International Journal of Biological, Life and Agricultural Sciences
ISSN: 2415-6612
Vol:5, No:6, 2011

Study of Water Relations, Chlorophyll and their Correlations with Grain Yield in Wheat (*Triticum aestivum* L.) Genotypes

Mokhtar Ghobadi, Saeed Khosravi, Danial Kahrizi and Firooz Shirvani

Abstract**—**The objective of this experiment was to study of water relations and chlorophyll in different wheat genotypes and their correlations with grain and biological yields. 21 genotypes of bread wheat were compared in a field experiment as randomized complete blocks design with four replications. The results showed that relative water deficit, relative water loss, excised leaf water retention, cell membrane stability, chlorophyll-a, chlorophyll-b, total chlorophyll, grain yield and biological yield were different significantly among wheat genotypes, but SPAD-chlorophyll index, relative water content and chlorophyll florescence were not. Significant correlations were not observed among above mentioned water relations and chlorophyll characteristics with grain yield, but there was a positive and significant correlation between biological yield and grain yield.

Keywords**—**Wheat, water relations, chlorophyll, yield

I. INTRODUCTION

WHEAT (*Triticum aestivum* L.) is an important cereal crop that ranks first globally and in Iran. Grain yield of wheat is usually determined by genetic and environmental factors. Physiologic characteristics of wheat are genetic factors that researchers pay great attention them nowadays.

Determination of water relation components at the whole plant or cellular level is important for determination of resistance of species or cultivars to environmental stresses such as drought, heat or salinity stresses [1].

Leaf water potential is considered to be a reliable parameter for quantifying plant water stress response. Singh *et al.*, (1990) observed significant differences in water potential among wheat genotypes under drought stress [2]. Sinclair and Ludlow (1985) proposed that leaf relative water content (RWC) was a better indicator of water status than water potential [3]. Among several methods used to characteristics internal plant water status, RWC is an integrative indicator [4] and was used successfully to identify drought resistant cultivars [5].

Relative water deficit (RWD), relative water loss (RWL) and excised leaf water retention (ELWR) are applied to study of water relations in crops, too [6, 7].

Cell membranes are one of the first targets of many plant stresses and it is generally accepted that the maintenance of their integrity and stability under water deficit conditions is a major component of drought tolerance in plants [8]. Selection for slow leaf electrolyte leakage under heat stress has been proposed as a method for increasing heat tolerance and heat resistance of several grain crops by enhancing membrane thermo-stability [9, 10]. The degree of cell membrane injury induced by drought stress or heat stress may be easily estimated through measurements of electrolyte leakage from the cells [8, 11].

Although a high correlation between the chlorophyll content and photosynthesis rate was not obtained [12], the assessment of photosynthetic pigments and consequently their relationships is an important indicator of senescence [13]. Chlorophyll loss is associated to environmental stress and the variation in total chlorophyll/carotenoids ratio may be a good indicator of stress in plants [14]. In addition, measuring gas exchange, water relations and chlorophyll content repeatedly on the same leaves in field may provide useful information on the relationship between these parameters [15].

The chlorophyll meter (or SPAD meter) is a simple, portable diagnostic tool that measures the greenness or the relative chlorophyll concentration of leaves. Compared with the traditional destructive methods, this equipment might provide a substantial saving in time, space and resources [16].

In the assessment of effects caused by high temperature or water deficit on the photosynthetic activity, chlorophyll florescence may be a safer indicator than net photosynthesis rate, because it is a practical and precise method. Net photosynthetic rate may be influenced by induced stomatal closure caused primarily by heat, by abscisic acid and by the dehydration of guard cells [17, 18].

This study was carried out to determination of water relations and chlorophyll in different wheat genotypes and their correlations together and with grain and biological yields.

II. MATERIALS AND METHODS

The field experiment was conducted at the Research Farm of Agricultural College, Razi University, Kermanshah,

Mokhtar Ghobadi is with Department of Agronomy and Plant Breeding, and Department of Biotechnology for Drought Resistance, College of Agriculture, Razi University, Kermanshah, Iran (corresponding author to provide phone: +98-831-8331723; fax: +98-831-8321083; e-mail: m.ghobadi@yahoo.com).

Danial Kahrizi is with Department of Agronomy and Plant Breeding, and Department of Biotechnology for Drought Resistance, College of Agriculture, Razi University, Kermanshah, Iran (e-mail: dkahrizi@yahoo.com).

