

## Comparative Study on the Effect of Soil Water Stress on Photosynthetic Function of Triticale, Bread Wheat, and Barley

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### ABSTRACT

In order to compare photosynthetic features of triticale, bread wheat, and barley under water deficit conditions, this experiment was carried out in Kurdistan province in western Iran. Four genotypes of triticale, three cultivars of bread wheat, and a new variety of barley were compared under well watered ( $\Psi_{\text{soil}} = -3$  bars) and soil water deficit ( $\Psi_{\text{soil}} = -12$  bars) conditions in a randomized complete block design (RCBD) with a split-plot arrangement. Biomass production at anthesis accompanied with gas exchanges and chlorophyll fluorescence of flag leaves, as indicators of photosynthetic function of plant, were determined in each treatment. The results showed that by imposing water deficit, photosynthetic rate ( $P_n$ ) was reduced in all studied genotypes irrespective of plant species. Under water deficit conditions, the highest  $P_n$  ( $6.86$  and  $5.90 \mu\text{mol m}^{-2} \text{s}^{-1}$ ) was found in bread wheat variety Pishgam and, triticale genotype No. 1, while the lowest value ( $3.63 \mu\text{mol m}^{-2} \text{s}^{-1}$ ) was found in barley variety Bahman. Similar trend was observed for the maximum quantum yield of primary photochemistry ( $F_v/F_m$ ) and performance index (PI). Significant positive correlations were found between biomass production at anthesis and both  $P_n$  ( $r = 0.83^*$ ) and  $F_v/F_m$  ( $r = 0.77^*$ ). Our data revealed the better performance of Pishgam than Alvand and Zarrin in the reduction rate of biomass at anthesis and photosynthetic features against soil water deficit conditions. Overall, triticale was less affected by water deficit in comparison with wheat and barley in terms of photosynthetic function as indicated by less reduction in  $P_n$ , PI, and  $F_v/F_m$ .

**Keywords:** Barley, Bread wheat, Chlorophyll fluorescence, Gas exchanges, Triticale.

### INTRODUCTION

Long-term dehydration in C3 plants impairs various physiological processes and, especially, inhibits photosynthesis (Chaves, 1991; Hura *et al.*, 2007) through decreased  $\text{CO}_2$  availability by limiting both stomatal and non-stomatal factors (Flexas *et al.*, 2004; Subrahmanyam *et al.*, 2006; Chaves *et al.*, 2009). These effects vary with the duration and intensity of drought, as well as by plant species (Lawlor and Cornic, 2002; Munns, 2002; Flexas *et al.*, 2004; Galme's *et al.*, 2007). Stomata close in response to drought, therefore, the supply of  $\text{CO}_2$  is limited, and the photosynthetic apparatus becomes

predisposed to increase energy dissipation and down-regulation of photosynthesis when the plant is subjected to high light and temperature (Chaves *et al.*, 2009). Although triticale has been shown to have higher dry matter accumulation and photosynthetic rate than wheat (Winzeler *et al.*, 1987; Bobodzhanov *et al.*, 1990) some studies have suggested that tetraploid wheat had higher water use efficiency than triticale (T300) under drought (Morant-Avice *et al.*, 1994).

Chlorophyll fluorescence has been used as a rapid technique to estimate the operating quantum efficiency of electron transport throughout PS II in leaves (Genty *et al.*, 1989) and its relationship with  $\text{CO}_2$  assimilation (Siebke *et al.*, 1997). The genetic

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variability of the net photosynthetic rate, its relative parameters, and chlorophyll fluorescence have been reported previously for small grain cereals (Sarraf *et al.*, 1989; Grzesiak *et al.*, 2003; Jiang *et al.*, 2006; Hura *et al.*, 2007; Grzesiak *et al.*, 2007). Although these three cereals have been separately investigated in terms of their chlorophyll fluorescence (Lu and Zhang, 1998; Hura *et al.*, 2007; Guo *et al.*, 2009), comparative studies have not been designed clearly for the assessment of any advantages of triticale under conditions of considerable water limitation.

A better understanding of the physiological traits that enable triticale plant to adapt to drought stress and maintain growth, development, and productivity during stress periods would help in breeding for drought resistance. We hypothesized that there were differences in physiological responses among triticale, bread wheat, and barley species under long-term drought stress. Therefore, the comparative study presented here was carried out under limiting water conditions in order to (i) compare the biomass performance and the changes in gas exchange and chlorophyll fluorescence, as methods to study both stomatal and non-stomatal limitations effects on photosynthesis and (ii) determine if any of these parameters may be useful as selection criteria for the breeding of different types of cereals for tolerance to water stress.

## MATERIALS AND METHODS

### Plant Materials and Growth Conditions

The experiment was conducted during the growing season of 2009-2010 at the Experimental Field of Grizeh Station in Kurdistan Province, west of Iran. The experimental design was a randomized complete block design (RCBD) with a split-plot arrangement. Two water regimes were used for the main plots, which comprised sub-plots with eight genotypes, including four spring triticale lines, three bread wheat cultivars, and one barley variety (Table 1), as follows: irrigation to achieve a soil water potential of -3 bars ( $\Psi_{\text{soil}} = -3$  bars) i.e. well watered treatment (WW), and irrigation to achieve a soil water potential of -12 bars ( $\Psi_{\text{soil}} = -12$  bars) i.e. water deficit treatment (WD). Plots were replicated three times. Irrigation was set according to the moisture retention curve of the soil. During rainy days, a mobile rain shelter for each block (6 × 10 × 2.5 m) was used in the WD treatment to prevent infiltration of the rain, consequently, the soil moisture was retained at a level of -12 bars. To limit the lateral penetration into the soil, a drainage ditch was dug around the rain shelter. A drip irrigation system with drip lines between the crop rows was used. Irrigation was applied from the developmental stage of

**Table 1.** The list of crop species and genotypes with their growth habit and source.

Crop	No	Variety/Pedigree	Growth habit	Source
Triticale	No. 1	ET-79-17 ARDI-1/TOPO 1419//ERIZO-9 cty 87352	Spring	CIMMYT <sup>a</sup>
	No. 2	Juanillo-092	Spring	CIMMYT
	No. 3	ET-82-15 RONDO/BANT_5//ANOAS_2/3/VICUNA_4	Spring	CIMMYT
	No. 4	ET-82-18 SRIER-29/FARS-1//MANATI-1	Spring	CIMMYT
Bread Wheat	No. 5	Alvand	Winter	SPII <sup>b</sup>
	No. 6	Pishgam	Winter	SPII
	No. 7	Zarrin	Winter	SPII
Barley	No.8	Bahman	Winter	SPII

<sup>a</sup> International Maize and Wheat Improvement Center.

