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Effect of Defoliation and Drought Stress on Yield Components and Chlorophyll Content of Wheat

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Abstract: In order to examine the effects of source restriction and drought stress on yield components, flag leaf chlorophyll content and Relative Water Content (RWC) of wheat cultivars, a greenhouse experiment was carried out at Shiraz University, Shiraz, Iran, during 2009-2010. The wheat cultivars including Shiraz, Bahar, Pishtaz, Sistan and Yavaros were sown in 5 kg plastic pots. The source manipulation treatments including control (C), defoliation of all leaves (D_1), defoliation of all leaves except the flag leaf (D_2) and defoliation of all leaves except the flag leaf and penultimate leaf (D_3), were imposed at anthesis. Results showed that source restriction decreased number of grain per spike significantly in Shiraz cultivar under drought stress condition. Maximum 100-grain weight was observed in C treatment of Yavaros and Shiraz under well-watered condition. Among the defoliation treatments under drought stress condition, D_2 in Pishtaz and D_1 in Shiraz had the highest (3.66 g) and lowest (2.71 g) 100 grain weight, respectively. In all cultivars drought stress decreased main shoot yield significantly but in Pishtaz and Sistan decreasing rate was less than the other cultivars. RWC in Shiraz decreased sharply from 92.1% in well-watered to 66.7% (27.5% reduction) under drought stress at 10 DAA. After anthesis, Pishtaz and Sistan maintained higher content of flag leaf chlorophyll (from 49.4 to 56.8 SPAD unit) under drought stress condition. Generally, selection and culture of cultivars that had small responses to defoliation might be a useful strategy in yield improvement of wheat in areas where the water availability is low.

Key words: 100-grain weight, relative water content, source restriction, wheat

INTRODUCTION

Wheat is the most food crops in South of Iran and Fars Province is the largest wheat producer in Iran (Emam *et al*., 2007). It plays a vital role in the national economy to decrease the gap between food production and food import in developing countries (Alam *et al*., 2008). The wheat grain yield mainly depends on the formation, translocation, partitioning and accumulation of assimilates during grain filling period (Emam and Seghatoleslami, 2005; Wang *et al*., 1997). Also, photosynthetic activity of source (leaves) and storage ability of the sink (grains) after anthesis are the main factors limiting wheat grain yield (Zhenlin *et al*., 1998; Ma *et al*., 1990).

Drought stress is an important limiting factor which can cause major loss in wheat productivity in arid and semi arid of Iran (Ahmadi *et al*., 2009). Growth of wheat grain is reduced depending upon degree of water stress and on the rate of stress development, thereby limiting final wheat yield (Plaut *et al*., 2004). In South of Iran such as Fars Province, limited rainfall and drought stress occurred frequently during the grain filling stage (Shekoofa and Emam, 2008). During the late season drought stress conditions, water potential in the rhizosphere becomes sufficiently negative and reduce water availability to suboptimal levels for crop development (Ahmadi and Joudi, 2007).

Dry matter production in wheat is highly related to sink-source relationships under different environments (Alam *et al*., 2008; Shekoofa and Emam, 2008). Manipulation of the source-sink ratio in wheat by artificial reduction in grain number per spike (Borras *et al*., 2004; Ma *et al*., 1990), defoliation (Bijanzadeh and Emam, 2010; Ahmadi *et al*., 2009) or use of chloromequat chloride (Shekoofa and Emam, 2008; Emam and Dastfal, 1997) has been trailed in several experiments to provide clear evidence that grain yield in wheat is mainly limited by the source strength, the sink capacity, or co-limited by both. Cruz-Aguado *et al*. (1999) concluded that final grain weight limited by the ability of the source to provide assimilation during grain filling period. In contrast, Borras *et al*. (2004) reported that under most conditions grain growth in wheat was more sink-limited.

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Ahmadi *et al*. (2009) suggested that the relative limitation of yield by source or sink is influenced by several factors and varies in different environments.