Firooz Shirvani is with Department of Agronomy and Plant Breeding, College of Agriculture, Razi University, Kermanshah, Iran.

during October 2006 to June 2007. Kermanshah (34º20' N latitude, 47º20' E longitude, elevation 1351 m above see level) is located in the west of Iran with the moderate-cold and semiarid zone.

The soil was clay texture with pH 7.6, N 0.12%, P_2O_5, K_2O, Mn, Fe, Zn and Cu were equal 11.1, 380, 5.4, 5.9, 1.01 and 2.3 $mg.kg^{-1}$, respectively. A basal application of 50 kg N ha^{-1} and 60 kg P_2O_5 ha^{-1} was given before sowing. 50 kg N ha^{-1} at the beginning of stem elongation (Zadox scale: 31) and 50 kg N ha^{-1} at booting stage (Zadox scale: 41) were applied, too. The source of N and P_2O_5 fertilizers were urea and triple-superphosphate, respectively.

21 genotypes of bread wheat (*Triticum aestivum* L.) were planted as a randomized complete blocks design with four replications. Each plot contained six rows, three m length and 20 cm space between two rows. Plant density was 400 plants per square meter. Plants were under irrigated conditions.

Measurements of plant water relations were made between 10:00 to 13:00 h. Relative water content (RWC) and other water relations were measured using flag leaves. Immediately after cutting at the base of lamina, leaves were sealed within plastic bags and quickly transferred to the laboratory. Fresh weights (W_F) of leaves were determined. Turgid weight (W_T) were obtained after soaking leaves in distilled water in test tubes for 16 to 18 h at room temperature (about 20 °C) and under the low light conditions of laboratory. After soaking, leaves were quickly and carefully blotted dry with tissue paper in preparation for determining turgid weight. Dry weight (W_D) were obtained after oven drying the leaf samples for 48 h at 70 °C. RWC was calculated from the equation of Schonfeld *et al.*, [19].

$$RWC\ (\%) = [(W_F - W_D) / (W_T - W_D)] * 100$$

Relative water deficit (RWD) was measured using below equation [6].

$$RWD\ (\%) = 100 - RWC$$

In order to measuring of relative water loss (RWL) after sampling (the same as RWC), flag leaves were located at 30 °C during 2 h (t), then they were weighted as wilted leaf weight (w_w). RWL was calculated from the below equation [7].

$$RWL = [(W_F - W_W) / W_D] / [t / 60]$$

Excised leaf water retention (ELWR) was measured from below equation. Leaf water retention weight (W_R) was obtained after soaking leaves in distilled water in test tubes for 3 h.

$$ELWR\ (\%) = [1 - ((W_F - W_R) / W_F)] * 100$$

Cell membrane stability (CMS) was obtained through measuring of cell electrolyte leakage [11]. Leaf chlorophyll content was obtained by portable chlorophyll meter (SPAD-502, Minolta, Japan) from ten individual flag leaves per plot [16]. The chlorophyll a and b (chl-a, chl-b) were determined spectrophotometrically at 650 and 665 nm, respectively, according to the equation exposed by wellburn [20]. The Chlorophyll Florescence was obtained with a MINI-PAM Modulated Fluorimeter (Walz, Germany) [17].

The total above ground dry matter (biological yield) and grain yield were obtained after physiological ripening from one square meter in the two middle rows of each plot.

Data were analyzed by ANOVA and means were testes by Duncan's multiple range test using MSTAT-C and SAS statistical analysis packages.

III. RESULTS AND DISCUSSION

Analysis of variance showed that significant differences were observed among wheat genotypes in respect of relative water deficit (RWD), relative water loss (RWL), excised leaf water retention (ELWR), cell membrane stability (CMS), chlorophyll a (Chl-a), chlorophyll b (Chl-b), total chlorophyll (chl-t), biological (BY) and grain (GY) yields. But the genotypes did not have significant differences in respect of relative water content (RWC), SPAD-chlorophyll index and chlorophyll florescence (Chl-f) (Table I).

