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RESEARCH ARTICLE

Morphological Traits Associated with Drought Stress Tolerance in Six Moroccan Durum Wheat Varieties Released Between 1984 and 2007

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Abstract

Drought is one of the main environmental factors affecting grain yield and plant architecture in durum wheat. The present work was conducted to evaluate the contribution of morphological traits above flag leaf node on grain yield of six Moroccan durum wheat varieties released between 1984 and 2007 and grown under two water regimes (irrigated and rainfed) during the 2015-2016 crop season. The following morphological traits were measured at anthesis: Flag leaf length (FLL), flag leaf area (FLA), peduncle length (PL), spike length (SL), spike area (SA), and green leaf area (GLA). Days from sowing to anthesis (DSA) and grain-filling period (GFP) were recorded. In addition, grain yield per plant (GYP) was evaluated at maturity. Analysis of variance showed the greater effect of water stress in explaining total variability of the studied traits. Mean comparisons indicated that water deficit significantly affected GYP and all morphological traits; it shortened DSA and GFP by 4 and 7 days, respectively. Moreover, GYP, DSA, and GFP increased in modern varieties. In contrast, FLL, FLA, and PL were reduced from old to modern varieties. Under irrigated conditions, correlation studies revealed that GYP was positively associated to GLA. Regarding the rainfed trial, GYP was positively correlated to FLL. Additionally, negative relationships were found between GYP and DSA in both water regimes. Stepwise regression highlighted the relative importance of each morphological traits on the stress susceptibility index, FLL was the most consistent trait entered the model followed by PL and FLA. In this study, some morphological traits above flag leaf node proved to be useful tools to select for grain yield in water-limited environments.

Key words : Durum wheat, morphological traits, grain yield, stress susceptibility index, Morocco

Introduction

Durum wheat (*Triticum turgidum* L. var. *durum*) is one of the cereals most grown worldwide and mainly used for pasta production, and other traditional foods such as couscous, flatbreads, and bulgur (Giraldo et al. 2016). In the Mediterranean Basin, durum wheat is grown in different climatic zones varying from cool and wet, to warm and dry (Royo et al. 2014). In the Mediterranean region, drought stress is a major limiting factor for crop production; water deficit in plants may lead to physiological disorders, such as a reduction in photosynthesis and transpiration (Araus et al. 2008; El Yamani et al. 2018; Farooq et al. 2009). In Morocco, durum wheat is one of the oldest cultivated cereals sown annually over 1.0 million hectares and produces around 1.2 million tons. However, in recent years, Morocco imports of durum wheat increased substantially in order to meet the growing demand for this commodity (Henkrar et al. 2016).

During the last decades, in several countries, breeders have attempted to produce modern varieties that are highly productive and widely adapted to contrasting environments. Durum wheat breeding programs began in Morocco at the beginning of the 20th century. By the 1970s, collaboration with CIMMYT (International Wheat Improvement Centre and Maize) and ICARDA (International Center for Agricultural Research in Dry Areas) allowed the introduction of germplasm characterized by earliness and containing the semi-dwarfing

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genes, responsible for plant height reduction (Nsarellah et al. 2005).

Getting a high yield will depend on the knowledge of both environmental and genetic yield-limiting factors (Reynolds et al. 2012). Comparisons of varieties bred in different periods (old, intermediate, and modern) can inform on the trend changes in agronomic characteristics and provide an estimate of breeding progress. Breeding programs for durum wheat were primarily focused on increased grain yield and early maturation to suit Mediterranean conditions where drought can occur during maturity (De Vita et al. 2007). Many works from several countries in Mediterranean region have provided a comparison of yield, agronomical, and morpho-physiological traits between old and modern durum wheat (Álvaro et al. 2008; De Vita et al. 2010; Giunta et al. 2019; Royo et al. 2008; Subira et al. 2015).

