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Physiological Performance of Soybean Cultivars Under Salinity Stress

K Ghassemi-Golezani[1*], M Taifeh-Noori[2], S Oustan[1], M Moghaddam[1], S Seyyed Rahmani[3]

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[1]Department of Agronomy and Plant Breeding, Faculty of Agriculture, University of Tabriz, Iran
[2]Islamic Azad University, Maragheh Branch, Iran
[3] Agricultural Products Insurance Fund, Iran
*Corresponding Author: Email: golezani@gmail.com

Abstract

Two experiments with factorial arrangements on the basis of randomized complete block design in three replications were conducted in 2007 and 2008, to evaluate chlorophyll content index (CCI), fluorescence of chlorophyll, proline content and grain yield of three soybean cultivars (Williams, Zan and L_{17}) under a non-saline (control) and three saline (3, 6 and 9 ds/m NaCl) conditions in the greenhouse. Six seeds were sown in each pot filled with 900 g perlite, using 144 pots in each experiment. After emergence, seedlings were thinned and four plants were kept in each pot. Zan had the highest leaf proline content, but the lowest *CCI*, chlorophyll fluorescence and grain yield per plant. However, these traits were statistically similar with those of L_{17} and Williams. The chlorophyll content index and fluorescence of chlorophyll in soybean leaves decreased with increasing salinity. Reduction in chlorophyll fluorescence due to salinity stress was related to the damage of chlorophyll under saline conditions. In contrast, leaf proline content increased as salinity increased. Mean grain yield per plant under salinity stress was much less than that under non-saline conditions. These reductions were closely related with low *CCI* and PS *II* activity (fv/fm) and high leaf proline content in soybean cultivars. It was concluded that soybean is a sensitive plant to salinity stress, but the extent of this sensitivity varies among cultivars.

Keywords: Chlorophyll content, Fluorescence, Proline, Salinity stress, Soybean

Introduction

Soybean is an important grain legume. The unique chemical composition of soybean has made it one of the most valuable agronomic crops worldwide (Thomas *et al.* 2003). Its protein has great potential as a major source of dietary protein. The oil produced from soybean is highly digestible and contains no cholesterol (Essa and Al-Ani 2001). Wide genetic variability exists among different cultivars of soybean (Shereen and Ansari 2001). Nevertheless, soybean production may be limited by environmental stresses such as soil salinity (Ghassemi-Golezani *et al.* 2009).

Soil salinity, resulting from natural processes or from crop irrigation with saline water, occurs in many arid and semi-arid regions of the world (Meloni *et al.* 2004). Most of the salt stresses in nature are due to Na^+ salts, particularly NaCl (Demirel 2005). High salinity lowers water potential and induces ionic stress, and results in secondary oxidative stress. Salinity is known to cause

changes in physiological processes in plants (Ganivea *et al.* 1998).

Netondo *et al.* (2004) reported that photosynthetic activity decreases when plants are grown under saline conditions leading to reduced growth and productivity. The reduction in photosynthesis under salinity can be attributed to a decrease in chlorophyll content (Jamil *et al.* 2007) and activity of photo-system *II* (Ganivea *et al.* 1998). Salinity can affect chlorophyll content through inhibition of chlorophyll synthesis or an acceleration of its degradation (Reddy and Vora 1986). Fluorescence of chlorophyll reflected the photochemical activities of photo-system *II* (Ganivea *et al.* 1998). Photochemical efficiency of photo-system *II* (fv/fm) could be reduced by salinity stress (Netondo *et al.* 2004, Jamil *et al.* 2007).

Plants have evolved complex mechanisms that contribute to the adaptation to osmotic stress caused by high salinity (Meloni *et al.* 2004). Osmotic adjustment has undoubtedly gained considerable recognition as a significant and effective mechanism of salinity tolerance in crop plants (Pakniyat and Armion 2007). In salt stressed plants osmotic potential of vacuole decreased by proline accumulation (Yoshiba *et al.* 1997). Several possible roles have been attributed to supra-optimal level of proline including osmoregulation under salinity, stabilization of proteins and prevention of heat denaturation of enzymes and conservation of nitrogen and energy for a post-stress period (Aloni and Rosenshtein 1984). This research was carried out to evaluate changes in chlorophyll content and fluorescence and proline accumulation in leaves of soybean cultivars in response to salinity stress. The consequences of such changes on crop yield were also investigated.

Materials and Methods

Seeds of three soybean cultivars (Williams, Zan and L_{17}) were obtained from Agricultural Research Institute, Moghan, Iran. Two experiments with factorial arrangements on the basis of randomized complete block design with three replications were conducted in 2007 and 2008 to investigate changes in chlorophyll content index (seven weeks) and fluorescence of chlorophyll (four times) in leaves and to determine proline content and grain yield of three soybean cultivars under a non-saline (control) and three saline (3, 6 and 9 ds/m NaCl) conditions. Six seeds were sown 3 cm deep in each pot, filled with 900 g perlite, using 144 pots in each experiment. Pots were then placed in the greenhouse. The temperature variation in the greenhouse was 17-34ºC and 13-28ºC during the first and second experiments, respectively. Tap water and saline solutions were added to the pots in accordance with the treatments to achieve 100% FC.

After emergence, seedlings were thinned to keep four plants in each pot. During the growth period, the pots were weighed and the losses were made up with Hoagland solution (EC = 1.3 dS/m). Perlites within the pots were washed every 25 days and non-saline and salinity treatments were reapplied in order to prevent further increase in electrical conductivity (EC), due to adding the Hoagland solution.

Leaf chlorophyll content index (CCI) was measured by a chlorophyll meter (CCM-200, Opti- Science, USA) in weekly intervals for seven weeks. After seedling establishment, a plant was marked in each pot and CCI of upper, middle and lower leaves was measured at each stage. Subsequently, mean CCI for each treatment and replicate at each developmental stage was calculated.

The chlorophyll fluorescence induction parameters were measured in leaves by a chlorophyll fluorometer (OS-30, OPTI-SCIENCES, USA) every 10 days from 30 to 60 days after sowing. Fluorescence emission was monitored from the upper surface of the leaves. Dark-adapted leaves (30 min.) were initially exposed to the weak modulate measuring beam, followed by exposure to saturated white light to estimate the initial (F0) and maximum (Fm) fluorescence values, respectively. Variable fluorescence (Fv) was calculated by subtracting F0 from Fm. The Fv/Fm ratio measures the efficiency of excitation energy capture by open PSII reaction centers, representing the maximum capacity of light-dependent charge separation in PSII (Rizza *et al.* 2001).

The proline content was determined spectrophotometrically according to Bates *et al.* (1973). In each experimental unit, 200 mg leaf samples were powdered in liquid nitrogen and were homogenized in 5 ml sulphosalcylic acid. Then, 2 ml acid ninhydrine and 2 ml glacial acetic acid were added to the extract. The samples were heated at 100 °C. The mixture was extracted with toluene and the free toluene was quantified spectrophoto-metrically at 520 nm.

At maturity, plants of each pot were separately harvested and grains were detached from the pods. Finally, grains were weighed and grain yield per plant for each treatment at each replicate was determined.

MSTATC software was used to analyze the data for CCI and chlorophyll fluorescence as factorial split plot and those for proline and grain yield as factorial. Means of the traits were compared at $p \leq 0.05$. Figures were drawn using Excel software.

Results

The results of analysis of variance showed highly significant ($P \leq 0.01$) effects of year, cultivar, salinity and measuring time on both chlorophyll content index (*CCI*) and fluorescence of chlorophyll. Means of *CCI* and fv/fm in 2007 experiment were higher than those in 2008. The CCI and fluorescence of chlorophyll in soybean leaves decreased with increasing salinity. L17 and Zan had the highest and the lowest *CCI* and fv/fm, respectively.

Mean *CCI* and chlorophyll fluorescence of soybean cultivars increased with progressing plant growth up to a point where maximum values was a chieved under non-saline and saline conditions (Figures 1 and 2). Maximum CCI of all cultivars under salinity treatments was obtained earlier than that under non-saline treatment (Figure 1), but maximum chlorophyll fluorescence under all treatments was achieved at almost similar stage (Figure 2). Thereafter, due to senescing of leaves, CCI and chlorophyll fluorescence started to decrease. Mean CCI and chlorophyll fluorescence at all developmental stages decreased as the salinity increased. In general, L17 and Williams had more *CCI* and chlorophyll fluorescence at different stages of growth and development, compared to Zan (Figure 1).

Leaf proline content and grain yield per plant were significantly ($P \leq 0.01$) affected by cultivar and salinity, but cultivar × salinity interaction was not significant for these traits ($P \leq 0.05$). Leaf proline content of soybean increased with increasing salinity. Proline content of Zan was significantly higher than that of Williams and L_{17}. However, proline content of the latter cultivars was similar (Table 2). Grain yield per plant significantly decreased as salinity increased. Zan had the

lowest grain yield per plant, but there was no significant difference in grain yield of L_{17} and Williams (Table 2).

Discussion

Decreasing chlorophyll content index (CCI) of soybean leaves with increasing salinity (Table 1, Figure 1) could be related to increasing the activity of chlorophyll degrading enzyme, chlorophyllase (Jamil *et al*. 2007), and the destruction of the chloroplast structure and the instability of pigment protein complexes (Singh and Dubey 1995). Similar results were reported for tomato (Lapina and Popov 1970), pea (Hamada and El-Enany 1994), alfalfa (Winicov and Seemann 1990), sunflower (Ashraf 1999), sorghum (Netondo *et al.* 2004), and wheat (El-Hendawy *et al.* 2005). Differences in CCI among cultivars (Table 1, Figure 1) indicate that this trait can be also influenced by genetic constitution.

Table 1. Means of chlorophyll content index (*CCI*) and fluorescence chlorophyll (fv/fm) of three soybean cultivars under salinity stress

Treatment		*CCI*	fv/fm
Year	1	13.66 [a]	0.779 [a]
	2	10.50 [b]	0.728 [b]
Salinity (dS m^{-1})	0	14.06 [a]	0.792 [a]
	3	12.81 [b]	0.768 [b]
	6	11.50 [c]	0.742 [c]
	9	9.97 [d]	0.713 [d]
Cultivar	L_{17}	12.63 [a]	0.764 [a]
	Zan	11.19 [b]	0.739 [b]
	Williams	12.43 [a]	0.758 [a]

Different letters for each factor in each column indicate significant difference at $p \leq 0.05$.

Table 2. Means of proline content and grain yield per plant for three soybean cultivars under salinity stress

Treatment		Proline content (mM/g)	Grain yield per plant (g)
Salinity (dS m^{-1})	0	19.40 [d]	1.250 [a]
	3	26.12 [c]	0.892 [b]
	6	39.28 [b]	0.516 [c]
	9	45.89 [a]	0.274 [d]
Cultivar	L_{17}	31.71 [b]	0.782 [a]
	Zan	35.36 [a]	0.651 [b]
	Williams	30.96 [b]	0.766 [a]

Different letters in each factor in each column indicate significant difference at $p \leq 0.05$.

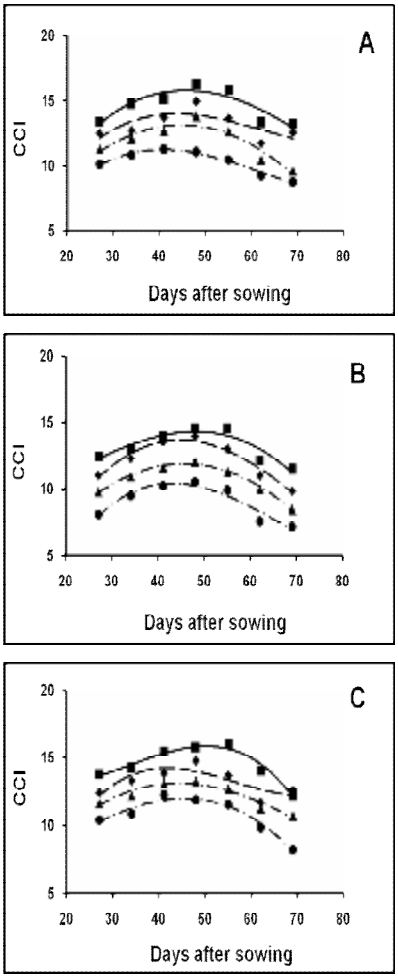
Reduction in fv/fm due to salinity stress (Table 1, Figure 2) is possibly related to the damage of chlorophyll under saline conditions (Ganieva *et al.* 1998). Ashraf (2004) found that ionic imbalance can also cause the reduction in fv/fm under high salinity conditions. Nasir khan *et al.* (2007) reported that the decrease in chlorophyll content and PS *II* activity have adverse effect on growth and grain yield of treated plants.

Increasing leaf proline content under salinity stress (Table 2) might be caused by the induction or activation of proline synthesis from glutamate or decrease in its utilization in protein synthesis or enhancement in protein turnover. Thus, proline may be the major source of energy and nitrogen during immediate post stress metabolism and accumulated proline apparently supplies energy for growth and survival, thereby inducing salinity tolerance (Gad 2005). Zan had the highest proline content (Table 2) and the lowest CCI and fv/fm (Table 1). Gad (2005) also reported that proline content was much higher in sensitive cultivar of tomato than in salt-tolerant.

Large reductions in grain yield per plant clearly show that soybean is a salt sensitive crop, but the extent of this sensitivity varies among cultivars (Table 2). Salinity can severely limit crop production because high salinity lowers water potential and induces ionic stress and results in a secondary oxidative stress (Shanon 1998). Reductions in grain yield as a result of salt stress have also been reported for some other crop species (Katerji *et al.* 1992, Ashraf 2004, Sohrabi *et al.* 2008). These reductions are closely related with low *CCI* and PS *II* activity (Table 1) and high leaf proline content (Table 2) in soybean cultivars.

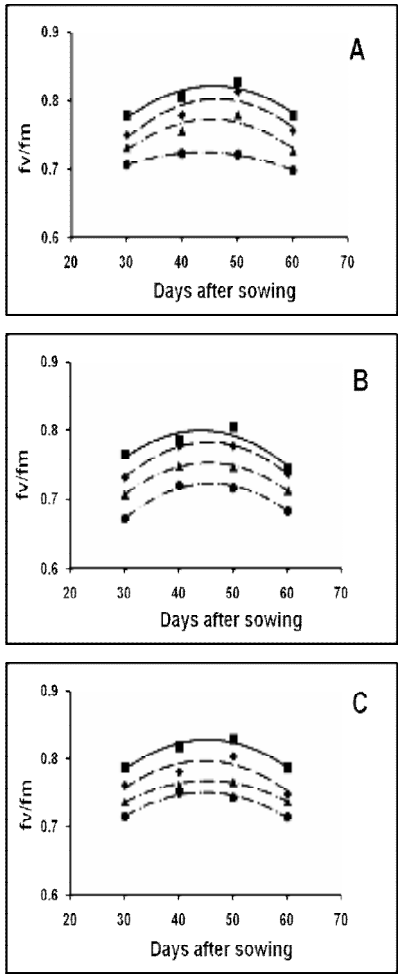
Conclusion

Salinity stress can considerably reduce chlorophyll content index and PS *II* activity and consequently grain yield per plant in soybean cultivars. These reductions enhance with increasing salinity. In contrast, leaf proline content increases due to NaCl salinity. In general, soybean is a sensitive crop to salinity stress, but the extent of this sensitivity varies among cultivars.



A: Williams B: Zan C: L17, — 0 ds/m – – 3 ds/m - - - 6 ds/m · · · 9 ds/m

Figure 1. Changes in chlorophyll content index (*CCI*) of soybean cultivars under non-saline (control) and saline conditions (means of two years).



A: Williams B: Zan C: L17, — 0 ds/m – – 3 ds/m - - - 6 ds/m · · · 9 ds/m

Figure 2. Changes in chlorophyll fluorescence (fv/fm) of soybean cultivars under non-saline (control) and Saline conditions (means of two years).

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Relationships Between Seedling Growth Rate and Yield of Maize Cultivars Under Normal and Water Stress Conditions

M Zharfa[1*], AA Maghsoudi Moud[2] and VR Saffari[3]

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[1]Graduate student, Department of Agronomy and Plant Breeding, Faculty of Agriculture, S.B. University of Kerman, Kerman, Iran
[2]Assistant Professor, Department of Agronomy and Plant Breeding, Faculty of Agriculture, S.B. University of Kerman, Kerman, Iran
[3]Assistant Professor, Department of Plant Production, Faculty of Agriculture, S.B. University of Kerman, Kerman, Iran
*Corresponding author. Email: maryam.zharfa@gmail.com

Abstract
Effects of water stress on root and leaf growth rates and their relationships with yield under normal and water stress conditions were examined at the Greenhouse and Research Field of Faculty of Agriculture, Kerman University, Iran, using seven maize cultivars including SC-404, SC-704, BC-666, TC-647, DC-370, Jeta and Kordona. During a period of 14 days, water stress at -0.4 MPa was imposed by application of PEG-6000 to the seedlings. The root and leaf growth were measured every day. Leaf and root growth rates were expressed as the slope of the line fitted to the data of length and time. Results showed that water stress inhibited root and shoot growth in all cultivars. Differences were found among cultivars in terms of root and leaf growth rates. Differences were greater under normal compared with water stress condition. In a field experiment during 2007- 2008 growing season, yield performances of the same cultivars were measured under water stress and normal conditions. Correlation coefficients of yield with root (0.54), (0.10) and leaf (0.79), (0.32), (0.91), (0.63) growth rates were stronger under normal compared with water stress condition. Higher grain yield (mean of 10 plants per plot) of cultivars with higher growth rates under normal condition may be attributed to the higher growth rates at the early stages of growth. It could be also indicated that the higher growth rate ability of cultivars decreases when they are exposed to water stress condition.

Keywords: Corn, Growth rate, Water stress, Yield

Introduction

Water stress adversely affects crop growth and yield in many regions of the world. (Teulat *et al.* 1997). Maintaining high water status plays an important role in tolerance to water stress and in yield stability of crop plants (Teulat *et al.* 1997). Different mechanisms such as developed root systems are involved in maintaining plant water status at high levels and plant normal growth and functioning depends on the amount of water supplied by the root system. Under water stress condition, reduced root and shoot growth in maize (Kolarovic *et al.* 2006) and also root growth in wheat (Blum *et al.* 1988, Galle *et*

al. 2002 and Akmal and Hirasawa 2004) were reported. Water stress was also shown to decrease root and shoot dry weight in wheat (Kerepesi and Galiba 2000), root and stem dry weight in soybean (Michalek and Browski 2005) and root weight in maize (Chammacho and Caraballo 1994, Ogawa *et al.* 2005, Grzesiak *et al*. 2007), rice (Cui *et al*. 2008) and triticale (Grzesiak *et al*. 2007). Water stress reduced relative growth rate of adventitious roots in maize and millet (Blum 1986) and elongation rate of roots in maize (Ogawa *et al*. 2006). Root length and number of roots were shown to decrease under water stressed environment in maize (Ogawa *et al.* 2005) and rice (Cui *et al*. 2008). Leaf growth rate and number of leaves which are considered as stress tolerance indicators were reduced under water stress condition (Ingram and Bartels 1996, Veselov *et al.* 2002). As the level of water stress increased, leaf growth rate in barley, maize and rice (Lu and Neumman 1998) and leaf area in maize (Sobrado 1986) were shown to be decreased. Leaf wilting was also reported in maize under severe water stress conditions (Lu *et al*. 2007).

Maize is usually grown in loamy soils in some parts of central region of Iran. Seedlings are, therefore, exposed to short term water stress as these types of soil often can not maintain the soil water content high enough to supply the required amount of water for normal growth of seedlings. Leaf rolling, usually observed by farmers, is, perhaps, the result of water stress as disappears right after irrigation. Maize hybrids with higher growth rates, particularly under water stress condition, may have advantages, because this could help them to stand vigorously at earlier stages of growth. The aims of this study were: i) to compare root and leaf growth rates of maize cultivars at the seedling growth stage under normal and water stress condition imposed by PEG solutions and ii) to evaluate the yield performance of cultivars under the same stress conditions and iii) to investigate the relationship between seedling growth rate and grain yield.

Materials and Methods

a) Seedling experiment: In order to compare seedling root and leaf growth rates under water stress and normal conditions, an experiment was carried out under controlled condition in the Faculty of Agriculture, Kerman University, using seven maize cultivars of SC-404, SC-704, BC-666, TC-647, DC-370, Jeta and Kordona (Table 9). After germination, seedlings were grown four days in a hydroponic medium to make possible non-destructive sampling, especially root measurements. PVC tubes (160mm diameter) were divided longitudinally, closed at both ends, and tested for any leakage of water before starting the experiment. To provide oxygen for root respiration, an air pump was connected to a net of pips with porous ceramic heads fixed at the bottom of the half tubes. Uniform seeds in terms of size, weight, and shape were selected for sowing. Selected seeds, however, were weighted up to four decimal digits before sowing. Rootlets of pre-germinated seeds were carefully passed through the holes made on Styrofoam plates (with 10mm thickness) and were fixed so that the growing coleoptile was directed upward and seminal roots downward. The plates were then floated on the surface of full strength Hoagland nutrient solutions with a pH adjusted to 6.5 (Table 10). Seeds were pretreated with a fungicide (Vitawax) before sowing. The half tubes were fixed on greenhouse benches and

Table1. Mean squares for length of root and the first (5 to 10) and second leaves (11 to 14) in maize seedlings grown under normal and water stress conditions

			Days after sowing										Dry matter *10^4	RWC *10^4
S.V.	df		5	6	7	8	9	10	11	12	13	14		
Stress	1	Root	7.61ns	29.92ns	91.88**	179.34**	238.43**	322.32**	405**	491**	631**	740.9**	30**	
		Leaf	0.81ns	8.07ns	59.01**	119.58**	175.81**	190.19**	455**	628**	886**	1122.5**	250**	640*
Error 1	3	Root	6.87	6.52	7.85	12.1	10.37	10.92	9.6	8.43	9.83	10.13	0. 8	
		Leaf	2.77	1.26	5.46	7.7	7.78	4.93	12.16	13.03	14.07	14.09	10	30
Cultivar	6	Root	4.98**	7.40**	9.11**	12.46**	15.26**	18.93**	21.8**	22.2**	25.4**	28.1**	3**	
		Leaf	2.29**	2.48**	5.81**	7.14**	7.70**	7.57**	21.3**	23.3**	24.9**	30.48**	35**	7ns
Interaction	6	Root	1.86*	2.27	2.43ns	3.44ns	3.82ns	6.11*	6.8*	7.26*	8.05*	8.91*	0. 7ns	
		Leaf	0.55ns	1.21ns	2.08**	2.32ns	3.15ns	4.41**	1.60 ns	1.54ns	1.52ns	2.55ns	4ns	3ns
Error 2	36	Root	0.75	1.01	1.41	1.85	2.16	2.34	2.46	2.79	3.26	3.73	0. 5	
		Leaf	0.55	0.74	0.83	1.29	1.64	1.79	3	2.89	3.23	4.4	3	4

*,**: Significant at 5 and 1% probability level, respectively; ns: Non-significant; RWC: Relative Water Content

filled with sufficient amount of solution. There were no symptoms of nutrient deficiency in plants during growth. The experimental design was split-plot based on randomized complete blocks with four replications. Water stress and normal conditions were arranged in whole half tubes as main plots and nine plants of each cultivar on Styrofoam plates as sub-plots. Water stress was imposed by application of PEG solution to the related tubes (158g PEG per litter). Seeds were planted in three rows per plate. The rows were spaced three cm apart with three seeds per row. Four days after seedling emergence, PEG-6000 stock solution was applied to the main plots until the solution water potential was reached to -0.4MPa (Michel and Kaufmann 1973). The calculated amount of PEG-6000 solution was applied gradually at one hour intervals so that the solution water potential was decreased by -0.1MPa (Lu and Neumman 1998). Air temperature ranged from 25 to 33˚C during the day and 18 to 23˚C during the night time. Humidity ranged from 40 to 55%. Light intensity was kept constant at 1400molm^{-1}s^{-1} during the day time. Root and leaf lengths were measured every day during a period of 14 days after sowing using a transparent ruler. Care was taken to avoid any damage to the seedlings during measurements. Seedlings were returned back to their place after each measurement. All measurements in a day were made within one hour.