Artificial defoliation in wheat might be changed the photosynthetic characteristics of remaining tissues (Bijanzadeh and Emam, 2010; Zhenlin *et al*., 1998). Zhu *et al*. (2004) suggested that after anthesis, source restriction could enhance net photosynthesis rate, stomatal conductance and chlorophyll content of wheat flag leaf. Also, Joudi *et al*. (2006) reported that source restriction by defoliation of winter wheat increased net photosynthesis rate and chlorophyll content of most leaves, however, the range of increase depended on type of cultivars.

Little information has been published about the effects of source restriction on Iranian wheat cultivars. This research was conducted in order to study the effects of defoliation on yield components, flag leaf chlorophyll content and RWC of five Iranian wheat cultivars under well-watered and drought stress conditions.

MATERIALS AND METHODS

A greenhouse experiment was conducted to examine the effect of defoliation intensity and drought stress on grain development, chlorophyll content and RWC of durum and bread wheat cultivars at the College of Agriculture, Shiraz University, Shiraz, Iran during 2009 and 2010 growing season. Iranian wheat cultivars including Shiraz, Bahar, Pishtaz, Sistan (as bread wheat) and Yavaros (as durum wheat) were sown in 5 kg pots filled with a silt loamy soil (sand, silt and clay: 7, 67.6 and 25.4%, respectively pH = 7.8 and EC = 0.43 dS m^{-1}) and 22 mg kg^{-1} nitrogen as urea was applied. The experimental design was a completely randomized with four replications. Ten seeds of each wheat cultivar were sown in each pot, on September 28th 2009 and at three-leaf stage thinned to six seedlings. The greenhouse temperature was 25°C (±5), with 70% (±10) relative humidity and light intensity varied in the range of 600-1000 µmol m^{-2} sec^{-1}.

Before sowing the seeds in pots, the Field Capacity (FC) of the soil was determined in the laboratory to set the irrigation regimes. Irrigation regimes were well-watered (100% FC) and drought stress (50% FC). Pots weighted every other day and irrigated according to 50% FC for drought stress treatment and 100% FC for well-watered treatment from anthesis until late season.

The source restriction treatments including control (C) in which the plants were intact, defoliation of all leaves (D1), defoliation of all leaves except the flag leaf (D2) and defoliation of all leaves except the flag leaf and penultimate leaf (D3), were imposed at anthesis on November 27th 2009. To avoid contribution of tillers in grain filling of main shoot, all plants were de-tillerd at anthesis (Emam, 2007; Slafer and Savin, 1994).

The SPAD meter is a hand-held spectrometer which measures light (650 nm) absorbed by single leaves and gives a non-destructive estimate of plant chlorophyll (Barraclough and Kyte, 2001). In this study, the SPAD chlorophyll meter (Opti-Sciences X. USA) was used to acquire a rapid estimate of flag leaf chlorophyll content at 10 and 20 Days after Anthesis (DAA). For the leaf relative water content (RWC) measurement immediately after cutting the flag leaf in each plant, leaf sealed within plastic bags and quickly transferred to the laboratory. Fresh weight was determined within 1 hour after excision. Turgid weight was obtained after soaking leaves in distilled water for 5-7 h at room temperature (about 25°C). After soaking, leaves were carefully blotted dry with tissue paper to determine turgid weight. Dry weight was obtained after oven drying the leaf sample for 72°C. Then, the RWC was calculated according to Beadle *et al*. (1993) using the equation:

$$\text{RWC} = \left(\frac{\text{Fresh weight-Dry weight}}{\text{Turgid weight-Dry weight}}\right)\times 100$$

Finally, crops were harvested at physiological maturity (January 18th, 2010) and oven-dried at 80°C, then yield components including the number of grains per spike, 100 grain weight, spike compactness (spikelet number per spike divided by spike length (Emam, 2007) and main shoot yield was determined. The data were subjected to analysis of variance by SAS software (SAS, 2000) and the means were separated using Fisher's LSD protected test at 0.05 probability level.