<sup>b</sup> Seed and Plant Improvement Institute of Iran.

stem elongation onwards. A water flow meter and a Time-Domain Reflectometry (TDR) were used to measure the amount of applied irrigation water and soil water content, respectively. Conventional cultural practices were implemented, including the usual sowing rate, fertilizer application, and post-emergence herbicide application, followed by hand-hoeing where necessary to control the weeds. Planting was performed on 23 October, and ample rainfall occurred on 28 October, which helped crop emergence. Each plot consisted of 6 rows, 20 cm apart.

### Measurements

The date of anthesis was recorded for each plot when 50% of the spikes of the main shoots had either visibly exerted anthers or when the anthers that had dehisced were observed through the palea (Estrada-Compuzano *et al.*, 2008). Three flag leaves were labeled and representative physiological traits, including gas exchanges, chlorophyll fluorescence parameters, water loss and relative water content were measured. To determine the biomass yield at anthesis (BYAA), 50 plants were harvested and oven dried at 80 °C for 48 hours and then weighed. The reduction in value of each variable by water deficit treatment, WD, which was taken as an approximate representation of its sensitivity to water stress, was calculated according to Blum *et al.* (1983), as follows:

$$\% \text{ reduction} = 100 \times (WW - WD) / WW \quad (1)$$

Where, *WW* and *WD* are the values of the variables under the control and stress conditions, respectively

### Leaf Gas Exchange

The leaf gas exchanges were measured on three labeled, fully expanded, flag leaves using a gas analyzer device (IRGA.ADC, LCA4.UK). The net photosynthesis rate (*P<sub>n</sub>*), transpiration rate per leaf area (*E*), stomatal conductance (*g<sub>s</sub>*), internal concentration of CO<sub>2</sub> (*C<sub>i</sub>*), and apparent carboxylation

efficiency (*P<sub>n</sub>/C<sub>i</sub>*) were determined at a light intensity of 1,200-1,600 μmol (photon) m<sup>-2</sup> s<sup>-1</sup> and ambient CO<sub>2</sub> concentration of 380-400 ppm.

### Chlorophyll Fluorescence Parameters

The same three flag leaves labeled to measure the gas exchanges were fully dark-adapted to determine chlorophyll fluorescence parameters using a plant stress meter (Handy PEA V1.3, U.K). Based on definitions of Yusuf *et al.* (2010) and Han *et al.* (2009), the following parameters were determined:

*F<sub>v</sub>/F<sub>m</sub>*= Maximum quantum yield of primary photochemistry, PS II;

*F<sub>v</sub>/F<sub>o</sub>*= Ratio of the variable fluorescence to the ground fluorescence (Maxwell and Johnson, 2000);

*PI*= Performance index (potential) for energy conservation from excitation to the reduction of PSI end acceptors;

*DIO/CS<sub>o</sub>*= Dissipated energy flux per excited cross section at *t*= 0,

*RC/CS<sub>m</sub>*= Maximum number of active reaction centers.

### Water Relations

Relative water content and water loss were determined to express water relations. Five flag leaves of each genotype from the WW treatments were sampled to determine the water loss (WL). The leaves were cut and weighed (*fw<sub>1</sub>*), allowed to desiccate in a dark room at 25°C and, after 24 hours, reweighed (*fw<sub>2</sub>*). The leaves were then oven dried at 80°C (*dw*) for 24 hours, and the water loss was calculated by the following formula (Grzesiak *et al.*, 2003):

$$WL = (fw_1 - fw_2) \times dw^{-1} \quad (2)$$

### Relative Water Content

Relative water contents (RWC) were determined for detached leaves. The relative water contents (RWC) were calculated from



flag leaf blades using the method devised by Mata and Lamattina (2001) with the help of the following equation:

$$RWC (\%) = (FW - DW) / (TW - DW) \times 100 \quad (3)$$

The fresh weight (FW) was measured immediately after excision, the full turgid weight (TW) was determined after rehydration of the leaves placing them in a test tube containing distilled water for 24 hours at 4°C in darkness, and the dry weight (DW) was determined after oven drying at 80°C for 48 hours.

### Statistical Analysis

All of the data were analyzed using one-way analysis of variance. The means  $\pm$  SE were used to compare the data. Simple correlation analyses were used to determine the relationships between the measured parameters.

## RESULTS

The total rainfall in the 2009-2010 growing season was 455 mm, compared to the long-term average of 450 mm. However, the rainfall pattern showed a normal distribution during the growth season. Approximately 40% of the seasonal rain occurred during autumn and 29% during winter, whereas 31% of the rain fell during spring, from the end of March to the middle of May. The rainfall was normally distributed, where no increase in competition for the limited resources (mainly water) occurred during the vegetative and early reproductive growth stages, except in the stress treatment, in which the plants were grown under a rain shelter.