Morphological traits not only affect stress tolerance to limiting soil moisture, but they also indicate how adaptive genotypes cope with water stress (Anjum et al. 2011; Nouri-Ganbalani et al. 2009). These traits have the important role in determining yield components and are used in breeding programs for improving grain yield and introducing commercial varieties (Mollasadeghi et al. 2011). Briggs and Aytenfisu (1980) and Kaul (1974) indicated that green tissues above the flag leaf node are the main contributors to the synthesis and production of carbohydrates required to fill the grains. In fact, flag leaves contribute about 40% of the carbohydrates for grain filling (Sharma et al. 2003). Different results about the relationship among traits above the flag leaf and grain yield are reported in the literature ranging from strong (Singh and Singh 1992) to inexistent (McNeal and Berg 1977) or depending on the environment (Villegas et al. 2007).

In order to quantify drought tolerance, the stress susceptibility index (SSI) was suggested by Fischer and Maurer (1978) based on grain yield loss under stressed environments as compared to favorable ones. This index has been previously used in bread wheat (Bansal and Sinha 1991; Cedola et al. 1994) and durum wheat (Khamssi and Najaphy 2012; Mohammadi 2012; Villegas et al. 2007). The morphological traits could be useful tools to select for low SSI, in order to get varieties with high-yielding and good stability. In this context, the objective of the present work was to assess the relationships between morphological traits above the flag leaf node with grain yield, and their relative contribution to SSI in a set of Moroccan durum wheat varieties released between 1984 and 2007 and grown under two water regimes during the 2015-2016 crop season.

Materiel and Methods

This work was carried out in a pot experiment during the 2015-16 crop season at the experimental station of the Polydisciplinary Faculty of Taza (34°12′ N 4°00′ W, 550 m a.s.l). Six durum wheat (*Triticum turgidum* L. var. *durum*) varieties were selected to represent the most widely grown germplasm in Morocco during the past 30 years. On the basis of the year of release (Table 1), these varieties were grouped in three periods, namely old (released between 1980 and 1990), intermediate (released between 1990 and 2000), and modern (from 2000 to 2010). The study region is characterized by a Mediterranean climate with humid winters and semiarid summers. In the 2015-2016 crop season at Taza (Fig. 1) we registered a total rainfall of 365 mm, of which 360 mm fell between sowing and anthesis. Mean maximum and minimum temperatures from sowing to anthesis were 22 and 7°C, respectively, while from anthesis to maturity they were 36 and 18°C, respectively.

Fifteen seeds of each variety were sown on December 22, 2015 in plastic 10 L pots filled with a mixture of field soil, peat, and sand (2:2:1, v/v/v) in a completely randomized block design with three replications. Pots were subjected to two water regimes (irrigated and rainfed). Irrigated pots were weighed on weekly basis, cumulative water loss was added to each pot to compensate for transpiration and evaporation. Days from sowing to anthesis (DSA) and grain-filling period (GFP) were recorded following Zadoks phenological scale (Zadoks et al. 1974). At flowering, three plants per pot were used to measure the flag leaf length (FLL), flag leaf area (FLA), peduncle length (PL), spike length (SL),

Table 1. Description of the six durum wheat varieties used in this study.

Varieties	Origin	Year of release
Olds		
	Marzak INRA Morocco	1984
	Karim INRA Morocco	1985
Intermediates		
	Ourgh INRA Morocco	1995
	Tarek INRA Morocco	1995
Moderns		
	Marouane INRA Morocco	2003
	Faraj INRA Morocco	2007

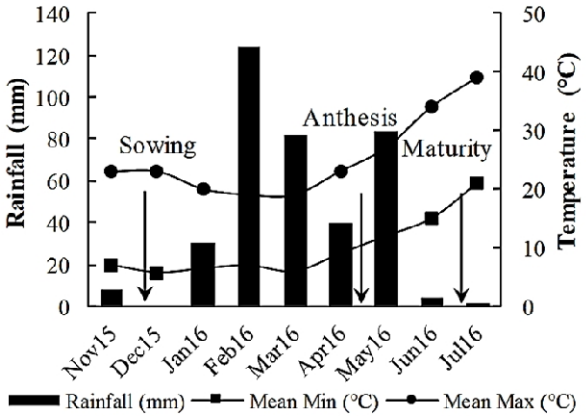

Fig. 1. Rainfall, minimum and maximum temperatures recorded during 2015-2016 crop season.

