

## Selectable physiological traits for yield improvement in durum wheat grown under water deficit condition

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### Abstract

Wheat varieties differ significantly in yield potential and selecting the right variety for a particular zone is the most important step in growing a profitable wheat crop. The selection of a cultivar is principally an economic decision, where the producer must find a balance between risk and yield potential. Measurements of different physiological processes of plant response to drought is an important information on the reactions of the plant intended to remove or to reduce the harmful effects of water deficit and targeting of specific physiological characters that is associated with maintained better yield may be more effective than direct selection for yield. In this study high and low yielding durum wheat varieties were selected and grown under rain fed condition in the 1<sup>st</sup> and 2<sup>nd</sup> settlement zone in southern part of Syria (Daraa Province). High yielding varieties were superior in all physiological traits (membrane stability index, relative water content, chlorophyll content and chlorophyll fluorescence) in both zones, particularly at post anthesis stage and same varieties were superior in yield and yield components. However, strong positive association between different physiological traits and various yield components particularly in the 2<sup>nd</sup> settlement zone as water shortage was more severe. Our findings indicated the importance of these physiological traits particularly relative water content in yield improvement under water deficit condition.

**Keywords:** wheat, membrane stability index, chlorophyll content, relative water content, chlorophyll fluorescence

### INTRODUCTION

Wheat (*Triticum aestivum* L.) is the most important cereal crop in the world (Zahid et al., 2003; Tunio et al., 2006). It is a staple food for more than 35% of the world population and it is also the first grain crops in most of the developing countries. Wheat has been reported to provide 73% of the calorie and protein requirements of the daily diet (Arif et al., 2010). It plays an important role in food security and poverty alleviation as a strategic crop and has an important role in economy (Khan et al., 2011). Abiotic stress, especially drought stress is a worldwide problem, seriously constraining global crop production (Pan et al., 2002). It is one of the major causes of crop loss worldwide, which commonly reduces average yield for many crop plants by more than 50% (Wang et al., 2003; Bayoumi et al., 2008). Recent progressive global climate change and increasing shortage of water resources has made this problem more serious.

The effects of drought on yield depend on severity and the stage of plant growth during which it occurs (Khakwani et al., 2011). The ability of improving wheat cultivars that are able to maximum use of existing water and drought tolerant is the main objectives of increasing yield potential in semi-arid and dry areas (Ghasemali et al., 2011). In plants, a better understanding of the morpho-anatomical and physio-biochemical characteristics of changes in drought resistance could be used to select or create new varieties of crops to obtain a better productivity under water stress conditions (Nam et al., 2001; Martinez et al., 2007). The improvement of tolerance to drought has been a principal goal of the majority of breeding programmes for a long time, as a water deficit in certain stages of wheat growth is common for many wheat growing regions of the world (Farshadfar, 2012). Plant improvement for drought resistance is complicated by the lack of fast, reproducible screening techniques and the inability to routinely create defined and repeatable water stress conditions where a large amount of genotypes can be evaluated efficiently (Naroui Rad et al., 2012). Selection efficiency could be improved if particular physiological and/or morphological attributes related to yield under a stress environment could be identified and employed as selection criteria for complementing traditional plant breeding (Acevedo, 1991). The present investigation was carried out to evaluate a set of high and low yielding durum wheat varieties for physiological traits associated with drought tolerance and yield as well as yield components.

## **MATERIALS AND METHODS**

### **Plant materials and growth conditions**

This study was conducted in the 1<sup>st</sup> settlement zone (Jellen Research Station, annual rainfall 400 mm) with high and low yielding durum wheat varieties grown usually in this area, that is Bohouth11 and Acsad65 respectively, and in the 2<sup>nd</sup> settlement zone (Izra Research Station, annual rainfall 291 mm) with high and low durum wheat varieties grown usually in this area, that is, Douma1 and Hourani respectively. Seeds were obtained from Crop Research Directorate, General Commission for Scientific Agricultural Research, Syria, and sown under rainfed conditions in the field on 20<sup>th</sup> Nov. 2011. Crops were sown at an adjusted rate of 300 viable seeds/m<sup>2</sup> in three replications. Normal agronomic practices were performed and relevant metrological parameters were obtained from the observatory at each research station and daily minimum and maximum temperature and rainfall were recorded. Chlorophyll content (chl), membrane stability index (MSI), relative water content (RWC), chlorophyll fluorescence  $F_v/F_m$  were estimated on the first fully expanded leaf (third from top) at vegetative stage and flag leaf at anthesis and post anthesis stage.

### **Chlorophyll content estimation**

The chlorophyll meter (SPAD meter) was used for chlorophyll estimation. The meter makes instantaneous and non-destructive readings on a plant based on the quantification of light intensity (peak wavelength: approximately 650 nm: red LED) absorbed by the tissue sample. A second peak (peak wavelength: approximately 940 nm: infrared LED) is emitted simultaneous with red LED to compensate the leaf thickness.