Data were subjected to analysis of covariance taking the initial seed weight as covariate. Since there was no significant effect of seed weight on seedling characteristics, analysis of variance was performed, without considering covariate, and means were compared using Duncan's multiple range tests. Linear regression analysis was performed on root and leaf length data as dependent and time as independent variables. Slopes of the regressed lines were considered as the root and leaf growth rates. The regression coefficients were then compared using t- test (Steel and Torrie 1980).

In each plot three leaf samples were taken from middle parts of the second leaf. Samples were weighted immediately (W_1) and incubated under darkness over a wet sponge for four hours. Leaf saturated weight was then measured after removing the excess water from leaf samples by a tissue paper (W_2). Samples were oven dried at 80˚C for 24 hrs and again were weighted (W_3). The relative water content (RWC) of a sample was then computed as follows:

$$RWC = \frac{W_1 - W_3}{W_2 - W_3} \times 100$$

Finally, seedlings were removed, divided into root and shoot parts, and oven dried. Root and shoot dry weights were then determined.

b) Field experiment

The same cultivars were grown in the experimental field of Shahid Bahonar University of Kerman on May 2008 under normal (irrigation at seven-day intervals) and water stress (irrigation at 15-day intervals) conditions. Again, the experiment was arranged in a split plot design based on three randomized complete blocks, with irrigation intervals and cultivars in main plots and sub-plots, respectively. In each plot, there were four rows, 70cm apart and 20cm space between the plants. Plots were supplied with sufficient amount of N-P-K fertilizers and were hand-weeded during the growth period. Plants in one square meter of

the central rows in each plot were harvested at physiological maturity and their grain yield was recorded. Data were subjected to analysis of variance. Cultivar means were compared using Duncan's multiple range tests.

Results

a) Root growth: The effect of water stress was highly significant on root length of seedlings, two days after application of PEG solutions (Table1). Cultivar effect on root and leaf lengths during the growth period was also highly significant. Cultivar by stress interaction was only significant on root length after ten days (Table1). At the end of the experiment, Jeta showed the highest root length (15.62cm) and TC-647 the lowest root length (10.44cm) among cultivars (Table 2).

b) Leaf growth: Lengths of the first and second true leaves were significantly affected by water stress after two days (Table1). Growth of the first and second leaves, 5 to 10 and 11 to 14 days after sowing, respectively, were significantly affected by water stress. Leaves were, generally, significantly affected by cultivar during the growth period (Table1). At the end of the experiment, Jeta and SC-404 showed the highest growth of the first leaf with 9.89 and 9.82 cm, respectively, while TC-647 with 7.22 cm showed the lowest growth (Table 3). In the case of second leaf, the highest and the lowest growth were recorded for SC-404 with 13.36 cm and TC-647 with 7.31 cm, respectively (Table 3).

c) Seedling dry matter: Water stress and cultivar effects on seedling dry weight were significant. Generally seedling dry matter was lower under stress as compared with non-stress condition (Table1). The highest and the lowest root dry matter were observed in SC-404 and TC-647 with 0.033 and 0.014g, respectively. In the case of leaf dry matter, the highest and the lowest values belonged to SC-404 (TC-647 with 0.095 and 0.034g, respectively) (Table 7).

d) Leaf relative water content: Water stress significantly decreased leaf RWC and it was 6.9% lower under water stress compared with normal condition. However, cultivar effect on RWC was not significant (Table1). The highest and the lowest values of RWC were found in Jeta (93.8%) and SC-404 (91.3%), respectively (Table 7).

e) Seedling growth rate: Under normal condition, root growth rate was significantly different among cultivars. However, the differences disappeared under water stress condition (Table 5). Generally, Jeta with 1.49 and DC-370 with 0.65cmday^{-1} showed the highest growth rates under normal and water stress conditions, respectively. On the other hand, TC-647 showed the lowest root growth rate under both normal and water stress conditions with 0.84 and 0.36cmday^{-1}, respectively (Table 4) (Figure 1).

The highest growth rate of the first true leaf was also found in Jeta (2.01cmday^{-1}), while the lowest rate was obtained in DC-370 (1.18cmday^{-1}). Under water stress condition SC-404 showed the highest and TC-647 showed the lowest growth rates with 0.96 and 0.54cmday^{-1}, respectively (Table 4) (Figure 1). Under normal condition, Jeta with 2.09 and TC-647 with 1.40cmday^{-1} showed the highest and the lowest growth rates of the second leaf, respectively. However, under water stress condition, SC-404 and Kordona with 0.96 and BC-666 with 0.37cmday^{-1} showed the highest and the lowest values, respectively (Table 4) (Figure 1).

Table 2. Mean values for root length during the growth period of maize cultivars

Cultivar	Days after sowing													
	1	2	3	4	5	6	7	8	9	10	11	12	13	14
SC404	4.36^{b}	4.36^{b}	5.23^{b}	5.94^{bc}	6.89^{bc}	7.96^{bc}	8.67^{bc}	9.93^{bc}	10.79^{bc}	11.65^{bc}	12.16^{bc}	12.66^{bc}	13.2^{bc}	13.91^{bc}
SC704	4.05^{b}	4.05^{b}	5.0^{b}	5.71^{b}	6.48^{b}	7.31^{b}	8.19^{b}	8.99^{b}	9.59^{b}	10.20^{b}	10.68^{b}	11.30^{b}	11.68^{b}	12.41^{b}
BC666	4.44^{b}	4.44^{b}	5.35^{b}	6.14^{bc}	6.93^{bc}	7.87^{bc}	8.91^{bc}	10.59^{bc}	10.59^{bc}	11.30^{bc}	11.87^{bc}	12.37^{bc}	12.37^{bc}	13.66^{bc}
TC647	3.39^{a}	3.39^{a}	3.95^{a}	4.54^{a}	5.98^{a}	5.98^{a}	6.58^{a}	7.88^{a}	7.88^{a}	8.51^{a}	8.92^{a}	9.47^{a}	9.84^{a}	10.44^{a}
DC370	4.59^{b}	4.59^{b}	5.50^{b}	6.40^{bc}	7.27^{bc}	8.49^{c}	9.49^{bc}	11.67^{c}	11.67^{c}	12.68^{c}	13.36^{c}	13.98^{c}	14.64^{c}	15.57^{c}
Jeta	4.49^{b}	4.49^{b}	5.67^{b}	6.61^{c}	7.65^{c}	8.88^{c}	9.76^{c}	10.82^{c}	11.792^{c}	12.85^{c}	13.57^{c}	14.14^{c}	14.82^{c}	15.62^{c}
Kordona	4.05^{b}	4.05^{b}	5.1^{b}	6.00^{bc}	7.10^{bc}	8.39^{bc}	9.25^{bc}	10.37^{bc}	11.29^{c}	12.19^{c}	12.92^{c}	13.57^{c}	14.12^{c}	10.44^{a}

Values within each column followed by the same letter are not significantly different at 0.05 probability level according to Duncan's test

Table 3. Length of first and second true leaves of maize cultivars during the growth period

cultivar	Days after sowing									
	First leaf					Second leaf				
	5	6	7	8	9	10	11	12	13	14
SC404	3.86^{bc}	4.73^{bc}	6.07^{bc}	7.83^{c}	8.98^{c}	9.82^{c}	9.01^{c}	10.40^{c}	11.79^{d}	13.36^{d}
SC704	3.0^{ab}	4.30^{ab}	5.34^{b}	6.51^{b}	7.36^{ab}	8.07^{ab}	6.07^{ab}	7.07^{ab}	8.32^{ab}	9.76^{b}
BC666	4.04^{c}	4.73^{bc}	6.22^{bc}	7.52^{bc}	8.18^{bc}	8.90^{bc}	7.59^{bc}	8.73^{bc}	9.65^{bc}	10.68^{bc}
TC647	2.5^{a}	3.57^{a}	4.04^{a}	5.22^{a}	6.18^{a}	7.22^{a}	4.c45^{a}	5.47^{a}	6.50^{a}	7.31^{a}
DC370	4.04^{c}	5.40^{c}	6.71^{c}	7.77^{bc}	8.37^{bc}	8.826^{bc}	9.02^{c}	10.03^{c}	11.02^{cd}	12.28^{cd}
Jeta	3.24abc	4.35^{ab}	5.89^{bc}	7.67^{bc}	8.93^{c}	9.89^{c}	7.41^{bc}	8.69^{bc}	10.38^{cd}	11.73^{bc}
Kordona	3.30^{abc}	4.51^{abc}	5.86^{bc}	7.06^{bc}	8.27^{bc}	9.49$_{bc}$	6.55^{b}	7.86^{b}	9.26^{bc}	10.73^{bc}

Values within each column followed by the same letter are not significantly different at 0.05 probability level according to Duncan's test

Table 4. Root and first and second leaf growth rates of maize cultivars under normal and water stress condition

Cultivar		Water stress	Normal
SC404	Root	Y=0.56X+4.133	Y=1.10X+1.40
	First leaf	Y=0.96X-0.99	Y=1.56X-4.20
	Second leaf	Y=0.96X-4.23	Y=1.92X-9.58
SC-704	Root	Y=0.45X+3.26	Y=.0.98X+2.26
	First leaf	Y=0.61X-0.33	Y=1.44X-3.43
	Second leaf	Y=0.71X-5.00	Y=1.74X-10.18
BC-666	Root	Y=0.52X+3.99	Y=1.06X+1.95
	First leaf	Y=0.71X+0.42	Y=1.40X-3.22
	Second leaf	Y=0.37X+0.81	Y=1.66X-7.99
TC-647	Root	Y=0.36X+2.44	Y=0.84X+2.06
	First leaf	Y=0.54X-0.81	Y=1.32X-3.58
	Second leaf	Y=0.53X-4.52	Y=1.40X-7.66
DC-370	Root	Y=0.65X+3.33	Y=1.25X+2.02
	First leaf	Y=0.75X+0.62	Y=1.183X-1.44
	Second leaf	Y=0.46X+1.75	Y=1.69X-7.45
Jeta	Root	Y=0.50X+3.73	Y=1.41X+1.86
	First leaf	Y=0.76X-0.81	Y=2.01X-6.76
	Second leaf	Y=0.77X-4.68	Y=2.09X-12.0
Kordona	Root	Y =0.51X+3.76	Y=1.37X+0.97
	First leaf	Y=0.87X-1.02	Y=1.6X-4.73
	Second leaf	Y=0.96X-4.23	Y=1.82X-10.63

Leaf growth rates of cultivars were more variable under water stress condition than under normal condition. The differences between cultivars in terms of the second leaf growth rate were also significant. The differences were again greater under stress compared with the normal condition.

f) Yield and yield components: Water stress significantly affected yield and its components. Cultivar effect on grain yield and its components was also significant (Table 6). The highest and the lowest grain yields were found in Jeta (with 6.17t/ha) and TC-647 (with 2.36 t/ha), respectively (Table 7). There was no interaction between watering condition and cultivar for grain yield (Table 6). The highest grain numbers were found in SC-704 (524.64), Jeta (515.27) and the lowest in TC-647 (267.5). Furthermore, the highest values of 1000 grain weight were found in SC-404 (169.03g) and Jeta (160.57g) and the lowest values in TC-647 (with 120.94g) (Table 7).

g) Correlations: Significant correlation coefficients were found between root and leaf growth rates. Generally, correlations under normal condition were stronger than water stress condition. Positive correlation coefficients were found between root growth rate and grain yield, though they were not significant at 5% probability level. However, the correlations were stronger under normal compared to water stress condition. Correlations of the first and the second leaf growth rates with yield were significant only under the normal condition. The correlation in the case of the second leaf was stronger than

Table 5. Calculated t-student values used for the comparison of maize seedlings growth rates under normal and water stress conditions

			SC-404	SC-704	BC-666	TC-647	DC-370	Jeta
SC-704	Root	Normal	2.69*					
		Stress	1.28 ns					
	First leaf	Normal	1.94ns					
		Stress	18.99**					
	Second leaf	Normal	2.44ns					
		Stress	7.03**					
BC-666	Root	Normal	0.93ns	-1.68ns				
		Stress	0.39 ns	-0.86 ns				
	First leaf	Normal	1.88ns	-0.03ns				
		Stress	13.07**	-7.76**				
	Second leaf	Normal	4.08*	1.29ns				
		Stress	24.07**	12.08**				
TC-647	Root	Normal	6.14**	2.75*	4.74**			
		Stress	2.53*	1.32 ns	2.10*			
	First leaf	Normal	2.94*	1.14 ns	1.16 ns			
		Stress	14.71**	2.71*	6.75**			
	Second leaf	Normal	7.52**	4.88**	4.13*			
		Stress	13.17**	5.06**	-6.65**			
DC-370	Root	Normal	-3.45**	-5.34**	-3.95**	-8.56**		
		Stress	-0.97 ns	-2.54*	-1.42 ns	-4.20**		
	First leaf	Normal	5.06**	3.46**	3.41**	2.14ns		
		Stress	9.17**	-7.74**	-2.05ns	-7.40**		
	Second leaf	Normal	3.78*	0.94ns	-0.44ns	-7.52**		
		Stress	19.60**	8.75**	-6.37**	3.01*		
Jeta	Root	Normal	-7.42**	-8.67**	-7.54**	-12.11**	-3.51**	
		Stress	0.72 ns	-0.54 ns	0.318 ns	-1.81 ns	1.83 ns	
	First leaf	Normal	-3.78**	-5.46**	-5.38**	-6.19**	-7.79**	
		Stress	8.21**	-8.10**	-2.70**	-7.75**	-0.63ns	
	Second leaf	Normal	-2.24ns	-4.36*	-5.92**	-8.88**	-5.69**	
		Stress	5.55**	-1.79ns	-16.06**	-7.36**	-12.12**	
Kordona	Root	Normal	-8.22**	-9.17**	-8.02**	-13.45*	-3.11**	1.15ns
		Stress	0.60 ns	-0.66 ns	0.20 ns	-11.93 ns	1.69ns	-0.11 ns
	First leaf	Normal	3.64**	-12.66**	-7.49**	-11.00**	-4.85ns	-4.10**
		Stress	-0.41ns	-2.33*	-2.26ns	-3.29*	-5.33**	3.39**
	Second leaf	Normal	1.27ns	-1.07ns	-2.44ns	-5.84**	-2.12ns	3.28*
		Stress	3.82*	-1.95ns	-11.65**	-6.28**	-9.15**	-0.52ns

*, **: Significant at 5 and 1% probability level, respectively. ns: Non-significant.

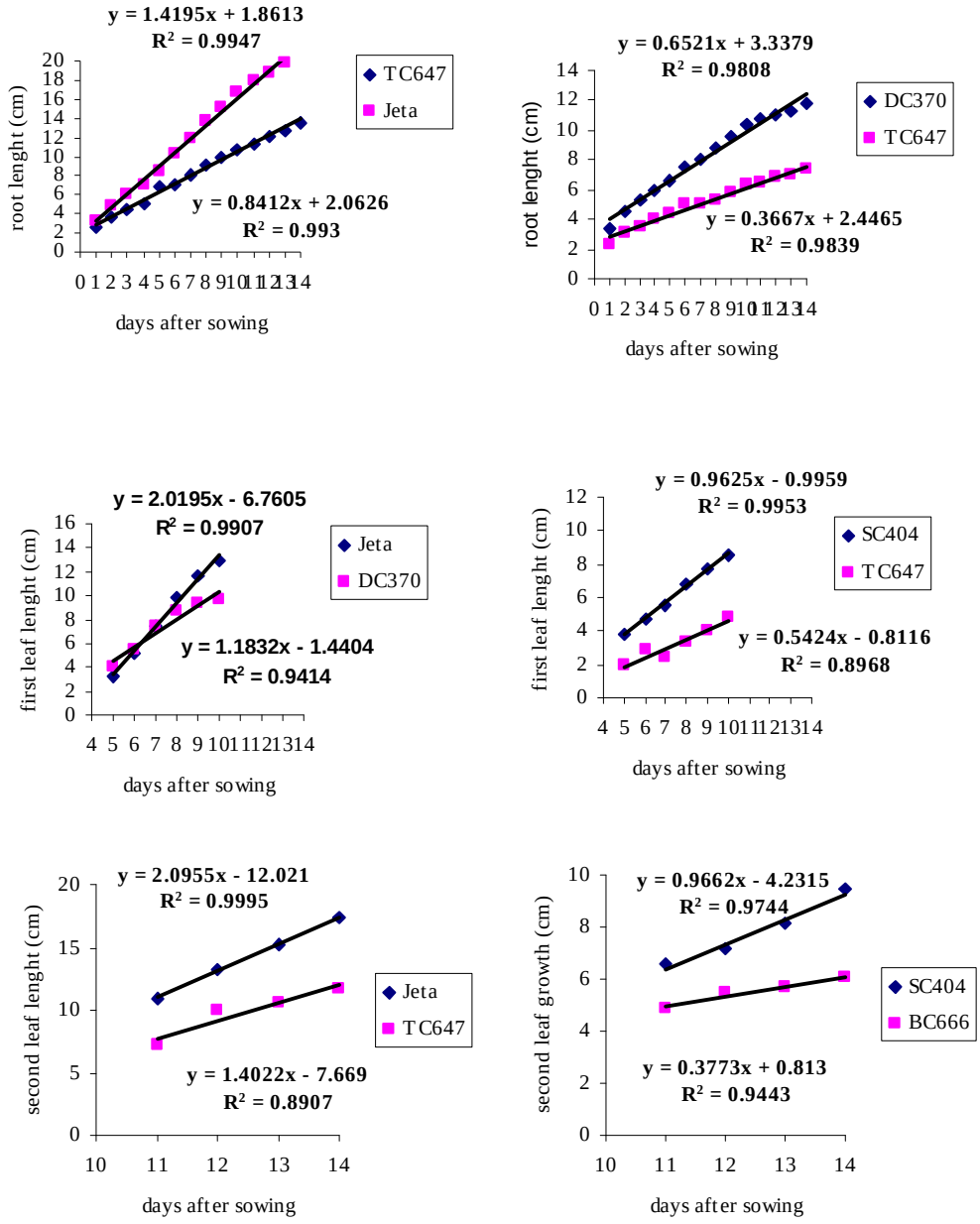

Figure 1. Linear regression lines showing the highest and the lowest growth rates of roots, first leaf and second leaf of maize seedlings grown under normal (left) and water stress (right) conditions

the case of the first leaf. On the other hand, mean seedling growth rate (roots and leaves) was significantly correlated with grain yield under normal condition (Table 8).

Discussion

This study showed the decline of root and leaf growth under water stress conditions. Retardation of growth under low water availability, was also reported by other workers in maize root (Vamerali *et al*. 2003, - Fan and Neumann 2004, Ogawa *et al*. 2006 and Kolarovic *et al*. 2006), leaf (Douglas and Paleg 1981, Sobrado 1986 and Lu and Neumann 1998) and other crop plants (Mian *et al*. 1993, Matsuura *et al*. 2000, Radhouane, 2007 and Aydi *et al*. 2008). It has been shown that water stress decreases the cell division and elongation rates and as a result decreases the growth rate of plants (Choi *et al*. 2000). Lower rates of cell division and cell elongation may be the cause of lower growth of roots and shoots under water stress condition. It was shown that leaf water potential decreases in water stressed plants due to decrease in leaf turgor pressure and as a result leaf elongation rate decreases (Hsiao 1973). Cell wall elasticity also decreases under water stress condition due to hormonal and hydraulic signals (Nilson and Orcut 1996). This in turn may reduce the final size of the cells (Nilson and Orcut 1996).

Reduced values of root and shoot dry matter under water stress condition was reported in soybean (Michalek and Browski 2005), maize (Ogawa *et al.* 2006) and wheat (Kerepesi and Galiba 2000). Water stress also reported to reduce stem dry matter in sensitive cultivars compared to the tolerant genotypes (Turkan, *et al.* 2005). In wheat, barley and bean, shoot dry matter was decreased under water stress condition (Samia 2008). Reduced shoot dry matter was also reported in maize (Chammacho and Caraballo 1994), and bean (Alyari *et al*. 2001). Reduced dry matter may be attributed to lower activities of photosynthetic enzymes under water stress condition (Abdalla and El-Khoshiban, 2007). Higher levels of triglycerides and sterylesters are shown in maize leaves under water stress environments (Douglas and Paleg 1981).

Higher RWC in water stressed plants may be the result of lower rates of water loss due to stomatal closure and more developed root systems (Valentovic *et al*. 2006). Lower levels of RWC have been reported in maize (Valentovic *et al*. 2006) and triticale (Kayden and Yagmur, 2008) when confronted with water stress.

Lower growth rates of leaves compared to the roots may be due to higher levels of water stress imposed on them. Since roots are in direct contact with solution, they may be exposed to lower levels of water stress. This may cause the roots to have higher growth rates. However, since roots have to penetrate into soil in the field, they may not show the same response as they did under hydro culture condition. Smith (1990) concluded that the difference between root and shoot growth under normal and water stress condition could be the result of accumulation of indigenous hormones and osmotic adjustment. In the water stress environment, the regular arrangement of microtubules in the cell walls changes and the level of ABA increases which limits cell growth (Lu *et. al*. 2007). On the other hand, higher concentration of osmolites

such as proline in the roots may cause the root cells to keep their turgor pressures at higher levels which in turn may cause the roots to growth more rapidly. It has been reported that ABA synthesized in root apical meristem in response to water stress causes high concentration of proline in elongation zone of roots (Mohammadkhani and Heidari 2008).

Reduced grain yield under water stress condition in maize has been reported by many researches (Zinselmei *et al*. 1995, Cakir 2004). Decrease in maize grain yield has been attributed to smaller and lower grains per ear which is the result of lower seed set under low tissue water content and lower translocation rate of assimilates. Reduction of the duration of grain filling period is also reported to be another reason for lower grain yield under water stress situation (Zinselmeier *et al*. 1995, Nesmith and Ritchie 1992). Water stress also advances anthesis soon before eggs are ready to accept them which in turn results in lower seed set and yield (Cakir 2004).

Grain yield was poorly correlated with root and leaf growth rates under water stress condition. Correlations were, however, stronger under normal condition and in the case of the second leaf growth rate were significant at 1% level of probability. Positive correlation between growth characteristics and yield has been found in some crop plants such as wheat (Mian *et al*. 1993) and pea (Ogbonnaya *et al*. 2003) under normal condition. It may be concluded that genotypic potential of cultivars in the water stress environment is limited so that their differences reduce under such condition and in turn results in weaker correlation coefficient under water stress condition. This may be considered as a turning point in the plant life cycle which is accompanied by accelerating investment rate in growth substances for later growth and yield. More studies are needed to confirm this at later growth stages.

Crop establishment in silt-loam soils which loose their water content in the sowing depth soon after irrigation is difficult. Water stress may even become more severe due to the retardation of irrigation. Cultivars with higher growth rate at early growth stages may be able to develop their root systems into deeper soil layers in order to avoid the detrimental effects of soil surface drying. More studies are needed to confirm the results in a wider range of soils and water stress conditions.