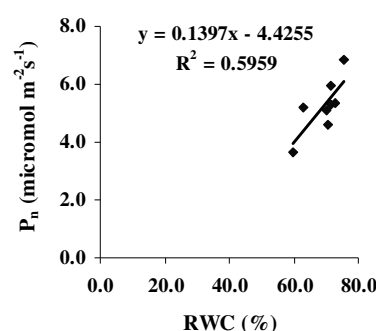
### Biomass Production at Anthesis

Water stress ( $\Psi_{\text{soil}} = -12$  bars) significantly decreased the biomass at anthesis in our

tested genotypes (Table 2). This reduction rate ranged from 17% in triticale No. 1 to 38% in barley variety Bahman. However, genotypes did not differ much in the dry matter production under soil water deficit, with the exception of Bahman. We compared the studied species based on their response to water stress using the parameter of the percent reduction for each trait. The mean biomass yield reductions due to water deficit were 26, 29, and 38% for, respectively, triticale, wheat, and barley.

### Water Loss and Relative Water Content

A significant variation ( $P \leq 0.05$ ) was found among the genotypes for the parameter of water loss (WL). Generally, the amount of WL in triticale ( $0.259 \text{ g g}^{-1} \text{ dw}$ ) was less than wheat ( $0.305 \text{ g g}^{-1} \text{ dw}$ ) and barley ( $0.557 \text{ g g}^{-1} \text{ dw}$ ) (Table 2), suggesting more relative water absorption or water maintenance in triticale when faced with drought. In terms of relative water content (RWC), a decrease occurred in all the genotypes under water stress, a sharp decline in barley (23%) was detected, which led to the lowest values for gas exchange and fluorescence, as described below. The rate of decline in Pishgam was less than the other two wheat varieties (Table 2). Under water deficit a significant relationship was found between RWC and  $P_n$  (Figure 1).



**Figure 1.** Relationship between photosynthetic rate ( $P_n$ ) and relative water content (RWC).

**Table 2.** Mean biomass yield at anthesis (BYAA), relative water content (RWC), leaf temperature (LT), and gaseous exchanges under well watered (WW,  $\Psi_{\text{soil}} = -3$  bars) and water deficit (WD,  $\Psi_{\text{soil}} = -12$  bars) conditions. The data are shown as Mean $\pm$ SE.

trait	treatment	No. 1	No. 2	No. 3	No. 4	Alvand	Pishgam	Zarrin	Bahman
BYAA (g plant <sup>-1</sup> )	WW	4.1 $\pm$ 0.12	5.1 $\pm$ 0.15	4.8 $\pm$ 0.17	4.8 $\pm$ 0.16	4.8 $\pm$ 0.11	4.5 $\pm$ 1.51	4.7 $\pm$ 0.15	3.5 $\pm$ 0.08
	WD	3.4 $\pm$ 0.19	3.4 $\pm$ 0.12	3.6 $\pm$ 0.05	3.5 $\pm$ 0.27	3.0 $\pm$ 0.43	3.7 $\pm$ 0.25	3.3 $\pm$ 0.05	2.2 $\pm$ 0.21
	% reduction	17	33	25	28	37	19	30	38
RWC (%)	WW	80.0 $\pm$ 6.55	83.5 $\pm$ 5.02	82.6 $\pm$ 3.99	80.2 $\pm$ 4.28	81.0 $\pm$ 5.28	79.5 $\pm$ 2.92	79.4 $\pm$ 1.93	77.4 $\pm$ 5.03
	WD	71.3 $\pm$ 0.17	71.3 $\pm$ 1.37	62.8 $\pm$ 0.71	72.8 $\pm$ 3.74	70.4 $\pm$ 2.28	75.4 $\pm$ 1.41	70.1 $\pm$ 3.25	59.8 $\pm$ 5.09
	% reduction	11	15	24	9	20	5	12	23
E (mmol m <sup>-2</sup> s <sup>-1</sup> )	WW	5.0 $\pm$ 0.26	53 $\pm$ 0.26	5.1 $\pm$ 0.69	5.6 $\pm$ 0.95	5.4 $\pm$ 0.77	5.9 $\pm$ 0.64	6.0 $\pm$ 0.58	4.9 $\pm$ 0.91
	WD	3.5 $\pm$ 0.58	3.19 $\pm$ 0.38	3.02 $\pm$ 0.13	3.32 $\pm$ 0.20	3.24 $\pm$ 0.45	4.29 $\pm$ 0.71	3.51 $\pm$ 0.44	2.8 $\pm$ 0.41
	% reduction	30	39	41	41	49	27	41	43
C <sub>i</sub> (ppm)	WW	250.7 $\pm$ 6.6	238.4 $\pm$ 12.9	244.8 $\pm$ 30.5	231.7 $\pm$ 13.5	234.7 $\pm$ 11.3	242.2 $\pm$ 9.8	260.1 $\pm$ 4.3	247.5 $\pm$ 19.2
	WD	192.7 $\pm$ 19.3	210.9 $\pm$ 13.1	202.4 $\pm$ 12.6	226.2 $\pm$ 20.8	217.4 $\pm$ 2.2	210.7 $\pm$ 10.1	224.1 $\pm$ 10.0	242.1 $\pm$ 9.4
	% reduction	23	12	17	9	9	13	14	2
P <sub>n</sub> ( $\mu$ mol m <sup>-2</sup> s <sup>-1</sup> )	WW	7.9 $\pm$ 0.87	8.8 $\pm$ 0.64	9.5 $\pm$ 1.59	10.6 $\pm$ 1.55	10.3 $\pm$ 0.59	11.8 $\pm$ 1.66	9.7 $\pm$ 0.02	7.9 $\pm$ 2.47
	WD	5.9 $\pm$ 1.26	5.3 $\pm$ 0.70	5.2 $\pm$ 0.33	5.4 $\pm$ 1.77	6.1 $\pm$ 1.70	6.9 $\pm$ 1.04	5.1 $\pm$ 1.19	3.6 $\pm$ 92
	% reduction	24	39	45	50	56	42	47	54
g <sub>s</sub> (mmol m <sup>-2</sup> s <sup>-1</sup> )	WW	150.0 $\pm$ 17.0	150 $\pm$ 5.0	180.0 $\pm$ 36.0	186.0 $\pm$ 29.6	186.0 $\pm$ 29.6	226.6 $\pm$ 18.0	216.1 $\pm$ 8.0	153.1 $\pm$ 31.1
	WD	80.0 $\pm$ 36	66.0 $\pm$ 14.0	63.0 $\pm$ 3.0	86.6 $\pm$ 42.5	80.0 $\pm$ 26.1	103.3 $\pm$ 31.1	93.3 $\pm$ 43.7	53.3 $\pm$ 12.3
	% reduction	47	56	65	54	71	54	57	65
MC (mmol m <sup>-2</sup> s <sup>-1</sup> )	WW	31.0 $\pm$ 1.0	37.0 $\pm$ 4.0	41.0 $\pm$ 11.0	46.6 $\pm$ 8.3	43.6 $\pm$ 1.0	49.4 $\pm$ 9.1	37.0 $\pm$ 0.001	38.1 $\pm$ 16.0
	WD	30 $\pm$ 3.0	25.0 $\pm$ 3.0	25.0 $\pm$ 2.9	23.9 $\pm$ 7.0	28.0 $\pm$ 8.3	30.1 $\pm$ 2.1	22.4 $\pm$ 7.0	15.4 $\pm$ 4.4
	% red	3	32	37	45	51	38	39	60
WL (g g <sup>-1</sup> dw)		0.22 $\pm$ 0.02	0.29 $\pm$ 0.01	0.26 $\pm$ 0.06	0.26 $\pm$ 0.03	0.31 $\pm$ 0.04	0.23 $\pm$ 0.001	0.36 $\pm$ 0.02	0.56 $\pm$ 0.09