### **Membrane stability index**

Membrane stability index was determined by recording the electrical conductivity of leaf leachates in double distilled water at 40 and 100°C (Deshmukh et al., 1991). Leaf samples (0.1 g) were cut into discs of uniform size and taken in test tubes containing 10 ml of double distilled water in two sets. One set was kept at 40°C for 30 min. and another set at 100°C in boiling water bath for 15 min and their respective electric conductivities were measured by Conductivity meter.

### **Relative water content**

Relative water content was determined by the method described by Barrs and Weatherly (1962). 100 mg leaf material was taken and kept in double distilled water in a petri-dish for two hours to make the leaf tissue turgid. The turgid weights of the leaf materials were taken after carefully soaking the tissues between the two filter papers. Subsequently this leaf material was kept in a butter paper bag and dried in oven at 65°C for 24 h and their dry weights were recorded and RWC was calculated.

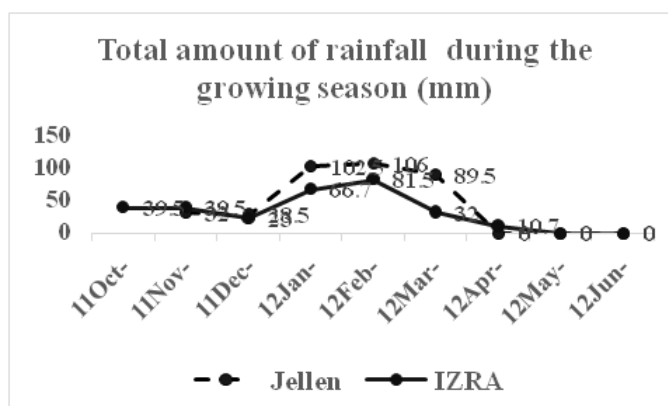
## Chlorophyll fluorescence

For the estimation of the polyphasic rise of fluorescence transients of intact leaves of non-stressed and water stressed plants were measured by a Plant Efficiency Analyzer (PEA, Handsatech Instruments Ltd., King's Lynn, UK) according to Strasser et al. (1995).

In mid June plants were harvested from m<sup>2</sup> and they were used for recording biological and grain yield, number of tillers/m<sup>2</sup>, grain number per ear, 1000 grain weight. Data were analyzed statistically and the analysis of variance (ANOVA) for factorial design at each stage was carried out using CoStat6.311 Cohort software and LSD values were also measured.

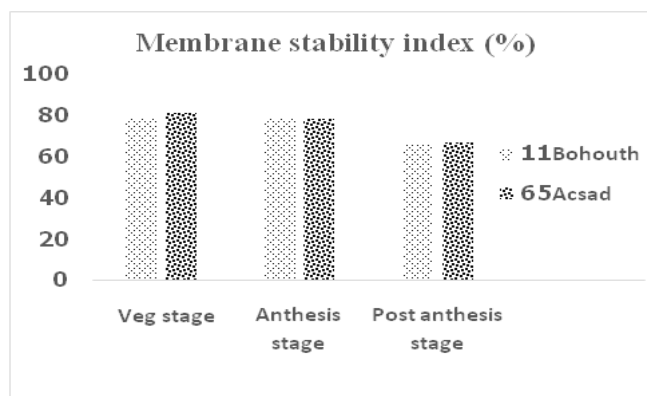
## RESULTS AND DISCUSSION

Rainfall was well distributed at the early stages of plant growth, which was sufficient enough for good establishment of the varieties before anthesis stage. The total amount of rain received in the 1<sup>st</sup> settlement zone were 358.8 mm, at the same time, only 292.9 mm were received in the 2<sup>nd</sup> settlement zone. Only 42.8 mm were received at the most sensitive stage (anthesis and grain filling stage) in the 2<sup>nd</sup> zone compared with 89.5 mm in the 1<sup>st</sup> zone, which may have adverse effect on growth and productivity particularly in the 1<sup>st</sup> settlement zone. In general, these amounts is not enough for ideal growth and yield in both zones for all varieties but the impact of water deficit stress imposed during anthesis and grain filling stage could be more sever particularly in the 2<sup>nd</sup> zone, the total amount of rainfall received in this area during the growing season is shown in Figure 1.



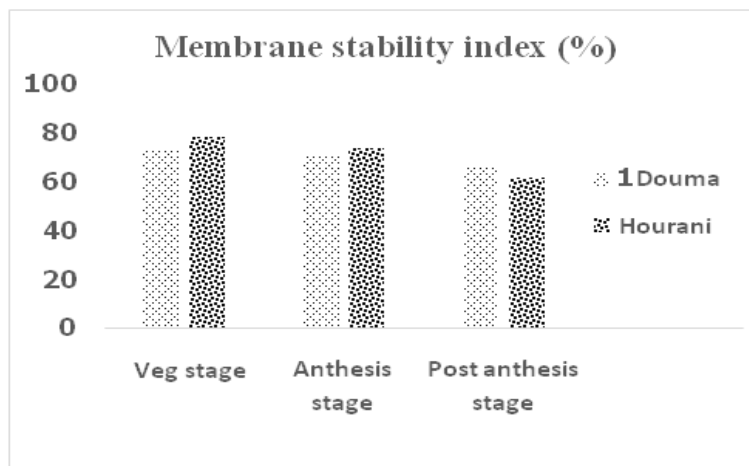
**Figure 1.** Total amount of rainfall (mm) in the 1<sup>st</sup> and 2<sup>nd</sup> settlement zone during the growing season.