Table 6. Mean squares for yield and yield components under normal and water stress conditions

S.V.	df	Ear no. per plant	Grain no. per ear	1000 grain weight	Grain yield
Stress	1	0.002^{ns}	297108^{**}	22204^{*}	100.9^{*}
Error 1	2	0.002	2877	1113	2.37
Cultivar	6	0.0009	57673^{**}	1804^{*}	10.18^{**}
Interaction	6	0.001^{ns}	7048^{ns}	486.48^{ns}	1.84^{ns}
Error 2	24	0.001	3074	699.296	1.32

*,**: Significant at 5 and 1% probability level, respectively; ns: Non-significant

Table 7. Root and leaf dry matter, relative water content (RWC) at seedling growth stage, yield and yield components of maize cultivars under study

Cultivar	Root dry matter (g)	Leaf dry matter (g)	RWC (%)	Ear no. per plant	Grain no. per plant	1000 grain weight (g)	Yield (t/ha)
Sc404	0.033d	0.095C	91.3a	1a	413.14b	169.03c	5.13cde
SC704	0.019ab	0.05a	92.8ab	1.01a	524.64c	140.62abc	5.44de
BC666	0.027bcd	0.073b	92.5ab	1a	380.83b	134.23abc	3.89bc
TC647	0.014a	0.034a	91.5ab	1a	267.5a	120.94a	2.36a
DC370	0.025bd	0.075bc	93.5ab	1.03a	299.30a	155.87bc	3.41ab
Jeta	0.031cd	0.085bc	93.8b	1.01a	515.27c	160.57bc	6.17e
Kordona	0.022ab	0.075bc	93.3ab	1.01a	426.91b	132.62ab	4.23bcd

Values within each column followed by the same letter are not significantly different at 0.05 probability level according to Duncan's test

Table 8. Correlation coefficients among root, shoot and seedling growth rates and grain yield under normal and water stress conditions

		Root growth rate		First leaf growth rate		Second leaf growth rate		Seedling growth rate	
		N	S	N	S	N	S	N	S
Root growth rate	N								
	S	0.54^{ns}							
First leaf growth rate	N	0.60^{ns}	-0.12^{ns}						
	S	0.62^{ns}	0.64^{ns}	0.38^{ns}					
Second leaf growth rate	N	0.76^{*}	0.39^{ns}	0.81^{*}	0.68^{ns}				
	S	0.40^{ns}	-0.02^{ns}	0.57^{ns}	0.65^{ns}	0.62^{ns}			
Seedling growth rate	N	0.71^{ns}	0.12^{ns}	0.96^{**}	0.54^{ns}	0.94^{**}	0.62^{ns}		
	S	0.77^{*}	0.44^{ns}	0.80^{*}	0.70^{ns}	0.99^{**}	0.58^{ns}	0.93^{**}	
Grain yield	N	0.56^{ns}	0.18^{ns}	0.79^{*}	0.42^{ns}	0.91^{**}	0.47^{ns}	0.88^{**}	0.90^{**}
	S	0.28^{ns}	0.09^{ns}	0.56^{ns}	0.32^{ns}	0.76^{**}	0.63^{ns}	0.69^{ns}	0.73^{ns}

*, **: Significant at 5 and 1% probability level, respectively. ns: Non-significant

Table 9. Maize cultivars characteristics used in the experiments

Cultivar	Grain yield (t/ha)	Response to abiotic stresses	Growing length (day)	1000 grain weight (g)
SC-704	10-12	Non- tolerant to salt and drought	145-150	450
TC-647	8-9	Non- tolerant to salt and drought	115-125	440
DC-370	8-10	Relatively tolerant to salt and drought	90-110	280-420
SC-404	9-11	Relatively tolerant to salt and drought	100-115	450
BC 666	10-12	Sensitive to drought and salt stress	120-140	400
Jeta	12-15	Relatively tolerant to salt and drought	120-140	300-400
Kordona	10-12	Relatively tolerant to salt and drought	120-140	300-400

Table10. Amount of chemical compounds used for making nutrient solutions according to Hogland

Volume of solution needed for 1 liter of nutrient solution (ml)	Molarity (mmol)	Molecular weight	Chemical compound
2	1000	115.3	NH4H2PO4
6	1000	101.11	Kno3
4	1000	236.15	Ca(NO3)2, 4H2O
1	1000	246.68	MgSO4, 7H2O
1	25	61.83	HBO3
1	50	76	KCl
1	2	287.54	ZnSO4, 7H2O
2.5	2	249.68	CuSO4, 5H20
1	2	169.02	MnSO4, H2O
1	2	162	H2MOO4
1	1000	373	NaEDTA
		278	FeSO4, 7H20

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The Effect of Water Stress on Remobilization of Pre-anthesis Stored Assimilates to Grains in Wheat

A Maghsoudi Moud[1*] and M Islami[2]

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[1]Assistant Professor, Dept. of Agronomy and Plant Breeding, Faculty of Agriculture, S.B. University of Kerman, Kerman, Iran.
[2]Graduate student, Dept. of Agronomy and Plant Breeding, Faculty of Agriculture, S.B. University of Kerman, Kerman, Iran.
* Corresponding author : E-mail: akubaru2@yahoo.com

Abstract

Five bread wheat (*Triticum aestivum* L.) cultivars including Kavir, Pishtaz, Niknejad, Omid and Roshan were evaluated in a split plot experiment based on randomized complete block design with three replications. Water stress and well-watered conditions were assigned to the main plots and varieties to the subplots. Water stress was imposed by withholding irrigation at the booting stage. During a period of 45 days, which spanned before and after anthesis, plants were sampled and changes in dry matter of peduncle and penultimate internodes were assessed. An increase in grain weight was accompanied with a decrease in peduncle and penultimate internodes dry matter, which could be attributed to the remobilization of stored assimilates to grains. Remobilization of stored assimilates was relatively higher under water-stress condition as compared with the well-watered environment. Under water stress condition, tall cultivars remobilized more assimilates than the dwarf genotypes. In the well-watered plots, remobilization percentage from peduncle was more than penultimate internodes in tall varieties as compared with the dwarf varieties. On the other hand, under water stress condition, more assimilates were remobilized from penultimate than the peduncle in dwarf varieties as compared to the tall genotypes. In the dwarf genotypes assimilates remobilization was reduced under water stress conditions while it was increased in the tall cultivars.

Keywords: Assimilate, Drought, Remobilization, Wheat

Introduction

Different physiological processes are involved in grain development and yield of wheat crop under water stress condition (Austin 1989, Richards 1996). Current photosynthesis, remobilization of assimilates stored in the vegetative organs and cell division are among the most important factors (Rawson and Hoestra 1969, Austin *et al*. 1977, Nicolas *et al.* 1985, Austin 1989, Davidson and Chevalier 1992). Sufficient amount of light, water and nutrient elements in the growing media are also

necessary for physiological processes to take place at optimum level (Levitt 1983). To reduce the detrimental effect of drought on yield, many plants including cereals store carbohydrates in the vegetative organs such as stems and leaves before reproductive stage and then remobilize them into the grains (Nicolas *et al*. 1985, Kobata *et al.* 1992, Kobata *et al.* 1994, Ehdaie and Waines 1996, Richards 1996). This feature has been proposed as a selection criterion for selecting more stable genotypes in terms of grain yield particularly under water stress condition (Blum *et al*. 1991, Regan 1993, Ehdaie and Waines 1996, Nicolas 1996). This may cause both biological and grain yield to increase under such a stressful condition.

Wheat is the most important crop plant grown in the semi-arid regions of Iran in which usually experiences water stress during grain filling period. Under such conditions, normal grains could be obtained if pre-anthesis stored carbohydrates in the stem can be remobilized efficiently (Blum *et al*. 1983a, Blum *et al*. 1983b, Ehdaie and Waines 1996). Pre-anthesis assimilates were shown to provide up to 27% of the final grain yield (Bedinger *et al*. 1977) and even more under dry environmental conditions (Pheloung and Siddique 1991). In wheat, genotypic variation has been found for translocation efficiency (Blum *et al.* 1991, Regan *et al.* 1993, Ehdaie and Waines 1996) though environmental conditions significantly affect genotypes ability to remobilize the stored assimilates.

Application of a chemical desiccant at post-anthesis stage can stop current photosynthesis. Therefore, tolerance to drought stress, in terms of translocation-based grain filling can be evaluated by comparison of cultivars without active leaves but with carbohydrate stores in their stems (Regan *et al.* 1993). In wheat, selecting such plants with large grains led to the improvement of grain weight (Regan *et al*. 1993).

There are few reports regarding the role of pre-anthesis stored assimilates on grain filling of wheat under water stress condition (Shakiba *et al*. 1996). This study was, then, performed to investigate the effect of water stress on remobilization of assimilates from main stem internodes to the grains in tall and dwarf varieties of wheat. For this purpose, several bread wheat cultivars, recommended for planting in dryland conditions of the central and southern part of Iran, were used in the experiment.

Materials and Methods

This experiment was conducted in the research field of Faculty of Agriculture, Kerman University with an average annual precipitation of 143.2 mm in 2004- 2005. Seeds of five bread wheat cultivars including three dwarf (Kavir, Pishtaz, Niknejad) and two tall (Omid and Roshan) genotypes which were recommended for planting in warm dryland conditions of central and southern parts of Iran, were sown in 5×2 square meter experimental plots. Two groups of such plots were considered as main plots and randomly assigned to two different water supplying regimes. During the period from seeding to the

beginning of stem elongation, all plots were normally irrigated and thereafter well-watered plots were continued to be irrigated whereas the water stressed plots not irrigated to provide water stress development in plants. Well-watered plots were irrigated at booting, flowering and grain filling stages. Plots were replicated three times, based on a randomized complete block design, each with a different randomly arrangement of sub-plots (five wheat cultivars). During a period of 45 days before and after anthesis, plants was sampled and divided into peduncle and penultimate internodes, ears and grains. These parts were then oven dried at 75°C for 72 hrs and weighted. Samplings were done at ear emergence, 50% flowering, early grain filling (milky stage), mid grain filling (dough stage) and physiological maturity. Each time six plants per plot were sampled. Dry matter changes of these parts-expressed as the sample means-before and after anthesis were then calculated. Positive differences between dry matter at two successive stages were attributed to the storage of assimilates and negative values to remobilization to the grains. Remobilization percentage (*R%*) was calculated using the following equation:

$$R\% = \frac{d_1 - d_2}{d_1} \times 100$$

In which, d_1 is the internode dry weight at the stage with highest value and d_2 is its dry weight at physiological maturity.

In order to determine the stress level imposed to the plants during the growth period right after the last irrigation in water stressed plots, segments of flag leaves were taken from both well-watered and water-stressed plants at flowering, milky and soft dough stages. All samples in a day were taken within one hour at 11:00-12:00 AM local time. Immediately after cutting, fresh leaf weight was measured (*FW*). Then, leaf samples were immediately saturated to full turgidity on the lower shelf of a lab refrigerator (about 8-10^{o}C) in darkness over a wet sponge for 4 hrs and again were weighted (*SW*). Finally, samples were oven dried at 80^{o}C for 24 hrs and weighted (*DW*). Leaf relative water content (*RWC*) was, then, calculated using the following equation:

$$\text{RWC} = \frac{\text{FW} - \text{DW}}{\text{SW} - \text{DW}} \times 100$$

Data were subjected to analysis of variance. Mean values were compared using Duncan's multiple range test.

Results and Discussion

a) Leaf RWC: At all three sampling stages, well-watered plants had higher *RWC* than water-stressed plants. Under both well-watered and water-stressed conditions, *RWC* was higher in the first than the second and third sampling stages Results also showed that at all stages Roshan and Omid (tall varieties) had the highest *RWC* as compared to the dwarf genotypes. Generally, Roshan indicated the highest *RWC* at all sampling stages under well-watered and Omid under water stress condition (Table 1). Several mechanisms has been proposed to be responsible for higher *RWC* under such conditions such as partially opened stomata, smaller leaf area, leaf rolling and osmotic adjustment (Jones & Lezenby 1988,

Saneoka, 1996), enhancement of senescence in extra leaves, leaf orientation on stem and leaf angel to stem (Humphreys 1981, Levitt 1983, Austin 1989).

b) Grain dry weight: During the sampling period, grain dry weight was increased up to maturity under both well-watered and water stressed conditions. However, almost in all cultivars grain dry weight was higher in well-watered as compared to the water-stressed condition. On the other hand, grain weight tended to increase more sharply under water stress condition particularly at later growth stages, as shown in Figure 1. This may be due to higher rates of assimilate remobilization from internodes to the grains under stress condition.

Table 1. Relative water content rates (%) of flag leaf at flowering, milky and soft dough sampling stages

	Well-watered					Water-stressed				
Stage	Roshan	Pishtaz	Kavir	Omid	Niknejad	Roshan	Pishtaz	Kavir	Omid	Niknejad
Flowering	97.4^{a}	95.9^{ab}	94.7^{bc}	93.0^{cd}	92.0^{d}	89.9^{e}	86.6^{f}	82.7^{g}	88.8^{e}	84.2^{g}
Milky	96.6^{a}	96.0^{a}	92.0^{b}	93.1^{b}	90.3^{b}	83.1^{d}	71.3^{f}	68.6^{c}	78.3^{e}	71.0^{f}
Soft dough	92.3^{a}	91.0^{ab}	89.0^{b}	89.3^{ab}	88.5^{b}	78.6^{c}	66.6^{e}	63.6^{f}	75^{d}	67.3^{e}

Differences between mean values within each sampling stage which are followed by the same letter are not significantly different at 5% level of probability.

c) Peduncle dry weight: Significant differences were found for peduncle dry weight under both well-watered and water-stressed conditions (Table 2). In all varieties, peduncle dry weight increased up to a maximum value and then decreased up to physiological maturity (Figures 1 & 2). In all varieties the increase in peduncle dry weight continued until the middle of the grain filling stage under well-watered condition, except Omid that had the highest dry weight in the early grain filling; thereafter dry weight decreased until physiological maturity (Figure 2). In both environments, dry weight of all varieties was then started to decrease until plants reached physiological maturity. The rate of decrease was more in water-stressed as compared with the well-watered condition. This may be the result of the adverse effect of water stress on photosynthesis, less amount of stored assimilates in stem or beginning of remobilization from peduncle to grains. Under water stress condition, Omid and Pishtaz at the flowering, Roshan and Kavir at the early grain filling and Niknejad at the middle of grain filling stages had the highest dry weight (Figure 1). Under well-watered condition due to higher amounts of stored assimilates, plants continued to store assimilates in peduncle for a longer

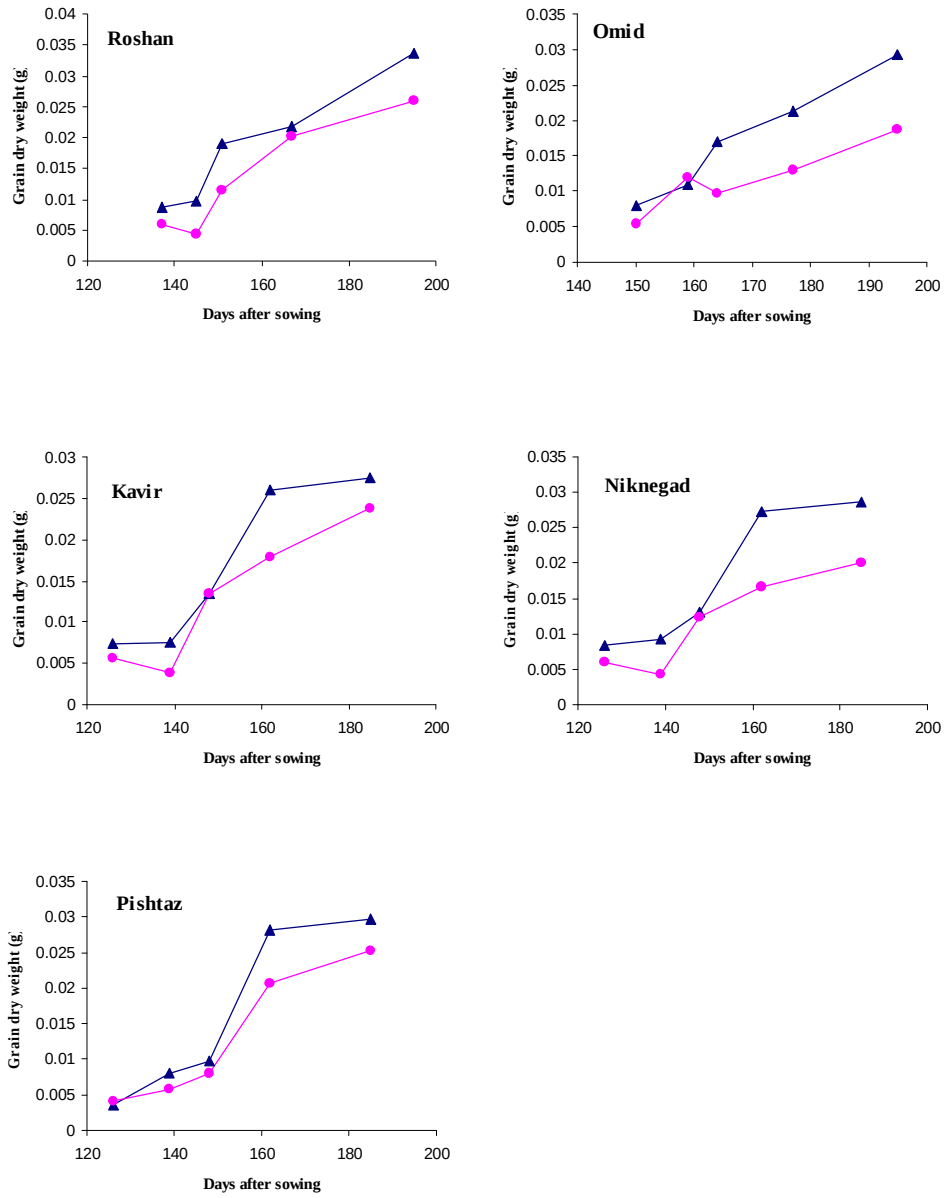


Figure 1. Time course of wheat cultivars grain dry weight change at different growth stages under well watered (triangles) and water-stressed (circles) conditions

period of time as compared with the water stress condition. Decrease in dry matter was started earlier in water-stressed plants due to more sink demand for assimilates and carbohydrates (Shakiba *et al*. 1996).

d) Penultimate internode dry weight: Effects of water condition, variety, and their interaction were significant on remobilization percentage of dry matter in the penultimate internode (Table 2). All varieties reached the highest value of penultimate dry weight during grain filling stage (Figures 3 & 4). In the well-watered condition, penultimate internode dry weight of all varieties

Table 2. Effects of water stress condition and variety on peduncle and penultimate internode dry weight during pre-and post-anthesis growth stages of wheat

SV	df	Ear emergence		Flowering		50% flowering		Mid grain filling		Maturity	
		Peduncle	penultimate	Peduncle	penultimate	Peduncle	penultimate	Peduncle	penultimate	Peduncle	penultimate
Water Stress	1	ns	*	**	*	**	ns	**	**	**	**
Variety	4	ns	**	ns	**	**	*	*	*	*	**
Interaction	4	ns	ns	ns	ns	**	ns	ns	*	**	**

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

was more than the water-stressed condition. Under water stress treatment, Roshan at the flowering and other varieties at the early grain filling stages had the highest penultimate dry weight and thereafter, started to decrease until physiological maturity (Figure 4). Under well-watered condition, Omid at the flowering and other varieties at the mid-grain filling stages had the highest penultimate internode dry weight and thereafter decreased in all varieties until physiological maturity (Figure 3). Under well-watered condition, plants were expected to continue to store assimilates in the penultimate internode for a longer period of time than water-stressed condition, which may be due to higher levels of relative water content. In the water stressed plants, the penultimate dry weight decreased which could be due to lower rates of hotosynthesis and higher rate of respiration and sinks demand for assimilates (Shakiba *et al.* 1996).

e) Peduncle remobilization percentage *(R%)*: Results showed that after reaching to its maximum dry matter, peduncle dry weight started to decline from grain filling to physiological maturity indicating that assimilates move to the grains. Effects of water stress, variety and water stress × variety interaction on the percent of remobilized assimilates were significant (Table 3). Roshan and Pishtaz, with 36.9% and 25.4%, showed the highest and lowest remobilization, respectively. The difference between water-stressed and well-

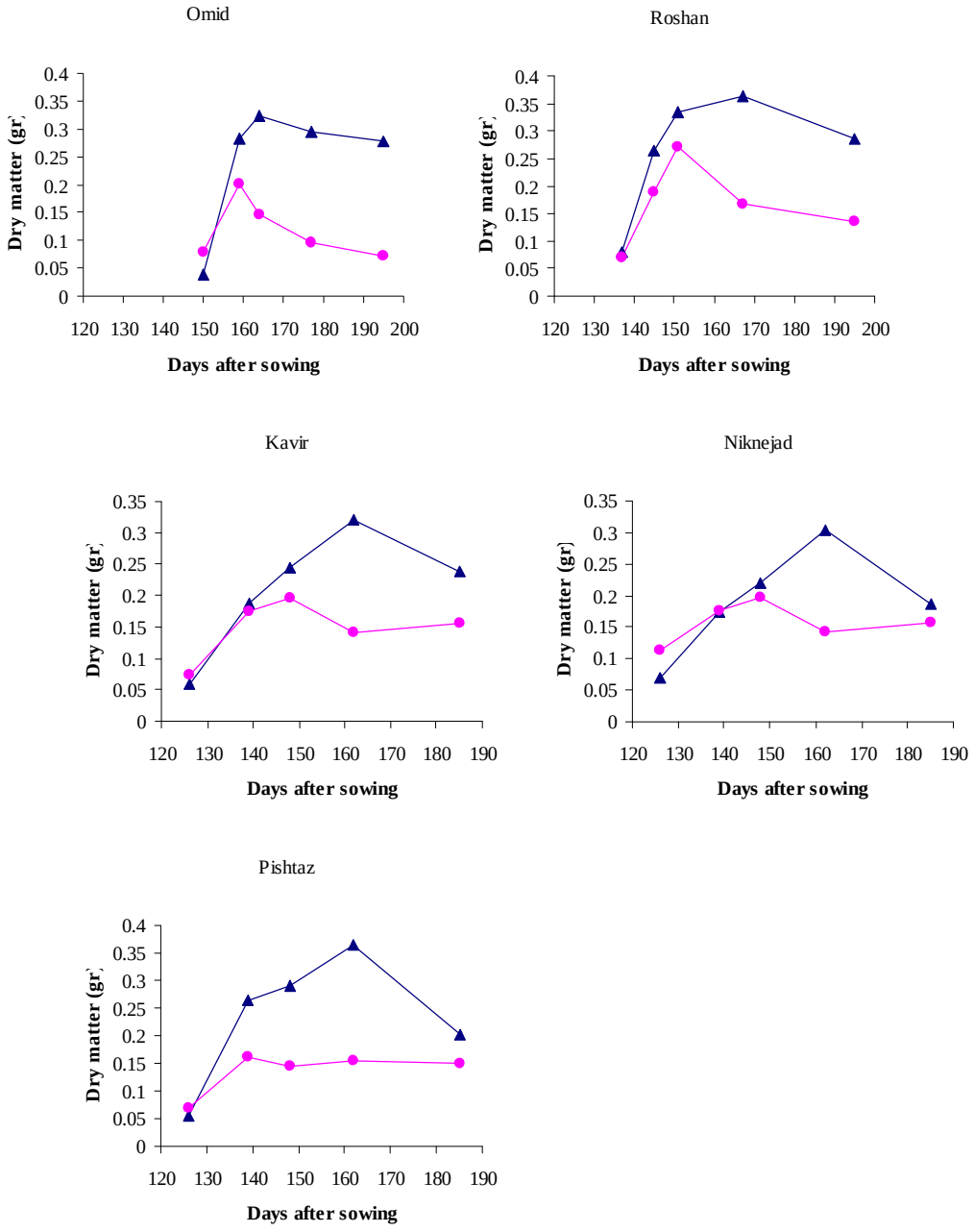


Figure 2. Time course of wheat cultivars peduncle dry weight change at different growth stages under well-watered (triangles) and warter stress (circles) conditions

watered conditions for the percentage of assimilates remobilization was also significant (data not shown). Mean values for water-stressed and well-watered conditions were 28.4% and 33.3٪, respectively (Figure 4). It has been reported that contribution of the pre-anthesis assimilates of stem to grain was greater under drought (46.6%) than well-watered (29.5%) field conditions (Pheloung and Siddique 1991). In the well-watered plants, assimilate remobilization ranged from 13.8٪ in Omid to 44.4٪ in Niknejad (Figure 4). On the other hand, under water stress condition assimilate remobilization was lowest in Niknejad (8.57%) and highest in Roshan (52.68%) and Omid (50.40%) (Figure 4). Under the well-watered condition, tall varieties (Omid and Roshan) had the lowest *R%* from peduncle to the grains while dwarf genotypes, particularly Niknejad, indicated the highest value. In the water stress condition, tall varieties showed the highest *R%* (Figure 4) and, generally, it was two to three times more than the *R%* observed in the plants under well-watered condition. *R%* declined in the dwarf varieties under the water stress condition. Obviously, photo-assimilates have to move in a longer distance in the tall varieties. This in turn may increase the hydraulic resistance of the phloem against the movement of photo-assimilates (Setter 1993). Therefore, increasing the hydraulic resistance may not be considered as the cause of decreased *R*% in the dwarf cultivars unless the increasing resistance is caused by reducing the diameter of the phloem elements (O'Brien *et al*. 1985). On the other hand, slower rate of photo-assimilate remobilization as a result of lower capacity of the dwarf varieties to store photo-assimilates produced before anthesis and lower sink demand for assimilates due to smaller grains and slower rate of endosperm cell division (Ober *et al*. 1991, Setter & Flannigan 2001) may also be considered as the cause of lower R%. Significant water stress × variety interaction indicated that *R%* in different varieties were different in well-watered and water stress conditions. It has been shown that, at first, assimilates mobilize from fully expanded leaves to enhance developing organs and this mobilization takes place separately in each stem and tiller (Rawson & Hoestera 1964). Leaf assimilates are then temporarily stored in the peduncle and penultimate internodes. Assimilates from flag leaf were shown to be used as the main source for remobilization to the grains (Rawson & Hoestera 1964). More partitioning of the flag leaf assimilates has been observed in tall varieties and at the beginning of enhanced kernel growth (Madore and Waines 1996) and a decline of dry weight was observed that is due to the movement of dry matter or respiration (Rawson & Evans 1971). Internode weight at anthesis and post-anthesis was higher in tall varieties as compared with semi dwarf genotypes (Nicolas *et al.* 1985, Madore and Waines 1996). In our experiment, tall varieties also showed higher peduncle biomass and more remobilization from this organ. Other researchers also reported the same results in wheat (Phelonge and Siddique 1991, Madore and Waines 1996, Ehdaei and Waines 1996). Austin *et al.* (1977) showed that remobilization

of stored assimilates to grains was higher in tall varieties in which stem and leaf dry weight decreasd rapidly after anthesis, so higher remobilization in the tall varieties was expected to be due to faster remobilization of assimilates to grains. Among the dwarf varieties, Kavir had the highest *R%* (40.9%) and Omid and Roshan had the lowest value in the well-watered than water stressed treatments (Figure 4).