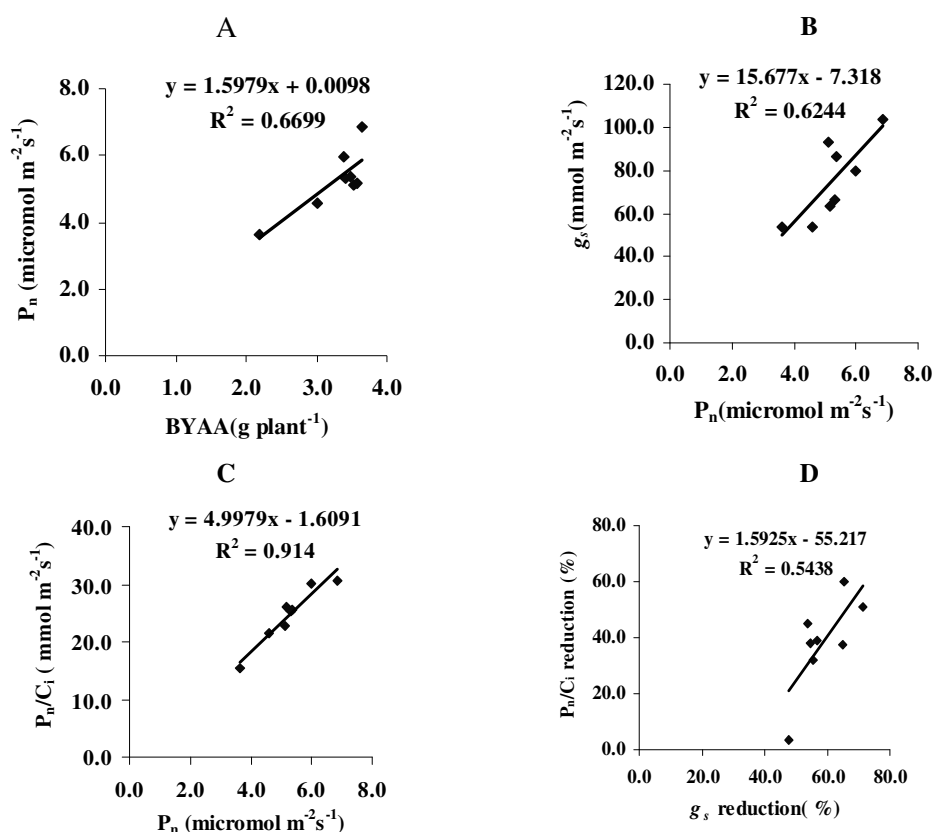


## Gaseous Exchanges

Leaf gas exchange parameters indicated that under soil moisture stress  $P_n$ ,  $g_s$ ,  $E$ ,  $C_i$ , and  $P_n/C_i$  declined in all genotypes tested. The mean decreases were 45%, 59%, 39%, 10%, and 37%, respectively (Table 2). By the imposition of water stress, the reduction of  $P_n$  ranged from 24% for triticale No. 1 to 54% for Bahman. However, on the average, these reductions were 39, 48, and 54% in triticale, wheat, and barley, respectively. In contrast to the triticale No. 1, which had a low  $P_n$  under normal condition ( $7.85 \pm 0.87 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ ), the Pishgam had higher  $P_n$  in both stress ( $11.8 \pm 1.66 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ ) and non-stress ( $6.86 \pm 1.04 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ ) conditions, suggesting high ability of photosynthetic machinery in Pishgam under different soil moisture regimes. This might have resulted in

more stability of this cultivar in the case of  $P_n$ . A significant and positive relationship was found between  $P_n$  and BYAA (Figure 2-A).

A gradual reduction in  $g_s$  (47-71%) and  $P_n/C_i$  (20-58%) was observed (Table 2) under water deficit. Average reduction of  $g_s$  in barley (65%) was more than wheat (61%) and triticale (55%). The lowest reduction of  $g_s$  was observed in triticale No. 1 (47%), and the highest was found in Alvand (71%), with similar pattern for  $P_n/C_i$ , suggesting an increased susceptibility of stomatal conductivity to water stress, resulting in an increase in the apparent mesophyll resistance, which can be differentiated among different types of cereals. However, under soil water deficit, Pishgam had the highest  $g_s$  ( $103.3 \pm 31.1 \text{ mmol m}^{-2} \text{ s}^{-1}$ ) and Bahman had the lowest ( $53.3 \pm 12.3 \text{ mmol}$



**Figure 2.** Correlations of photosynthetic rate,  $P_n$  and biomass yield at anthesis, BYAA (A),  $P_n$  and stomatal conductance,  $g_s$  (B),  $P_n$  and apparent carboxylation efficiency,  $P_n/C_i$  (C), % reduction of  $P_n/C_i$  and % reduction of  $g_s$  (D) among four genotype of triticale, three genotypes of bread wheat and one genotype of barley under water deficit ( $\Psi_{\text{soil}} = -12 \text{ bar}$ ).