RESULTS

Effects of defoliation on wheat yield and yield components: In all cultivars defoliation treatments had no significant effect on number of grain per spike under well-watered condition while, source restriction decreased number of grain per spike significantly in Shiraz cultivar from 29.2 in C to 27.1 in D1 and in Yavaros from 27.4 in C to 26.1 in D1 treatment, under drought stress condition (Table 1). On the other hand, Bahar, Pishtaz and Sistan cultivars had the least reduction of number of grain per spike under defoliation and drought stress conditions (Table 1).

Maximum 100-grain weight was observed in C treatment of Yavaros (4.43 g) and Shiraz (4.04 g) under well-watered condition (Table 1). In all cultivar 100-grain weight decreased under drought stress compared to well-watered condition in a similar level of defoliation. However, decreasing rate of 100-grain weight in Shiraz

Table 1: Effect of defoliation on yield components of wheat cultivars under well-watered and drought stress condition

Wheat cultivars	Irrigation status	Defoliation treatments	No. of grain per spike	100-grain weight (g)	Main shoot yield (g)
Shiraz	Well-watered	C[a]	31.1	4.04	1.26
		D1	30.3	3.66	1.11
		D2	30.1	3.91	1.14
		D3	30.3	3.81	1.15
	Drought	C	29.2	3.02	0.88
		D1	27.1	2.71	0.73
		D2	27.3	2.75	0.75
		D3	27.6	2.80	0.77
Bahar	Well-watered	C	28.6	3.78	1.08
		D1	28.1	3.81	1.07
		D2	28.1	3.86	1.08
		D3	28.3	3.72	1.05
	Drought	C	27.9	3.50	0.92
		D1	27.3	3.53	0.96
		D2	27.1	3.52	0.95
		D3	27.0	3.59	0.97
Pishtaz	Well-watered	C	31.3	3.88	1.21
		D1	30.9	3.81	1.18
		D2	30.8	3.89	1.19
		D3	31.0	3.90	1.21
	Drought	C	30.1	3.51	1.08
		D1	29.1	3.55	1.06
		D2	29.3	3.66	1.07
		D3	29.4	3.56	1.04
Sistan	Well-watered	C	32.8	3.73	1.22
		D1	32.6	3.66	1.19
		D2	32.4	3.70	1.20
		D3	32.8	3.65	1.20
	Drought	C	32.3	3.44	1.10
		D1	32.1	3.41	1.09
		D2	32.2	3.40	1.09
		D3	32.2	3.42	1.08
Yavaros	Well-watered	C	30.8	4.43	1.36
		D1	30.6	4.14	1.26
		D2	30.4	4.23	1.27
		D3	30.5	4.26	1.29
	Drought	C	27.4	3.71	1.01
		D1	26.1	3.22	0.84
		D2	26.2	3.33	0.87
		D3	26.1	3.41	0.88
[b]LSD			1.1	0.21	0.10

[a]Control (C), Defoliation of all leaves (D1), Defoliation of all leaves except the flag leaf (D2) and Defoliation of all leaves except the flag leaf and penultimate leaf (D3). [b]Least significant differences for comparison of treatments

and Yavaroos cultivars was significantly more than the other cultivars. Among the defoliation treatments under drought stress condition, D2 in Pishtaz and D1 in Shiraz had the highest (3.66 g) and lowest (2.71 g) 100-grain weight, respectively (Table 1).

In all cultivars drought stress decreased main shoot yield significantly but in Pishtaz and Sistan decreasing rate was less than the other cultivars (Table 1). The main shoot yield in Shiraz cultivar declined drastically by source restriction from 1.26 in C to 1.11 g in D1 under well-watered and from 0.88 in C to 0.73 g in D1, under drought stress condition. In all cultivars except the Shiraz and Yavaros, the main shoot yield was not affected by defoliation treatments and Shiraz and Yavaros cultivars was more sensitive to drought stress than the other cultivars (Table 1).

