

# Gas Exchange Responses of Different Wheat (*Triticum aestivum* L.) Cultivars to Water Stress Condition

ALI AKBAR MAGHSOUDI MOUD<sup>1</sup> AND TOHRU YAMAGISHI<sup>†</sup>

Department of Agronomy and Plant Breeding, College of Agriculture, Shahid Bahonar University, Kerman, Iran

<sup>†</sup>Graduate School of Agricultural and Life Science, Department of Agricultural and Environmental Biology, The University of Tokyo, Tokyo 113, Japan

<sup>1</sup>Corresponding author's e-mail: [akubaru2@yahoo.com](mailto:akubaru2@yahoo.com)

## ABSTRACT

The objectives of this study were to compare photosynthesis (Pn), transpiration (T) and instantaneous water use efficiency (iWUE) and their correlations across different wheat cultivars under water stress condition. Seeds of six different wheat cultivars were sown in pots. Plants were transferred to a growth cabinet at the late stem elongation period and water stress was applied by withholding water. Pn was measured at the beginning and the end of the stress period, while CO<sub>2</sub> (write this as corrected) concentration was changed to different values. Transpiration (T), intercellular CO<sub>2</sub> concentration (Ci) and stomatal conductance (g) were also measured. Carboxylation efficiency (CE) was calculated as the slope of the fitted line between Pn and Ci at low levels of CO<sub>2</sub>. Significant differences were found at both times between cultivars, when CE was compared among cultivars. All parameters were reduced at the end of the stress period except iWUE (g) was highly correlated with T at the beginning, while it was correlated with Pn as well at the end of the stress period. iWUE was negatively correlated with Ci and Ci/Ca under both conditions, while it was negatively correlated with g. Generally it can be concluded that differences in iWUE was correlated to that of Ci rather than to that of Pn. Cultivar BR 10 had sensitive stomata to drought condition, because of higher reduction in (g) at the end of the stress period. On the other hand Asakaze had less sensitive stomata to dry condition as it kept higher (g) in terms of absolute value at the end of the stress period.

**Key Words:** wheat; Gas exchange; Water stress

## INTRODUCTION

Plants exposed to adverse environmental conditions show some characteristics changes to maintain their physiological functions at normal level (Blum, 1996). Drought is one of the important stresses, which plants often experience during their life cycle. When plants are exposed to drought, several mechanisms become active in order to sustain their water status at a level high enough for physiological processes to be normal (Bartels *et al.*, 1996; Blum, 1996). Osmotic adjustment is considered as one of the mechanisms that could maintain high turgor in droughted plants (Ludlow *et al.*, 1990). As a result stomata could be kept opened and gas exchange normally took place. On the other hand the role of abscisic acid (ABA) in maintaining the stomatal closure during stress periods has been well documented (Hall *et al.*, 1994). Stomatal sensitivity to reduced soil water availability could also change the response of the plants to water stress condition (Garrity *et al.*, 1982). However, a physiological antagonism between carbon assimilation (which requires sufficient stomatal conductance) and maintenance of water status (which requires adverse condition arise) (Stanhil, 1985) even if the plants adapt to drought by other mechanisms such as reducing leaf area (Belaygue *et al.*, 1996) or tillering (Davidson & Chevalier, 1987). Therefore, the plants, which

can sustain higher carbon assimilation while keeping almost lower transpiration or higher water use efficiency (WUE), would be of higher value under dry condition particularly if the water resources were limited. For many years plant breeders have focused on the problem of how to develop more efficient crops in water use (Hall *et al.*, 1994). As an easy and rapid measurement, carbon isotope discrimination ( $\Delta$ ) has been proposed as a surrogate of direct measurement of water use efficiency (WUE), which is virtually time consuming and difficult to be measured for a relatively high number of lines (Garrity *et al.*, 1982; Condon *et al.*, 1990; Hall *et al.*, 1994).