Remobilization in the tall varieties was two or three times higher in the water stress condition as compared with the well-watered condition. A decline was observed in the dwarf varieties. Among dwarf varieties, the highest *R%* was observed in Kavir and Niknejad under water stress and well-watered conditions, respectively. Rawson and Evans (1971) reported the higher possibility of lower storage and susceptibility to environmental stresses in modern dwarf varieties.

Table 3. Effects of water stress condition and variety on peduncle and penultimate internode remobilization percentage

SV	df	Remobilization percentage	
		peduncle	penultimate
Water stress	1	*	**
Variety	4	*	ns
Interaction	4	**	**

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

f) Penultimate internode remobilization percentage *(R%)*: Data analysis showed that withholding irrigation caused significant decrease in penultimate dry matter in all varieties. Water stress and water stress × variety interaction were significant for the percent of remobilized assimilates (Table 3). Considering penultimate internode, Kavir and Omid, with 47.9% and 39.1%, showed the highest and the lowest remobilization percentage, respectively. Under well-watered and water-stressed conditions mean *R%* were 39.4% and 50.4%, respectively. Under the well-watered condition *R%* from penultimate internode ranged from 53.5% in Niknejad to 16.5% in Omid (Figure 5). In the water-stressed plants, however, it ranged from 61.7% in Omid to 39.3% in Pishtaz (Figure 5). In Omid, Roshan and Kavir, *R%* from penultimate in the water-stressed condition was more than the well-watered condition. However, it was lower in Pishtaz and Niknejad. Omid and

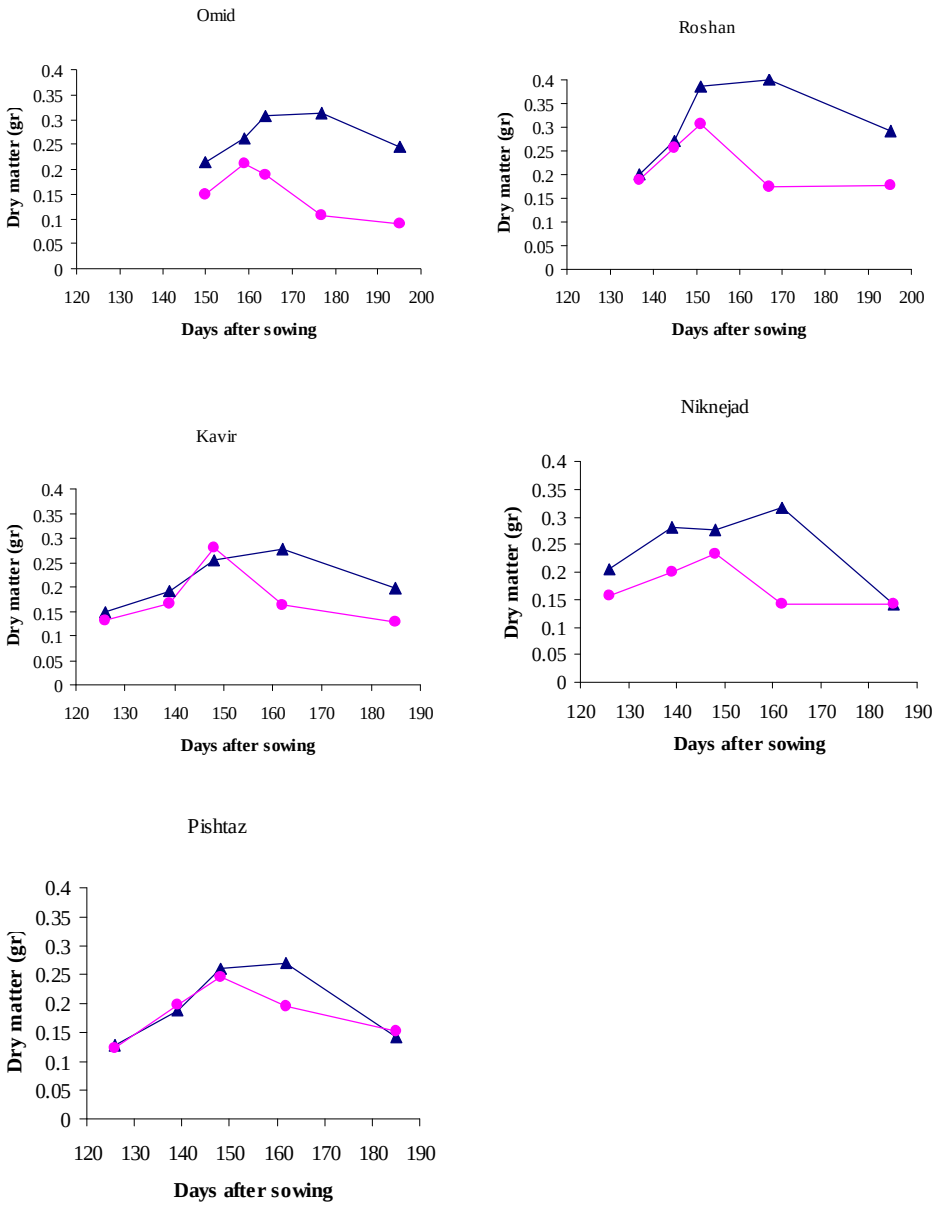
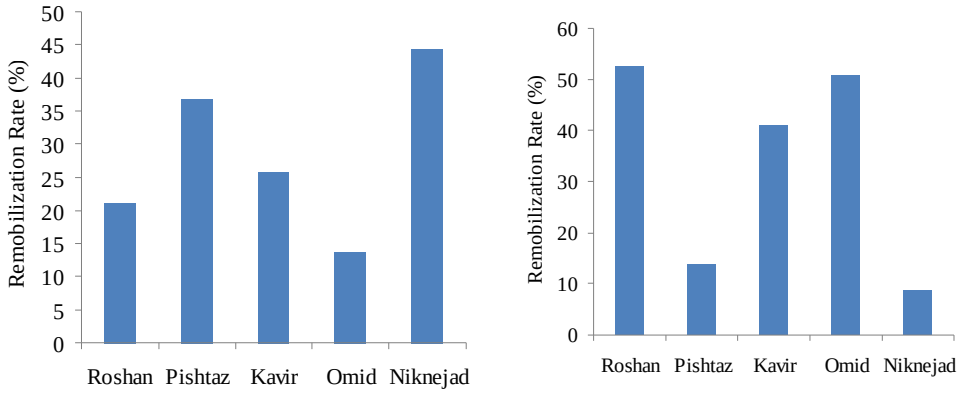
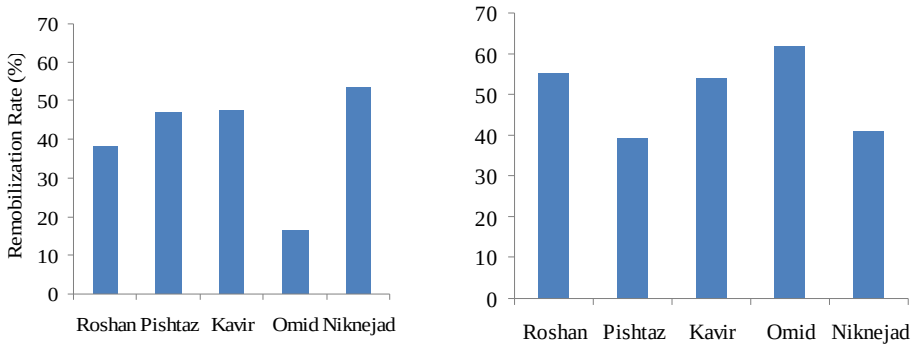


Figure 3. Time course of wheat cultivars penultimate internode dry weight change at different growth stages under well watered (triangles) and water-stressed (circles) conditions



FigureFigureure 4. Stored assimilates remobilization (%) from peduncle to grains in wheat cultivars grown under well-watered (left) and water-stressed (right) conditions

Roshan (tall varieties) had the highest *R%* in the water-stressed environment. Omid and Roshan, with 61.7% and 55.5%, had the highest *R%* under water-stressed condition. *R%* increased to more than three folds in Omid. In tall varieties, *R%* from peduncle and penultimate internodes were higher. Madore and Waines (1996) also reported the same results. In the case of Omid and Roshan, remobilization rate under well-watered condition decreased to less than 50% of the water stress condition. In the water-stressed condition, *R%* decreased in the dwarf varieties but increased in the tall genotypes. In the dwarf varieties, *R%* from penultimate was more than peduncle under both conditions. The same results were observed in the tall varieties although the differences between the two sources were low. Nicolas *et al.* (1985) found that sources were low. Nicolas *et al.* (1985) found that sources were low. Nicolas *et al.* (1985) found that three days before flowering,assimilates start to accumulate in the penultimate internode and reach their maximum values nine days after anthesis. At this period source of assimilates precedes sink demand. Davidson and Chevaliar (1992) concluded that among internodes, penultimate was the strongest internode to reserve assimilates, coming from leaf blades, and later remobilization of them to the ear. Mador and Waines (1996) reported that mean *R%* from the penultimate internode was more than peduncle showing its higher contribution to grain filling. As the photosynthetic machinery of plants is working at its highest capacity, the penultimate internode



FigureFigureure 5. Stored assimilates remobilization (%) from penultimate internode to grains in wheat cultivars grown under well-watered (left) and water-stressed (right) conditions

dry weight, after full elongation, starts to increase indicating the availability of assimilates needed for both plant structure and reservation in stem tissues. Remobilization of stored dry matter from the second internode to the top of the stem and ear then makes the internodes dry weight to decrease. Respiration is also responsible for stem dry matter decrease particularly because air and plant canopy temperature begin to increase later in the growing season, which coincides with the grain filling stage (Rawson & Evans 1971, Austin 1977, Austin 1989, Phelonge & Siddique 1991, Davidson & Chevaliar 1992). Current leaf photosynthesis can not compensate for such high levels of respiration particularly under stress condition (Phelonge & Siddique 1991). Under such conditions, remarkable amounts of dry matter are expected to come from pre-anthesis storages to fill the grains (Gent 1994). Varieties showed a faster decrease in internode dry weight under water stress condition. Both peduncle and penultimate internodes had a role in remobilization. Higher *R%* of the tall varieties as compared with the dwarf genotypes could be due to higher levels of water status which is reflected in the higher flag leaf *RWC*, higher photosynthetic activity and efficiency of remobilization from both internodes (Rawson & Evans, 1971, Madore and Waines 1996). Although remobilization was observed under both growing conditions but it was more under the water stress than the well-watered condition.

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Chemical Composition, Yield and Yield Components of Two Wheat Cultivars in Response to Salt Stress

H Sadeghi[1*] and Y Emam[2]

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[1]Assistant Professor of College of Agriculture, Shiraz University, Shiraz, Iran
[2] Professor of College of Agriculture, Shiraz University, Shiraz, Iran
* Coresponding author : E-mail sadeghih@shirazu.ac.ir

Abstract

In most southern provinces of Iran, soil salinity is a growing problem, particularly in irrigated agricultural areas, and has been found to reduce wheat yield, dramatically. To investigate the effect of sodium chloride on two wheat (*Triticum aestivum* L.) cultivars, four levels of salinity: 0, 4, 8 and 12 dS/m, were employed as a factorial experiment arranged in a randomized complete block design with four replications in a controlled environment of the greenhouse during 2006-2007. The results indicated that increasing salinity from 0 to 12 dS/m, decreased the emergence percentage significantly. Two cultivars of Kavir and Shiraz responded differently to salinity, so that Kavir showed a significantly higher emergence rate. This cultivar also had greater shoot potassium content. Number of tillers and leaves per plant and, also, plant height were decreased upon increasing salinity level. The shoot sodium content was, also increased by increasing the salinity level in both cultivars. However, sodium content of Kavir in comparison with Shiraz, was lower, probably due to Na^+ exclusion mechanisms in this cultivar. The highest grain number and phytomass was obtained from Kavir at the lowest salinity level. Phytomass and grain yield were, also significantly decreased as the result of salinity. Less adverse effect of salinity on Kavir indicates that this cultivar might be suitable for saline soils, an object which worth more investigation.

Keywords: Potassium, Salinity, Sodium, Wheat, Yield components

Introduction

In most southern provinces of Iran, salinity is a growing problem particularly in irrigated agricultural areas with rising water tables, poor water quality and/or deficient soil drainage. Soil salinity has reduced wheat yield usually when values of electrical conductivity were above 6 dS/m throughout the root zone (Munns *et al*. 2006).

Salt stress is one of the most important abiotic stresses affecting natural productivity and causes significant crop loss worldwide. For plants, the sodium ion (Na^+) is harmful, whereas the potassium ion (K^+) is an essential ion. The cytosol of plant cells normally contains 100–200 mM of K^+ and 1–10 mM of Na^+ (Taiz and Zeiger 2002); this Na^+/K^+ ratio is optimal for many metabolic functions in cells. Physico-chemically, Na^+ and K^+ are similar cations. Therefore, under the typical NaCl-dominated salt environment in nature, accumulation of high Na^+ in the cytosol, and

thus high Na^+/K^+ ratios, disrupts enzymatic functions that are normally activated by K^+ in cells (Bhandal and Malik 1988, Tester and Davenport 2003, Munns *et al.* 2006). Therefore, it is very important for cells to maintain a low concentration of cytosolic Na^+ or to maintain a low Na^+/K^+ ratio in the cytosol under NaCl stress (Maathuis and Amtmann 1999).

In wheat, it has been showed that the two responses occur sequentially, giving rise to a two-phase growth response to salinity (Munns 1993). For example, comparison of two genotypes with contrasting rates of Na^+ uptake and long-term differences in salt tolerance (Schachtman *et al*. 1991), showed that both genotypes had similar growth reduction for the four first weeks in 150 mM NaCl, and it was not until afterwards that a growth difference between the genotypes was clearly observed (Munns *et al*. 1995). However, within two weeks, dead leaves were visible on the more sensitive genotype and the rates of leaf death of old leaves were clearly greater on the sensitive than on the tolerant genotype. Once the number of dead leaves increased above about 20% of the total, plant growth slowed down and many individuals started to die (Munns *et al*. 1995). Improved salt tolerance of crops can lessen the leaching requirement, and so lessen the costs of an irrigation scheme, both in the need to import fresh water and to dispose of saline water (reviewed by Pitman and Läuchli 2002). Salt-tolerant crops have a much lower leaching requirement than salt-sensitive ones. In dry-land agriculture, improved salt tolerance can increase yield on the saline soils.

In most southern provinces of Iran, where the rainfall is low and the salt remains in the subsoil, increased salt tolerance will allow plants to extract more water. Salt tolerance may have its greatest impact on crops growing on soils with natural salinity, when all of the other agronomic constraints have been overcome (e.g. disease resistance and nutrient deficiency); subsoil salinity remains a major limitation to agriculture in all semi-arid regions as most southern provinces of Iran. Even where clearing of land in higher rainfall zones has caused water-tables to rise and salt to move, improved salt tolerance of crops will have a place. The introduction of deep-rooted perennial species is necessary to lower the water-table, however, salt tolerance will be required not only for the 'de-watering' species, but also for the annual crops that follow, as salt will be left in the soil when the water-table is lowered (Francois *et al*. 1994).

Wheat is a moderately salt-tolerant crop (Maas and Hoffman 1977). One of the two new cultivars of wheat, used in the present study, Kavir, is an improved genotype recommended for saline areas in most southern provinces of Iran, However, the salt tolerance mechanisms of these varieties have not been studied in detail. The objective of the present study was to quantify plant growth, yield and yield components of the two wheat cultivars in relation to various concentrations of NaCl. In addition, NaCl effect on the chemical composition of the plant organs was investigated.

Materials and Methods

Site, treatment application and data collection

This experiment was conducted to evaluate the effect of four levels of salinity (0, 4, 8 and 12 dS/m) on two wheat cultivars (Kavir, a relatively salt tolerant genotype and Shiraz, a salt sensitive cultivar). The desired

salinity levels were developed by mixing the required amount of NaCl and $CaCl_2$ (5:1) in soil before filling the pots (0, 2.16, 4.32, 8.64 g/kg soil). The wheat crop was sown on 17 November 2006 and harvested on 29 April 2007.The experiment was carried out in a greenhouse at the College of Agriculture, Shiraz University, Shiraz, Iran (52° 46'E, 29° 50'N, altitude 1810 m asl), on a fine mixed, mesic Typic Calcixerpets soil with air temperature in the range of about 25 to 30 °C and light intensity in the range of about 600–1000 µmol m^{-2} s^{-1}, as a factorial experiment arranged in a randomized complete block design with four replications. Soil properties are shown in Table 1. Pre-germinated seeds were sown in 5 L perforated plastic pots filled with fertilized (50, 25 and 25 N, P and K mg kg^{-1}, respectively) soil and were kept in concrete tanks filled with tap water according to Maas *et al*. (1986). The level of water was maintained at 3 cm below the soil surface for two days. Ten seeds of each cultivar were sown in each pot, thinned to five seedlings at two-leaf stage. The pots were kept flooded thereafter for the rest of the experiment. The emergence percentage and number of leaves per plant were recorded throughout the experiment. Plants were harvested and threshed manually. The data regarding grain number, straw yield, grain weight, spikes per plant, tillers per plant and shoot length were recorded (Wilhelm *et al*. 1989).

Table 1. Soil properties (0-30 cm) before plant sowing

Year	OC (%)	pH	Sand (%)	Silt (%)	Clay (%)	Soil texture	EC (dSm^{-1})	P (mg kg^{-1})	K (mg kg^{-1})	Total N (%)
2005-06	0.83	7	7	66.7	26.3	Silty loam	0.05	16.5	476	0.08

Sodium and potassium measurements

Dried samples were ground to a fine powder and about 0·1 g was transferred to a test tube containing 10 mL of 0·1 N acetic acid, and heated in a water bath at 80 °C for 2 h. The extracted tissue was cooled at room temperature and left overnight, and then filtered using filter paper number 40. Sodium and potassium concentrations were then determined using an atomic absorption spectrometer (Munns and James 2003).

Proline measurements

Fresh flag leaf tissue (0·5 g) was ground in liquid nitrogen and then extracted in 20 ml of hot water for 30 min with moderate shaking. The homogenate was centrifuged at 5000 g for 10 min. The proline concentration was quantified by application of the ninhydrin acid reagent method as described by Bates *et al*. (1973) using L-proline as a standard.

Statistical analysis

Statistical analysis for each variable was performed based on a randomized complete block design model using SAS software (SAS Institute 1985). Means were compared by Duncan's multiple range test at $p \leq 0.05$.

Results and Discussion

Effect of sodium chloride on growth and morphological characteristics

Salinity had significant effect on morphological traits of both cultivars. The results indicated that increasing salinity from 0 to 12 dS/m, decreased emergence

percentage significantly. The two cultivars (Kavir & Shiraz) responded differently to salinity and Kavir showed significantly higher emergence rate. Number of tillers and leaves per plant and, also the plant height were decreased upon increasing salinity level (Table 2), which is in agreement with the finding of Abdullah *et al*. (1978). It was found that Kavir was superior to Shiraz as far as the salinity tolerance characteristics (as shown in Table 2) were concerned. Kingsbury *et al*. (1984) showed that the major difference between two lines of wheat in salinity tolerance was their different response to specific ion effects, at the level of the organ, tissue, cell, and sub-cellular entities. Superior compartmentation of toxic ions by the more salt-tolerant line, presumably in the vacuole, might have enabled it to maintain its cytoplasmic metabolic apparatus in a stable and more nearly normal state than the sensitive line. Therefore, a measure of true cytoplasmic toleration of salt maybe needed. The first phases of the growth response results from the effect of salt outside the plant i.e. the salt in the soil solution (the osmotic stresses) reduces leaf growth as shown in Table 2. Indeed, salts themselves do not build up in the growing tissues at concentrations that inhibit growth, as the rapidly elongating cells can accommodate the salt that arrives in the xylem within their expanding vacuoles. Thus, the salt taken up by the plant does not directly inhibit the growth of new leaves (Munns 1993).

The second phase of the growth response results from the toxic effect of salt inside the plant. The salt taken up by the plant

Table 2. Means of main effects and their interaction for morphological traits

Treatment	Emergence percent	Leaves per plant	Tillers per plant	Plant height (cm)	Spikes per plant
Cultivar					
(V_1) Shiraz	58.58 a	5.91 a	1.33 a	30.46 a	1.16 a
(V_2) Kavir	64.41 a	7.66 a	1.99 a	32.66 a	1.41 a
Salinity (dS/m)					
(S_0) 0	94.00 a	13.83 a	3.00 a	53.17 a	2.50 a
(S_1) 4	93.67 a	10.00 b	2.50 a	44.67 b	1.83 b
(S_2) 8	55.00 b	3.33 c	1.16 b	28.50 c	083 c
(S_3) 12	3.33 c	-[+]	-	-	-
Cultivar *Salinity					
V_1S_0	92.67 a	13.33 a	2.66 ab	48.33 ab	2.33 ab
V_1S_1	94.00 a	8.33 b	2.00 bc	48.33 ab	1.66 bc
V_1S_2	47.67 c	2.00 cd	0.66 d	25.33 d	0.66 de
V_1S_3	0.00 e	0.00 d	0.00 d	0.00 d	0.00 d
V_2S_0	95.33 a	14.33 a	3.33 a	58.00 a	2.66 a
V_2S_1	93.33 a	11.67 a	3.00 a	41.00 bc	2.00 ab
V_2S_2	61.33 b	4.66 c	1.66 c	31.67 cd	1.00 cd
V_2S_3	6.66 d	-	-	-	-

Means at each column for each source, followed by similar letters are not significantly different using Duncan's multiple range tests ($p \leq 0.05$).

