text{No.of germinated seeds}}{\text{Days of first count}} + \frac{\text{No.of germinated seeds}}{\text{Days of final count}}$$

***Time to 50% germination*:** Time taken to 50% germination (T_{50}) was calculated described by Farooq *et al*. (2005).

$$T_{50} = ti + \left[\frac{\{(N+1)/2) - (ni)\}}{(nj - ni)} \right] \times (tj - ti)$$

Where N is the final number of germinated seeds and ni and nj cumulative number of seeds germinated by adjacent counts at times ti and tj when ni<N+1/2<nj.

***Growth and yield*:** The plant height, number of grains per spike, 100-grain weight and grain yield per plant were recorded following standard procedures.

***Biochemical analysis*:** The flag leaves of wheat cultivars were randomly selected from each treatment to measure their antioxidants status, at three critical growth stages (leaf initiation, booting and heading) and freezed at -20°C. For determination of enzymatic antioxidants, leaf samples were extracted in 50 mM phosphate buffer (pH 7.8). The extract was centrifuged at 15000 rpm at 4°C and the supernatant was used for assay of peroxidase (POD), catalase (CAT) (Chance and Maehly, 1955) and super oxide dismutase (SOD) (Giannopolitis and Ries, 1997). Total phenolic contents (TPC) were determined by adopting the method described by Waterhouse (2001). Ascorbate contents were determined following the protocol of Ainsworth and Gillespie (2007). Chlorophyll contents ("*a*" and "*b*") were determined only at heading stage after extraction in 80% acetone following the method of Nagata and Yamashta (1992) by using Nano-spectrophotometer (UV-4000, O.R.I. Germany). Leaf potassium (K^+) contents were estimated by using the protocol of USDA Laboratory Staff, (1954) after oven drying leaf samples at 70°C and digesting leaf powder in concentrated nitric acid (HNO_3) and perchloric acid ($HCLO_4$) at 2:1 ratio (Di-acid mixture) according to the method adapted by Rashid (1986).

***Grain SDS-PAGE*:** The extraction of grain crude protein for SDS-PAGE was done by taking 0.5 gram grain sample from each wheat cultivar grown at 100% & 50% FC and ground

in 2 mL extraction buffer (50 mM Tris-HCL, pH 7.5) to make the slurry. The crude extracts were centrifuged at 17000 rpm for 15 min. The supernatant was used for 1D SDS-page. Protein profiling through 1D SDS-page was carried out by the modified method of Laemmli's (1970). The protein of the crude extract was estimated using the Bradford method (Bradford, 1976) and equal amount of protein extract (10 µg) loaded onto each well. The samples of wheat cultivars were run along with molecular weight marker of 10 to 200 KDa as a standard at 60 V for 3 h. The polyacrylamide gel was stained using CBB R250 for 4 h and destaining was carried out using glacial acetic acid, methanol and water in ratio 2:5:53 respectively.

***Statistical analysis*:** Data were computed and analyzed using Fisher's analysis of variance technique and LSD test ($p<0.01$) was used to compare differences among the mean values (Steel *et al*., 1997). Moreover, Microsoft Excel Program 2013 was used for the graphical description of data.

RESULTS

Water deficit levels affected the germination attributes of nine wheat cultivars (Table 1) but the significance ($p<0.01$) of the impact variable with respect to final germination percentage, germination index (GI) and time to 50% germination (T_{50}). Results demonstrated that AARI-11 took maximum days for final germination percentage and GI under both non-stress and drought conditions (Table 1). The interaction effect of T_{50} was non-significant but among the cultivars, AARI-11 obtained maximum T_{50} at 50% FC while Millat-11 took minimum time in this regard. Growth and yield components showed significant decrease in all the wheat cultivars under water deficit stress as compared to non-stressed well-watered condition (Table 2). Maximum plant height was observed in AARI-11 at 50% FC followed by its respective values under 100% FC. Results described that AARI-11 performed better exhibiting the maximum

Table 1. Germination parameters of wheat cultivars under drought stress conditions (50% & 100% Field Capacity).