Instantaneous water use efficiency (iWUE), which is the ratio between the net photosynthesis and transpiration could be considered as a short-term measurement of water use efficiency (Ehleringer *et al.*, 1993). WUE at plant and even at canopy level is determined by (iWUE). Relative changes of (iWUE) under stress compared to control conditions may also shows the degree of stress tolerance of the plants. If plants grown under the same environmental conditions and the samples taken from the same organs of the same growth stages, this parameter could also be considered as an estimate of WUE. It has been shown that (iWUE) changed with plant growth. WUE at the plant level also showed to be strongly related to (iWUE) (Turner, 1997). With the available technology measurement of (iWUE) and

the parameters like Pn and T, which is quite easy at any growth stage particularly with ones, which can control the cuvette air humidity and leaf temperature. The objective of this study was to compare the gas exchange characteristics and their correlation with (iWUE) across different wheat cultivars under water stress conditions.

## MATERIALS AND METHODS

Six wheat cultivars that represent a wide range of genetic variation were selected. Table I shows some features of the cultivars used in this study. Experiments were started in a glasshouse and continued in growth cabinet. Wagner pots (1/2000) were filled with 3000 g of an air-dried soil (12% water content). Four seeds were sown directly in each pot at 3 cm depth. Cultivars were seeded at different date in order to coincide their phenological stage with each other. 10 pots prepared for each genotype. Initially pots were irrigated with little water until the tillering stage. As a result plants produced few tillers ranged from one to two. All measurements that will be described later were done only on the main tiller leaves. Pots were weighted regularly and watered with half strength Hogland's solution to keep the soil water content at field capacity. Plants transferred to a growth cabinet after they reached to the late stem elongation period. Experimental conditions were 13 h photoperiod at 25°C with 680  $\mu\text{mol m}^{-2} \text{s}^{-1}$  light intensity, 11 h night at 20°C, RH 60% day and 65% night. After the plants adapted to the new growing conditions water stress was applied by withholding water for a period of 16 days. In order to reduce the evaporation from the soil and extend the period of imposing water stress to the plants, pots were covered with a layer of aluminum foil and the soil surface was covered with a 4 cm layer of perlite.

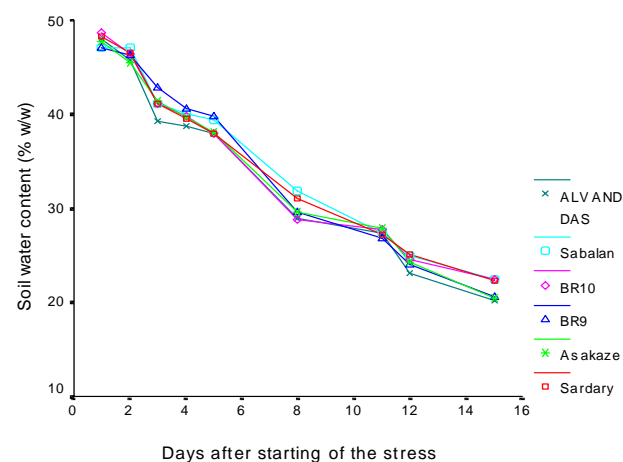
Photosynthesis and transpiration rates were measured using a Ciras-I system at the beginning and the end of the stress period. It should be mentioned that the soil water content was at the field capacity at the beginning and around 40% of the field capacity at the end of the stress period with no significant differences between cultivars (Fig. 1). Ciras-I was connected to a PC and using the remote control software of the system  $\text{CO}_2$  concentration was changed to different values step-wisely, while the light intensity was kept constant at 1000  $\mu\text{mol m}^{-2} \text{s}^{-1}$ , leaf temperature at 25°C and relative humidity at 50%. In addition Ciras-I also measured intercellular  $\text{CO}_2$  concentration ( $C_i$ ) and stomatal conductance. Instantaneous water use efficiency was calculated as the ratio between photosynthesis and transpiration. All adjustments were done according to Ciras-I operator's manual.