[+] No plant growth due to salinity

concentrates in the old leaves. Continued transport of salt into transpiring leaves over a long period of time, eventually results in very high Na^+ and Cl^- concentrations, and the leaves died as it was observed in our experiment (see Table 2 and 4). The cause of the injury is probably due to the salt load exceeding the ability of the cells to compartmentalize salts in the vacuole. Salts then would rapidly build up in the cytoplasm and inhibit enzyme activity (Munns 1993). Alternatively, they might build up in the cell walls and dehydrate the cell (Flowers *et al*. 1991). However, Mühling and Läuchli (2002) found no evidence for this in maize cultivars that differed in salt tolerance

Relationship between salinity and yield components

The results revealed that the highest grain number and phytomass was obtained from Kavir at the lowest salinity level (Table 3). Phytomass and grain yield were, also decreased upon salinity, significantly. Yield reduction was attributed, primarily to the reduced spike weight and individual seed weight rather than spike number (Table 3). This finding confirms the results of Francois *et al*. (1989). The straw yield was more sensitive to salinity than was the grain yield **(**Table 3).

Our results also suggest that estimates of grain yield might bring another complexity to the salinity response, not just because the crops must be grown in controlled environments for long periods of time, but also due to the complexity of the converting shoot biomass into the grain. A low level of salinity may not reduce grain weight even though the leaf area and phytomass is reduced **(**Table 3), the fact that grain yield may not decrease until a given ('threshold') salinity is reached (Maas and Hoffman 1977).

Table 3. Means of main effects and their interaction for yield and yield components of two wheat cultivars

Treatment	No. of grains per plant	Grain weight per plant (g)	Grain yield per plant (g)	Phytomass (g)	Leaf area at anthesis (cm^2)	Straw weight (g)	Spike weight (g)
Cultivars							
(V_1) Shiraz	9.75 a	0.18 a	1.75 b. + -+-	3.31 b	4200 b	1.38 b	2.90 b
(V_2) Kavir	12.66 a	0.17 a	2.15 a	4.02 a	4700 a	1.75 a	3.60 a
Salinity (dS/m)							
(S_0) 0	19.17 a	0.43 a	8.24 a	11.67 a	5700 a	3.09 a	11.20 a
(S_1) 4	15.00 ab	0.25 a	3.75 b	6.21 b	5150 b	2.21 b	5.78 b
(S_2) 8	10.67 b	0.04 b	0.43 c	1.36 c	3100 c	0.96 c	0.98 c
(S_3) 12	-[+]	-	-	-	-	-	-
Cultivars* Salinity							
V_1S_0	14.00 ab	0.54 a	7.56 a	10.75 a	5350 a	2.65 b	10.35 a
V_1S_1	14.33 ab	0.20 bc	2.86 b	5.31 b	5200 ab	2.25 b	4.91 b
V_1S_2	10.67 bc	0.0 1 c	0.11 c	0.79 c	2800 d	0.63 d	0.39 c
V_1S_3	-	-	-	-	-	-	-
V_2S_0	24.33 a	0.33 ab	8.03 a	11.89 a	5750 a	3.53 a	11.49 a
V_2S_1	15.67 ab	0.30 abc	4.70 b	7.18 b	5210 b	2.18 b	6.78 b
V_2S_2	10.67 bc	0.07 bc	0.75 c	2.11 c	3400 c	1.29 c	1.71 c

Means at each column for each source, followed by similar letters are not significantly different using Duncan's multiple range tests ($p \leq 0.05$).

[+] No plant growth due to salinity

Effect of sodium chloride on the chemical composition

Our results showed that Kavir had greater shoot potassium content (Table 4). The shoot sodium concentration was also increased by increasing the salinity level in both cultivars; however, the sodium content of Kavir in comparison with Shiraz, was lower, probably due to Na^+ exclusion mechanisms in this cultivar (Table 4). The increase in Na^+ and Cl^- and decrease in K^+ content of wheat grains suggest that the effect of salinity on the physiological phenomenon is due to changes in the ionic content of the plants (Abdullah *et al*. 1978). Other approaches to improve salt tolerance in wheat are based on the mechanisms for salt tolerance, using physiological traits to select within the germplasm. In wheat, salt tolerance is associated with low rates of transport of Na^+ to shoots, with high selectivity for K^+ over Na^+ (Gorham *et al*. 1987, 1990). Correlations between grain yield and Na^+ exclusion from leaves, along with the associated enhanced K^+/Na^+ discrimination, have been shown in wheat (Chhipa and Lal 1995, Ashraf and O'Leary 1996, Ashraf and Khanum 1997), although the relationship may not hold across all genotypes (Ashraf and McNeilly 1988, El-Hendawy *et al*. 2005), showing that Na^+ exclusion is not the only mechanism of salt tolerance (Colmer *et al*., 2006).

There is a strong correlation between salt exclusion and salt tolerance in many species (reviewed by Läuchli, 1984; Munns and James 2003). Figure 1 shows the negative relationship between leaf Na^+ concentration and salt tolerance of Kavir. In general, Kavir, which was characterized with the lowest Na^+ concentrations, produced greater dry matter than the Shiraz cultivar (Table 4). This low-Na^+ genotype had fewer injured leaves, and a greater proportion of living to dead leaves, as observed during the experiment. The effect on growth was probably due to a better carbon balance in the genotype with less Na^+. Similar relationship between shoot dry matter and leaf Na^+ was found in a population from the cross between high- and low-Na^+ genotypes (Munns and James 2003).

The results showed that there was a significant difference among different salinity levels for proline content of the two cultivars, and Kavir had greater proline content (Table 4). The proline content in both cultivars was also increased by increasing the salinity level (Table 4). Moradi and Ismail (2007) stated that it has been repeatedly inferred, but not yet proven, that there might be a relationship between salt tolerance and the accumulation of proline and other metabolites for osmotic adjustment. However, Colmer *et al*. (1995) suggested that the increase in proline concentration may not be associated with salinity tolerance. Indeed, elevated proline levels may also confer additional regulatory or osmo-protective functions under salt stress, such as its role in the control of the activity of plasma membrane transporters involved in cell osmotic adjustment in barley roots (Cuin and Shabala 2005).

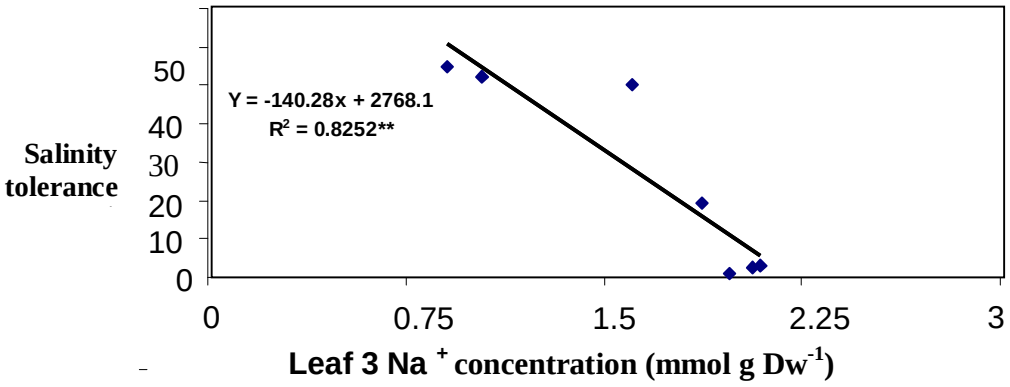

Figure 1. Relationship between salinity tolerance (% growth of the control) and leaf Na^+ concentration in the Kavir cultivar. Na^+ concentrations were measured on the third leaf after 10 d in 150 mM NaCl and shoot biomass after 24 d. Values are expressed as a percentage of shoot biomass in the control condition (R^2=0.8252). All values are based on means (n=5).

Table 4. Mean comparison of main and interaction effects of chemical composition of two wheat cultivars

Treatment	K^+ (mmol per Kg)	Proline (µ g/g)	Na^+ (mmol per Kg)
Cultivars			
(V_1) Shiraz	222.70 b	0.25 b	157.10 b
(V_2) Kavir	435.50 a	0.34 a	13.80 a
Salinity (dS/m)			
(S_0) 0	319.40 c	0.25 d	94.10 d
(S_1) 4	410.70 b	0.27 b	87.30 b
(S_2) 8	586.50 a	0.41 a	160.50 a
(S_3) 12	-	-	-
V_1S_0	287.20 d	0.25 d	141.14 d
V_1S_1	209.00 d	0.26 b	168.80 b
V_1S_2	394.90 c	0.33 a	318.40 a
V_1S_3	-	-	-
V_2S_0	351.70 c	0.29 d	46.80 de
V_2S_1	612.30 b	0.30 ab	5.80 e
V_2S_2	778.10 a	0.37 a	2.50 e

Means at each column for each source, followed by similar letters are not significantly different using Duncan's multiple range tests ($p \leq 0.05$).

+ No plants growth due to salinity

Conclusion

Our results indicated that the two cultivars, Kavir & Shiraz, responded differently to salinity, so that Kavir showed significantly higher emergence rate. This cultivar (Kavir) also had greater shoot potassium content. Number of tillers and leaves per plant and also plant height were decreased in both cultivars upon increasing salinity. The shoot sodium content in both cultivars was also increased by increasing the salinity level; however, the sodium content of Kavir, compared to Shiraz, was lower probably due to Na^+ exclusion mechanisms in this cultivar. The results also revealed that the highest grain number and phytomass was obtained from Kavir at the lowest salinity level. Phytomass and grain yield were, also decreased upon salinity significantly. Overall, it appeared that less adverse effect of salinity on Kavir cultivar may make it more suitable for growth in saline soils. This subject is worthy of further explorations.

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Karyotypic Study in Some Iranian Local Onion Populations

R Paknia[1] and G Karimzadeh[1*]

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[1]Plant Breeding and Biotechnology Department, Faculty of Agriculture, Tarbiat Modares University, Tehran, Iran
[*]Corresponding author Email: Karimzadeh_g@modares.ac.ir

Abstract

A karyotypic study was performed on 12 Iranian local onion (*Allium cepa* L.) populations. A number of mitotic cells at metaphase stage for each population were prepared. Chromosomes of suitable mitotic cells were counted and various parameters, including long arm (L), short arm (S), total length of chromosome (TL), relative length of chromosome (RL), arm ratio (AR), r-value, total chromosome volume (TCV) and centromeric index (CI), were measured. All populations were diploid with 2n=2x=16 and they were differentiated by their karyotype formula and parameters. Chromosome length varied in the populations and the highest quantity of chromatinic material was found in BehdashtShahrud onion. The onion populations under study occupied classes 1A and 2A of Stebbins' karyotype classification, indicating the presence of a primitive symmetrical karyotype in these populations. The mean chromosome length ranged from 8.54 to 11.97 µm. Haploid genome length was in the range of 67.79 to 96.65 µm and mean centromeric index (CI) of complements varied from 41.1 to 43.7%.The chromosome types were detected as mostly metacentrics "m" and some submetacentrics "sm", showing the karyotype formula of 8m (two populations), 7m+1sm (eight populations) and 6m+2sm (two populations). The cluster analysis using chromosomal parameters and based on Ward's minimum variance algorithm assigned the populations into two groups.

Keywords: *Allium cepa*, Centromeric index, Chromosome type, Karyotype, Onion

Introduction

The genus *Allium* includes agronomically useful species. The taxonomic position of onion and related genera has long been a matter of controversy. In early classifications of angiosperms, *Allium* species were placed in the Liliaceae but later, they were more often included in the Amaryllidaceae on the basis of their florescence structure. *Alliumcepa* is one of the oldest cultivated vegetables, recorded for over 4000 years (Fritsch and Friesen 2002, Phillip and Jenderek 2003). It is a biennial plant growing vegetatively one year and produces its seeds in the second year after prior exposing to a period of cold temperatures (Kovatch 2003). The *Allium* genus has great economic significance because it includes several important vegetable crops and ornamental species. All plant parts of onions except the seeds may be consumed by human, and wild species are exploited by local inhabitants. Onion is propagated by seeds, bulbs or sets (small bulbs).

Currently, there are different opinions about the number of species in this genus (Xingjin *et al.* 2000). Estimations indicate about 750 species in the genus *Allium* (Steam 1992) and 650 more synonymous species names exist (Gregory *et al.* 1998). The species of section *cepa* are diploid (2n=2x=16) although the occasional occurrence of individual tetraploid bulbs has been reported contrary to what is found in other *Allium* groups. The chromosomes are either metacentric or submetacentric which differs somewhat in their length; only the satellite chromosome pair is subtelocentric (subacrocentric) and the satellites being attached to the short arms. Most species of the section *cepa*have very small dot like satellite, as in other subgroups of this genus (Fritsch and Friesen 2002).

In spite of the cytological similarities between the species of section *cepa*, there are strong crossing barriers between them preventing interspecific gene flow even where sympatric distribution of two species occurs. Although in modern breeding programs, many classical cultivar groups have been crossed, the boundaries between the different taxa are becoming blurred, making it difficult to place materials within the scheme (Fritsch and Friesen 2002). To expand the genetic variation of onion, onion has been crossed with other *Allium* species, *e.g.A. sativum* (Yamashita*et al.* 2002). Since onion is regarded as an important crop worldwide, for decades there have been well-established onion breeding and seed production programs in the world, developing short intermediate and long-day cultivars. Today, almost 90% of the cultivars used come from local breeding programs (Martinez *et al.* 2000, Paknia *et al.* 2007). Chromosome identification is essential for biotechnological studies including genome analysis, somatic hybridization and ploidy manipulation (Yamamoto and Tominaga 2004). Karyotype features allow individual species to be distinguished. Thus, chromosome variation, although not always large, has accompanied evolutionary divergence of the taxa studied, a general phenomenon observed in both the plant and animal kingdoms (Acosta *et al.* 2005). Knowledge of karyotype relationships is an important prerequisite for effective plant genetic and breeding studies (Martinez-Gomez *et al.* 2003) and also provides valuable information related to the mechanism of genome evolution (Wilkinson 1994). Widely different karyotypes have been found among plants with the same chromosome number, and the relationships between karyotypes are not easily inferred from the morphology of the chromosomes (Brighton 1976). Distinguishing chromosomes based on the length may be charged with significant error due to variation in chromosome contraction within the chromosome complement in either one or different cells (Bajer 1959).

Hybrids obtained from the pollination of *Allium cepa*by other species were easily identified due to the presence of acrocentric chromosomes characteristic for the paternal genotypes (Keller *et al.* 1996). The onion breeding is slow process primarily due to the biennial nature of this outcrossing species (Alan *et al.* 2003). Alteration in chromosome

number and structure raises question about the origin, extent and evolutionary relationships of chromosome variants. Cytological variation also needs to be taken into account when conservation strategies are planned for the restoration of depleted populations, or the establishment of new ones, as the mixing of cytotypes can lead to hybrids (Murray and Young 2001). Cytotaxonomic studies provide taxonomic insight, at different hierarchical levels, not only through the determination of chromosome numbers, but also through the elaboration and comparison of karyotypes, in addition to the analysis of interphase nuclei and of chromatinic condensation standards in prophase. Cytotaxonomic studies can also contribute to discussions on evolutionary trends through chromosome changes.

The aims of this study were i) to quantify the cytological variation among 12 Iranian local onion populations, using karyotype analysis and ii) to establish karyologic relationship among the populations.

Materials and Methods

Seeds of 12 Iranian local onion populations (Table 1) obtained from Seed and Plant Improvement Institute (SPII), Karaj, Iran, were germinated on damp filter paper in petri dishes at room temperature. For the analysis of somatic chromosomes, 1-1.5 cm long fresh root tips were collected from rapidly growing germinating seeds. Different protocols for pretreatment were tested and the best result was obtained from 8 mM of 8-hydroxyquinoline in darkness at room temperature for 2.5 h. Sample roots were subsequently washed three times with distilled water (each 5 min) at room temperature. They were then fixed in Carnoy's fixative (glacial acetic acid: ethanol; 3:1) for overnight at 4°C. After thorough washing with distilled water, accesses transferred to 70% (v/v) aqueous ethanol and stored in a refrigerator until use. Hydrolysis was carried out with 1 M HCl for 8-12 min at 60°C. Thereafter, root tips were stained in 2% (w/v) aceto-carmine for 7-8 h at 20°C according to Ostergren and Haneen (1962). The stained root tips were squashed in a drop of 45% (v/v) acetic acid. The slides were frozen in liquid nitrogen to permit coverslip removal and permanently mounted.

At least five well-spread metaphase plates from different individuals were analyzed per population. The best metaphasic plates were photographed, using a camera attached to the BX50 Olympus microscope and then scanned at 200-resolution and loaded in Photoshop 8.0. Chromosome morphology was described using nomenclatures proposed by Levan *et al.* (1964). For numerical characterization, arm ratio (AR) of each pair, relative length of chromosome (RL), ratio between the longest and the shortest chromosome pair (r-value), form percentage (%F) and total form percentage (%TF) were calculated.. AR, RL, r-value, %F and %TF were calculated as follows:

$$AR = \frac{L}{S};\quad RL = \frac{TL}{\sum TL};\quad r-value = \frac{S}{L};$$

$$\%F = \frac{S}{TL}\times 100;\ \%\mathrm{TF} = \frac{\sum \mathrm{S}}{\sum \mathrm{TL}}\times 100$$

The karyotype symmetry classes of Stebbins (1971) were further quantitatively differentiated into finer karyotype evolutionary gradations, the parameter of chromosome-dispersion (Lavania and Srivastava 1999). The values of Dispersion Index (DI) for a given karyotype were estimated from the following equations:

$$CG = \frac{S\bar{x}}{TL\bar{x}} \times 100$$

Where *CG* is the centromeric gradient, $S\bar{x}$ is the length of median short arm and $TL\bar{x}$ is the total length of median chromosome.

$$CV = \frac{SD}{\bar{X}} \times 100$$

Where *SD* is standard deviation, $\bar{X}$ is mean chromosome length and *CV* is coefficient of variation for chromosome length.

DI = Proportionate measure of *CG* with respect to *CV*

Total volume of chromosome (TVC) was estimated, using the following formula:

$$TVC = \pi r^2 \times TL$$

Where π is the ratio of a circle's circumference (= 3.14), "r" and TL are the average chromosome radius and total chromosome length, respectively.

To estimate karyotype asymmetry, two numerical parameters were used according to Romero-Zarco (1986) method as follows:

$$A1 = \frac{\sum_{1}^{n} \frac{S\bar{x}}{L\bar{x}}}{n}$$

Where $S\bar{x}$ and $L\bar{x}$ are the mean length of the short and long arms of each pair of homologs, respectively, n is the number of homologs and $A1$ is intrachromosomal index.

$$A2 = \frac{S}{\bar{X}}$$

Where S and $\bar{X}$ are standard deviation and mean chromosome length, respectively and $A2$ is interchromosomal index.

The resultant data were first examined for normality test and then analyzed according to a completely randomized design with five replications of metaphase cells. Tukey's test was carried out for population mean comparisons (Coulaud *et al.* 1999, Guillermo Seijo and Fernandez 2003; Mahdavi and Karimzadeh 2010, Karimzadeh *et al.* 2011). Cluster analysis was performed using Ward's minimum variance method and squared Euclidean distance coefficient. Principal component analysis (PCA) was carried out to differentiate the studied populations based on karyotype parameters (Jolliffe 1986).

Results and Discussion

Karyotype formula and parameters for 12 Iranian local onion populations are summarized in Table 2. Figure 1 illustrates the mitotic metaphases and Figure 2 demonstrates their respective idiograms. All populations were diploid with 2n=2x=16 chromosomes. The chromosome number (2n=2x=16) identified for Iranian local populations in the present study was in agreement with the number reported by Xingjin *et al.* (2000) and Fritsch and Friesen (2002). In our best knowledge, this is the first published cytological work n Iranian local population.

Analysis of variance indicated significant differences among populations for most of the karyological parameters (Table 3). Means are shown in Table 4. Chromosome variation among populations of the same species has been observed in many plant groups (Maffei *et al.* 1999). Among the populations studied, the highest total chromosomes length, the longest chromosomes, shortest chromosome, centromeric index (CI) and total chromosome length were observed in BehdashtShahrud population (11.97, 6.93, 5.046, 0.442 and 96.65 µm, respectively) and the lowest values were detected in White Khomein. White Sary onion had higher total volume (34.5 μm^3) and White Ghom possessed the largest dispersion index (6.974) among the populations examined. Karyotype asymmetry was determined for all populations in which TF%, S% and Romero-Zarco (1986) indices showed higher degree of symmetry; the karyotypes of BehdashtShahrud population were the most asymmetric. Total F% analysis showed that symmetrical karyotype had median to nearly median chromosome with a moderate fluctuation in TF% values (from 40.51% in Red Ray to 43.46% in Red Neyshabur). The maximum asymmetry value is 50% where all chromosomes are metacentric (Huziwara 1962). The gradual alterations to shifting of TF% values may be due to the chromosomal abnormalities. The structural alterations in chromosome morphology, as well as the variations of secondary constricted chromosomes may be due to chromosome duplication or translocations between chromosomes with or without secondary constrictions at a very early stage of evolution (Das 1991, Das *et al.* 1998, Mohanty *et al.* 2004).

In our study, the mean chromosome length ranged from 8.54 to 11.97 µm. Haploid genome length varied from 67.79 to 96.65 µm (Table 4). The mean centromeric index in the complement differed from 41.1% to 44.2% (Table 4). DI has been found as a useful parameter to differentiate quantitatively the closely related karyotypes belonging to the same class of symmetry (Lavania and Srivastava 1999). In this study, the higher values of DI indicated higher levels of karyotype specialization. The CV% estimated for the homology of chromosome/chromosome arms, spread over populations, is considered to serve as a general guiding parameter to detect the extent of gradual variation (Table 2). As a whole, karyotypes of the populations examined had predominance of either "m" (centromere at median region) or "sm" (sub-metacentric) chromosome types (Table 2). In other words, the most common haploid formula among Iranian local *Allium cepa* populations was 7m+1sm (eight populations), followed by 6m+2sm, (two populations) and 8m (two populations) (Table 2). Unal *et al.* (1997) with the analysis of *Allium enginii* karyotypes identified 16 somatic chromosomes (2n=2x=16) with total haploid chromosome length of 80.29 µm. The chromosome length was changed from 8.43 to 11.83 µm and the arms ratio was ranged between 1.02 and 1.46. All of the eight haploid chromosomes of *Allium enginii* were metacentrics (Unal *et al.* 1997),

while in our study, the *Allium cepa* populations showed differed chromosome types.

Stebbins (1971) found that increased karyotype asymmetry is associated with increased morphological specialization. In general, the karyotypes of onion populations in our study were mostly symmetrical and fell in Stebbins 1A category of symmetry, except for Ramhormoz, Red Ray and White Sary populations which fell in 2A category (Table 2). Romero-Zarco (1986) indices permitted the detection of slight differences among populations. These indices (A1 and A2) showed small ranges of between-population variations (Table 1). The karyotype asymmetry of the studied populations was very similar, in particular the values of the A1 intrachromo-somal index parameter, the highest value (0.317) was found in the Red Ray population. The diversity was found in the presence, or in the absence, of chromosomes with satellites on either short or long arm. Satellites were detected in one or two chromosome pairs in the karyotype of onion populations tested. Most of populations possessed a satellite on 6L, except in GholyGesehZanjan in which the satellite was not detected (Table 2, Figure 2).

Genetic relationship among populations was assessed by cluster analysis. Grouping based on Ward's minimum variance algorithm and squared Euclidean distance coefficient assigned the populations into two clusters (Figure 3). Cluster I consisted of nine populations (Dorche Isfahan, holyGesehZanjan, Ramhormoz, White Ghom, Red AzarShahr, White Khomein, White Neyshabur, Red Ray, RedNeyshabur) and Cluster II contained three populations (White Kashan, BehdashtShahrud, White Sary). This clustering,based on cytogenetic data, showed close relationship between three populations (White Kashan, White Sary, BehdashtShahrud), supporting morphological characteristics of the genus (Paknia *et al*. 2007). These populations appeared to be very similar, locating closely to each other in a single cluster, while other cluster showed to be distant.

The principal component analysis (PCA) based on karyotypic parameters showed that the first two principal components account for the 84.7% of the total variations and they were projected in a two-dimensional graphic (Figure 4). The first component (46.7%) emphasized the position of the centromer, while the second component (38%) accentuated variation in complement length. The arrangement of populations based on PCA was fully agreed with the result of cluster analysis. Differences in karyotype formula and asymmetry indices found among onion populations suggest that structural changes may have contributed to the diversification of the studied populations.