GP	Field Capacity	Fareed-06	Millat -11	Miraj-08	AARI-11	Lasani-06	AAS-11	Shafaq-06	Sahar-06	Punjab-11	Mean
Germination	D1	57.78 b.d	33.33 d	51.11 cd	82.22 ab	66.66 a.c	77.77 ab	86.66 a	46.66 cd	84.44 a	75.31 a
%age	D2	80.00 ab	48.89 cd	77.78 ab	88.89 a	82.22 ab	88.89 a	48.89 cd	80.00 ab	82.22 ab	65.18 b
	Mean	68.89 ab	41.11 c	64.44 b	85.55 a	74.44 ab	83.33 a	67.77 ab	63.33 b	83.33 a	
LSD values (0.01)	Field capacity, Cultivars and Field capacity×Cultivars 8.4126, 17.846 and 25.238 respectively										
Germination	D1	4.71 d	4.94 d	5.17 cd	9.29 ab	6.26 cd	5.60 cd	4.36 d	7.10b.d	6.04 cd	6.30 a
Index	D2	6.46 b.d	4.79 d	6.31 cd	9.51a	7.99 a.c	4.60 d	6.76 a.d	5.20 cd	5.07 d	5.94 a
(GI) (Days)	Mean	5.58 bc	4.86 c	5.74 bc	9.40 a	7.13 b	5.10 c	5.56 bc	6.15 bc	5.56 bc	
LSD values (0.01)	Field capacity, Cultivars and Field capacity×Cultivars 0.6034, 1.2801 and 1.8103 respectively										
Time to 50%	D1	14.66 ab	12.00 ab	15.66 ab	18.00 a	15.00 ab	15.33 ab	16.66 a	16.66 a	15.33 ab	15.48 a
Germination	D2	15.66 ab	9.00 b	16.66 a	13.66 ab	15.00 ab	15.66 ab	16.66 a	16.33 a	13.33 ab	14.66 a
(T50) (Days)	Mean	15.16 a	10.50 b	16.16 a	15.83 a	15.00 a	15.50 a	16.66 a	16.50 a	14.33 ab	
LSD values (0.01)	Field capacity, Cultivars and Field capacity×Cultivars 1.4393, 3.0533 and 4.3180 respectively										

Means not sharing the same letters differ significantly at 1% probability level. D_1= 50% Field capacity D_2= 100% Field capacity.

Table 2. Seedlings growth and yield components of wheat cultivars under drought stress conditions (50% & 100% Field Capacity).