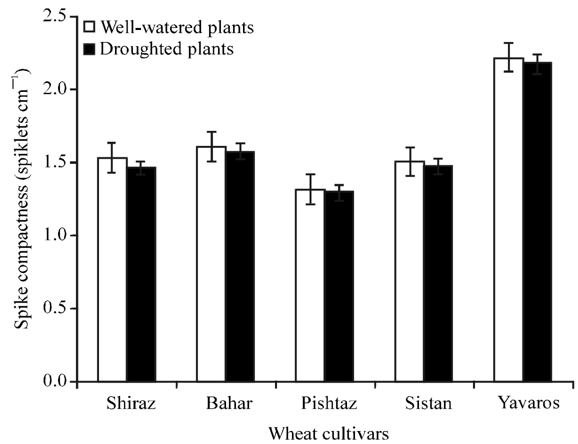

Fig. 1: Spike compactness of five wheat cultivars under well-watered and drought stress conditions

In both of the irrigation regimes in Bahar, Pishtaz and Sistan cultivars there were no significant differences among D1, D2 and D3 treatments and penultimate leaf had no significant effect on main shoot yield compared to D2 treatment (Table 1). In Shiraz cultivar, D1 treatment decreased main shoot yield from 1.11 g under well-watered condition to 0.73 under drought stress (34% reduction) while in Pishtaz, the main shoot yield of D1 declined from 1.18 g to 1.06 (10% reduction).

The spike compactness of five wheat cultivars was shown in Fig. 1. Results showed that, in all cultivars drought stress had no significant effect on spike compactness compared to well-watered. Under well-watered and drought stress conditions Pishtaz and Yavaros cultivars had the lowest (1.32 and 1.30 spikelet cm^{-1}) and highest (2.22 and 2.19 spikelet cm^{-1}) spike compactness, respectively. Also, Pishtaz and Sistan cultivars had large awns 8.4 and 8.1 cm, respectively and awn photosynthesis might have contributed to grain filling under defoliation (data not shown).

Effects of defoliation on RWC and chlorophyll content: At 10 DAA, in Shiraz, Bahar and Yavaros cultivars drought stress caused a significant reduction in flag leaf RWC at the similar level of defoliation treatments compared to well-watered condition (Table 2). The RWC of Shiraz and Yavaros in D2 and D3 treatments was higher (from 90.3 to 92.6%) than the other cultivars under well-watered condition however, RWC in Shiraz decreased sharply from 92.1% in well-watered to 66.7% (27.5% reduction) under drought stress and in Yavaros from 92.6 to 70.4% (23.9% reduction) at 10 DAA. Similar trend was obtained for RWC at 20 DAA and Pishtaz and Sistan cultivars had the highest RWC compared to other cultivars under drought stress (Table 2).

Table 2: Effect of defoliation on Relative Water Content (RWC) and flag leaf chlorophyll content of five wheat cultivars at 10 and 20 Days after Anthesis (DAA)

Wheat cultivars	Irrigation status	Defoliation treatments	RWC (%) 10DAA	RWC (%) 20DAA	Chlorophyl content (SPAD unit) 10DAA	Chlorophyl content (SPAD unit) 20DAA
Shiraz	Well-watered	C[a]	89.4	76.2	56.1	54.1
		D2	90.3	79.1	58.3	55.2
		D3	92.1	75.2	56.8	54.1
	Drought	C	64.3	54.1	35.1	32.7
		D2	66.7	55.3	38.2	33.9
		D3	67.1	50.1	37.2	33.5
Bahar	Well-watered	C	87.7	79.2	57.9	55.7
		D2	88.1	80.6	58.3	56.4
		D3	86.4	78.3	56.2	54.3
	Drought	C	79.6	61.2	46.1	40.3
		D2	72.3	63.1	48.2	42.1
		D3	71.1	62.8	47.1	41.8
Pishtaz	Well-watered	C	81.1	77.3	55.5	53.5
		D2	83.0	78.6	58.6	51.6
		D3	83.6	78.3	58.1	50.1
	Drought	C	79.2	71.5	50.5	49.4
		D2	80.6	72.3	56.8	50.7
		D3	80.5	72.7	56.6	51.4
Sistan	Well-watered	C	86.3	74.2	53.1	50.4
		D2	87.2	73.2	57.1	52.1
		D3	86.4	72.7	56.2	53.2
	Drought	C	79.1	68.3	50.1	49.1
		D2	83.2	66.2	53.1	48.3
		D3	81.1	66.9	52.7	50.7
Yavaros	Well-watered	C	88.3	79.6	58.3	50.3
		D2	92.6	80.1	59.2	50.2
		D3	92.2	79.8	59.1	51.1
	Drought	C	69.3	56.1	40.1	36.1
		D2	70.4	57.3	42.7	35.3
		D3	77.3	51.2	41.6	36.1
[b]LSD(0.05)			10.2	9.70	6.30	5.10