In conclusion, the main object of the present report was to select onion populations with the most homology in chromosomal variations for the purpose of crossing in plant breeding programs. Crosses of Dorche Isfahan, White Ghom, White Khomein, Red Ray and White Kashan populations by GholyGesehZanjan or Ramhormoz, Red AzarShahr, White Neyshabur, RedNeyshabur and White Saryare, therefore suggested for obtaining the higher genetic variation. This type of analysis can help breeders choosing diverse

parents for heterosis breeding programs aimed at varietal improvement.

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Estimation of Combining Ability and Gene Effects in Forage Maize (*Zea mays* L.) Using Line × Tester Crosses

J Mosa Abadi[1], S Khavari Khorasani[2], B Syah Sar[3], S Movafeg[2] and M Golbashy[4*]

Received: 7 June 2010 Accepted: 17 December 2010
[1] MSc student of Agronomy & Plant Breeding, Zabol University, Zabol, Iran
[2] Seed and Plant Improvement Division, Khorasan Razavi Agriculture Research and Natural Resources Institute, Mashhad, Iran
[3] Department of Agronomy and Plant Breeding, Zabol University, Zabol, Iran
[4] PhD student of Nano-Biotechnology, University of Tehran, Tehran, Iran
[*]Corresponding author Email: mgolbashy@ut.ac.ir

Abstract

Determination of gene effects and combining abilities is a critical stage in maize hybrid breeding. In the present study, 20 S_6 lines as female and three S_6 inbred lines (K18, K19 and K1264/5-1) as tester were crossed and the resulting test cross progenies were evaluated in a randomized complete block design with three replications in 2008. During the growing period, several agronomic characters including forage yield were measured. Effects of lines and testers were significant on all the characters except ASI and days to physiological maturity for lines and ASI for testers. This indicated the importance of additive gene effect in controlling most of the traits under investigation. Inbred lines L5, L14 and L1 were identified as good general combiners for forage yield because they showed significant positive GCA for this trait. L14 was superior compared with L1 and L5 because of significant positive GCA for most of the agronomic characters. These lines, especially L14, have potential additive gene effects to be utilized in the breeding programs. T3 tester showed favorable additive gene effects for forage yield, its components, early maturity and shorter stature. The highest forage yield (79.040 t/ha) with large positive SCA belonged to L5 × T3 combination. L2 × T3 and L15 × T2 crosses were other desirable combinations. Additive genetic variance was substantially higher than dominance genetic variance for all of the traits except days to physiological maturity. This indicated that additive gene effects were more prominent than dominance effects in controlling forage yield and some other agronomic traits in relation to the studied S_6 inbred lines. Therefore, narrow sense heritability estimates closely resembled the broad sense heritability values except for phenological characters. Medium to high narrow sense heritability estimates enable to select for favorable additive gene effects among the studied lines.

Keywords: GCA, Inbred line, Maize, SCA, Testcross, Tester

Introduction

Development of new hybrid varieties in maize requires information about genetic structure of the parental lines and their progenies. This information can be derived from different mating designs such as diallel (Hayman 1954, Jinks 1954, Griffing 1956) and line × tester (Kempthorne 1957) crosses. Venkatesh *et al.* (2001) used line × tester method to evaluate the progeny of 42 test crosses (21 lines and 2 testers) in order to decrease number of lines at the early stage of screening. Line × tester method has been used in various studies (e.g. Hossein and Aziz 1998, Petrovice 1998, Mankir *et al.* 2004, Wali *et al.* 2010, Hefny 2010) to determine general combining ability (GCA) and specific combining ability (SCA) of the lines under study. Petrovice (1998) suggested that combination of lines with significant positive or negative GCA can lead to positive and significant SCA in their test crosses. However, Hossein and Aziz (1998) showed that parents with high GCA for a trait do not give necessarily a high SCA for the same trait. Riboniesa and Efren (2008) classified white inbred lines of maize into two heterotic groups using yield combining ability effects.

Line × tester analysis is also helpful in estimating genetic variance components and types of gene effects (Singh and Chaudhary 1985). Venkatesh *et al.* (2001) using line × tester method found significant differences between lines, testers and line × tester combinations indicating the contribution of both additive and non- additive (dominance) gene actions in controlling grain yield. Hede *et al.* (1999) crossed 23 tropical maize inbred lines with four broad based synthetic testers and evaluated the progenies in six environments. Analysis of variance showed significant GCA and SCA for grain yield. Konak *et al.* (1999) in a 6× 4 line × tester analysis reported that additive gene action was more prominent in controlling plant height and number of kernel rows, however, grain yield, 100 seed weight, ear height, ear length and time to maturity were mainly affected by dominance effects. Petrovice (1998) also obtained the similar results for number of kernel rows, grain yield, 100 seed weight and ear height. Chokan (1999) evaluated the progenies of a line × tester cross in maize at normal and high plant densities. Significant additive genetic variances were observed for kernel number per row and number of kernel rows under high plant density. For other traits, including grain yield, additive and dominance genetic variances were significant in both conditions. Degree of dominance for most traits was in the range of overdominance. Jha and Khera (1992) in a factorial mating system using five testers as female and 16 S_3 lines as male parents in maize under two environments reported significant variation for all the components. For grain yield, SCA and SCA× environment interaction were more important than GCA and its interaction with environment, indicating the role of non-additive gene action in controlling grain yield. Although both additive and dominance type of gene action has been documented in maize, but dominance gene effect was reported more important than the additive type, especially for grain yield.

The objectives of this study were to estimate GCA, SCA and the gene effects for forage yield

and related characters in maize using hybrids produced by the line × tester mating system.

Materials and Methods

The experiment was conducted in Khorasan Razavi Agriculture Research and Natural Resources Center, Iran, in 2008 using 60 maize test crosses. A set of 20 S_6 inbred lines as female were crossed with three inbred lines (T1=K18, T2=K19, T3=K1264/5-1) as male parents or testers in three separate fields in 2007. The resulting test crosses were evaluated in 2008 using a randomized complete block design with three replications. Each test cross progeny was planted in a row with 4.5 m length and between-row and within-row spacing of 75 and 16.5 cm, respectively. During the growing season, plant height, ear height, stem diameter, number of leaves above ear, total number of leaves, number of ears per plant were measured randomly on 10 competitive plants in each plot. In addition, days to silking, days to anthesis, anthesis-silking interval (ASI) and days to physiological maturity were reported. Furthermore, all competitive plants from a plot were cut to the ground level at dough stage and after adjusting for moisture level, the forage yield was recorded on the basis of kilograms per hectare of harvested area. The collected data were analyzed by SAS (Version 9.1) program.

GCA and SCA and standard errors of the estimates were determined by the following formula (Singh and Chaudhary 1985):

$GCA\ (Lines) = Y_{i..}/rt - Y_{...}/rlt$

$GCA\ (Testers) = Y_{.j.}/rl - Y_{...}/rlt$

$SCA = Y_{ij.}/r - Y_{i..}/rt - Y_{.j.}/rl + Y_{...}/rlt$

$SE\ (GCA\ for\ line) = (Me/rt)^{1/2}$

$SE\ (GCA\ for\ tester) = (Me/rl)^{1/2}$

$SE\ (SCA) = (Me/r)^{1/2}$

$SE\ (GCA_i - GCA_{i'})\ line = (2Me/rt)^{1/2}$

$SE\ (GCA_j - GCA_{j'})\ tester = (2Me/rl)^{1/2}$

$SE\ (SCA_{ij} - SCA_{i'j'}) = (2Me/r)^{1/2}$

Where, $Y_{i..}$= Total of the ith line, $Y_{.j.}$= Total of the jth tester, $Y_{...}$= Grand total, r, l and t = number of replications, lines and testers, respectively, SE= Standard error of the estimate and Me= Error mean square

Additive genetic variance (σ^2_A), dominance genetic variance (σ^2_D), narrow sense heritability (h^2_N), broad sense heritability (h^2_B) and average degree of dominance were estimated as below (Singh and Chaudhary 1985):

$\sigma^2_A = (4/1+F)\ \sigma^2_{gca}$

$\sigma^2_D = (2/1+F)^2\ \sigma^2_{sca}$

$h^2_N = \sigma^2_A / \sigma^2_P$

$h^2_B = (\sigma^2_A + \sigma^2_D) / \sigma^2_P$

$Average\ degree\ of\ dominance = (2\sigma^2_D / \sigma^2_A)^{1/2}$

Where, σ^2_{gca}= Estimate of GCA variance, σ^2_{sca}= Estimate of SCA variance, σ^2_P= Estimate of phenotypic variance (plot mean basis) and F= Inbreeding coefficient, which was considered as unity because both lines and testers were inbred. Lines were considered as random and testers as fixed factors. Therefore, additive genetic variance was only calculated from σ^2_{gca} of the lines.

Results and Discussion

Analysis of variance showed significant differences between test crosses for all ofthe traits except ASI (Table 1). Effects of lines and testers were also significant for all the measured traits except for ASI (both lines and testers) and

Table 1. Analysis of variance for agronomic traits of maize test crosses

Sources of variation	Degrees of freedom	Mean squares										
		Ear height	Plant height	Stem diameter	Anthesis-silking interval	Days to anthesis	Days to silking	Forage yield	Number of leaves above ear	Number of leaves	Number of ears/plant	Days to Physiological maturity
Replication	2	616.87**	962.62**	14.81**	0.206^{ns}	2.93^{ns}	3.02^{ns}	851.13**	0.138^{ns}	5.36**	0.053*	28.85
Cross	59	261.40**	558.40**	5.51**	0.747^{ns}	18.09**	18.33**	166.96**	0.127**	1.01**	0.025**	62.56**
Line	19	480.22**	838.58**	5.11**	0.974^{ns}	13.05**	12.85**	191.82**	0.691**	1.72**	0.034*	34.48^{ns}
Tester	2	2169.05**	6791.49**	89.17**	0.339^{ns}	308.53**	328.65**	1603.88**	0.681**	8.55**	0.088*	735.8**
Line ×Tester	38	51.59^{ns}	90.26^{ns}	1.31^{ns}	0.655^{ns}	5.32*	4.74*	78.90^{ns}	0.098*	0.27^{ns}	0.017^{ns}	41.17**
Error	118	56.43	71.99^{+}	1.45^{+}	0.578	3.16^{+}	2.98	70.50^{+}	0.061	0.32	0.014	21.88

* , **: Significant at 0.05 and 0.01 probability levels, respectively ns: Non-significant

+: Error mean squares (with 117 degrees of freedom) excluding non-additivity variance

for days to physiological maturity (lines) which indicates the existence of genetic variability among lines and testers in terms of general combining ability. However, mean squares for testers were substantially larger than lines for most of the traits under study. Line × tester interaction was only significant for number of leaves above ear, days to silking, days to anthesis and days to physiological maturity suggesting that dominance gene action was also involved in governing these traits.

Table 2 shows the estimates of GCA for lines and testers and their SCA. L5, L14 and L1 Inbred lines showed significant positive GCA for forage yield whereas L1 had significant positive GCA for total number of leaves and number of leaves above ear. L15 also showed significant positive GCA for stem diameter, ear height and days to anthesis and significant negative GCA for ASI. L14 was superior over L1 and L5 having significant positive GCA for most of the agronomic characters including total number of leaves, number of leaves above ear, number of ears per plant, stem diameter, plant height and ear height. Thus, these three inbred lines, especially L14, have potential to be utilized for producing synthetic maize varieties and for other breeding purposes.

Among the testers, T3 showed significant positive GCA for total number of leaves, number of leaves above ear, number of ears per plant, forage yield and significant negative GCA for days to anthesis, days to silking, days to physiological maturity, stem diameter, plant height and ear height (Table 2.). Therefore, this tester had favorable additive genes for forage yield and its components and, also, additive genes for early maturity and shorter stature. T1 and T2 had significant positive GCA for days to anthesis, days to silking and days to physiological maturity indicating that these testers had additive genes for late maturity. On the other hand, for plant height and ear height positive and significant GCA was observed for T2, while negative and significant for T1. The existence of considerable diversity among testers for GCA of different characters justified the use of these genotypes for testing the GCA of S_6 inbred lines under study.

Promising crosses are selected based on *per se* performance, standard heterosis and SCA effects. The highest forage yield (79.040 t/ha) belonged to L5 × T3 combination (Table 3). This combination had also high positive SCA for forage yield. The superiority of L5 × T3 hybrid can be attributed to its higher leaf number, days to physiological maturity and more ears per plant (data not shown). Higher forage yield and SCA for forage yield were also observed in L1 × T2, L2 × T3 and L15 × T2. L15 × T2 had also large positive SCA for number of leaves above ear. Furthermore, negative SCA of days to anthesis and days to silking were observed for L5 × T3, L2 × T3 and L15 × T2, while positive SCA of these characters were determined for L1 × T2. However, none of the SCAs for these combinations were significant. Early and medium-maturing forage maize hybrids are desirable in the area of study in order to decrease the risk of early autumn cold stress. Therefore, for traits such as days to anthesis and silking, negative GCA or SCA are preferred. Thus, on the basis of forage yield and maturity the genotypes L5 × T3, L2 × T3 and L15 × T2 may be regarded as promising hybrids and

Table 2. Estimates of general combining ability of maize inbred lines and testers

Line	Number of leaves	Number of leaves above ear	Number of ears/plant	Forage yield	Days to silking	Days to anthesis	ASI	Stem diameter	Plant height	Ear height	Days to Physiological maturity
L1	0.70	0.36	0.04	6.36	-0.19	0.12	-0.32	0.60	0.45	-0.22	0.244
L2	0.13	-0.08	0.08	-5.25	0.03	0.01	0.02	0.55	-9.92	-10.86	-0.978
L3	-0.66	-0.31	-0.07	-1.43	-0.64	-0.77	0.13	0.24	-10.95	-11.09	-2.200
L4	-0.33	-0.61	0.07	0.09	-1.31	-1.54	0.24	0.89	-15.93	-1.17	-1.422
L5	0.26	0.04	0.02	10.04	0.92	1.46	-0.54	1.10	3.38	5.79	2.689
L6	-0.64	-0.32	-0.06	-9.45	-2.86	-3.10	0.24	-1.00	-22.36	-13.44	-1.867
L7	-0.24	0.15	-0.06	-2.55	-0.53	-0.77	0.24	-0.01	4.45	-7.67	1.578
L8	0.69	0.21	0.09	2.34	0.47	0.68	-0.21	0.34	6.53	1.23	-0.200
L9	0.32	0.00	-0.05	-0.33	0.92	1.23	-0.32	-0.82	4.45	0.40	0.244
L10	-0.28	-0.12	0.01	-0.30	-0.31	-0.43	0.13	-0.76	-0.89	1.37	0.244
L11	0.53	0.64	0.08	0.72	-0.75	-0.43	-0.32	0.12	1.95	-0.77	-3.867
L12	0.02	-0.01	-0.08	-5.08	-1.97	-1.88	-0.09	-0.61	-11.13	-5.08	-2.533
L13	-0.18	-0.15	-0.03	-2.53	-0.08	-0.10	0.02	-1.14	4.53	5.57	-1.644
L14	0.53	0.26	0.10	7.44	1.03	1.23	-0.21	1.72	19.06	12.69	3.022
L15	-0.08	0.01	0.06	0.21	1.03	1.12	-0.09	0.07	5.84	7.56	1.133
L16	-0.08	0.05	-0.04	-1.02	2.36	1.46	0.91	0.41	0.75	-3.70	3.467
L17	-0.87	-0.38	-0.06	3.80	1.36	0.90	0.46	-0.35	2.60	2.49	2.022
L18	0.10	-0.01	-0.01	-2.73	0.70	0.79	-0.09	-0.53	6.11	7.21	-0.200
L19	-0.10	0.16	-0.09	-3.41	-0.19	0.12	-0.32	-0.72	0.49	-2.26	-0.533
L20	0.15	0.10	0.05	3.15	0.03	-0.10	0.13	-0.09	10.63	11.95	0.800
SE(GCA)	0.189	0.082	0.039	2.799	0.575	0.593	0.253	0.401	2.828	2.504	1.56
SE(GCAi-GCAi')	0.267	0.116	0.056	3.958	0.814	0.838	0.358	0.568	4.000	3.541	2.21
Tester											
T1	-0.05	-0.10	-0.036	-4.44	0.50	0.51	-0.01	0.35	-8.31	-4.23	1.77
T2	-0.35	-0.01	-0.004	-1.24	2.05	1.97	0.08	1.01	11.92	6.88	2.27
T3	0.40	0.11	0.040	5.68	-2.55	-2.48	-0.07	-1.36	-3.69	-2.66	-4.03
SE(GCA)	0.073	0.032	0.015	1.084	0.223	0.229	0.098	0.155	1.095	0.970	0.60
SE(GCAj-GCAj')	0.103	0.045	0.022	1.533	0.315	0.325	0.139	0.220	1.549	1.371	0.85

ASI: Anthesis-silking interval

Table 3. Forage yield and specific combining ability of several characters for line × tester combinations of maize under study

Cross	Forage yield	Specific combining ability				Cross	Forage yield	Specific combining ability			
		Forage yield	Days to silking	Days to anthesis	No. of leaves above ear			Forage yield	Days to silking	Days to anthesis	No. of leaves above ear
L1×T1	51.952	-5.140	-0.722	-0.506	-0.154	L11×T1	50.445	-1.005	-1.500	-1.950	0.035
L1×T2	66.615	6.328	1.061	1.028	-0.046	L11×T2	55.897	1.252	0.617	1.250	0.076
L1×T3	66.019	-1.187	-0.339	-0.522	0.200	L11×T3	61.317	-0.247	0.883	0.700	-0.111
L2×T1	41.954	-3.531	0.389	0.605	0.046	L12×T1	43.154	-2.495	0.056	-0.172	0.113
L2×T2	43.060	-5.621	1.172	1.138	-0.013	L12×T2	55.813	6.968	-0.494	-0.306	0.020
L2×T3	64.752	9.152	-1.561	-1.745	-0.033	L12×T3	51.290	-4.473	0.439	0.478	-0.133
L3×T1	45.981	-3.319	-0.944	-0.283	0.146	L13×T1	53.730	5.530	0.500	1.050	-0.110
L3×T2	53.821	1.325	1.506	0.917	0.087	L13×T2	55.390	3.994	-0.717	-1.083	0.065
L3×T3	61.408	1.994	-0.561	-0.633	-0.233	L13×T3	48.791	-9.524	0.217	0.033	0.045
L4×T1	47.579	-3.159	-1.611	-1.839	-0.221	L14×T1	62.020	3.846	0.056	-0.283	0.179
L4×T2	56.038	2.104	1.172	1.028	0.220	L14×T2	57.463	-3.906	-0.161	0.583	-0.180
L4×T3	61.908	1.055	0.439	0.811	0.000	L14×T3	68.348	0.060	0.106	-0.300	0.000
L5×T1	58.145	-2.625	-0.167	0.161	0.002	L15×T1	48.145	-2.790	1.722	2.161	-0.210
L5×T2	58.433	-5.532	1.283	1.028	-0.057	L15×T2	62.485	8.354	-1.161	-1.306	0.131
L5×T3	79.040	8.156	-1.117	-1.189	0.056	L15×T3	55.486	-5.564	-0.561	-0.856	0.078
L6×T1	40.723	-20.047	0.278	0.050	-0.343	L16×T1	50.876	1.167	0.056	-0.172	0.057
L6×T2	46.820	-17.145	-0.606	-0.750	0.298	L16×T2	54.138	1.233	1.172	0.361	-0.135
L6×T3	49.603	-21.281	0.328	0.700	0.045	L16×T3	57.424	-2.400	-1.228	-0.189	0.078
L7×T1	45.956	-2.227	-1.722	-1.617	0.103	L17×T1	55.821	1.289	0.722	0.383	0.179
L7×T2	48.855	-2.524	0.394	0.250	-0.075	L17×T2	58.371	0.643	-1.161	-0.750	-0.146
L7×T3	63.049	4.751	1.328	1.367	-0.028	L17×T3	62.715	-1.932	0.439	0.367	-0.033
L8×T1	55.243	2.170	0.278	-0.394	0.090	L18×T1	48.457	0.452	-1.278	-1.506	0.013
L8×T2	53.932	-2.337	-0.272	0.472	-0.135	L18×T2	50.340	-0.861	0.172	0.694	-0.046
L8×T3	63.354	0.167	-0.006	-0.078	0.045	L18×T3	58.529	0.409	1.106	0.811	0.034
L9×T1	48.560	-1.838	0.500	1.050	0.268	L19×T1	54.163	6.843	1.611	1.494	0.046
L9×T2	54.715	1.121	0.950	0.917	0.176	L19×T2	42.264	-8.251	-2.272	-2.306	-0.213
L9×T3	61.230	0.717	-1.450	-1.967	-0.444	L19×T3	58.842	1.408	0.661	0.811	0.167
L10×T1	50.246	-0.189	0.389	0.383	-0.143	L20×T1	61.455	7.578	1.389	1.383	-0.098
L10×T2	54.928	1.298	-0.161	-0.417	0.031	L20×T2	49.136	-7.936	-2.494	-2.750	-0.057
L10×T3	59.440	-1.109	-0.228	0.033	0.111	L20×T3	64.349	0.358	1.106	1.367	0.156
SE (mean)	4.848						4.848				
SE (SCA)		4.848	0.997	1.026	0.143			4.848	0.997	1.026	0.143
SE (SCAij – CAi'j')		6.856	1.409	1.451	0.202			6.856	1.409	1.451	0.202

should be evaluated further for forage yield and other desirable characters.

Estimates of various genetic parameters are presented in Table 4. Negative estimates of genetic components of variance for some characters were set to zero based on expected mean squares. Negative estimates of genetic components can be derived from unsuitable genetic and statistical model, insufficient sampling of original population, sampling error and improper experimental design (Mather and Jinks1982, Roy 2000). Although dominance genetic variance was present for some characters under study, additive genetic variance was much larger than dominance genetic variance for all of the traits except days to physiological maturity indicating that additive gene effects were more important than dominance effects in controlling forage yield. Therefore, the estimates of narrow sense heritability were very close to those of broad sense except for phenological characters. Narrow sense heritability estimates ranged from 0.17 for days to physiological maturity to 0.84 for plant height. Narrow sense heritability of forage yield was medium (0.51). Medium to high narrow sense heritability estimates suggest the possibility of selecting for additive gene effects among the studied lines.

Except for days to physiological maturity, average degree of dominance was in the range of incomplete dominance. Large over-dominance value (2.14) for days to physiological maturity may be due to the result of correlated gene distributions among parents so that partial dominance appears as over-dominance (Hayman 1954).

Ferret *et al.* (1991) reported that for stover and whole plant dry matter yield (DMY) in two dent maize populations additive gene effect was the most important gene action. Moreno-González *et al.* (2000) in a study using European flint (F) and U.S. corn belt dent (D) populations for forage use, reported non-significant average heterosis for stover DMY. In contrast, all populations had high significant heterosis for ear DMY, and six populations had significant heterosis for whole plant DMY. In addition, overall average heterosis was significant for ear and whole plant DMY. Therefore, they stated that most of the contribution to heterosis of the whole plant DMY was provided by the ear fraction rather than the stover fraction. Similarly, based on the results of Boppenmaier *et al.* (1992), stover fraction of DMY had higher effect on heterosis than the grain in maize hybrids. Moreno-González *et al.* (2000) suggested that dominance gene action for the ear DMY fraction may be present in all F × F, F × D and D × D types of crosses, whereas dominance gene action for the stover DMY fraction may be present in the F × D crosses. Therefore, they concluded that breeding strategies for silage hybrids should use populations from the F × D heterotic pattern.