Y and YP	Field Capacity	Fareed-06	Millat -11	Miraj-08	AARI-11	Lasani-06	AAS-11	Shafaq-06	Sahar-06	Punjab-11	Mean
Plant height	D1	56.46 d	40.43 ef	54.23 d	83.43 a	35.00 g	42.00 e	34.00 g	36.16 fg	45.33 e	47.45 b
(cm)	D2	63.16 c	41.16 ef	45.00 e	74.33 b	54.33 d	53.50 d	41.66 e	44.00 e	32.40 g	49.95 a
	Mean	59.81 b	40.80 e	49.61 c	78.88 a	44.66 d	47.75 cd	37.83 e	40.08 e	38.86 e	
LSD values (0.01)	Field capacity, Cultivars and Field capacity×Cultivars 1.8094, 3.8383and 5.4281 respectively										
Number of	D1	52.55 b.e	35.11 gh	38.00 fg	64.66 a	51.55 b.e	43.00 e.g	51.88 b.e	44.67 d.g	37.88 fg	46.59 a
grains/ spike	D2	55.33 a.d	25.22 h	52.88 a.e	61.00 ab	57.11 a.c	36.55 f.h	44.11 d.g	47.55 c.f	42.55 e.g	46.92 a
	Mean	53.94 bc	30.16 e	45.44 cd	62.83 a	54.33 ab	39.77 d	47.99 b.d	46.11 b.d	40.22 d	
LSD values (0.01)	Field capacity, Cultivars and Field capacity×Cultivars 4.0236, 8.5353 and 12.071 respectively										
100-grain	D1	3.33 de	1.53 h	3.41 de	4.64 ab	2.10 f.h	4.49 bc	2.59 ef	2.45 fg	2.27 f.h	2.98 b
weight (g)	D2	3.70 cd	1.74 gh	2.51 fg	5.43 a	3.34 de	4.54 b	3.65 d	3.50 d	2.32 f.h	3.41 a
	Mean	3.51 b	1.63 e	2.96 bc	5.04 a	2.72 cd	4.52 a	3.12 bc	2.97 bc	2.29 d	
LSD values (0.01)	Field capacity, Cultivars and Field capacity×Cultivars 0.2470, 0.5239 and 0.7409 respectively										
Grain yield/	D1	3.30 fg	1.63 g	3.86 d.f	9.40 b	2.76 fg	4.00 d.f	3.46 e.g	2.66 fg	4.30 d.f	3.93 b
plant (g)	D2	5.40 c.e	2.43 fg	5.50 c.e	12.33 a	2.53 fg	6.56 c	5.83 cd	5.40 c.e	6.90 c	5.87 a
	Mean	4.35 bc	2.03 e	4.68 bc	10.86 a	2.65 de	5.28 bc	4.65 bc	4.03 cd	5.60 b	
LSD values (0.01)	Field capacity, Cultivars and Field capacity×Cultivars 0.6874, 1.4583 and 2.0623 respectively										

Means not sharing the same letters differ significantly at 1% probability level. D_1= 50% field capacity D_2= 100% field capacity.

number of grains per spike at 50% FC but greater values were observed in 100-grain weight and grain yield per plant under 100% FC whereas least values for these attributes were obtained in Millat-11 during both irrigation treatments (Table 2). As far as enzymatic activity is concerned, a significant increasing trend was observed from leaf initiation to booting and heading stages in all the wheat cultivars under drought stress and well-watered conditions (Table 3). The POD, CAT and SOD activities were increased in wheat cultivars from vegetative to reproductive stages and reached

Table 3. Status of enzymatic antioxidants of wheat cultivars at leaf initiation, booting and heading stages under drought stress conditions (50% & 100% Field Capacity).