Data were subjected to the analysis of variance and correlation coefficients were computed. Mean values were compared when the F values were significant using the method of Duncan. Carboxylation efficiency of the cultivars was calculated between near zero to 100 ppm intercellular  $\text{CO}_2$  concentration. In that range highly significant linear

**Table I. Origin, drought resistance and plant size of the six wheat cultivars that were used in this study**

Cultivar	Country	Drought resistance in terms of grain yield	Plant height
Alvand	Iran	Tolerant	Semi-tall
Asakaze	Japan	Sensitive	Dwarf
BR10	Mexico	Non-tolerant	Semi-dwarf
BR9	Brazil	Tolerant	Tall
Sabalan	Iran	Tolerant	Tall
Sardary	Iran	Tolerant	Tall

**Fig. 1. Changes of soil water content during the stress period. Differences were not significant at each sampling time as compared with the corresponding LSD**



responses were found between the two parameters for all cultivars. Photosynthesis was considered as its maximum value near 950 ppm ambient  $\text{CO}_2$  concentration.

## RESULTS AND DISCUSSION

F test showed that line slopes (CE) are significantly different from each other among wheat cultivars ( $P \leq 0.003$ ). At the beginning CE was higher in Alvand, BR9 and Sabalan, while it was lower in Asakaze and Sardary and intermediate in BR 10 (Table II). Genotypic variation in CE among wheat cultivars, which is due to different levels of carboxylation enzymes and their activity and other related factors has already shown to be existing (Condon *et al.*, 1990). Table II also shows that significant genotypic variations exist among wheat cultivars in gas exchange characteristics both at the beginning and the end of the stress period. The same results have been obtained among different wheat cultivars elsewhere (Condon *et al.*, 1990; Morgan & LeCain, 1991). It has been claimed that semi-dwarf wheat lines have higher gas exchange rate due to higher stomatal density and higher ratio of total mesophyll cell area to leaf area ( $A_{\text{mes}}/A$ ) compared to their tall isogenic lines (LeCain *et al.*, 1989). Therefore, higher Pn and (g) rates in Asakaze maybe due to the dwarfing genes. For different gas exchange characteristics cultivars rank

were changed so that there were no correlation between each of the characteristics at the beginning and the end of the stress period (results are not shown). Generally, all parameters were reduced at the end of the stress period except (iWUE), which was increased (Tables II & III). It has been shown that photosynthetic water use efficiency (Pn/g) increased with water stress severity (Stanhill, 1985). Increased WUE under dry condition is showed to be commonly occurring in crop plants.

Since (iWUE) =  $P_n/T$ , theoretically the greater the  $P_n$  and the smaller the  $T$ , the higher the (iWUE). However correlation coefficient between  $P_n$  and (iWUE) was positive only at the beginning and with  $T$  was negative at both times as expected they were not significant (Table IV). In order to remove the confounding effect of differences in (g) and CE on these relations, partial correlation coefficients between characters was calculated when (g) and/or CE were kept constant (Table V). Results showing that when g is kept constant  $P_n$  effect on (iWUE) was positively high and greater than negative effect of  $T$  particularly at the beginning of the stress period. However when CE was kept constant negative effect of  $T$  on (iWUE) was higher than that of  $P_n$ . This may confirm what is expected from the theory that naturally water vapor loss is limited less than  $CO_2$  up-take. However if CE was not a limiting factor, photosynthesis would be limited less than transpiration. Practically it is not possible to increase the  $P_n$  without an increase in  $T$  since both are sharing the same pathway. However results also showing that to increase the (iWUE) cultivars with higher CE and lower g should be preferred. Significant negative correlation was found between  $C_i/C_a$  and (iWUE) at both the beginning and the end of the stress period even when (g) and/or CE were kept constant (Tables VI & V). These results also suggest that cultivars with higher ratio of  $C_i/C_a$  may have higher values of water use efficiency.