Significant differences in membrane stability index were recorded between varieties in both zones, however, low yielding variety Acsad65 were superior at vegetative stage in the 1<sup>st</sup> zone that is, 81.2%, while no significant differences were recorded at anthesis and post anthesis stage between high and low yielding varieties (Figure 2).



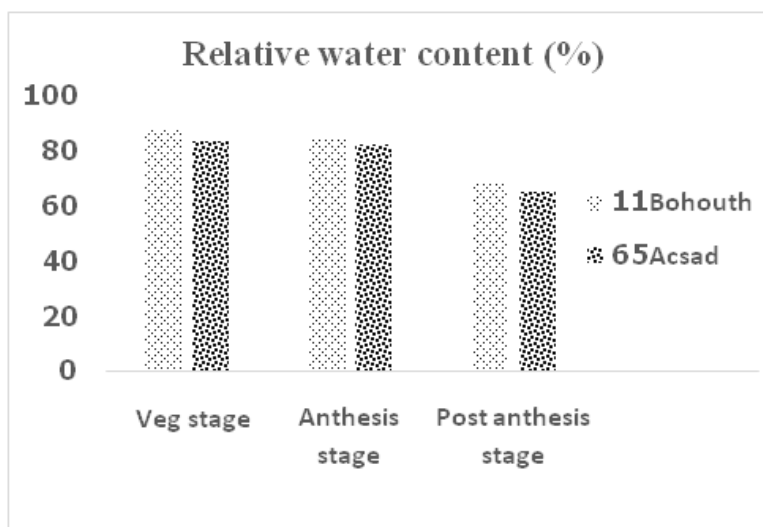
**Figure 2.** Membrane stability index (%) of high and low wheat yielding varieties (Bohouth11 and Acsad65) respectively in 1<sup>st</sup> settlement zones at vegetative, anthesis and post anthesis stages, LSD value: 1.8, 1.9 and 1.2 respectively.

In the 2<sup>nd</sup> zone Hourani showed highest membrane stability index value at vegetative and anthesis stage, while at post anthesis stage Douma1 were more superior and showed highest value, that is, 65.7% (Figure 3).



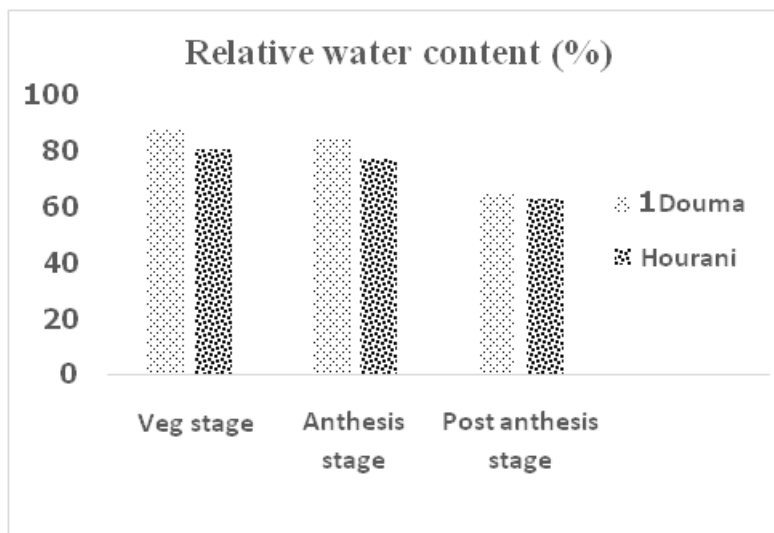
**Figure 3.** Membrane stability index (%) of high and low wheat yielding varieties (Douma1 and Hourani) respectively in 2<sup>nd</sup> settlement zones at vegetative, anthesis and post anthesis stages, LSD value: 2.3, 2.2 and 2.8 respectively.

It is well documented that the maintenance of cell membrane integrity and stability under water deficit conditions is a major component of drought tolerance in plants (Bajjii et al., 2001). The results from electrolyte leakage measurements at anthesis and post anthesis stages in our experiment showed that membrane integrity was higher for high yielding varieties compared to low yielding varieties particularly in the 2<sup>nd</sup> settlement zone where water deficit was more severe. Membrane stability is a widely used criterion to assess crop drought tolerance, since water stress caused water loss from plant tissues which seriously impairs both membrane structure and function (Buchanan et al., 2000). This character were used successfully by many wheat breeder for evaluating drought stress tolerance and it is associated with drought tolerance and yield stability under water stress deficit conditions (Almeselmani et al., 2012).