**Table 3.** Mean fluorescence parameters under well watered (WW,  $\Psi_{\text{soil}} = -3$  bars) and water deficit (WD,  $\Psi_{\text{soil}} = -12$  bars) conditions. The data are shown as Mean $\pm$ SE. Negative values represent the percent increase by water deficit.

trait	treatment	No. 1	No. 2	No. 3	No. 4	Alvand	Pishgam	Zarrin	Bahman
$F_v/F_m$	WW	0.75 $\pm$ 0.02	0.78 $\pm$ 0.001	0.73 $\pm$ 0.01	0.76 $\pm$ 0.01	0.74 $\pm$ 0.01	0.78 $\pm$ 0.01	0.79 $\pm$ 0.01	0.79 $\pm$ 0.01
	WD	0.74 $\pm$ 0.02	0.73 $\pm$ 0.02	0.72 $\pm$ 0.01	0.73 $\pm$ 0.02	0.67 $\pm$ 0.05	0.76 $\pm$ 0.02	0.69 $\pm$ 0.02	0.67 $\pm$ 0.03
	% reduction	1	6	1	3	11	4	13	15
$F_v/F_o$	WW	3.1 $\pm$ 0.47	3.5 $\pm$ 0.001	2.7 $\pm$ 0.14	3.2 $\pm$ 0.29	2.9 $\pm$ 0.25	3.6 $\pm$ 0.23	3.8 $\pm$ 0.28	3.8 $\pm$ 0.29
	WD	2.9 $\pm$ 0.38	2.5 $\pm$ 0.31	2.6 $\pm$ 0.16	2.8 $\pm$ 0.31	2.2 $\pm$ 0.47	2.5 $\pm$ 0.47	2.3 $\pm$ 0.26	2.2 $\pm$ 0.42
	% reduction	6	28	2	12	25	29	24	39
$PI$	WW	17.7 $\pm$ 2.11	25.4 $\pm$ 1.93	13.59 $\pm$ 0.42	19.43 $\pm$ 2.5	22.7 $\pm$ 5.03	36.1 $\pm$ 0.38	30.15 $\pm$ 4.40	18.93 $\pm$ 1.28
	WD	16.9 $\pm$ 1.08	18.4 $\pm$ 1.88	12.13 $\pm$ 0.45	11.5 $\pm$ 0.17	14.8 $\pm$ 2.68	34.1 $\pm$ 2.32	8.01 $\pm$ 1.08	3.88 $\pm$ 0.44
	% reduction	5	28	11	38	35	6	72	79
$DIo/CS$	WW	19.9 $\pm$ 0.39	19.4 $\pm$ 0.96	24.5 $\pm$ 1.14	19.2 $\pm$ 0.29	19.9 $\pm$ 1.8	20.8 $\pm$ 0.71	18.9 $\pm$ 1.56	20.2 $\pm$ 0.15
	WD	20.5 $\pm$ 0.63	23.3 $\pm$ 0.72	24.2 $\pm$ 2.42	24.3 $\pm$ 0.02	22.3 $\pm$ 1.20	21.4 $\pm$ 0.52	31.9 $\pm$ 2.25	35.9 $\pm$ 1.1
	% reduction	0	-21	0	-26	-8	-2	-72	-78
$RC/CSm$	WW	273.6 $\pm$ 37.6	353.5 $\pm$ 14.8	273.0 $\pm$ 8.4	242.2 $\pm$ 27.5	352.5 $\pm$ 32.6	442.0 $\pm$ 26.2	404.7 $\pm$ 34.1	380.5 $\pm$ 13.0
	WD	232.8 $\pm$ 24.0	275.3 $\pm$ 17.5	274.0 $\pm$ 33.2	212.2 $\pm$ 8.3	286.5 $\pm$ 14.2	401.8 $\pm$ 25.4	268.3 $\pm$ 8.43	183.1 $\pm$ 16.9
	% reduction	13	21	0	12	19	9	33	52

$\text{m}^{-2} \text{s}^{-1}$ ), suggesting more susceptibility of this barley genotype to soil moisture limitation.

The studied species exhibited significant reduction in transpiration rate,  $E$ , by soil water deficit conditions. A relatively high level of transpiration rate under stress condition ( $4.29 \pm 0.71 \text{ mmol m}^{-2} \text{s}^{-1}$ ) concomitant with low reduction (27%) was found for Pishgam. A similar trend was observed for triticale No. 1 (Table 2). The distinct decrease of  $E$  (49 and 43%) was observed in Alvand and Bahman, respectively. On the average, triticale and wheat transpired more than barley under both stress (15 and 25%) and non-stress (7 and 15%) conditions, respectively.

The examined genotypes had relatively significant differences in  $C_i$  in their response to soil water deficit. The  $C_i$  reduction by water stress ranged from 2% for Bahman to 23% for triticale No. 1 (Table 2). However, the reduction in barley was the least (2%), followed by wheat (12%) and triticale (15%). Under stress condition, the least  $C_i$  was observed for triticale No.1 ( $192.7 \pm 19.3$  ppm) and the highest was found in Bahman ( $242.1 \pm 9.4$  ppm).