It has been suggested that changes in genotypes ranking for  $\Delta$ , which is related to  $C_i/C_a$ , under different water stress levels could be used as an effective detector of stomatal sensitivity among wheat genotypes (Condon *et al.*, 1990). i.e. the lower the  $C_i/C_a$  the greater the sensitivity of the stomata to the stress condition.

It has been argued that osmotic adjustment causes the plants to maintain higher stomatal conductance at lower leaf water potential (Ludlow & Muchow, 1990). Stomatal conductance is a key factor influencing gas exchange properties of the leaf. However other stomatal characteristics such as stomatal sensitivity (Garrity *et al.*, 1982) and also ABA accumulation may also change the response of the stomata to water stress (Turner, 1997). Even some plants may have inherently lower stomatal conductance, whereas stomata of other plants respond only to the stress conditions (Ludlow & Muchow, 1990). For example more sensitive stomata to water stress caused the cowpea plants to avoid desiccation better than mungbean, soybean and pigeonpea (Lawn, 1982).

**Table II. Photosynthesis (Ph), transpiration (T), stomatal conductance (g), intercellular  $CO_2$  concentration ( $C_i$ ), carboxylation efficiency (CE), and instantaneous water use efficiency (iWUE) in wheat cultivars**

Cultivar	Ph $\mu\text{molm}^{-2}\text{s}^{-1}$	T $\text{mmolm}^{-2}\text{s}^{-1}$	g $\text{mmolm}^{-2}\text{s}^{-1}$	$C_i$ ppm	CE $\text{molm}^{-2}\text{s}^{-1}\text{p}^{-1}$	iWUE $\mu\text{molmmol}^{-1}$
<b>Beginning</b>						
Asakaze	17.3c	6.02b	763b	291b	0.097	2.87e
BR10	16.6d	5.89c	754b	297a	0.119	2.81f
Alvand	16.4f	3.97f	377d	260f	0.158	4.13a
BR9	20.3a	6.18a	820a	284d	0.139	3.28c
Sabalan	18.6b	4.7e	518c	271e	0.130	3.95b
Sardary	14.8f	4.74d	525c	288c	0.082	3.11d
<b>End</b>						
Asakaze	11.2a	2.25a	207a	255a	0.039	4.97c
BR10	4.5f	0.86f	60f	234c	0.031	5.23b
Alvand	9.5e	1.56e	120e	224d	0.048	5.94a
BR9	10.2c	2.16b	181b	253a	0.038	4.72d
Sabalan	10.4b	2.1d	165d	239b	0.048	4.95c
Sardary	9.55d	2.13c	168c	253a	0.034	4.47e

Means followed by different alphabet are different at 5% level of significance based on Duncan's multiple range test

**Table III. Percent change in photosynthesis (Pn), transpiration (T), instantaneous water use efficiency (iWUE), stomatal conductance (g), carboxylation efficiency (CE), intercellular  $CO_2$  concentration ( $C_i$ ) and its ratio to the ambient  $CO_2$  concentration ( $C_i/C_a$ ) at the end compare to the beginning of the stress period**

Cultivar	Ph	T	iWUE	g	$C_i$	$C_i/C_a$	CE
Alvand	57.9	39.3	143.8	32.0	86.3	85.8	30.4
Asakaze	64.7	37.4	173.2	27.2	87.6	87.3	40.2
BR10	27.1	14.6	186.1	8.0	79.0	78.8	26.1
BR9	50.2	35.0	143.9	22.1	89.3	88.6	27.3
Sabalan	55.9	44.7	125.3	31.9	88.4	88.6	36.9
Sardary	64.5	44.9	143.7	32.0	87.8	87.5	41.5