**Figure 4.** Relative water content (%) of high and low wheat yielding varieties (Bohouth11 and Acsad65) respectively in 1<sup>st</sup> settlement zones at vegetative, anthesis and post anthesis stages, LSD value: 2.6, 1.1 and 1.3 respectively.

Bohouth11 showed highest relative water content values at vegetative, anthesis and post anthesis stages that is, 87.3, 83.7 and 67.8% respectively in the 1<sup>st</sup> zone (Figure 4), while in the 2<sup>nd</sup> zone Douma1 showed highest values at all growth stages that is, 85.2, 81.7 and 64.2% respectively (Figure 5).

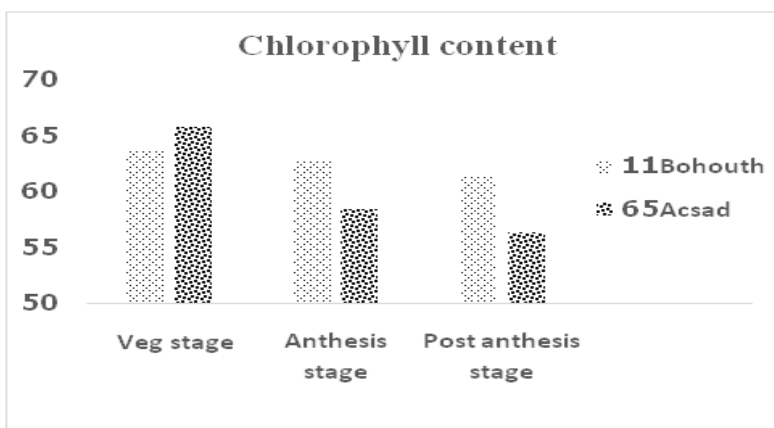


**Figure 5.** Relative water content (%) of high and low wheat yielding varieties (Douma1 and Hourani) respectively in 2<sup>nd</sup> settlement zones at vegetative, anthesis and post anthesis stages, LSD value: 3.6, 3.2 and 1.5 respectively.

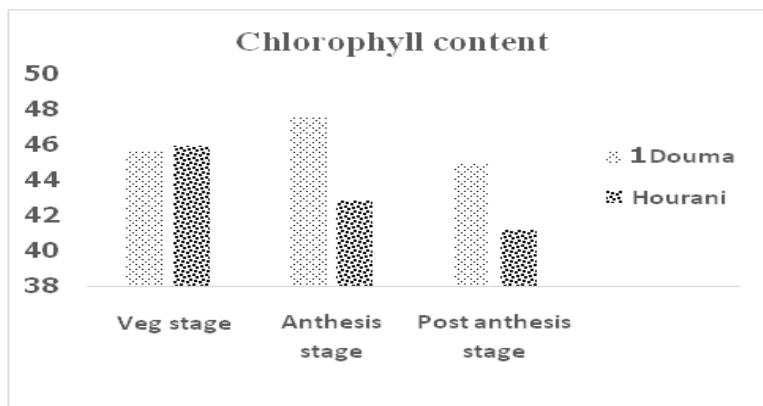
However, in the 1<sup>st</sup> zone Bohouth11 were superior compared with Acsad65 by 5, 2 and 4% at vegetative, anthesis and post anthesis stages respectively, while in the 2<sup>nd</sup> zone Douma1 were more superior compared with hournai by 9, 8 and 3% respectively at various growth stages. The differences in the 2<sup>nd</sup> zone were highly significant compared with the 1<sup>st</sup> zone.

It is also reported that high relative water content is a resistant mechanism to drought, and is the result of more osmotic regulation or less elasticity of tissue cell wall (Ritchie et al., 1990). According to Almeselmani et al. (2011) RWC indicates the water status of the cells and has significant association with yield and stress tolerance. The differences in RWC in wheat leaves may also be due to differences in the ability of the tested varieties to accumulate and adjust osmotically to maintain tissue turgor and hence physiological activities. According to Shamsi (2010) difference in RWC of cultivars that are under drought stress may be due to the differences in their ability to absorb more water from soil or the ability of the stomata to reduce the loss of water. However, high yielding varieties were more superior in this character particularly in the 2<sup>nd</sup> zone at productive stages.

Significant differences in chlorophyll content were recorded between Bohouth11 and Acsad65 in the 1<sup>st</sup> zone. However, Acsad65 were superior at vegetative stage, while Bohouth11 showed highest values at anthesis and post anthesis stages in this zone that is, 62.7 and 61.4 respectively (Figure 6). However, Bohouth11 were superior by 7 and 8% respectively compared by Acsad65 at anthesis and post anthesis stage.