### Fluorescence Parameters

The reduction of the maximal quantum yield of primary photochemistry,  $F_v/F_m$ , varied from 1% for genotype No. 1 to 15% for Bahman (Table 3). Most of this variation was due to differences between crop species. Generally, in triticale genotypes except No.2,  $F_v/F_m$  had less sensitivity to drought compared to the other two species, as indicated by insignificant reduction of its value by the water-stress condition (Table 3). However, the value of this parameter in triticale under water stress treatment was higher than that of wheat and barley, with the exception of Pishgam. This suggested less damage by water deficit to photosynthetic apparatus in the triticale genotypes and Pishgam in comparison with the other genotypes of wheat and barley. In our experiment, a significant correlation was observed between the yield biomass at

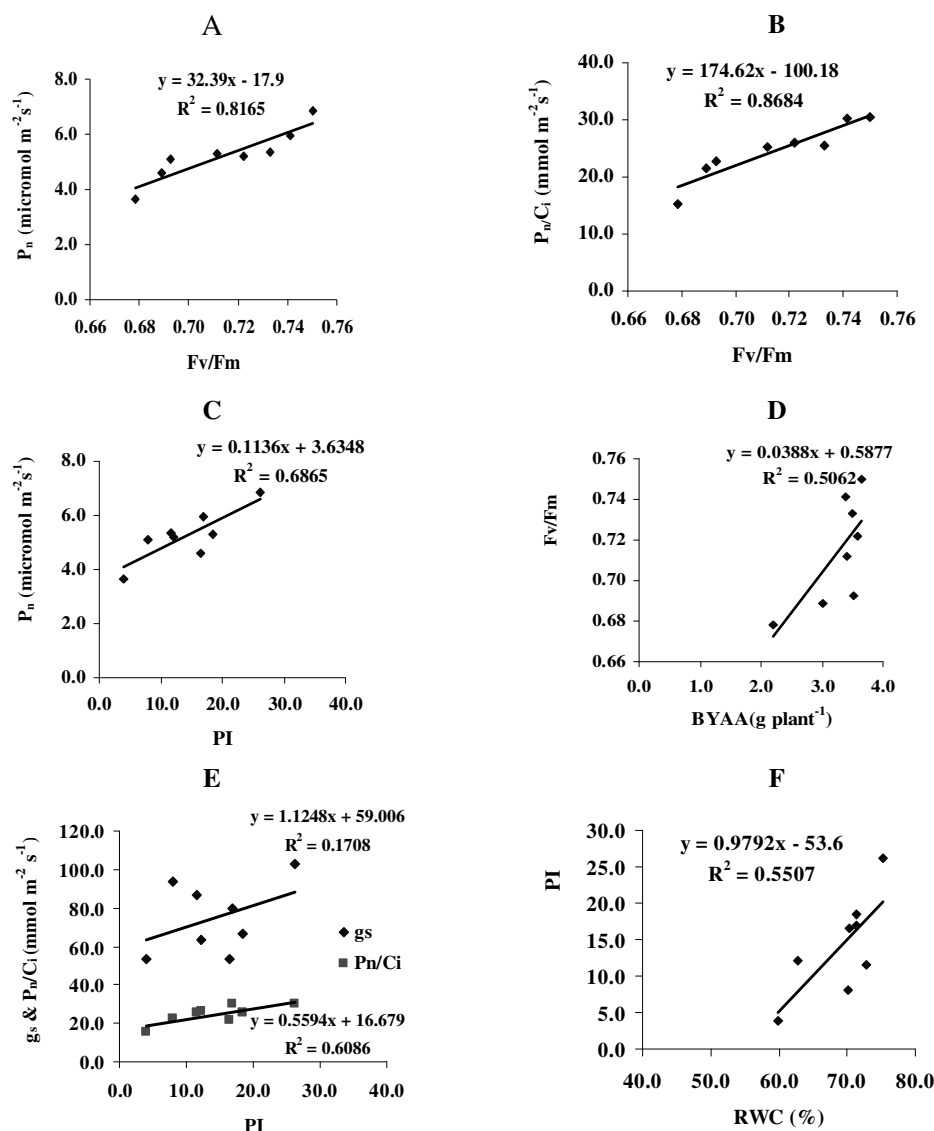


anthesis and the  $F_v/F_m$  under the stress treatment (Figure 3-D).

The results showed a smaller increase for the  $D_{I_o}$  in triticale (13%) than wheat (27%) and barley (78%) (Table 3). A distinct decrease of the  $F_v/F_o$  was noted for barley (40%) and wheat (26%) in contrast to the lowest value (11%) in triticale (Table 3). Under stress treatment, the highest values of

$F_v/F_o$  were observed in both triticale No. 1 and Pishgam ( $2.93 \pm 0.38$  and  $2.52 \pm 0.47$ , respectively) and the lowest values were found in Alvand and Bahman ( $2.15 \pm 0.47$  and  $2.20 \pm 0.42$ , respectively).

Overall, the water stress caused less damage to PS II in triticale than wheat or barley (Table 3), as indicated by the smaller reduction of the PI in the former (21%)



**Figure 3.** Correlations of maximum quantum yield of primary photochemistry, PS II,  $F_v/F_m$  and stomatal conductance,  $g_s$  (A),  $F_v/F_m$  and apparent carboxylation efficiency,  $P_n/C_i$  (B),  $F_v/F_m$  and biological yield at anthesis,  $BYAA$  (D), photosynthesis rate,  $P_n$  and performance index,  $PI$  (C),  $PI$ ,  $g_s$  and  $P_n/C_i$  (E) and relative water content,  $RWC$  and  $PI$  (F) among four genotypes of triticale, three genotypes of bread wheat and one genotype of barley under water deficit ( $\Psi_{soil} = -12$  bar).



against larger reduction for wheat (38%) and barley (79%). The results revealed that the  $PI$  was higher under normal conditions than under stress, as evidenced by the significant correlation between the  $RWC$  and  $PI$  (Figure 3-F).

## DISCUSSION

The results of this study showed that water deficit affected the growth of barley more than that of bread wheat and triticale. In triticale, genotype No. 1 had the lowest biomass yield reduction, whereas No. 2 showed a large decrease when the moisture was limited. In the case of wheat, Pishgam performed better than the other two varieties, Alvand and Zarrin, when confronted with water stress. These results suggest a high variation within triticale and wheat genotypes under water-limiting conditions, an observation that is in agreement with the findings of Grzesiak *et al.* (2003) and Izanloo *et al.* (2008). The better performance of wheat genotypes compared with barley under water deficit indicated that barley would not necessarily perform better than wheat under limited environments, as reported by Mariano Cossani *et al.* (2009).