spike area (SA), and green leaf area (GLA) Additionally, grain yield per plant (GYP) was determined at maturity. Stress susceptibility index (SSI) was calculated for each variety using the following relationship given by Fischer and Maurer (1978):

$$SSI = \frac{1-(GYPr\ /\ GYPi)}{1-(GYPmr\ /\ GYPmi)}$$

where GYPr and GYPi are the mean grain yields per plant of a given variety under rainfed and irrigated conditions, respectively. GYPmr and GYPmi are the mean grain yields per plant of all varieties under rainfed and irrigated conditions, respectively.

Combined analyses of variance (ANOVA) were carried out over periods of release and water regimes. Least significant difference (LSD) values were calculated at the 5% probability level. Correlation coefficients among the studied traits were calculated separately for the two water regimes. To elucidate the relative contribution of some morphological traits to determine the SSI, a stepwise regression was performed by using the variety means across water regimes. Principal component analyses (PCA) were performed on the basis of a correlation matrix constructed over the mean data of all replicates. All statistical analyses were carried out by using the STATGRAPHICS package version XVI (Statpoint Technologies, Inc., Virginia, USA).

Results

Analyses of variance

Results of the combined ANOVA showed that water regime was much more important than the period of release in explaining the variation of the studied traits (Table 2). Over 75% of total variance was assigned to water regime effect. In contrast, the period of release effect was mainly exhibited for DSA and PL accounting for 20 and 29% of total variance, respectively. The magnitude of water regime × period of release interaction was of minor importance and accounted for about 11% of the total variance, except for DSA and GFP where it explained 47 and 21%, respectively.

Effect of water regimes

Mean values of the investigated traits between water regimes are summarized in Table 3. Our results showed a significant reduction in GYP and all of the morphological traits under rainfed conditions. Water deficit reduced GYP by triple and FLA by half while decreases in FLL, PL, SA, and GLA were about 30%. DSA and GFP were shortened by 4 and 7 days, respectively, in the rainfed trial.

Effect of periods of release

In addition, mean comparison among periods of release

Table 2. Mean squares of the combined analyses of variance for days sowing-anthesis (DSA), grain-filling period (DSA), grain yield per plant (GYP), flag leaf length (FLL), flag leaf area (FLA), peduncle length (PL), spike length (SL), spike area (SA), and green leaf area (GLA) in six durum wheat varieties released in different periods (old, intermediate, and modern) grown under two water regimes (irrigated and rainfed) during 2015-2016.

Source of variation	d.f.	DSA	GFP	GYP	FLL	FLA	PL	SL	SA	GLA
Water regime (WR)	1	84.0**	506.3***	21.30 ***	134.2***	1125.6***	173.8***	3.063**	24.50***	1617.5***
Period of release (PR)	2	55.5**	76.3**	1.75***	18.8*	60.5*	78.6**	0.563*	2.13*	3.0
WR x PR	2	131.1***	156.3***	2.57***	1.2	17.4	5.6	0.363	0.33	184.5*
Replicate	2	2.4	2.6	0.02	2.3	5.3	3.6	0.083	0.26	32.6
Residuel	28	7.1	9.2	2.54	3.9	13.0	8.4	0.167	0.45	49.2
Total	35									

* Significant at 0.05 probability level; ** Significant at 0.01 probability level; *** Significant at 0.001 probability level.