TABLE I
ANALYSIS OF VARIANCE FOR STUDIED TRAITS IN WHEAT GENOTYPES (MEAN SQUARES)

Source of variations	Degree of freedom	RWC	RWD	RWL	ELWR	CMS	SPAD
Replication	3	378.549	4.195	0.002	66.756	0.010	21.634
Genotype	20	67.803^{ns}	17.604**	0.050**	117.567*	0.076**	11.826^{ns}
Error	60	98.208	3.670	0.013	67.872	0.010	10.199
CV (%)		14.18	28.55	7.27	14.81	4.99	6.96

THE CONTINUATION OF TABLE I

Source of variations	Degree of freedom	Chl-a	Chl-b	Chl-t	Chl-f	BY	GY
Replication	3	0.055	0.017	0.032	0.009	432602.964	609.556
Genotype	20	0.550**	0.620**	0.392**	0.018^{ns}	256300.462**	206504.248**
Error	60	0.064	0.029	0.021	0.019	113872.348	29794.656
CV (%)		12.76	11.33	12.84	11.00	20.20	14.06

Ns, * and **: Non-significant, significant at 5 and 1 % probability levels, respectively.

The results of this experiment demonstrated the presence of genetic diversity among used wheat genotypes in respect of physiologic characteristics such as water retentions, chlorophyll, biologic and grain yields. Mean comparisons showed that genotype 21 had the highest RWD (38.3 %), RWL (55.6 %) and CMS (227 µmhos.cm^{-1}), genotype 8 had the highest ELWR (60.5 %), genotype 20 had the highest Chl-a (0.021 mg.g^{-1}), genotype 2 had the highest Chl-b (0.027 mg.g^{-1}) and Chl-t (0.042 mg.g^{-1}) and genotype 15 had the highest BY (3877 g.m^{-2}) and GY (1890 g.m^{-2}) (Table II).

The correlations of RWL, RWD, ELWR, CMS, Chl-a, Chl-b, Chl-t with BY and GY were not significant. There was a positive and significant correlation between BY and GY (r = 0.479). The correlations of CMS with RWD, RWL and ELWR were positive but non-significant. RWD had a positive correlation with RWL and a negative correlation with ELWR. There was a positive and non-significant correlation between RWD and ELWR. Positive but non-significant correlation was observed between Chll-a and Chl-b. Chl-t had positive and significant correlations with Chl-a (r = 0.795) and Chl-b (r = 0.772) (Table III).