Both additive and non-additive gene effects for plant height were reported by Konak *et al.* (1999), Lee and Shung (1995) and Menkir *et al.* (2004), leaf number by Jha and Khera (1992), days to silking by Neastares *et al.* (1999) and Mendoza *et al.* (2000) and days to anthesis by Lopes *et al.* (1995) and Menkir *et al.* (2004). Esmaili *et al.* (2005) reproted additive gene effects for plant height, days to anthesis, days to physiological maturity and leaf no. Higher non-additive gene effects was obtained for days to silking by several researchers (Konak *et al.*

Table 4. Estimates of additive genetic variance, dominance genetic variance, narrow sense heritability, broad sense heritability and average degree of dominance for the maize characters under study

Estimates of parameters	Ear height	Plant height	Stem diameter	Anthesis-silking interval	Days to anthesis	Days to silking	Forage yield	Number of leaves above ear	Number of leaves	Number of ears/plant	Days to Physiological maturity
σ^2_A	94.18	170.35	0.81	0.098	2.20	2.19	26.96	0.14	0.31	0.004	2.8
σ^2_D	0.00	6.09	0.00	0.026	0.72	0.59	2.8	0.012	0.00	0.001	6.43
h^2_N	0.83	0.84	0.63	0.31	0.66	0.58	0.51	0.81	0.74	0.41	0.17
h^2_B	0.83	0.88	0.63	0.39	0.88	0.74	0.55	0.88	0.74	0.52	0.58
Average degree of dominance	-	0.27	-	0.73	0.65	0.73	0.46	0.41	-	0.71	2. 14

1999, Nestares *et al.* 1999, Esmaili *et al.* 2005), although some researchers indicated the contribution of additive gene effects for this character (Rissi and Hallauer 1991, Jha and Khera 1992). Petrovice (1998) indicated the importance of non-additive gene action in governing plant height, while others (Jha and Khera 1992, Konak *et al.* 1999) showed the prominence of additive gene action for the this trait. Furthermore, Hefny (2010) reported the greater role of additive gene effects in controlling days to anthesis. The different results can be due to different experimental materials and environmental conditiona or the use of different methods for estimating genetic parameters (Konak *et al.* 1999).

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Grain Filling Rate and Duration in Bread Wheat Under Irrigated and Drought Stressed Conditions

AG Sanjari Pireivatlou[1*], RT Aliyev[2] and B Sorkhi Lalehloo[3]

Received: 18 April 2010 Accepted: 1 October 2010
[1]Agricultural and Natural Resources Research Station of Ardabil, Ardabil, Iran
[2]Azerbaijan National Academy of Science, Genetic Resources Institute, Baku, Republic of Azerbaijan
[3]Seed and Plant Improvement Institute (SPII), Karaj, Iran
*Corresponding author Email: a.sangarip@yahoo.com

Abstract

Eleven wheat cultivars were evaluated at 10-day intervals, beginning from anthesis, under irrigated and drought stress conditions during 2006-2007. The effects of irrigation, genotype and date of harvest were significant for most of the studied characters. Water deficit decreased pre-and post-anthesis assimilation rate, grain weight per spike, grain number per spike and 1000 grain weight about 5.7, 24.5, 21.2, 15.7 and 6.4 %, respectively. Mobilization, mobilization efficiency and contribution of pre-anthesis assimilates to kernels were considerably increased under drought stress condition. Grain weight, grain growth rate and contribution of current assimilates to grain filling decreased under drought stress about 7.18, 22.1 and 29.6 %, respectively. However, the effective grain filling period was considerably increased in the stressed plants. Grain filling rate was correlated with the accumulation of dry matter at maturity, grain weight per spike and grain number per spike in the irrigated and drought stressed environments (r=0.87** and 0.53, r=0.87** and r=0.62*, r=0.75* and r=0.63*, respectively). A negative correlation was found between effective grain filling period and grain yield/spike under irrigation (r =-0.65*) and drought stress (r =-0.76**) conditions. Furthermore, positive correlations between grain filling rate and grain yield were obtained in the irrigated and drought stressed environments (r = 0.87** and r = 0.62*, respectively). It seems that accumulation of pre-anthesis assimilates (mainly under drought stress), short effective grain filling period and high grain filling rate are major factors for producing higher grain yield in wheat under both irrigated and drought stress conditions.

Keywords: Drought stress, Effective grain filling period, Grain filling rate, Wheat

Introduction

Genetic variability exists among winter cereal genotypes in response to environmental factors such as drought stress condition, and thus, selection of genotypes for each production area is feasible (Gooding *et al.* 2003). Based on Santiveri *et al.* (2002), grain dry weight at maturity was determined by the rate of dry

weight accumulation and the length of dry weight accumulation period. The assimilate supply problem may be compounded by further reductions in photosynthesis due to water stress and suggests that the reduction in grain number caused by stress at this critical period is proportional to the reduction in leaf-area development and hence, in available assimilate during about 25 days before anthesis (Fischer 1980). Grain growth and development in wheat depend on C from three sources: current assimilates produced by photosynthesis in leaves and stems, mobilization of the stored carbohydrates and N containing compounds within these organs and their subsequent transport to the spike and growing kernels, and assimilates produced by the spike (Bradford and Hasio 1982, Sanjari Pireivatlou and Yazdasepas 2009). Van Herwaarden *et al.* (1998) showed that under dry conditions, the apparent contribution of stored assimilates could be 75-100% of grain yield, compared with 37-39% under rainfall conditions. In fact, a high correlation was found between the use of non-structural carbohydrates stored in stems and grain yield under drought conditions (Gavuzz *et al.* 1997). Wheat crops grown in dry land areas may depend more on the stem reserves for grain filling than crops grown under well-watered conditions (Ehdaie *et al.* 2006). There are two components involved in the extent of contribution of stored reserves to grain yield in wheat; the first is the ability to store assimilates in the stem and the second is the efficiency of the crop to mobilize and translocate the reserved materials to the grains. The second component is a function of sink strength in a genotype, which depends on the number of grains per spike and mean grain weight (Ehdaie and Waines 1996). Stored reserves and their contribution to grain can be estimated by measuring post-anthesis changes in the internode dry matter (Hunt1997, Cruz-Aguado *et al*. 2000), changes in internode water soluble carbohydrate content during grain-filling period (Blum *et al.* 1994), or by difference between shoot dry weight at anthesis and at maturity excluding the grains (Flood *et al*. 1995). Grain filling duration seems to be more affected by environmental factors than grain filling rate (Wiegand and Cuellar 1981, Royo *et al*. 2000).

Genetic variation for the duration of grain filling has been reported for wheat (Bruckner and Frohberg 1987). However, final grain weight has also been suggested to be proportional to grain filling rate (Wiegand and Cuellar 1981), because grain filling duration is largely influenced by temperature under terminal stress conditions. Therefore, selection of genotypes with high grain filling rates appears to be a successful strategy for increasing grain yield (Van Sanford 1985, Bruckner and Frohberg 1987, Knott and Gebeyehou 1987), especially for regions where grain filling duration is restricted by high temperatures (Wiegand and Cuellar 1981, Bruckner and Frohberg 1987). Lack of relationship between grain yield and grain filling duration has been reported for wheat (Nass and Reisser 1975, Van Sanford 1985, Bruckner and Frohberg 1987). Grain growth of field crops is initially slow, enters a linear phase where the growth rate is fast and then slows down toward maturity (Yoshida 1981). Cho *et al.* (1988) divided rice grain filling duration into three phases: lag phase of five days from heading, linear increasing

phase of 5–20 days after heading and late grain filling period thereafter. However, grain filling patterns frequently demonstrate genotypic variations in many cereal crops. Yoshida (1981) suggested that effective grain filling duration, where grain growth is linear, is more important than the duration of ripening from the date of heading to the time when maximum grain weight is attained (Yashida 1981). The rate of dry matter accumulation by kernels was considerably decreased by water deficit in wheat cultivars (Plaut *et al*. 2004). Przulj and Momellovie (2003) reported that in the years with favorable growing conditions during vegetative growth, the main portion of dry matter was stored before anthesis, while in the years with unfavorable conditions during the same period significant amounts of dry matter were accumulated during grain filling as well. The more adapted cultivars continued the accumulation of dry matter and nitrogen during the grain filling period, while in the less adapted ones the main portion accumulated before anthesis (Przulj and Momellovie 2003). Austin *et al.* (1977) reported that a major quantity of pre-anthesis dry matter was used for sinks other than kernel and found that only 73 % of the losses of vegetative weight were used for kernel growth. Respiratory (Rawson and Evans 1971) and dead leaf losses (Bidenger *et al*. 1977) can account for the rest of losses. Modern cultivars seem to be relatively less sink-limited during post-anthesis than their predecessors (Shearman *et al.* 2005), as the semi dwarf wheat seems less sink-limited than the traditional tall wheat.

The major purposes of this study were to understand the accumulation rates of dry matter during pre-anthesis and the effects of grain filling rate and grain filling duration on grain yield in bread wheat genotypes under irrigated and drought stressed conditions.

Material and Methods

Eleven diverse wheat cultivars (Table 1) were evaluated under two water treatments (well watered and drought stress) in 2006-2007. Each experiment was laid out in a randomized complete block design with three replications. Seeds were planted on 15th October 2006, in a clay loam soil at Agricultural and Natural Resources Research Station of Ardabil (38^{o} 15´ N, 48^{o} 20´ E, with an elevation about 1350 m above sea level), Iran. Plants in the normal condition were irrigated five times from planting until they reached physiological maturity (i.e. at planting, tillering, booting, anthesis and milk stages). In the normal condition, plants received 672.4 mm of water (406 mm irrigation + 266.4 mm rain year-1) and those in drought stress condition received only 266.4 mm seasonal rainfall during the 2006-2007 growing year. However, 78.7% of this rainfall was received before booting stage, 19.6 % between booting and heading stage (4.0-4.9 sub-stage of Zadoks), and 1.7% fell between heading and early grain filling period (6.0 to 7.0 sub-stage of Zadoks). Based on the climatic data of Ardabil region, a period of drought was occurred during grain filling stage of wheat in June and July. The absolute maximum and mean temperatures in the grain filling period were 30˚C and 8.0˚C, respectively. Average temperatures were optimum during grain filling period (data not shown).

Each plot consisted of two rows, 2 m in length. Inter-row spacing was 30 cm and kg

Table 1. The pedigree, origin and growth habit of wheat cultivars under study

No	Pedigree	Origin	Growth habit
1	Siosson/M-73-4/3/Bez-2B/Cgn/Veratza	Iran	Spring and semi dwarf
2	MV17	Hungary	Winter and semi dwarf
3	Kal/Bb//Cj"s"/3/Hork"s"/4/Mv17/5/Gascogne/3/P101/Anza//1-66-49	Iran	Winter and semi dwarf
4	Gaspard/6/Bow"s"/Crow"s"/5/Omid/4/Hys//Drc*2/7c/3/2*Rsh	Iran	Winter and semi dwarf
5	Mv-92-2854//Rsh*2/10120	Iran	Winter and semi dwarf
6	ID800994w/Vee//F900K/3/Pony/Opata	9^{th} EYT^{+}	Winter and semi dwarf
7	Es14//Sitta//Agri/Nac	9^{th} EYT^{+}	Spring and semi dwarf
8	Sardari (landrace)	Iran	Facultative and tall
9	Agri/Nac//Atilla	Iran	Facultative and semi dwarf
10	Siossons	France	Winter and semi dwarf
11	Landrace	Azerbaijan	Spring and tall

+International Elite Yield Trial, CIMMYT

interplant spacing was 3 to 5 cm. The land was fallowed in the previous year and 100 kg ha^{-1} urea was utilized at the tillering stage. All cultural practices (i.e., hoeing, weeding, fertilization, etc.) were practiced uniformly, except for irrigation which was based on the respective treatments. Data for grain growth rate (GGR), effective grain filling period (EGFP), mobilization of reserves (MDM), mobilization efficiency (ME), contribution of pre-anthesis assimilates (CPAA) and contribution of current assimilates (CCA) *viz*., plant height, plant weigh, grain weight per spike, grain number per spike, *etc*. were collected by harvesting five spikes from main stems and primary tillers (Gooding *et al*. 2003) at 10-day intervals (five times) from anthesis to physiological maturity.

The following parameters related to dry matter mobilization were estimated in this research:

1. Pre-anthesis and post-anthesis dry matter accumulation (mg per plant).
2. Mobilization of dry matter (mg per plant) = dry matter at anthesis – dry matter (leaf + culm + chaff) at maturity.
3. Mobilization efficiency (%) = Mobilization of dry matter /dry matter at anthesis ×100.
4. Contribution of assimilates to grain (%) = Mobilization of dry matter /grain weight ×100 (Papakosa and Gagianas 1991).

Effective grain filling period= grain dry weight at maturity/ linear grain growth rate (Santiveri *et al*. 2002).

Analysis of variance (ANOVA) was performed for each character in each experiment. The combined ANOVA was also performed for irrigation experiments. Associations between characters were examined by calculating correlation coefficients. Means were compared using the LSD test (Steel *et al*. 1997).

Results and Discussion

The combined ANOVA showed significant effect of irrigation, date of harvest, genotype and genotype × irrigation interaction on most of the studied characters (data are not shown).

Heading time of wheat cultivars was reduced under drought stress condition as compared with the irrigated condition (161.5 and 159.8 days,

respectively). Sardary, a tall landrace, had the earliest heading time, whereas a tall landrace from Azerbaijan republic, had the latest heading time under both well watered and drought stress conditions.

Drought, on the average, reduced the aboveground dry matter by 5.7 and 24.5% at anthesis and maturity, respectively. Furthermore, significant differences were found between genotypes in terms of this character at anthesis under drought stress condition. The landrace from Azerbaijan Republic had the highest aboveground dry matter (3170 mg) and the genotype No. 6 had the lowest amount (1555 mg) under this condition (Table 2). Thereby contribution of pre-anthesis assimilates to grain under drought stress condition was higher in the landrace of Azerbaijan Republic (88.5%) than the genotype No. 6 (59.3%) (Table 2). It seems that the landrace from Azerbaijan reserved high amount of non-structural assimilates in the pre-anthesis growth stage and mobilized them into grains under drought stress condition. Ehdaie and Waines (1996) also reported the same results for Iranian landraces.

There were no significant differences among genotypes for dry matter at maturity and grain weight per spike under drought stress condition. Genotypic variation for this trait was considerably decreased at maturity under drought stress condition (Table 2). Although the genotypic differences were not significant at this condition, however, the highest and lowest aboveground dry matter reduction belonged to genotypes No. 9 and landrace of Azerbaijan Republic (50.2% and 19.1%, respectively). It is assumed that this reduction from anthesis to maturity was due to the utilization of assimilates for kernel development. The average contribution of pre-anthesis dry matter to kernel weight was 30.7% and 55.9% in the irrigated and drought stress conditions, respectively (Table 2). Some genotypes, such as No. 9 and No. 8, lost much more dry matter from anthesis to maturity in the irrigated condition (38.7 and 34.6 %, respectively). Thus, a major quantity of pre-anthesis dry matter was used for sinks other than kernels. Austin *et al.* (1977) found that only 73 % of the loss of vegetative weight were attributed to kernel growth. Respiration (Rawson and Evans 1971) and dead leaves (Bidenger *et al*. 1977) could be responsible for the rest of the loss. The highest reduction of grain weight per spike (48.9%) under drought stress condition was also found in the genotype No. 6. However, grain weight per spike was increased (5.1%) in the genotype No. 3 under drought stress condition.

Drought reduced grain number per spike by 15.7%. Fisher (1980) also reported the reduction in grain number per spike under water stress environment. However, the amount of reduction varied for different genotypes. The highest reduction (46.1%) was found in the genotype No. 6 in contrast with the genotype No. 3, in which grain number per spike was increased under drought stress condition (9.7%). On the other hand, grain number per spike in irrigated and drought stress conditions were similar in relation to the landrace of Azerbaijan Republic.
Mobilization of dry matter and mobilization efficiency were increased under drought stress condition by 60.1% and 74.9%, respectively. However, genotypes responded differently to drought for the amount of pre-anthesis dry matter translocation and translocation efficiency.

Table 2. Translocation of dry matter (TDM), contribution of pre-anthesis assimilates to grain (CPA) and translocation efficiency (TE) of the stem internodes in bread wheat genotypes under well watered and drought stress conditions during 2006-2007.

	Irrigated condition								Drought stress condition							
No.	Vegetative organs (above grand dry matter) (mg)		Yield (mg/ plant)	Grain/ spike	1000 grain weight (g)	TDM (mg)	TE (%)	CPA (%)	Vegetative organs (above grand dry matter) (mg)		Yield/ spike (mg)	Grain/ spike	1000 grain weight (g)	TDM (mg)	TE (%)	CPA (%)
	Anthesis	Maturity							Anthesis	Maturity						
1	2990	5440	2677	69.0	38.7	227	7.6	8.5	2725	4424	2220	65.5	33.9	521	19.1	23.5
2	2800	5734	2672	63.0	42.3	-262	0.0	0.0	2400	3395	1612	47.0	34.2	617	25.7	38.3
3	3050	4004	1730	41.0	41.7	776	25.4	44.8	2750	3600	1818	45.0	39.9	968	35.2	53.2
4	2305	4972	2213	55.0	40.8	-454	0.0	0.0	2549	3534	1762	45.5	37.0	577	22.6	32.7
5	3000	4202	1988	53.0	38.1	786	26.2	39.5	2555	3275	1562	43.5	37.1	841	32.9	53.8
6	2630	4290	2370	58.5	41.1	710	27.0	30.0	1555	2047	1210	31.5	38.4	718	46.2	59.3
7	2743	4022	1940	43.0	44.4	661	24.1	34.1	2975	3903	1712	43.5	39.2	784	26.3	45.8
8	2710	3182	1408	33.0	41.7	936	34.5	66.5	2455	2625	1285	31.0	41.9	1115	54.4	86.8
9	2285	2977	1580	46.0	34.3	888	38.5	56.2	2200	2260	1165	30.5	36.7	1105	50.2	94.8
10	2235	3844	2037	57.5	35.0	428	19.1	21.0	2565	3688	1825	47.0	38.4	702	27.4	38.5
11	3855	5217	2185	47.0	46.6	823	21.3	37.7	3170	3377	1798	47.0	39.1	1591	50.2	88.5
Mean	2691.2	4353.1	2072.7	51.5	40.4	501.7	20.3	30.7	2536.3	3284.4	1633.5	43.4	37.8	803.4	35.5	55.9
LSD 5%	996.9	2156	925.1	22.3	11.29	-	-	-	903.2	2411	1160	24.7	14.25	-	-	-
Changes under drought stress relative to irrigated condition									-5.7	-24.5	-21.2	-15.7	-6.4	60.1	74.9	82.1

Contribution of pre-anthesis dry matter to kernels was increased under drought stress by 82.1%. Mobilization of pre-anthesis assimilates, mobilization efficiency and contribution of pre-anthesis assimilates to kernels under drought stress condition, were higher in the drought tolerant variety of Sardary, Agri/Nac/Atilla and the landrace of Azerbaijan as compared with other varieties. These genotypes had dry matter mobilization of 1115, 1105, 1591 mg, mobilization efficiency of 54.4, 50.2, 50.2% and pre-anthesis assimilate contribution of 86.8, 94.8, 88.5%, respectively (Table 2). Van Herwaarden *et al*. (1998) reported that under dry conditions in the field, the apparent contribution of stored assimilates could be 75-100% of the grain yield as compared with 37-39% under high rainfall conditions. According to the results obtained, genotype No. 9, with 94.8 % of dry matter contribution to grain, produced 1165 mg grain per spike, whereas genotype No. 1, with 23.5 % of assimilate contribution to grain, produced 2220 mg grain per spike (Table 2). This shows that genotypes with larger amount of assimilate contribution to grain tended to have lower grain yields. The coefficient of `determination (R^2) between mobilization of pre-anthesis assimilates and grain yield was only 0.07, which means that the mobilization of dry matter was not useful for high grain yield production. The relationship between the contribution of pre-anthesis assimilates to grain yield and drought tolerance is not well understand, as also reported by Przulj and Momcilovic (2003). Drought, on the average, decreased dry matter accumulation in kernels by 6.4%, however, reduction varied in different genotypes. Genotype No. 2, showed highest reduction (19.1 %) but, in Sardary, Agri/Nac/Atilla and Siosson the rate of dry matter accumulation by kernels increased under drought stress condition (Table 1), as also reported by Plaute *et al*. (2004).

Drought stress decreased grain weight at maturity by 7.18 %. However, the reduction varied in different wheat genotypes. The highest reduction was occurred in genotype No. 2, by 19.1%, whereas in genotypes No. 8, 9 and 10 the grain weight at maturity increased by 0.48, 8.2 and 9.7 %, respectively. Grain growth rate and contribution of current assimilates to developing grains were also decreased under drought stress condition by 22.1 and 29.6%, respectively. The highest reduction was observed in genotype No. 2 (44.9%) and the rate of grain growth in genotype No. 5 was substantially increased (32.3%) under drought stress condition. The mean values observed for effective grain filling period and grain filling rate suggest that there was compensation between both traits especially in genotypes No. 1 and No. 5 under drought stress condition and No. 1 and No. 11 under irrigated condition, where the highest rate but the shorter duration were found (Table 3). In fact, a negative phenotypic correlations (r =-0.73** and r =-0.84** under irrigated and drought stress conditions, respectively) were observed between these two traits across the genotypes. The grain filling rate in genotypes No, 1, 2, 4, 6, 8, 9 and 11 decreased, whereas the effective grain filling period increased under drought stress condition in contrast with the irrigated condition. The amount of grain filling under drought stress condition in genotypes No. 3, 5 and 10 were increased, whereas the duration

Table 3. Grain weight (GW) at maturity, linear grain growth rate (LGGR), effective grain filling period (EFGP), contribution of pre-anthesis assimilates (CPAA) and contribution of current assimilates (CCA) in wheat genotypes under irrigated and drought stress conditions

Genotype	Irrigated condition					Drought stress condition				
	GW (mg)	LGGR (mg/grain /day)	EGFP	CPAA (%)	CCA (%)	GW (mg)	LGGR (mg/grain /day)	EGFP	CPAA (%)	CCA (%)
1	38.7	60.2	0.64	8.5	91.5	33.9	41.1	0.85	23.5	67.5
2	42.3	48.8	1.16	0.0	100.0	34.2	26.9	1.27	38.3	67.3
3	41.7	33.0	1.35	44.8	55.2	39.9	38.0	1.18	53.2	46.8
4	40.8	43.9	0.96	0.0	100.0	34.0	33.4	1.31	32.7	67.3
5	38.1	34.1	1.11	39.5	60.5	37.1	45.1	1.07	53.8	46.2
6	41.1	49.6	0.82	30.0	70.0	38.4	30.0	1.29	59.3	40.7
7	44.4	33.5	1.45	34.1	65.9	39.2	28.5	1.38	45.8	54.2
8	41.7	27.6	1.52	66.5	33.5	41.9	24.6	1.94	86.8	13.2
9	33.9	23.8	1.47	56.2	43.8	36.7	20.1	2.15	94.8	5.2
10	35.0	41.2	1.88	21.0	79.0	38.4	28.6	1.36	38.5	61.5
11	46.6	57.6	0.81	37.7	62.3	39.1	36.6	1.15	88.5	11.5
LSD 5%	11.30	30.45	0.82	30.7	-	14.25	28.1	1.03	55.9	-
Mean	40.4	41.2	1.20	30.75	62.2	37.5	32.1	1.36	55.9	43.8
Changes under drought stress relative to irrigated conditions						-7.18	-22.1	13.3	81.8	-29.6

of grain filling was decreased as compared with the irrigated condition. In conclusion, the higher rates were accompanied by the shorter duration across the genotypes studied, as also reported by Gooding *et al*. (2003).

Data about grain growth was well described by the logistic model proposed by Darroch and Baker (1990), because the coefficients of determination obtained were higher than 98% in most cases (Figures 1 to 14). Grain filling duration depended strongly on the environment, which accounted for 22.1% of the total variation. Probably genotypic variations were partially responsible for the observed differences in the grain filling duration. A reduction of 46.7% in grain filling duration was observed in the latest flowering genotype (No. 11) as compared with the earliest genotype, No. 8 (Table 3).

Differences between genotypes for grain filling duration were significant (Table 3). There was no evidence that grain filling in primary tillers suffered more from the stress than in main stem. For example coefficient of determinations between grain weight and grain number were 0.71 and 0.66 under irrigated and drought stress conditions, respectively (Figures 1 and 2). Under irrigated condition, the grain weight of the winter genotypes was 39.8 mg, whereas in the spring genotypes this ranged between 44.4 mg for the genotype No. 7 (Es14//Sitta//Agri/Nac), and 46.6 mg for the genotype No. 11 (Landrace of Azerbaijan Republic) (Table 3) whereas, on the average, the same grain weight was found in the winter and spring genotypes, as reported by Santiveri *et al*. (2002).