[a]Control (C): Defoliation of all leaves (D1), Defoliation of all leaves except the flag leaf (D2) and Defoliation of all leaves except the flag leaf and penultimate leaf (D3). [b]Least significant differences for comparison of treatments

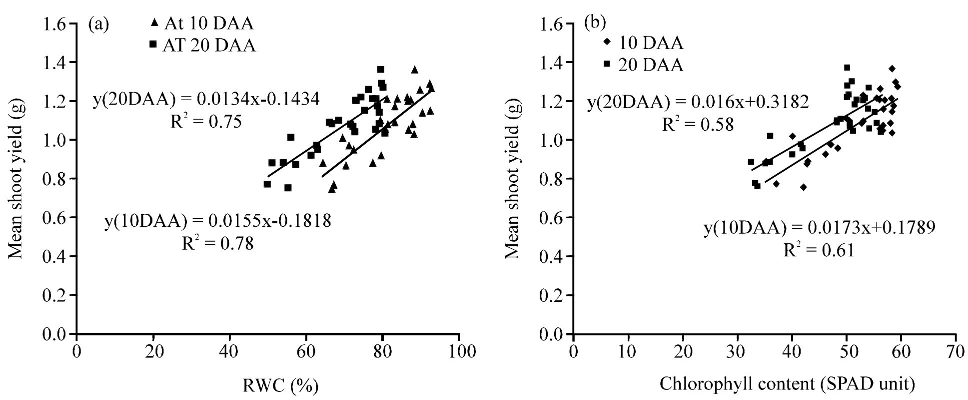

Fig. 2: Relationship between (a) main shoot yield of wheat and RWC and (a) flag leaf chlorophyll content (b) at 10 and 20 Days after Anthesis (DAA)

In each irrigation regime in all cultivars, flag leaf chlorophyll content was not affected by source restriction while, drought stress in Shiraz, Bahar and Yavaros caused a significant decreasing trend of chlorophyll content at 10 and 20 DAA (Table 2). After anthesis, Pishtaz and Sistan maintained higher content of flag leaf chlorophyll (from 49.4 to 56.8 SPAD unit) under drought stress condition. The relationship between RWC and main shoot yield at 10 ($R^2 = 0.78$) and 18 DAA ($R^2 = 0.75$) was highly significant (Fig. 2a). Also, a significant positive relationship was obtained between flag leaf chlorophyll content and main shoot yield at 10 DAA ($R^2 = 0.61$) and at 20 DAA ($R^2 = 0.58$) (Fig. 2b).

DISCUSSION

In all wheat cultivars the number of grain per spike was not affected by source restriction under well water condition. However, defoliation and drought stress decreased gradually the number of grain per spike of Shiraz and Yavaros cultivars after anthesis (Table 1). Zhenlin *et al*. (1998) reported that removing of all leaves declined partially the grain number of wheat 3 to 6%. In a study with 20 cultivars and lines of wheat Alam *et al*. (2008) asserted that number of grain per spike decreased significantly by removal of all leaves after anthesis.

It appears that 100 grain weight of Shiraz and Yavaros were more sensitive to source restriction and drought stress compared to the other cultivars (Table 1). Similar to our results, Ahmadi *et al*. (2009) drought stress reduced grain weight of Ghods wheat cultivar, significantly. Sadras (2007) in a review on evolutionary aspects of the trade off between seed size and number in crops declared that wheat grain weight is more conservative than grain number. In our study, the variation in 100 grain weight was low between defoliation treatments in all cultivars except the Shiraz (Table 1).