Table 3. Mean values of days sowing-anthesis (DSA), grain-filling period (DSA), grain yield per plant (GYP), flag leaf length (FLL), flag leaf area (FLA), peduncle length (PL), spike length (SL), spike area (SA), and green leaf area (GLA) in six durum wheat varieties released in different periods (old, intermediate, and modern) grown under two water regimes (irrigated and rainfed) during 2015-2016.

	DSA (Days)	GFP (Days)	GYP (g)	FLL (cm)	FLA (cm^2)	PL (cm)	SL (cm)	SA (cm^2)	GLA (cm^2)
Water regimes									
Rainfed	88.9 b	20.0 b	0.619 b	14.2 b	11.2 b	18.8 b	6.15 b	6.59 b	42.8 b
Irrigated	92.0 a	27.5 a	2.158 a	18.0 a	22.5 a	23.3 a	6.73 a	8.24 a	56.2 a
Periods of release									
Olds	88,8 b	21,8 b	1.201 b	17.5 a	19.2 a	23.4 a	6.3 b	7.2 b	49.3 a
Intermediates	89,7 b	22,9 b	1.267 b	15.6 b	16.4 ab	21.3 a	6.7 a	7.9 a	49.2 a
Moderns	92,9 a	26,6 a	1.700 a	15.2 b	14.7 b	18.3 b	6.3 b	7.2 b	50.1 a

Means for each character followed by the same letter are not significantly different according to the LSD test at $P < 0.05$.

Table 4. Correlations coefficients among days sowing-anthesis (DSA), grain-filling period (DSA), grain yield per plant (GYP), flag leaf length (FLL), flag leaf area (FLA), peduncle length (PL), spike length (SL), spike area (SA), and green leaf area (GLA) of six durum wheat varieties released in different periods (old, intermediate, and modern) grown under two water regimes (irrigated and rainfed) during 2015-2016.

	DSA	GFP	GYP	FLL	FLA	PL	SL	SA	GLA
Irrigated									
DSA	-	-0.625	-0.749	0.938**	0.799	0.436	-0.026	-0.295	-0.796
GFP			0.878*	-0.607	-0.638	-0.762	-0.584	-0.257	0.915*
GYP				-0.656	-0.721	-0.735	-0.135	0.200	0.912*
FLL					0.885*	0.535	0.122	-0.203	-0.667
FLA						0.822*	0.164	-0.140	-0.595
PL							0.453	0.167	-0.554
SL								0.890*	-0.301
SA									-0.024
GLA									-
Rainfed									
DSA	-	-0.519	-0.778	-0.635	-0.382	-0.608	0.283	-0.167	-0.760
GFP			0.251	0.086	-0.325	-0.059	-0.006	0.084	0.496
GYP				0.922*	0.342	0.618	-0.807	-0.257	0.679
FLL					0.467	0.631	-0.739	-0.071	0.758
FLA						0.918**	-0.083	0.106	0.536
PL							-0.356	-0.129	0.673
SL								0.687	-0.273
SA									0.263
GLA									-

* Significant at 0.05 probability level; ** Significant at 0.01 probability level; *** Significant at 0.001 probability level.

(Table 3) showed an increase in GYP (0.30-0.42 g/plant), DSA (88-92 days), and GFP (22-27 days) from old to modern varieties. In contrast, a significant decrease was observed in FLL (17.5-15.2 cm), FLA (19.2-14.7 cm^2), and PL (23.4-18.3 cm) from old to modern varieties. No clear differences were obtained among varieties concerning SL: SA and GLA.

Relationships among the studied traits

Correlation analyses for different water regime among studied traits are shown in Table 4. Under irrigated conditions, significant associations were detected between DSA and FLL (r = 0.938**), GFP and GLA (r = 0.915*), and GLA and GYP (r = 0.912*). In addition, a positive relationship was found between GYP and GFP (r = 0.878*). Regarding rainfed trial, GYP was positively correlated to FLL (r = 0.922*). At both water regimes, GYP was negatively related with DSA.