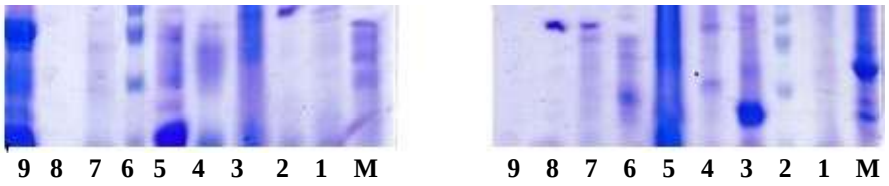
Antioxidant		Field capacity	Fareed-06	Millat-11	Miraj-08	AARI-11	Lassani-06	AAS-11	Shafaq-06	Sahar-06	Punjab-11	Mean	Mean
POD	Leaf	D1	6.08 m.p	2.52 w.z	2.80 v.y	23.33 b	1.27 z.c	8.85 g.i	11.45 f	5.99 m.q	1.83 y.b	7.13 d	5.42 c
(mmol	Initiation	D2	4.06 s.u	0.04 c	0.66 bc	17.46 d	0.97 a.c	2.04 x.b	2.37 w.a	3.41 t.x	2.48 v.z	3.72 f	
min^{-1} mg	Booting	D1	8.21 g.k	3.84 t.v	6.54 l.o	28.53 a	4.57 q.t	11.53 f	13.68 e	7.54 i.l	4.75 p.t	9.91 b	8.03 b
$protein^{-1}$)		D2	6.58 l.o	2.88 u.y	3.73 t.w	18.58 d	3.59 t.w	5.36 o.s	4.57 q.t	5.39 o.s	4.77 p.t	6.16 e	
	Heading	D1	9.09 gh	5.71 n.r	8.84 g.i	29.77 a	6.72 l.o	13.89 e	14.870 e	9.38 g	7.72 h.l	11.78 a	10.03 a
		D2	8.44 g.j	4.54 r.t	5.51 n.r	20.56 c	6.68 l.o	7.34 k.m	6.85 k.n	7.93 h.l	6.71 l.o	8.28 c	
		Cultivars	7.08 d	3.26 g	4.68 e	23.04 a	3.97 f	8.17 c	8.96 b	6.61 d	4.71 e		
LSD values (0.01) Field capacity, cultivars, critical growth stages individual and three way interaction 0.4778, 0.5852, 0.3379 and 1.4335 respectively													
CAT	Leaf	D1	2.84 u.z	4.20 r.w	6.08 o.s	10.35 i.l	3.94 s.x	2.44 u.z	1.10 yz	1.23 yz	4.42 r.v	4.07 e	3.26 c
(µ mol	Initiation	D2	0.52 z	1.85 w.z	2.15 v.z	3.42 t.y	1.50 x.z	2.69 u.z	2.32 u.z	1.94 w.z	5.73 p.t	2.46 f	
min^{-1} mg	Booting	D1	8.11 l.p	7.22 n.q	13.32 f.h	16.78 b.d	10.88 h.k	9.10 k.n	7.99 l.p	8.32 l.o	7.43 n.p	9.91 c	8.98 b
$protein^{-1}$)		D2	6.46 o.r	4.77 q.u	7.88 m.p	10.84 i.k	8.11 l.p	7.77 m.p	9.21 k.n	7.95 l.p	9.45 j.n	8.05 d	
	Heading	D1	14.55 c.g	11.67 h.j	17.67 b	25.67 a	14.22 e.g	13.33f.h	14.55 c.g	11.45 h.k	15.997b.e	15.45 a	14.48 a
		D2	12.77 f.i	9.89 j.m	15.21 c.f	16.89 bc	12.55 g.i	15.92b.e	14.22 e.g	9.66 j.n	14.44 d.g	13.50 b	
		Cultivars	7.54 de	6.60 e	10.38 b	13.99 a	8.53 cd	8.54 c	8.23 cd	6.76 e	9.58 b		
LSD values (0.01) Field capacity, cultivars, critical growth stages individual and three way interaction 0.8169, 1.0005, 0.5776 and 2.4506 respectively													