**Table IV. Pearson correlation coefficients of gas exchange characteristics across wheat cultivars**

	Pn $\mu\text{molm}^{-2}\text{s}^{-1}$	T $\text{mmolm}^{-2}\text{s}^{-1}$	iWUE $\mu\text{molmmol}^{-1}$	g $\text{mmolm}^{-2}\text{s}^{-1}$	$C_i$ ppm	$C_i/C_a$ ppm/ppm
<b>Beginning</b>						
T	0.471					
iWUE	0.159	-0.790				
g	0.490	0.998**	-0.777			
$C_i$	-0.106	0.797	-0.982**	0.787		
$C_i/C_a$	-0.090	0.809	-0.987**	0.798	0.997**	
CE	0.494	-0.282	0.709	-0.256	-0.707	-0.722
<b>End</b>						
T	0.937**					
iWUE	-0.229	-0.553				
g	0.927**	0.979**	-0.521			
$C_i$	0.455	0.704	-0.879*	0.750		
$C_i/C_a$	0.468	0.717	-0.884*	0.761	0.998**	
CE	0.373	0.336	-0.030	0.354	0.262	0.227

\*\*\* Correlation is significant at the 0.05 and 0.01 levels respectively.

Data obtained in this study shows that CV Alvand has inherent closed stomata and keeps them partially opened under water stress condition. Stomata are very sensitive in BR 10 since conductance was reduced considerably more in

**Table V. Partial correlation coefficients of gas exchange characteristics and water use efficiency (iWUE) across wheat cultivars when stomatal conductance (g) (part A) and carboxylation efficiency (CE) (part B) were kept constant at the beginning and the end of the stress period**

		iWUE $\mu\text{molmm}^{-2}\text{s}^{-1}$	Pn $\mu\text{molm}^{-2}\text{s}^{-1}$	T $\text{mmolm}^{-2}\text{s}^{-1}$	Ci/Ca
<b>Part A</b>					
<b>Beginning</b>	Pn	0.983**			
	T	-0.478	-0.467		
	Ci/Ca	-0.967**	-0.917*	0.403	
	CE	0.838*	0.735	-0.595	-0.889*
<b>End</b>	Pn	0.798			
	T	-0.246	0.385		
	Ci/Ca	-0.881*	-0.978**	-0.220	
	CE	0.194	0.128	-0.057	-0.069

		Pn $\mu\text{molm}^{-2}\text{s}^{-1}$	T $\text{mmolm}^{-2}\text{s}^{-1}$	iWUE $\mu\text{molmm}^{-2}\text{s}^{-1}$	g $\text{mmolm}^{-2}\text{s}^{-1}$
<b>Part B</b>					
<b>Beginning</b>	T	0.731			
	iWUE	-0.312	-0.872		
	g	0.733	0.999**	-0.873	
	Ci/Ca	0.443	0.911*	-0.973**	0.917*
<b>End</b>	T	0.929*			
	iWUE	-0.234	-0.576		
	g	0.916*	0.977**	-0.546	
	Ci/Ca	0.424	0.698	-0.901*	0.747

\*.Significant at 0.05 and \*\*. Significant at 0.01 level

response to the water stress. On the other hand, Asakaze had inherent opened stomata and stomatal conductance remained high in this cultivar. Stomata were opened in BR 9 under wet condition, while they were partially closed under stress condition. Among the other cultivars, stomatal aperture in Sabalan and Sardary was relatively intermediate in terms of absolute value at both the beginning and the end of the stress period. However, lower ratio of Ci/Ca in Sabalan may suggest higher sensitivity of the stomata in this cultivar. In a companion paper we found that cultivars BR 9, Alvand and Sabalan to be capable for osmoregulation, while Asakaze, BR 10 and Sardary were not. Stomatal adjustment under stress condition in cultivars Alvand and BR 9 may be attributed to their ability for osmoregulation. On the other hand sensitivity of stomata in BR 10 could be due to lack of osmoregulation capability in guard cells. However the response of Asakaze could not be explained by the changes in osmotic pressure under stress condition. Another possibility is the higher levels of ABA accumulation and also higher number of stomata per unit of leaf area as explained in the introduction for semi-dwarf cultivars.