TABLE II
MEAN COMPARISONS OF STUDIED TRAITS IN WHEAT GENOTYPES

Genotype	RWD (%)	RWL (%)	ELWR (%)	CMS (µmhos.cm^{-1})	Chl-a (mg.g^{-1})	Chl-b (mg.g^{-1})	Chl-t (mg.g^{-1})	BY (g.m^{-2})	GY (g.m^{-2})
1	31.1^{cde}	43.3^{abc}	57.0^{a}	128^{bcde}	0.003^{cdefg}	0.006^{bcde}	0.009^{efgh}	2349^{e}	798^{g}
2	34.1^{abc}	39.8^{abc}	57.2^{a}	66^{h}	0.015^{a}	0.027^{a}	0.042^{a}	2821^{bcde}	1085^{ef}
3	34.8^{ab}	41.0^{abc}	54.4^{ab}	91^{efgh}	0.007^{cd}	0.018^{a}	0.025^{b}	2913^{bcde}	1310^{bcdef}
4	22.3^{e}	33.8^{bc}	53.7^{abc}	79^{fgh}	0.002^{efghi}	0.004^{defg}	0.006^{ghij}	2448^{e}	1077^{ef}
5	27.4^{e}	33.4^{bc}	17.9^{d}	91^{efgh}	0.001^{ghi}	0.002^{hi}	0.006^{ghij}	2559^{de}	1025^{fg}
6	30.6^{de}	34.1^{bc}	59.4^{a}	106^{cdefg}	0.003^{cdefg}	0.005^{bcdef}	0.008^{efgh}	2585^{de}	1046^{fg}
7	30.9^{de}	27.5^{c}	60.0^{a}	111^{cdef}	0.001^{ghi}	0.007^{bc}	0.008^{efgh}	2750^{bcde}	1108^{ef}
8	26.3^{f}	17.3^{d}	60.5^{a}	131^{bcde}	0.004^{cdef}	0.007^{bc}	0.011^{def}	2559^{de}	1218^{cdef}
9	34.9^{ab}	27.9^{c}	59.0^{a}	131^{bcde}	0.001^{ghi}	0.003^{fgh}	0.004^{ijk}	2597^{de}	1155^{def}
10	31.4^{de}	28.5^{c}	55.2^{ab}	151^{bc}	0.012^{ab}	0.020^{a}	0.032^{ab}	2513^{e}	1097^{ef}
11	34.4^{abc}	37.5^{abc}	52.4^{abc}	161^{b}	0.002^{efghi}	0.004^{cdefg}	0.006^{ghij}	3491^{ab}	1492^{bc}
12	37.5^{a}	38.5^{abc}	59.4^{a}	73^{gh}	0.002^{efghi}	0.004^{cdefg}	0.006^{ghij}	2919^{bcde}	1164^{def}
13	32.4^{de}	31.2^{bc}	57.6^{a}	153^{bc}	0.005^{cde}	0.009^{b}	0.015^{cd}	3353^{ab}	1244^{bcdef}
14	31.2^{cde}	38.3^{abc}	57.0^{a}	156^{bc}	0.005^{cde}	0.001^{i}	0.006^{ghij}	3327^{ab}	1370^{bcde}
15	28.2^{e}	35.4^{bc}	59.5^{a}	79^{fgh}	0.002^{efghi}	0.003^{fgh}	0.005^{hijk}	3877^{a}	1890^{a}
16	32.5^{de}	48.1^{ab}	55.1^{ab}	104^{defg}	0.002^{efghi}	0.006^{bcde}	0.011^{defg}	2428^{e}	1224^{cdef}
17	25.7^{e}	48.7^{abc}	50.7^{abc}	109^{bcdef}	0.001^{ghi}	0.002^{ghi}	0.003^{k}	3525^{ab}	1510^{b}
18	30.8^{de}	31.8^{bc}	56.2^{ab}	98^{defg}	0.003^{cdefg}	0.002^{ghi}	0.007^{fghij}	3223^{abcd}	1434^{bcd}
19	35.7^{ab}	33.0^{bc}	40.6^{c}	65^{h}	0.005^{cde}	0.019^{a}	0.024^{bc}	3200^{abcd}	1301^{bcdef}
20	31.1^{cde}	55.2^{a}	42.7^{bc}	110^{cdef}	0.021^{a}	0.002^{hi}	0.021^{bc}	2773^{bcde}	1143^{ef}
21	38.3^{a}	55.6^{a}	53.8^{abc}	227^{a}	0.004^{cdef}	0.007^{bc}	0.012^{de}	2647^{cde}	1088^{ef}

Mean followed by the same letter(s) in each column are not significantly different at 5% probability level according to Duncan's Multiple Range Test.

TABLE III
CORRELATION COEFFICIENT AMONG STUDIED TRAITS.

	RWD	RWL	ELWR	CMS	Chl-a	Chl-b	Chl-t	BY	GY
RWD	1								
RWL	0.422^{ns}	1							
ELWR	0.028^{ns}	-0.126^{ns}	1						
CMS	0.109^{ns}	0.021^{ns}	0.184^{ns}	1					
Chl-a	0.135^{ns}	0.224^{ns}	0.200^{ns}	0.131^{ns}	1				
Chl-b	0.111^{ns}	-0.122^{ns}	0.266^{ns}	-0.075^{ns}	0.349^{ns}	1			
Chl-t	0.158^{ns}	0.116^{ns}	0.126^{ns}	-0.075^{ns}	0.795**	0.772**	1		
BY	-0.110^{ns}	0.041^{ns}	0.100^{ns}	-0.092^{ns}	-0.090^{ns}	-0.193^{ns}	-0.183^{ns}	1	
GY	-0.138^{ns}	0.048^{ns}	0.201^{ns}	-0.205^{ns}	-0.107^{ns}	-0.094^{ns}	-0.126^{ns}	0.479*	1

Ns, * and **: Non-significant, significant at 5 and 1 % probability levels, respectively.

REFERENCES

[1] N. C. Turner, “Crop water deficits: a decade of progress” Adv. Agron. vol. 39, pp. 1-51, 1986.

[2] M. singh, J. P. Srivastava, and A. Kumar, “Effect of water on water potential components in wheat genotypes” Indian J. Plant Physiol., vol. 33, pp. 312-317, 1990.