**Figure 6.** Chlorophyll content (SPAD reading) of high and low wheat yielding varieties (Bohouth11 and Acsad65) respectively in 1<sup>st</sup> settlement zones at vegetative, anthesis and post anthesis stages, LSD value: 1.7, 2.6 and 2.4 respectively.

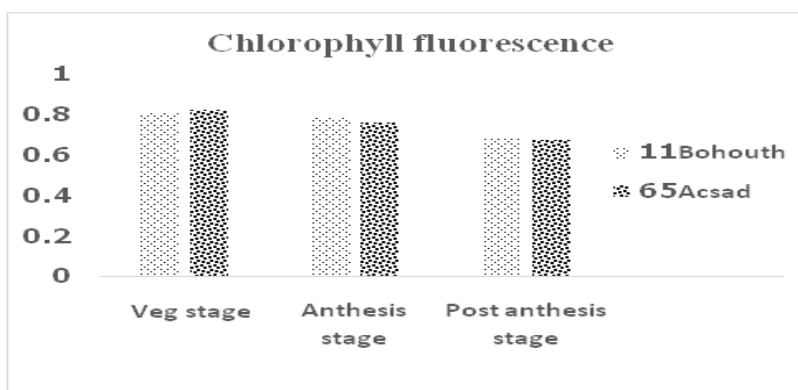


**Figure 7.** Chlorophyll content (SPAD reading) of high and low wheat yielding varieties (Douma1 and Hourani) respectively in 2<sup>nd</sup> settlement zones at vegetative, anthesis and post anthesis stages, LSD value: 1.8, 2.8 and 1.9 respectively.

In the 2<sup>nd</sup> zone no significant differences in chlorophyll content were recorded at vegetative stage between low and high yielding varieties, but at anthesis and post anthesis stage Douma1 showed highest values that is, 47.6 and 44.9 respectively. However, Douma1 were superior by 10 and 8% compared by Hourani at anthesis and post anthesis stage respectively (Figure 7).

Chlorophyll is one of the major chloroplast components for photosynthesis and relative chlorophyll content has a positive relationship with photosynthetic rate and flag leaf chlorophyll content is an indicator of the photosynthetic activity and its stability for the conjugation of assimilate biosynthesis (Bijanazadeh and Emam, 2010). Chlorophyll content is one of the major factors affecting photosynthetic capacity. Reduction or no-change in chlorophyll content of plant under drought stress has been observed in different plant species and its intensity depends on stress rate and duration (Arjenaki et al., 2012). Our achieved results showed that high yielding varieties were superior in this character at anthesis and post anthesis stages in both zones.

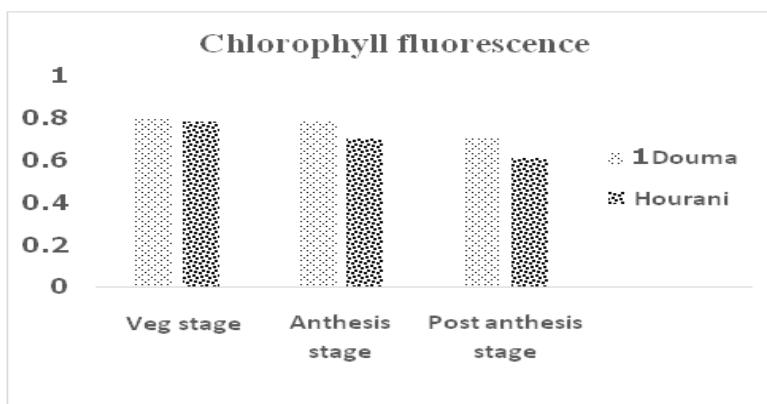
Highest chlorophyll fluorescence values were recorded in Acsad65 at vegetative stage in the 1<sup>st</sup> zone, while Bohouth11 were more superior at anthesis and post anthesis stages that is, 0.79 and 0.68 respectively. However, Bohouth11 were more superior by 4 and 2% respectively compared by Acsad65 at anthesis and post anthesis stage respectively (Figure 8).



**Figure 8.** Chlorophyll fluorescence ( $F_v/F_m$  ratios) of high and low wheat yielding varieties (Bohouth11 and Acsad65) respectively in 1<sup>st</sup> settlement zones at vegetative, anthesis and post anthesis stages, LSD value: 0.04, 0.027 and 0.08 respectively.