Our data showed considerable variation (24-56%) among our tested genotypes in the reduction of the net photosynthetic rate,  $P_n$ , under water stress, irrespective of the plant species. There are genotypic variations in the effect of drought stress on  $P_n$  (Johnson *et al.*, 1987; Matin *et al.*, 1989). At the whole-plant level, the effect of water stress has been perceived as reduction in photosynthesis (Blum, 1996; Mwanamwenge *et al.*, 1999; Inoue *et al.*, 2004; Ghaderi *et al.*, 2011). Even though triticale had lower  $P_n$  values under WW conditions in comparison with wheat (Singh and Singh, 2001) and barley, the reduction by water deficit in this species was lower (39% against 48 and 54%, respectively) indicating the better acclimation of this new

crop in relation to photosynthesis inhibition caused by water stress.

Monitoring gas exchange in plants is a common approach, with stomatal conductance ( $g_s$ ) reported as one of the most sensitive indicators of stress under drought for C3 species (Medrano *et al.*, 2002). However, it has been reported that decreasing leaf water content initially induces stomatal closure (Pasban Eslam, 2011), imposing a decrease in the supply of  $CO_2$  to the mesophyll cells and consequently, results in a decrease in the rate of leaf photosynthesis (Lawlor and Cornic, 2002). A genetic variation in this case has been reported for triticale (Grzesiak *et al.*, 2003; Hura *et al.*, 2007) and wheat (Loggini *et al.*, 1999).

The  $C_i$  reduction by water stress was 9.8% in all tested species, but was not statistically significant. Generally, lower  $C_i$  should be accompanied by lower stomatal conductance ( $g_s$ ). However, in barley, despite more decline in the  $g_s$ , the  $C_i$  was unaffected by water deficit conditions (Table 2), suggesting an inability of photosynthesis machinery to utilize internal  $CO_2$  (Luo, 1991; Ahmadi and Siosemardeh, 2005) and non-stomatal factors predominating over stomatal factors in the limitation of  $CO_2$  assimilation activity (Rivelli *et al.*, 2002). The significant relationship ( $P \leq 0.01$ ) between the reduction of the  $g_s$  and  $P_n/C_i$  (Figure 2-D) in the drought stress treatment revealed that, under unfavorable conditions accompanied by stomatal limitations, some physical (solubility of  $CO_2$ , surface area of the apoplastic, and symplastic routes of  $CO_2$ ) and metabolic components (aquaporins and carbonic anhydrase) (Pinheiro and Chaves, 2010), in particular, the activity, quantity, and regeneration of Rubisco, would be impaired. This could induce a non-stomatal limitation, such as mesophyll resistance. Such a limitation has also been reported in other studies (Flexas *et al.*, 2008; Chaves *et al.*, 2009).

Both stomatal and non-stomatal limitations were more pronounced in barley than the other two crops because of high



reduction of  $g_s$  (65%),  $P_n/C_i$  (60%) and  $P_n$  (54%) in this crop. There were two different scenarios between the genotypes with low sensitivity to water deficit, e.g. triticale No. 1 and Pishgam. Low reduction of  $P_n/C_i$  (3%) in triticale No. 1 against more reduction in Pishgam (38%) accompanied by higher reduction of  $P_n$  (42%) revealed the fact that Pishgam was more sensitive to non-stomatal limitation of photosynthesis in comparison with triticale No. 1. However, higher correlation between the  $P_n$  and  $P_n/C_i$  (Figure 2-C) rather than the  $g_s$  (Figure 2-B) under this condition suggests that the  $g_s$  might play an important role in the high  $P_n$  under well watered or mild drought stress, but, under severe drought stress, the high  $P_n$  is related more to the maintenance of a higher capacity for mesophyll photosynthesis (Farquhar and Sharkey, 1982; Johnson *et al.*, 1987; Rekika *et al.*, 1998; Shangquan *et al.*, 1999).

During water deficit conditions, the activity of the photosynthetic apparatus was lower, especially for some varieties of wheat (Alvand and Zarrin) and barley (Bahman) than that for the control, resulting in lower values of such parameters as  $F_v/F_m$  and  $F_v/F_o$  (Table 3). As described by Baker and Rosenquist (2004) and Hura *et al.* (2007), under stress treatment, the stomatal limitation on photosynthesis was accompanied with decrease in the rate of consumption of ATP and NADPH for  $CO_2$  assimilation that could result in decrease in the rate of linear electron transport and, consequently, in  $F_v/F_m$ . However, the distinct decrease in gas exchanges parameters, for instance  $g_s$ , that occurred in some genotypes (e.g. No. 1, No. 3 and Pishgam) when only relatively small drop of  $F_v/F_m$  were found may suggest that damage of PSII in this genotypes was less marked (Wu and Bao, 2011). On the other hand, in these genotypes, despite considerable stomatal limitation, the  $F_v/F_m$  was not significantly changed, likely due to the buffering effects of photorespiration as an electron sink, which maintained the rate of electron flow similar to that of non-stress conditions (Flexas *et al.*, 2002; Ort and

Baker, 2002). The low or lack of sensitivity of the  $F_v/F_m$  in different types of cereals has also been observed in other studies (Kocheva *et al.*, 2004; Hura *et al.*, 2007).

Under stress conditions, a significant correlation was found between the  $F_v/F_m$  and biomass yield at anthesis (Figure 3-D). Unlike stress free conditions (data not shown), the significant positive correlation (Figure 2-B) between the  $F_v/F_m$  and  $P_n/C_i$  and the strong relationship (Figure 3-A) between the  $F_v/F_m$  and  $P_n$  under water stress indicated that chlorophyll fluorescence under water limiting condition acts as a non-stomatal factor in limiting photosynthesis apparatus (Hura *et al.*, 2007).