[3] T. R. Sinclair, and M. M. Ludlow, “Who taught plants thermodynamics? The unfulfilled potential of plant water potential” Aust. J. Plant Physiol., vol. 33, pp.213-217, 1985.

[4] L. R. Parsons, and T. K. Howe, “Effects of water stress on the water relations of Phaseolus vulgaris and the drought resistant phaseolus acutifolius” Physiol. Plant, vol. 60, pp. 197-202, 1984.

[5] M. A. Matin, J. H. Brown, and H. Ferguson, “Leaf water potential, relative water content, and diffusive resistance as screening techniques for drought resistance in barley” Agron. J., vol. 81, pp. 100-105, 1989.

[6] C. Tourneux, A. Devaux, M. R. Combacho, P. Mamani, and J. F. Ledent, “Effect of water shortage on six potato genotypes in the highlands of Bolivia, II: Water relations, Physiologcal Parameters” Agronomy, vol. 23, pp.181-190, 2003.

[7] R. C. Yang, S. Jana, and J. M. Clarke, “Phenotypic diversity and associations of some potentially drought responsive characters in durum wheat” Crop Sci., vol. 31, pp. 1484-1491, 1991.

[8] M. Bajji, J. M. Kinet, and S. Lutts, “The use of the electrolyte leakage method for assessing cell membrane stability as a water stress tolerance test in durum wheat” Plant Growth Regulation, pp.1-10, 2001.

[9] A. Blum, N. Klueva, and H. T. Nguyen, “ Wheat cellular thermotolerance is related to yield under stress” Euphytica, vol. 117, pp. 117-123, 2001.

[10] S. Thiaw, and A. E. Hall, “Comparison of selection for either leaf-electrolyte-leakage or pod set in enhancing heat tolerance and grain yield of cowpea” Field Crops Res., vol. 86, pp. 239-253, 2004.

[11] H. U. Rahman, S. A. Malik, and M. Saleem, “Heat tolerance of upland cotton during the fruiting stage evaluated using cellular membrane thermostability” Field Crops Res., vol. 85, pp.149-158, 2004.

[12] R. P. Marini, “Do net gas exchange rates of green and red peach leaves differ?” Horticulture, vol. 21, pp. 118-120, 1987.

[13] S. B. Brown, J. D. Houghton, and G. A. F. Hendry, “Chlorophyll breakdown. In: Scheer, H. (Ed.), Chlorophyllus” CRC Press, Boca Raton, pp. 465-489, 1991.

[14] G. A. F. Hendry, and A. H. price, “Stress indicators: chlorophylls and carotenoids. In: G. A. F. Hendry, and J. P. Grime (Eds.), Methods in Comparative Plant Ecology” Chapman & Hall, London, pp. 148-152, 1993.

[15] H. Schaper, and E. K. Chacko, “Relation between extractable chlorophyll meter reading in leaves of eight tropical and subtropical fruit-tree species” J. Plant Physiol. Vol. 138, pp. 674-677, 1991.

[16] A. T. Netto, E. Campostrini, J. G. Oliveira, and R. E. Bressan-Smith, “Photosynthetic pigments, nitrogen, chlorophyll a florescence and SPAD readings in coffee leaves” Scientia Horticulture, vol. 104, pp. 199-209, 2005.

[17] E. S. Costa, R. Bressan-Smith, J. G. Oliveira, and E. Campostrini, “Chlorophyll a fluorescence analysis in response to excitation irradiance in bean plants (phaseolus vulgaris L. and Vigna unguiculata L. Walp) submitted to high temperature stress” Photosynthetica, vol. 41(1), pp. 77-82, 2003.

[18] Y. Yamane, Y. Kashino, H, Koile, and K. Satoh, “Increase in the fluorescence F0 level reversible inhibition of photosystem II reaction center by high temperature treatments in higher plants” Photosynth. Res., vol. 57, pp. 57-64, 1997.

[19] M. A. Schonfeld, R. C. Johnson, B. F. Carver, and D. W. Mornhinweg, “Water relations in winter wheat as drought resistance indicator” Crop Sci., vol. 28, pp. 526-531, 1988.

[20] A. R. Wellburn, “The spectral determination of chlorophyll a and b, as well as total carotenoids, using various solvents with spectrophotometers of different resolution” J. Plant Physiol., vol. 144, pp. 307-313. 1994.