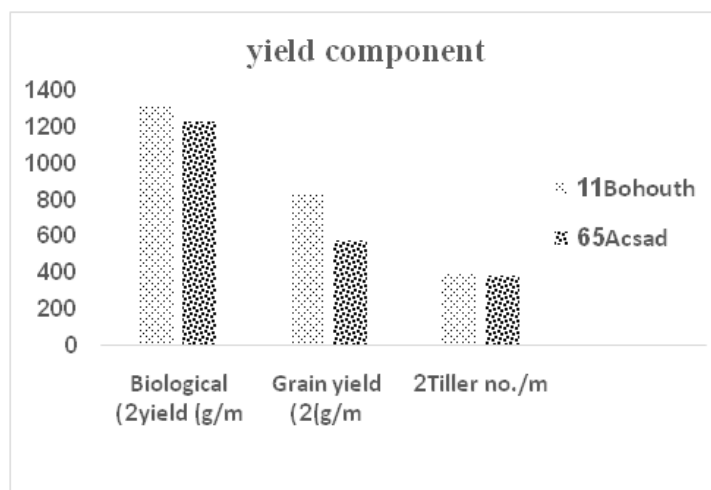
In the 2<sup>nd</sup> zone no significant differences were recorded between Douma1 and Hourani at vegetative stage, while at anthesis and post anthesis stage Douma1 were more superior and showed higher values compared with

Hourani that is, 0.79 and 0.7 respectively. However, Douma1 were more superior by 11 and 13% respectively compared by Hourani at anthesis and post anthesis stage (Figure 9).



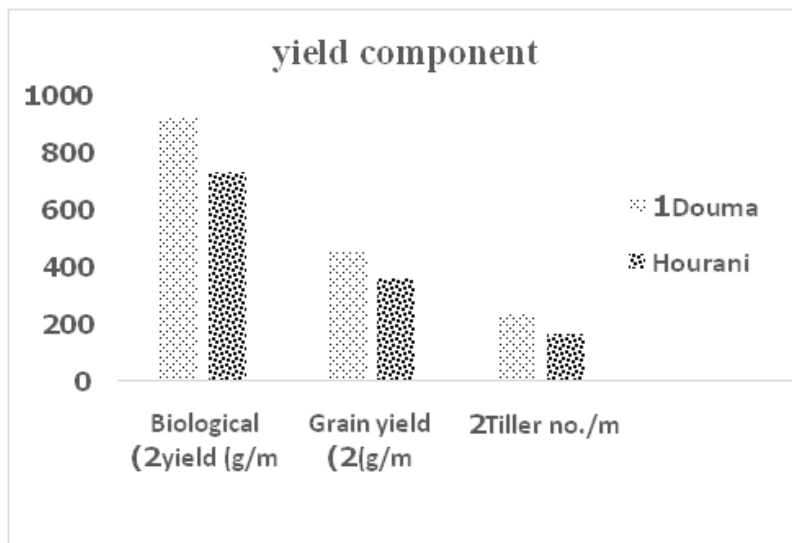
**Figure 9.** Chlorophyll fluorescence ( $F_v/F_m$  ratios) of high and low wheat yielding varieties (Douma1 and Hourani) respectively in 2<sup>nd</sup> settlement zones at vegetative, anthesis and post anthesis stages, LSD values: 0.052, 0.068 and 0.046 respectively.

Chlorophyll fluorescence analysis is a sensitive indicator of tolerance of the photosynthetic apparatus to environmental stress (Maxwell and Johnson, 2000). Flagella et al. (1995) and Almeselmani et al. (2011) also reported that drought tolerant cultivars showed a smaller decrease in photosynthetic efficiency ( $F_v/F_m$  ratios). According to Zivcak et al. (2009) chlorophyll fluorescence measurement appears very promising for screening of genotypes for improved tolerance to drought and high temperature. Data recorded in this experiment showed that high yielding varieties performed much better in both zones.



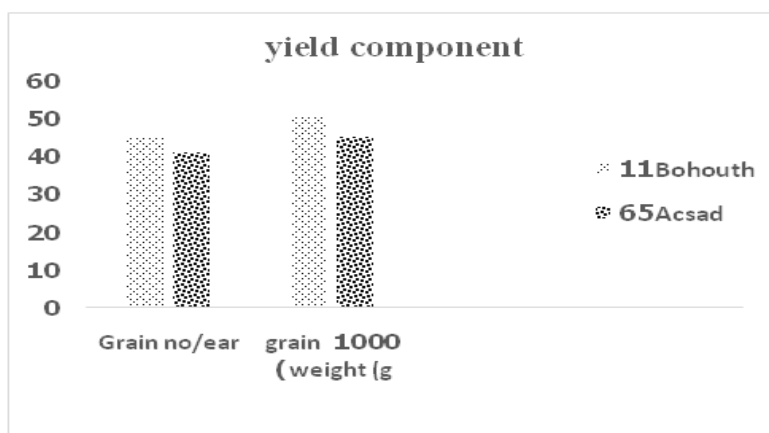
**Figure 10.** Biological yield (g), grain yield (g) and tiller number/m<sup>2</sup> of high and low wheat yielding varieties (Bohouth11 and Acsad65) respectively in 1<sup>st</sup> settlement zones, LSD values: 48, 37 and 18 respectively.