SOD	Leaf	D1	18.22 p.y	7.44 yz	18.33 p.y	69.09 ab	21.95 m.v	21.47 n.v	44.48 e.g	29.96 i.o	20.24 o.w	27.91 c	25.94 c
(IU min^{-1}	Initiation	D2	24.06 k.u	4.46 z	8.14 x.z	53.86 c.e	11.08 v.z	27.96 i.q	25.80 k.t	25.12 k.t	35.21 g.k	23.97 d	
mg^{-1}	Booting	D1	25.66 k.t	15.22 t.z	25.10 k.t	73.11 ab	26.66 j.s	27.54 i.r	48.12 d.f	31.46 i.n	26.67 j.s	33.28 b	30.57 b
protein)		D2	23.83 l.u	9.54 w.z	15.78 s.y	57.33 cd	16.43 r.y	31.45 i.n	31.77 i.n	27.56 i.r	37.10 f.j	27.86 c	
	Heading	D1	28.77 i.q	17.99 q.y	29.21 i.p	76.90 a	28.56 i.q	33.67 g.l	53.33 c.e	38.10 f.i	32.89 h.m	37.71 a	34.78 a
		D2	25.81 k.t	13.66 u.z	21.11 n.v	62.48 bc	18.76 p.x	32.88 h.m	34.88 g.l	34.10 g.l	42.99 e.h	31.85 b	
		Cultivars	24.39 d	11.38 f	19.61 e	65.46 a	20.57 de	29.16 c	39.73 b	31.05 c	32.52 c		
LSD values (0.01) Field capacity, cultivars, critical growth stages individual and three way interaction 3.7221, 4.5586, 2.6319 and 11.166 respectively													
AsA	Leaf	D1	86.14 m.p	49.50 za	71.48 wx	101.69 cd	78.91 r.t	93.24 g.k	64.59 y	92.47 h.k	71.74 v.x	78.86 c	79.80 c
(m.	Initiation	D2	85.48 n.p	41.19 b	64.09 y	99.09 c.f	84.84 n.q	97.05 d.h	86.14 m.p	91.79 i.l	77.09 s.u	80.75 b	
mole g^{-1})	Booting	D1	92.44 h.k	51.77 z	74.88 t.w	106.77 ab	82.66 o.r	97.33 c.g	68.32 xy	100.00 c.e	80.44 q.s	83.84 a	82.96 b
		D2	88.89 k.n	50.22 za	67.43 xy	97.22 d.h	82.22 p.r	93.45 g.k	87.04 l.o	94.88 f.j	77.34 s.u	82.07 b	
	Heading	D1	92.11 i.k	45.65 ab	76.14 s.w	110.21 a	84.55 n.q	100.21 c.e	73.33 u.w	102.06 bc	76.54 s.v	84.53 a	84.78 a
		D2	87.10 l.o	53.76 z	64.78 y	101.77 cd	90.60 j.m	94.66 f.j	92.44 h.k	96.33 e.i	83.78 o.q	85.02 a	
		Cultivars	88.69 c	48.68 g	69.80 f	102.79 a	83.96 d	95.99 b	78.65 e	96.26 b	77.82 e		
LSD values (0.01) Field capacity, cultivars, critical growth stages individual and three way interaction 1.6062, 1.9672, 1.1358 and 4.8186 respectively													