The timing and duration of the stresses coincided broadly with the different phases of grain development. The period from day 1 to 10 appeared to cover a lag phase, associated with cell division, and the start of the linear phase of grain growth. The period from day 11 to 40 broadly corresponded to the linear phase of grain growth in the irrigated plants (Figure 2). When stress was applied during this period (Figure 2), day 40 occurred after the end of the linear phase of grain filling. The period from day 41 to 50 occurred after maximum dry matter content was attained and therefore, coincided with the maturation period. All stresses imposed before the end of linear phase of grain growth reduced final mean grain weight (Table 3, Figure 2). This reduction was most marked when drought stress occurred between days 40 and 50 after anthesis (Figure 2). On the average, the effect of drought stress between days 11 and 40 was approximately additive (Figure 2), as reported earlier (Gooding *et al*. 2003). Drought stress also reduced grain number per spike (Table 3).

The period of lag phase was from day 1 to 20 and from day 20 to 40 and broadly corresponded to the linear phase of grain growth, when stress was imposed during this period (Figure 3), and day 40 occurred after the end of the linear phase of grain filling. The period from day 41 to 50 occurred after maximum dry matter content was attained and therefore, coincided with the maturation period in the genotype No. 1. All stresses imposed before the end of the linear phase of grain growth, reduced final mean grain weight by

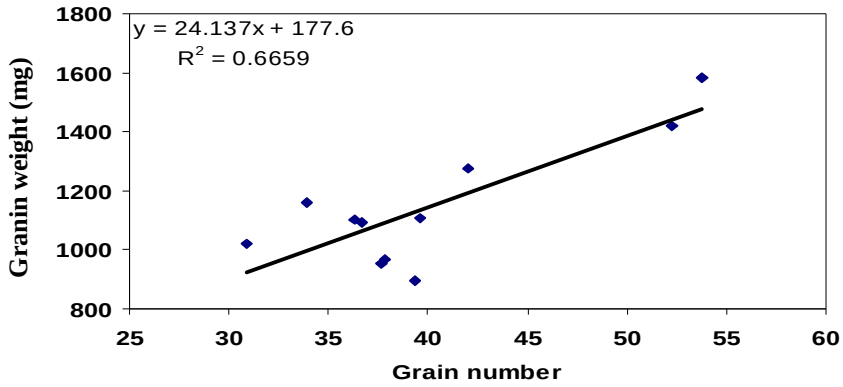

Figure 1. Relationship between grain weight and grain number on wheat genotypes under irrigated condition

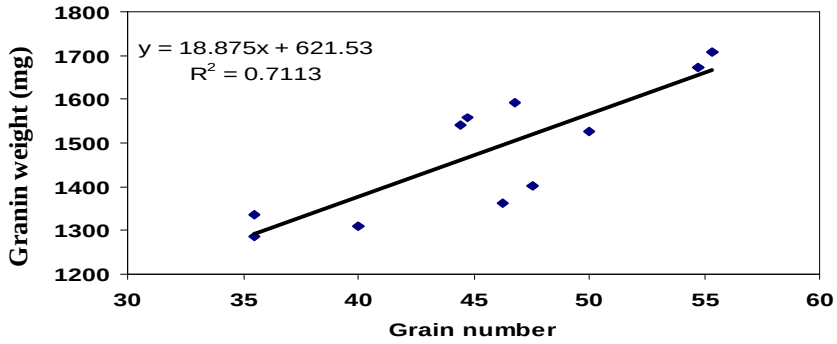

Figure 2. Relationship between grain weight and grain number on wheat genotypes under rainfed condition

12.4% (Table 3, Figure 3). This reduction was mostly marked when drought stress occurred between days 40 and 50 after anthesis (Figure 3). The effect of drought stress between days 20 and 40 has been reported to be additive (Gooding *et al*. 2003) as it was in our experiment (Figure 3).

All stresses applied before the end of the linear phase of grain growth reduced final grain weight (Table 3, Figures 4 to 12). This reduction was considerable when drought stress occurred between days 1 and 20 after anthesis (Figures 4 to 12). Effect of drought stress between days 21 and 40 was approximately additive upon exposure to the stress condition (Figures 4 to 12). Reductions in the final grain weight were mostly attributable to an earlier end to the grain filling when stress was applied and day 40 occurred after the end of the linear phase of grain filling. All stresses imposed before the end of the linear phase of grain growth reduced final grain weight by 12.4, 19.1%, 4.3%, 16.7%, 2.6%, 6.6%, 11.7% and 16.1% for the genotypes No. 1, 2, 3, 4, 5, 6, 7 and 11, respectively (Table 3, Figures 4, 5, 6, 7, 8, 9, 10 and 14). This reduction was most marked when drought stress occurred between days 40 and 50 after anthesis. On the average, the effect of drought stress between days 11 and 40 was additive across genotypes.

In the genotypes No. 8 and 10, the period of lag and linear grain growth were from day 1 to 20 and from day 21 to 40, respectively. The linear grain growth in the genotypes No. 8 and 10 were more additive under drought stress than irrigated condition so that grain filling continued until the end of physiological maturity and no reduction was observed in grain weight. In the genotype No. 9, the period from days 1-10 was lag period under both irrigated and drought condition. The linear grain growth was more additive under drought stress than irrigated conditions (Figure 11).

Grain filling rate was negatively and significantly correlated (r= -0.68) with the contribution of assimilates to developing kernels in the irrigated condition. Under irrigated and drought stress conditions, phenotypic correlations of grain filling rate with grain weight per spike (r= 0.87, r= 0.62) and grain number per spike (r= 0.71, r= 0.63) were significant. Furthermore, a significant positive correlation coefficient between grain filling rate and accumulation of dry matter at maturity (r=0.87) was observed in the irrigated environment. The observed values for effective grain filling period and grain filling rate suggested the compensation between these traits in both drought stress and irrigated plants. In fact, a significant negative phenotypic correlation was observed between these traits under irrigated and drought stress conditions (r =−0.73, r=-0.84, respectively). The same result was also reported by Goodong *et al*. (2003).

Mobilization of dry matter from vegetative organs to developing kernels was significantly correlated with accumulation of post-anthesis assimilate (r= -0.72), grain weight per spike (r= -0.74), grain number per spike (r= -0.73) and contribution of pre-anthesis assimilate to kernels (r= 0.95) in the irrigated environment. However, this character was only significantly correlated

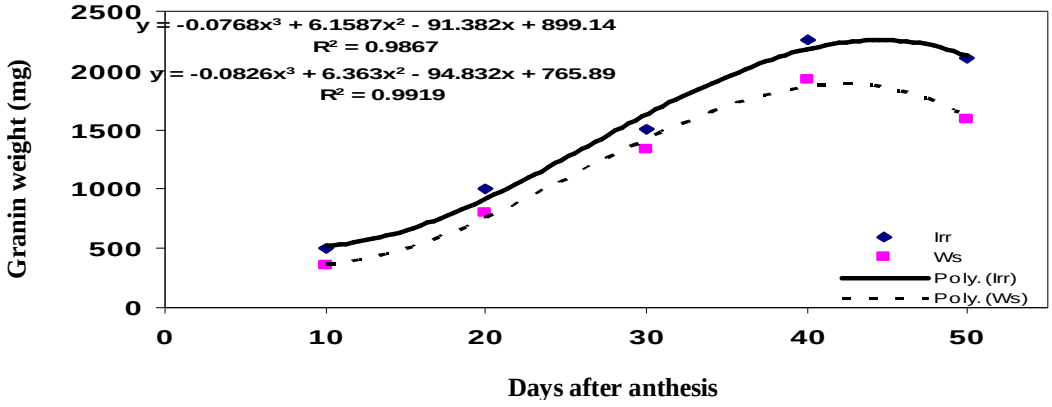

Figure 3. Changes in grain weight of wheat genotypes under irrigated and drought stress conditions

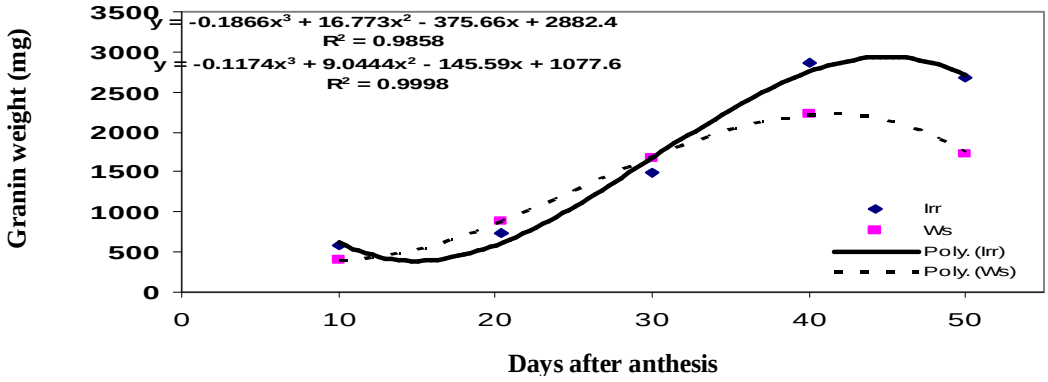

Figure 4. Effective grain filling in the wheat genotype No. 1 under irrigated and drought stress conditions

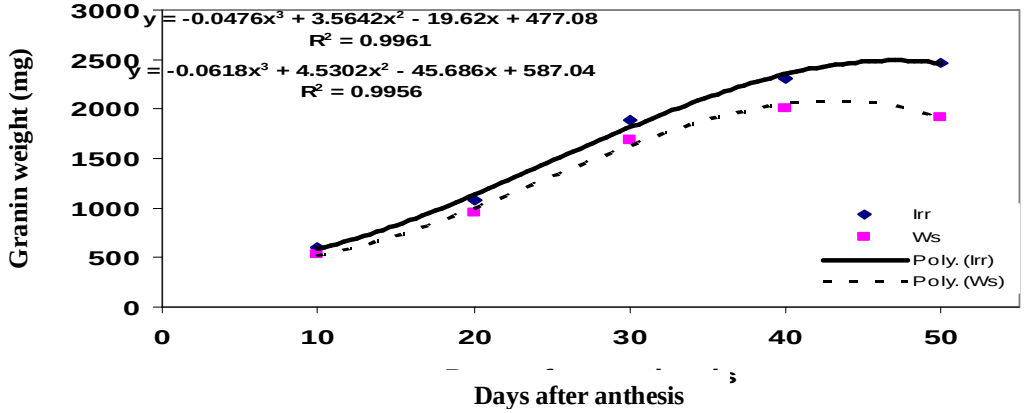

Figure 5. Effective grain filling in the wheat genotype No. 2, under irrigated and drought stress conditions

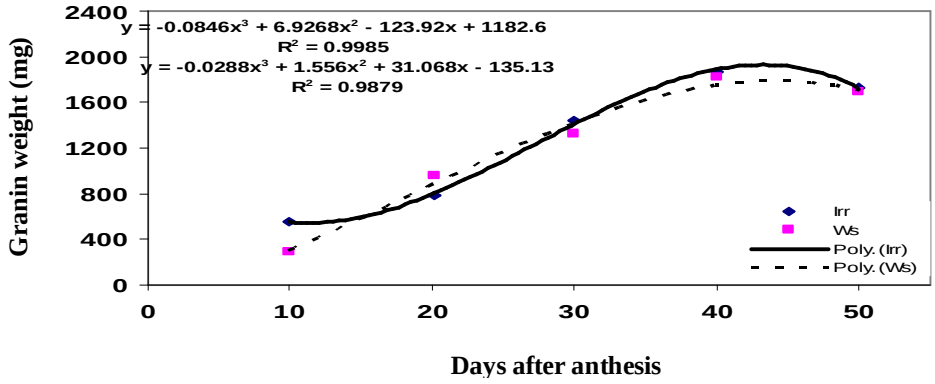

Figure 6. Effective grain filling in the wheat genotype No. 3, under irrigated and drought stress conditions

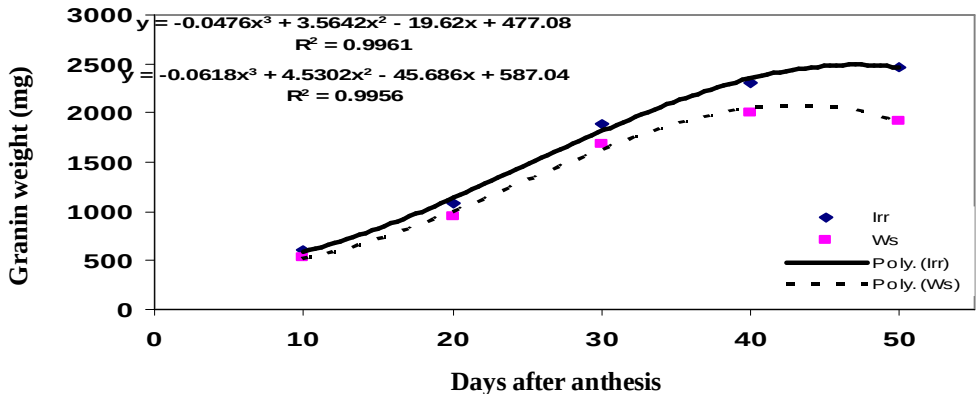

Figure 7. Effective grain filling in the wheat genotype No. 4, under irrigated and drought stress conditions

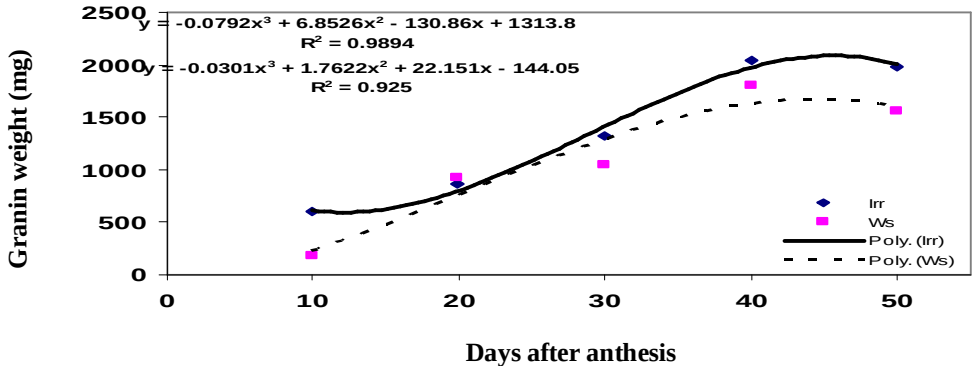

Figure 8. Effective grain filling in the wheat genotype No. 5, under irrigated and drought stress conditions

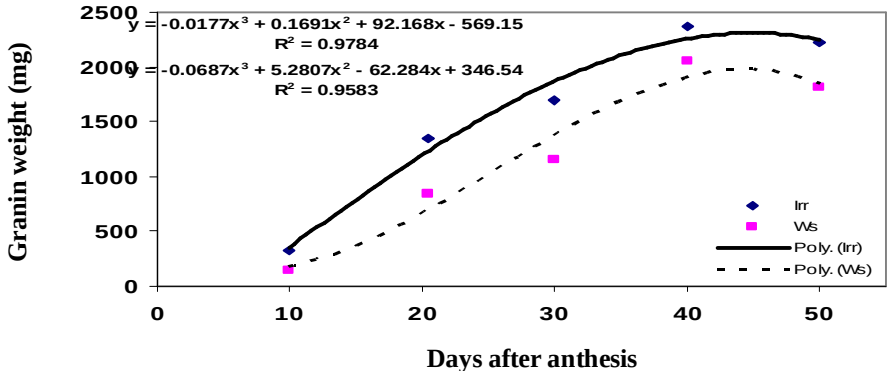

Figure 9. Effective grain filling in the wheat genotype No. 6 under irrigated and drought stress conditions

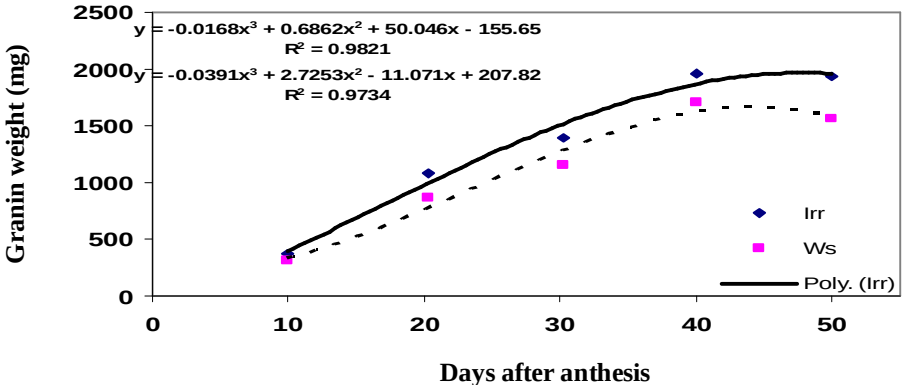

Figure 10. Effective grain filling in the wheat genotype No. 7 under irrigated and drought stress conditions

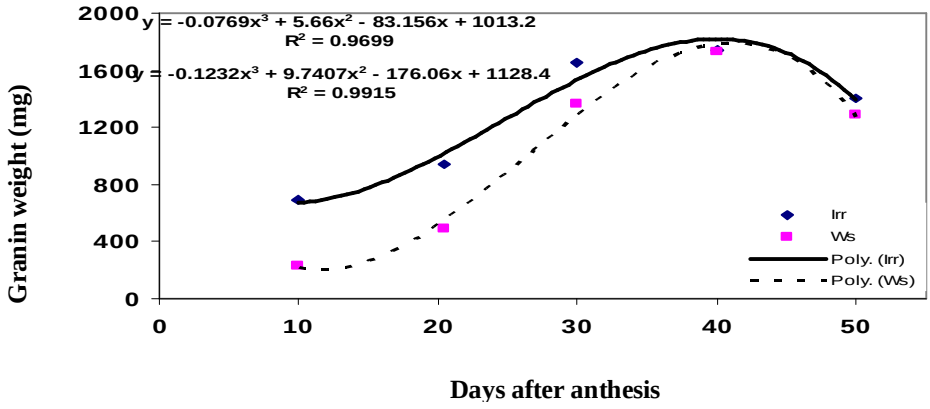

Figure 11. Effective grain filling in the wheat genotype No. 8 under irrigated and drought stress conditions

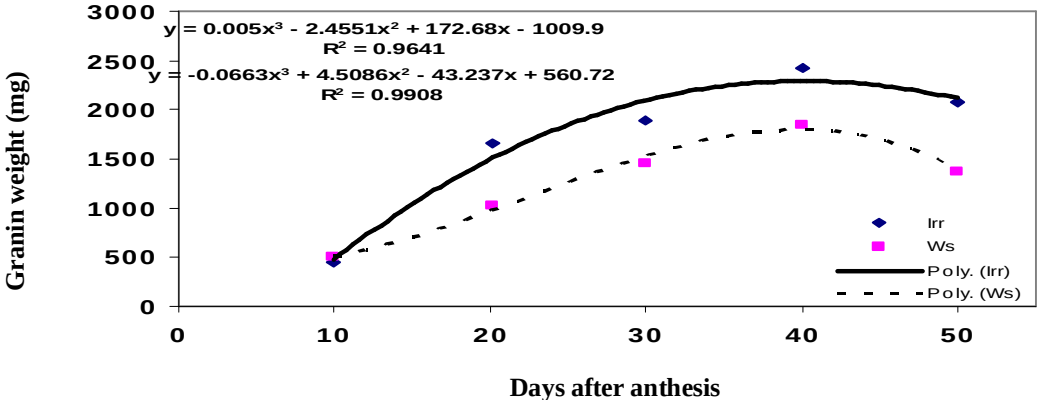

Figure 12. Effective grain filling in the wheat genotype No. 9 under irrigated and drought stress conditions

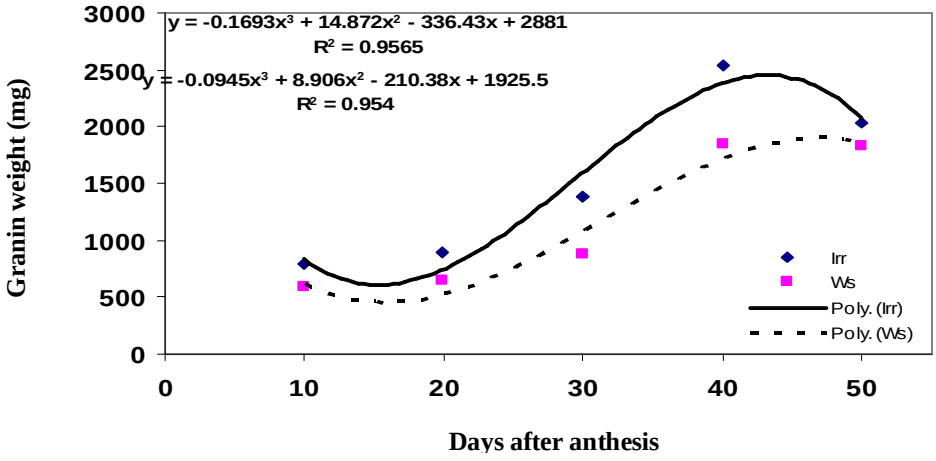

Figure 13. Effective grain filling in the wheat genotype No. 10 under irrigated and drought stress conditions

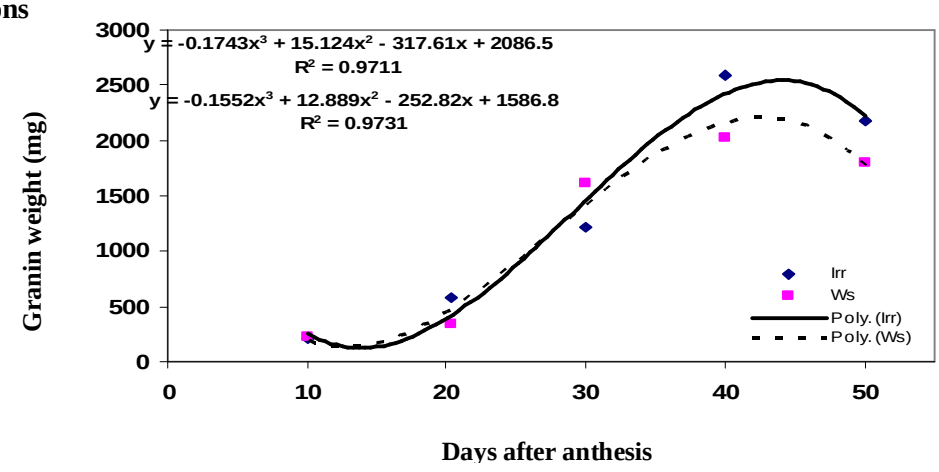

Figure 14. Effective grain filling in the wheat genotype No. 11 under irrigated and drought stress conditions

with the contribution of pre-anthesis assimilate to kernels (r=0.87) and grain weight (r= 0.61) under drought stress condition. Gavuzzi *et al.* (1997) reported high correlation of grain yield with the storage of non-structural carbohydrates in the drought condition.

Significant phenotypic correlation coefficients of effective grain filling period with grain weight (r= -0.70, r= -0.67), accumulation of pre-anthesis assimilates (r= -0.6, r= -0.76) and grain filling rate (r= -0.73, r= -0.84) were observed under irrigated and drought stress conditions, respectively. Effective grain filling period was also significantly correlated with accumulation of post-anthesis assimilates (r= -0.79) and grain number per spike (r= 0.68) in in the drought stressed plants. In addition, a phenotypic correlation coefficient of 0.56 was found between grain number per spike and accumulation of pre- anthesis assimilates under drought stress condition. According to Fischer (1980), who reported the same results, the assimilate supply problem may be compounded by further reductions in photosynthesis due to water stress, so it was suggested that the reduction in seed number caused by stress at this critical period is proportional to the reduction in leaf area development and hence, in available assimilates during approximately 25 days before anthesis.

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