Highest biological yield were recorded in Bohouth11 in the 1<sup>st</sup> zone and Douma1 in the 2<sup>nd</sup> zone that is, 1312 and 918 g respectively. However, Bohouth11 were superior compared to Acsad65 by 7% while Douma1 were superior by 21% compared to Hourani. Significant differences were recorded between varieties in both zones in grain yield and highest values were recorded in Bohouth11 and Douma1 that is, 831 and 456 g respectively. However, Bohouth11 were superior by 31% compared to Acsad65, and Douma1 superior by 22% compared to Hourani. No significant differences in tiller number/m<sup>2</sup> were recorded between high and low yielding varieties in the 1<sup>st</sup> zone, while in the 2<sup>nd</sup> zone highest value were recorded in Douma1 that is, 387 and 229 respectively and our recorded data showed that Bohouth11 were superior by 3% compared by Acsad65, while Douma1 were superior by 30% compared by Hourani (Figures 10 and 11).



**Figure 11.** Biological yield (g), grain yield (g) and tiller number/m<sup>2</sup> of high and low wheat yielding varieties (Bohouth11 and Acsad65) respectively in 1<sup>st</sup> settlement zones, LSD values: 67, 71 and 41 respectively.

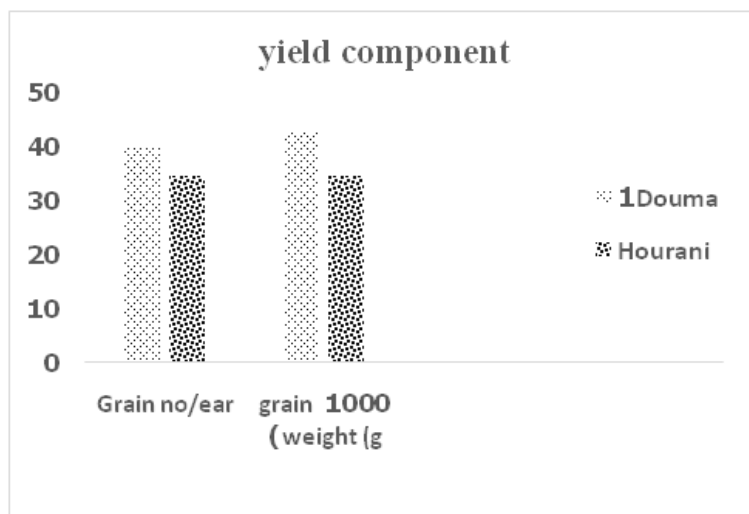
High yielding variety Bohouth11 in the 1<sup>st</sup> zone and Douma1 in the 2<sup>nd</sup> zone showed highest grain number/ear that is, 44.7 and 39.8 respectively. However, Bohouth11 were superior by 8% compared to Acsad65, while Douma1 were superior by 14% compared to Hourani. Also significant differences were recorded between varieties in 1000 grain weight highest value were recorded in Bohouth11 that is, 50.6 g in the 1<sup>st</sup> zone and in Douma1 that is, 42.9 g in the 2<sup>nd</sup> zone. This indicates that Bohouth11 were superior by 11% compared to Acsad65 in the 1<sup>st</sup> zone and Douma1 were superior by 20% compared to Hourani in the 2<sup>nd</sup> zone (Figures 12 and 13).



**Figure 12.** Grain number/ear and 1000 grain weight (g) of high and low wheat yielding varieties (Bohouth11 and Acsad65) respectively in 1<sup>st</sup> settlement zones, LSD values: 1.8 and 3.2 respectively.

The negative effect of drought stress on yield and yield performance has been well documented as a major problem in many developing countries of the world (Guo et al., 2004; Passioura, 2007; Almeselmani et al., 2012). Ashraf (1998) indicated that plant produces their maximum biomass under adequate water supply, whereas moisture stress causes a marked decrease in plant biomass production. Drought stress can reduce grain yield, Edmeades et al. (1994) estimated an average yield loss of 17 to 70% in grain yield due to drought stress. Sukhorukov (1989) reported reduction in grains/ear and 1000 grain weight revealed low yield in wheat under drought conditions. Khan et al. (2005) and Qadir et al. (1999) observed that 1000 grain weight of wheat was reduced mainly due to increasing water stress. The decrease in 1000 grains weight may be due to disturbed nutrient uptake efficiency and photosynthetic translocation within the plant (Iqbal et al., 1999) that produced shriveled grains due to hastened maturity.





**Figure 13.** Grain number/ear and 1000 grain weight (g) of high and low wheat yielding varieties (Bohouth11 and Acsad65) respectively in 1<sup>st</sup> settlement zones, LSD values: 2.8 and 4.7 respectively.

This is likely due to the shortage of moistures which forces plant to complete its grain formation in relatively lesser time (Riaz and Chowdhry, 2003). Drought stress reduced the number of gain/spike and grain yield (Saleem, 2003), while kernel weight is negatively influenced by high temperatures and drought during ripening (Atefeh et al., 2011). Under water stress, the decrease in seed set and grain growth in wheat has been reported by several workers (Morgan, 1980; Saini and Aspinall, 1982; Ahmadi and Baker, 1999; Khan et al., 2012).