Means not sharing the same letters differ significantly at 1% probability level.

Table 4. Status of non-antioxidants and mineral contents of wheat cultivars at leaf initiation, booting and heading stages under drought stress conditions (50% & 100% Field Capacity)

Antioxidants & minerals		Field capacity	Fareed-06	Millat-11	Miraj-08	AARI-11	Lassani-06	AAS-11	Shafaq-06	Sahar-06	Punjab-11	Mean	Mean
TPC	Leaf	D1	1.15 y	2.87 s.y	6.19 o	18.17 e.g	1.54 xy	4.00 p.v	4.73 o.s	10.24 l.n	4.03 p.v	5.88 d	4.32 c
(mg g^{-1})	Initiation	D2	2.40 t.y	1.94 v.y	3.49 r.x	4.33 o.t	2.40 t.y	2.07 u.y	2.43 t.y	4.05 o.v	1.70 w.y	2.76 f	
	Booting	D1	4.98 o.s	4.77 o.s	8.64 n	20.23 e	3.93 p.v	4.10 o.u	5.75 op	13.33 i.k	5.23 o.r	7.88 c	6.18 b
		D2	3.67 p.x	3.23 r.y	4.22 o.u	8.99 mn	2.33 t.y	3.78 p.w	4.76 o.s	5.68 o.q	3.56 q.x	4.47 e	
	Heading	D1	19.97 ef	14.67 hi	23.65 d	64.77 a	13.56 ij	16.77 gh	31.21 b	27.73 c	24.58 d	26.32a	21.48 a
		D2	11.23 kl	12.01 j.l	17.97 fg	33.11 b	10.87 lm	13.56 ij	16.91 g	19.74 ef	14.33 i	16.6 b	
		Cultivars	7.23 e	6.58 ef	10.69 c	24.93 a	5.77 f	7.38 e	10.97 c	13.46 b	8.90 d		
LSD values (0.01) Field capacity, cultivars, critical growth stages individual and three way interaction 0.7184, 0.8798, 0.5080 and 2.1551 respectively													
K^+	Leaf	D1	0.88 k.n	0.35 s.u	1.16 h.k	1.79 ab	0.54 p.t	0.38 s.u	0.72 l.q	1.50 c.g	0.73 l.q	0.90 d	0.76 c
contents	Initiation	D2	0.721 l.q	0.39 r.u	0.68 m.r	1.31 e.h	0.24 u	0.31 tu	0.55 o.t	0.51 q.u	0.88 k.n	0.62 e	
(mg g^{-1})	Booting	D1	1.34 e.h	0.74 l.q	1.43 d.h	1.83 a	0.99 i.l	0.51 q.u	0.81 l.p	1.47 c.g	0.74 l.q	1.09 b	1.04 b
		D2	1.21 g.j	0.76 l.q	1.25 f.i	1.59 a.e	0.85 l.n	0.54 p.t	0.64 n.s	0.78 l.q	1.33 e.h	0.99 c	
	Heading	D1	1.57 a.e	0.84 l.o	1.60 a.e	1.75 a.c	0.95 j.m	0.85 l.n	0.87 k.n	1.54 a.f	0.79 l.q	1.20 a	1.15 a
		D2	1.37 d.h	0.76 l.q	1.43 d.h	1.52 b.f	0.79 l.q	0.78 l.q	0.84 l.o	0.75 l.q	1.67 a.d	1.10ab	
			1.186 bc	0.64 ef	1.26 b	1.63 a	0.73 e	0.56 f	0.74 e	1.09 cd	1.02 d		
LSD values (0.01) Field capacity, cultivars, critical growth stages individual and three way interaction 0.0981, 0.1202, 0.0694 and 0.2943 respectively													
Leaf Chlorophyll		D1	0.84 cd	0.91 c	0.16 h	2.53 a	0.51 e.g	0.84 cd	1.23 b	0.64 d.f	1.43 b	1.01 a	
"*a*" (mg g^{-1})		D2	0.19 h	0.18 h	0.61 d.f	0.77 c.e	0.26 gh	0.11 h	0.64 d.f	0.12 h	0.49 fg	0.37 b	
		Cultivars	0.51 c	0.55 c	0.38 c	1.65 a	0.38 c	0.47 c	0.93 b	0.38 c	0.96 b		
LSD values (0.01) Field capacity, Cultivars and Field capacity×Cultivars 0.0868, 0.1841 and 0.2603 respectively													
Leaf Chlorophyll		D1	0.39 gh	0.33 hi	1.23 de	2.08 a	1.10 de	1.37 cd	1.67 bc	0.04 i	0.99 ef	1.02 a	
"*b*" (mg g^{-1})		D2	0.05 i	0.10 hi	0.66 g	1.86 ab	1.57 bc	1.17 de	0.06 i	0.67 fg	1.04 e	0.80 b	
		Cultivars	0.22 d	0.21 d	0.94 c	1.97 a	1.34 b	1.27 b	0.86 c	0.35 d	1.01 c		
LSD values (0.01) Field capacity, Cultivars and Field capacity×Cultivars 0.2303, 0.1086 and 0.3257 respectively													

Means not sharing the same letters differ significantly at 1% probability level.