The main shoot yield decreased in Shiraz and Yavaros 42% and 38% respectively in D_1 treatment under drought stress compared to well-watered condition (Table 1). Singh and Singh (2002) showed that source restriction reduced 30 to 40% yield of wheat cultivars. Bijanzadeh and Emam (2010) declared that in Shiraz cultivar, defoliation of all leaves decreased main shoot yield by 40.75% and this demonstrated that Shiraz was sensitive to source restriction under well-watered condition. Generally, genetic diversity was observed among wheat cultivars when were imposed to source restriction and drought stress.

The low spike compactness in Pishtaz cultivar (Fig. 1) might be associated with more light penetration to the spike of this cultivar. It appeared that when the compactness of the spike is lower the solar radiation could be used more efficiently by the spike components (Bijanzadeh and Emam, 2010). Also, the lower responses of Pishtaz and Sistan to changes in assimilate availability by defoliation after anthesis might suggest that grain yield of theses cultivars is more regulated by sink rather than source (Table 1) and photoassimilate translocation from the spike components could support yield under source reduction (Ahmadi and Joudi, 2007; Joudi *et al*., 2006).

Water status of wheat cultivars was evaluated by the determination of flag leaf RWC in C, D2 and D3 treatments (Table 2). The variation in RWC was observed among wheat cultivars under drought stress condition and Pishtaz and Sistan maintained higher RWC in flag leaf than the other cultivars. In each cultivar and irrigation regime, RWC was not affected by defoliation treatments. Possible explanation to no significant effects of defoliation on flag leaf RWC is that, while reducing transpiration, the defoliation inevitably increased bare soil evaporation as soil was exposed following source restriction (Ahmadi and Joudi, 2007). In contrast, Bijanzadeh and Emam (2010) reported that wheat cultivars with low transpiration rates might conserve higher Relative Water Content (RWC) in their leaves under water deficit conditions. Therefore, source restriction by removal of transpiring leaves which are less effective in grain filling could be one way to decrease water loss during grain filling period of wheat. Similar to present results, Rohi and Mardeh (2008) declared that there were a positive relationship between flag leaf RWC and wheat yield under moisture stress condition.

Flag leaf chlorophyll content is an indicator of the photosynthetic activity and its stability for the conjugation of assimilate biosynthesis. Present results showed that flag leaf chlorophyll content was not affected by source restriction while, drought stress caused a significant reduction in chlorophyll content of Shiraz and Yavaros cultivars. Theses results were in agreement with Ahmadi and Joudi (2007) who asserted that changes in chlorophyll content was low in Ghods wheat cultivar by defoliation treatment. In contrast, in the greenhouse experiment Barraclough and Kyte (2001) reported that chlorophyll content of winter wheat (CV. Hereward) decreased significantly under drought stress. In present study, it appears that accelerated senescence of remaining leaf of Shiraz and Yavaros cultivars caused a significant reduction of chlorophyll content under drought stress compared to well-watered condition (Table 2). One of the most important factors regulating leaf senescence at the whole plant is the sink-source relationship after anthesis especially under drought stress condition (Rajcan and Tollenaar, 1999; Yin *et al*., 1998).

CONCLUSION

It concluded that Pishtaz and Sistan cultivars, did not show a consistent trend toward a reduction in number of grain per spike, grain weight and main shoot yield if the availability of assimilate reduced. In contrast, yield and yield components of Shiraz and Yavaros cultivars were sensitive to defoliation and drought stress and demonstrated that in these cultivars reduced source size by defoliation was associated with reduced sink development. Also, flag leaf chlorophyll content and

RWC of Shiraz and Yavaros, were sensitive to drought stress while, Pishtaz and Sistan maintained higher content of flag leaf chlorophyll at late season drought stress. Pishtaz and Sistan wheat cultivar, responded to source manipulation slightly and it appears that theses cultivars were tolerant to late season drought stress and source restriction. Probably, spike structure of Pishtaz cultivar might be affect yield and yield components of this cultivar positively under source restriction and drought stress. Generally, selection and culture of cultivars such as Pishtaz and Sistan that had small responses to defoliation after anthesis might be a useful strategy in yield improvement particularly in regions where the water availability is low in the grain filling period.

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