Under progressive soil drying condition like what is occurring in Mediterranean regions, reduced stomatal conductance may improve the yield, because it lowers the probability of exhausting the stored soil water before crop maturity. Cultivar Alvand had higher (iWUE) under both wet and dry conditions. If this could maintain over whole

growth period under such progressive soil drying condition, it maybe expected that cultivars like Alvand could maintain their advantages over the others in terms of WUE. However, cultivars like BR 9, which could respond to the stress by other mechanisms may also remain useful. Cultivar BR 10, which closes its stomata may be useful under more dry condition. Conversely concerning the sensitivity of the stomata, cultivars like Asakaze have no advantages, because they will exhaust the stored soil water under wet condition and have no mechanism of transpiration control under dry condition.

The data used in this study were obtained under constant leaf temperature condition and confirm what is expected from the theory in the relationship between stomatal conductance and WUE. However, it should be considered that under field condition, stomatal closure is usually accompanied by rising in the leaf temperature, which in turn could increase air-leaf water vapor deficit and therefore decrease WUE.

## REFERENCES

- Bartels, D.A. Furini, J. Ingram and F. Salamini, 1996. Responses of plants to dehydration stress: a molecular analysis. *Pl. Growth Regulation*, 20: 111–8
- Belaygue, C, J. Wery, A. A. Cowan and F. Tardieu, 1996. Contribution of leaf expansion rate of leaf appearance and stolon branching to growth of plant leaf area under water deficit in white clover. *Crop Sci.*, 36: 1240–6
- Blum, A., 1996. Crop responses to drought and the interpretation of adaptation. *Pl. Growth Regulation*, 20: 135–48
- Condon, A.G., G.D. Farquhar and R.A. Richards, 1990. Genotypic variation in carbon isotope discrimination and transpiration efficiency in wheat. Leaf gas exchange and whole plant studies. *Australian J. Plant Physiol.*, 17: 9–22
- Davidson, D.J. and P.M. Chevalier, 1987. Influence of polyethyleneglycol-induced water deficit on tiller production in spring wheat. *Crop Sci.*, 27: 1185–7
- Ehleringer, J.R., A.E. Hall and G.D. Farquhar, 1993. Introduction: Water use in relation to productivity. In: Ehleringer, J.R., A.E. Hall and G.D. Farquhar (eds.), *Stable Isotopes and Plant Carbon-water Relations*, Pp: 3–7. Academic Press, San Diego
- Garrity, D.P., C.Y. Sullivan and D.G. Watts, 1982. Changes in grain sorghum stomatal and photosynthesis response to moisture stress across growth stages. *Crop Sci.*, 24: 441–6
- Hall, A.E., R.A. Richardes, G.C. Wright and G.D. Farquhar, 1994. Carbon isotope discrimination and plant breeding. *Pl. Breed. Rev.*, 12: 81–113
- Lawn, R.J., 1982. Respose of four grain legumes to water stress in south-eastern Queensland. I Physiological respose mechanisms. *Australian J. Plant Physiol.*, 33: 481–96
- LeCain, D.R., J.A. Morgan and G. Zerbi, 1989. Leaf anatomy and gas exchange in nearly isogenic semidwarf and tall winter wheat. *Crop Sci.*, 29: 1246–51
- Ludlow, M.M. and R.C. Muchow, 1990. A critical evaluation of traits for improving crop yields in water-limited environments. *Adv. Agron.*, 43: 107–53
- Morgan, J.A. and D.R. LeCain, 1991. Leaf gas exchange and related leaf traits among 15 winter wheat genotypes. *Crop Sci.*, 31: 443–8
- Stanhil, G., 1985. Water use efficiency. *Adv. Agron.*, 39: 53–85
- Turner, N.C., 1997. Further progress: In crop water relations. *Adv. Agron.*, 58: 293–338

(Received 25 October 2005; Accepted 18 November 2006)