100% Field capacity (Non-reduced) 50% Field capacity (Non-reduced)

1. Fareed-06, 2. Millat -11 3. Miraj-08 4. AARI-11 5. Lasani-06 6. AAS-11 7. Shafaq-06 8. Sahar-06 9. Punjab-11

Figure 2. Protein profiling of wheat cultivars in response to drought stress conditions.

up to its maximum at heading under 100% FC and 50% FC, respectively (Table 3). The performance of AARI-11 was better for activities of POD, CAT, SOD, AsA and TPC contents (Table 4) which were highest at heading stage as compared to leaf initiation stage at 50% water regime than 100% and lowest in Millat-11. TPC was lowest in Lassani-06 at booting and heading but Fareed-06 showed its lowest values at leaf initiation stage. The highest chlorophyll contents "*a*" and "*b*" at heading stage were observed in AARI-11 while other varieties showed variable results under both water regimes (Table 4). Comparing the data regarding leaf K^+ contents, wheat cultivars showed significant results among two field capacity levels at leaf initiation and booting stages but maximum values were observed at heading stage under both watered treatments (Table 4). Electrophorogram showed the grain proteinaceous bands of different wheat varieties under 100% FC and 50% FC (Fig. 2). The results depicted variations in bands on both water levels (well-watered and stressed watered) from 10 to 60 KDa but the major band was shown at 75-76 KDa in AARI-11 under 50% FC whereas no clear bands were obtained in gel for grain protein of plants grown at 100% FC.

DISCUSSION

Deficiency of water affected the germination of studied wheat cultivars as compared to well-watered condition. Highest final germination percentage and germination index observed in AARI-11 might be due to its genetic tolerance to face the scarcity of water, germination potential and vigor of seeds (Khakwani *et al*., 2011). Drought stress imposed negative effects on the anthesis stage which led to minimum number of grains per spike in Millat-11 and AAS-11 but AARI-11 was least affected and produced the largest number of grains (Shirazi *et al*., 2014). Cultivar AARI-11 recorded maximum 100-grain weight and grain yield per plant. It may be due to efficient photosynthetic activities by increased nutrients uptake and more Chlorophyll "*a"* and "*b"* contents in plant leaves at the flowering stage (Baque *et al*., 2006) which maintained the green appearance in plant leaves (Moucheshi *et al*., 2012). On the other hand, Millat-11, Punjab-11 and Lasani-11 showed least outputs. Siddique *et al*. (2000) observed the similar results and suggested, it might be due to the shrinkage of grain size under shortage of water.

Superoxide dismutase, catalase and peroxidase enzymes are the important antioxidants which help the plants to tolerate drought like situation. In AARI-11, higher enzymatic (SOD, CAT, POD) activities especially at reproductive stages might be due to more H_2O_2 scavenging system which is actively involved in detoxification of oxidative stress induced by water deficiency (Nazarli and Faraji, 2011; Mafakheri *et al*., 2011; Saed-moucheshi *et al*., 2012). Ascorbic acid (AsA) plays a significant role in non-enzymatic antioxidant which protects the plant from the damaging impact of reactive oxygen species. Maximum production of AsA at vegetative and reproductive stages of all wheat cultivars might have triggered the antioxidant system for effectively defense against ROS and improve the photosynthetic activities in plants (Amira and Qados, 2014). The reduction in the total phenolic contents (TPC) at vegetative (leaf initiation) and reproductive (booting and heading) stages caused the maximum breakdown of photosynthetic pigments like chlorophyll contents in the wheat cultivars under drought stress. The maximum concentration of K^+ contents in AARI-11 at all critical stages might have increased the stomatal conductance whereas Millat-11 as a sensitive variety showed the lowest values of K^+ contents not only in drought stress but also in normal watered condition (Yasmeen *et al*., 2013).

The major band of grain protein in AARI-11 under stressed condition of 50% FC illustrated high proteolytic activity and genetic tolerance to face the drought conditions as compared to 100% FC. Millat-11 proved the sensitive wheat cultivar by producing the negligible protein banding pattern under both well-watered and stressed conditions. This result is related to the work of Shuaib *et al*. (2010) so it is concluded that the observed protein of 76-78 KDa might be glutenin in AARI-11 wheat cultivar.

***Conclusion*:** Taking in conjunction the results of the present study, it is concluded that AARI-11 showed prominent performance at various critical growth stages and proved as a tolerant wheat cultivar under water deficit condition.

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