There was a negative association between MSI and all yield components at all growth stages in the 1<sup>st</sup> zone and this association was high particularly at vegetative stage. In the 2<sup>nd</sup> zone there was a negative association between yield components and MSI at vegetative stage (Table 1), however, strong positive association between MSI and grain yield at anthesis and post anthesis stage (Tables 2 and 3). There was a strong positive association between relative water content, yield and yield components in both zones at various growth stages. The highest associations were recorded at post anthesis stage in the 2<sup>nd</sup> zone. With regard to chlorophyll content negative association was observed between chlorophyll content, yield and yield components in both zones at vegetative stage, also negative association was recorded in the 2<sup>nd</sup> zone at vegetative and anthesis stage, while positive association was recorded at post anthesis stage, however, the association was stronger in the 1<sup>st</sup> zone (Tables 1, 2 and 3). There was a negative association between chlorophyll fluorescence and various yield components at vegetative and post anthesis stage. The association was positive and stronger in the 2<sup>nd</sup> zone particularly at post anthesis stage.

**Table 1.** Association between MSI, RWC, chlorophyll content and chlorophyll fluorescence, yield and yield components at vegetative stage.

	MSI		RWC		CHL		Chl Flu	
	1 <sup>st</sup> zone	2 <sup>nd</sup> zone	1 <sup>st</sup> zone	2 <sup>nd</sup> zone	1 <sup>st</sup> zone	2 <sup>nd</sup> zone	1 <sup>st</sup> zone	2 <sup>nd</sup> zone
Biological yield	-0.89**	-0.98**	0.93**	0.98**	-0.83**	-0.67*	-0.68*	0.66*
Grain yield	-0.92**	-0.99**	0.94**	0.99**	-0.88**	-0.63*	-0.7**	0.74**
Tiller number/m <sup>2</sup>	-0.81**	-0.99**	0.97**	0.99**	-0.73**	-0.68*	-0.68*	0.64*
Grain number/ear	-0.67*	-0.96**	0.81**	0.97**	-0.84**	-0.69*	-0.28	0.54*
1000 grain weight	-0.75**	-0.97**	0.97**	0.98**	-0.61*	-0.64*	-0.68*	0.77**

**Table 2.** Association between MSI, RWC, chlorophyll content and chlorophyll fluorescence, yield and yield components at anthesis stage.

	MSI		RWC		CHL		Chl Flu	
	1 <sup>st</sup> zone	2 <sup>nd</sup> zone	1 <sup>st</sup> zone	2 <sup>nd</sup> zone	1 <sup>st</sup> zone	2 <sup>nd</sup> zone	1 <sup>st</sup> zone	2 <sup>nd</sup> zone
Biological yield	-0.1	-0.99**	0.97**	0.98**	0.92**	-0.41*	0.99	0.98**
Grain yield	-0.14	0.97**	0.84**	0.97**	0.91**	-0.46*	0.99	0.98**
Tiller number/m <sup>2</sup>	-0.91**	-0.95**	0.87**	0.95**	0.81**	-0.36*	0.93	0.94**
Grain number/ear	0.43*	-0.97**	0.68**	0.97**	0.96**	-0.26	0.95	0.95**
1000 grain weight	-0.18	-0.92**	0.79**	0.94**	0.81**	-0.45*	0.88	0.96**

**Table 3.** Association between MSI, RWC, chlorophyll content and chlorophyll fluorescence, yield and yield components at post anthesis stage.

	MSI		RWC		CHL		Chl Flu	
	1 <sup>st</sup> zone	2 <sup>nd</sup> zone	1 <sup>st</sup> zone	2 <sup>nd</sup> zone	1 <sup>st</sup> zone	2 <sup>nd</sup> zone	1 <sup>st</sup> zone	2 <sup>nd</sup> zone
Biological yield	-0.49*	0.98**	0.87**	0.99**	0.97**	0.85**	-0.42*	0.99**
Grain yield	-0.57*	0.98**	0.86**	0.99**	0.98**	0.82**	-0.53*	0.99**
Tiller number/m <sup>2</sup>	-0.67*	0.98**	0.95**	0.97**	0.92**	0.79**	-0.28	0.96**
Grain number/ear	-0.37	0.98**	0.72**	0.98**	0.97**	0.85**	-0.59*	0.94**
1000 grain weight	-0.51*	0.92**	0.99**	0.95**	0.85**	0.73**	-0.16	0.97**

## CONCLUSION

The goal of crop physiologists is to identify different physiological traits which could be manipulated to increase grain yields particularly under water deficit condition. Most difficulties encountered in the identification of accurate drought tolerance traits were due to the fact that wheat is cultivated under very different climatic conditions and faces very different drought scenarios worldwide. Knowledge of the changes in physiological traits associated with genetic gains in yield potential is essential to improve understanding of yield-limiting factors and to inform future breeding strategies. Knowledge of changes associated with advances in crop productivity is essential for understanding yield limiting factors and developing strategies for future genetic improvement. Our observation emphasizes on the importance of all these study characters particularly at post anthesis stage.

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