Moreover, photo inhibition is considered to be more accurately identified by an increase in the  $DIO$  (Force *et al.*, 2003). Under water stress, energy dissipation rises with an increasing degree of PSII injuries resulting in an increase in the  $DIO$  (Rapacz and Wozńiczka, 2009). High increase in the  $DIO$  in barley compared to low decrease in triticale means that PS II was less damaged in the later species.

The parameter of  $F_v/F_o$  decreased in comparison to the well-irrigated plants, which has been associated with the donor part of PS II (Hura *et al.*, 2007), especially in the water-splitting system. As mentioned by Maxwell and Johnson (2000), this parameter is more sensitive to change. It was confirmed by more variation of  $F_v/F_o$  than  $F_v/F_m$  as shown in Table 3.

The performance index,  $PI$  is one of the chlorophyll fluorescence parameters that can provide useful and quantitative information about the state of plants and their vitality (Oukarroum *et al.*, 2007). As mentioned, the  $PI$  decreased after water stress. This parameter is sensitive to change in either the antenna properties, trapping efficiency, or electron transport beyond  $Q_A$  (Oukarroum *et al.*, 2007). The decrease in the  $PI$  observed in the water deficit treatment may be related to damage in both PSII reaction center and antenna, as well as electron transfer disturbance (Rapacz and Wozńiczka, 2009). The slight reduction of

the *PI* in triticale No. 1 and Pishgam may explain the slight decrease in the photosynthetic machinery that, consequently, reduced the sensitivity to drought in these genotypes. In fact, the performance index is closely related to the final outcome of plant's activity, such as growth or survival under stress conditions (Yusuf *et al.*, 2010). The variability of the *PI* in response to water deficit on the basis of the reduction rate ranged from 5 to 79%, which was more than that of  $F_v/F_m$ , suggesting a higher sensitivity of the former to drought compared to the latter. This was in agreement with the results of Oukarroum *et al.* (2007) who reported limited differences in the  $F_v/F_m$  compared to the *PI* and a higher effectiveness of the latter parameter in the evaluation of the susceptibility of genotypes to environment with considerable water limitation. The significant correlation between the *PI* and the  $P_n/C_i$  under water deficit condition in conjunction with the lack of relationship between the *PI* and  $g_s$  (Figure 3-E) also confirmed our abovementioned inference that non-stomatal limitations have considerable impact on the photosynthetic activity under limiting water conditions. Finally, the significant relationship between  $P_n$  and the *PI* (Figure 3-C) also emphasized the reliability of the *PI* for screening genotypes with high photosynthetic activity in reproductive stage under moisture-stress conditions.

In conclusion, stomatal and non-stomatal inhibition to  $P_n$  under stress condition may vary with genotypes as well as plant species. High leaf  $P_n$ ,  $RWC$ ,  $g_s$ ,  $P_n/C_i$ ,  $F_v/F_m$  and *PI* appear to be involved in triticale No. 1 and Pishgam regardless of their species. However, these traits are associated with better performance of these genotypes under water deficit condition. On average, triticale had a lower reduction in photosynthetic traits under water deficit in comparison with wheat and barley. The better performance of Pishgam wheat variety in reducing stomatal and non-stomatal limitations confirms

higher adaptability of Pishgam for water limiting conditions in Iran.

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## بررسی مقایسه ای اثر تنش رطوبت خاک بر عملکرد فتوسنتزی تریتیکاله، گندم و جو

۱. روحی، ز. طهماسبی سروستانی، س. م. ع. مدرس ثانوی، ع. سی و سه مرده

### چکیده

به منظور مقایسه خصوصیات فتوسنتزی تریتیکاله، گندم و جو این بررسی در کردستان واقع در غرب ایران انجام شد. برای این منظور چهار ژنوتیپ تریتیکاله، سه رقم گندم و یک واریته جدید جو در شرایط خوب آبیاری (آبیاری در پتانسیل رطوبتی خاک ۳- بار) و تنش کم آبیاری (آبیاری در پتانسیل رطوبتی خاک ۱۲- بار) در یک طرح بلوک های کامل تصادفی با آرایش کرت های خرد شده مقایسه شدند. مقدار زیست توده در گلدهی همراه با تبادلات گازی و فلورسانس کلروفیل برگ پرچم به عنوان شاخص های عملکرد فتوسنتزی گیاه در هر تیمار اندازه گیری شد. نتایج بیانگر کاهش سرعت فتوسنتز،  $P_n$  بر اثر تنش رطوبتی در تمامی ژنوتیپ های مورد مطالعه صرف نظر از نوع گونه گیاهی بود. بیشترین میزان  $P_n$  ( $6/86$  و  $5/90$  میکرومول بر متر مربع بر ثانیه) به ترتیب در رقم گندم پیشگام و لاین شماره یک تریتیکاله و کمترین آن ( $3/63$  میکرومول بر متر مربع بر ثانیه) در رقم جو بهمن مشاهده شد. روند مشابهی برای حداکثر عملکرد کوانتومی فتوسیستم دو،  $F_v/F_m$  و شاخص کارایی،  $PI$  وجود داشت. همبستگی مثبت و معنی داری بین زیست توده موجود در گلدهی با  $P_n$  ( $r = 0/83^{**}$ ) و  $F_v/F_m$  ( $r = 0/77^{**}$ ) مشاهده گردید. رقم گندم پیشگام از وضعیت بهتری نسبت به ارقام گندم الوند و زرین از لحاظ درصد کاهش بیوماس در گلدهی و خصوصیات فتوسنتزی در شرایط تنش برخوردار بود. بطور متوسط تریتیکاله حساسیت کمتری نسبت به تنش رطوبتی در مقایسه با گندم و جو از لحاظ عملکرد فتوسنتزی و کاهش در مقادیر  $P_n$ ،  $PI$  و  $F_v/F_m$  از خود نشان داد.