In addition to correlation studies, principal component analysis (PCA) was performed on mean values across two water regimes of the nine traits shown in Fig. 2. The first two PCA axes accounted for 80% of the total variance: 60 and 20 for axes 1 and 2, respectively. PC1 clearly separated the irrigated trial in the negative direction and rainfed trial in the opposite direction. Values of all parameters were higher under irrigated conditions. Moreover, there was a clear separation of DSA toward the positive direction of PC1 which interacted negatively with GYP, GLA, and GFP in the other side of this axis. Additionally, a positive relationship appeared between all morphological traits positioned on the negative side of PC1. No clear separation among modern, intermediate and old varieties was detected on both axes.

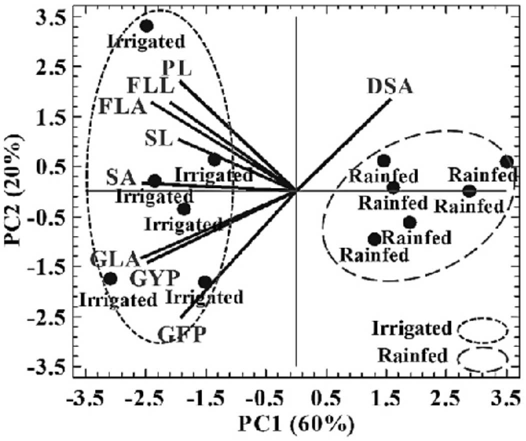

Fig. 2. Principal component analysis (PCA) projections on axes 1 and 2 accounting for 80 of total variance. The eigenvalues of the correlation matrix are symbolized as vectors representing traits that most influence each axis, DSA: Days sowing-anthesis, GFP: Grain filling period, GYP: Grain yield per plant, FLL: Flag leaf length, FLA: Flag leaf area, PL: Pedoncule length, SL: Spike length, SA: Spike area, GLA: Green leaf area. The 12 points representing varieties mean for each water regimes (Rainfed and irrigated).

Contribution of morphological traits to SSI

To explain the importance of some morphological traits in determining the SSI, a stepwise regression was carried out across the two water regimes, considering SSI as a dependent variable, and all morphological traits as independent variables (Table 5). Stepwise regression indicated that FLL was the trait most related to SSI, since it explained 83.5% of the SSI variation. PL and FLA entered the equation jointly with FLL and accounted for 12 and 4.5% of SSI variability, respectively. Mean values for each variety were calculated over water regimes and the three traits that entered in the regression model were plotted against SSI (Fig. 3). This later was negatively correlated with FLL (r = 0.803*), FLA (r = 0.790*), and PL (r = 0.755*) demonstrating that higher values for these traits resulted in lower SSI and thus greater resistance to drought.

Discussion

Analyses of variance showed the high influence of water regime on all studied traits with lower effect of period of release and interaction water regime × period of release. The water regime effect explained 75% of the studied traits variability, Similar results have been reported by Royo et al. (2010) in 191 durum wheat accessions tested at nine sites of four Mediterranean countries. Genotype effect was of lower magnitude although statistically significant for all traits thus confirming what was reported in other works (Álvaro et al. 2008; Mohammadi 2016; Moragues et al. 2006).

According to our results, water stress reduced grain yield per plant probably due to a reduction of its three main components (spikes per m^2, grains per spike, and grain weight) as previously reported by (García del Moral et al. 2003, 2005; Rharrabti et al. 2001; Subira et al. 2015).

Similarly, mean values for all the morphological traits decreased under stress conditions. This is in agreement with the findings by Khamssi and Najaphy (2012) and Ayed et al. (2014). Moreover, Farooq et al. (2009) and Kamrani (2015) reported that drought stress reduces leaf size and peduncle length. Recently, Selim et al. (2019) found that water stress strongly affected the sizes of photosynthetic organs.

In our work, a significant rise in GYP from old to modern varieties was revealed. This increase could be attributed to a higher number of fertile florets per spikelet in modern varieties compared to the old ones as reported by Royo et al. (2007), as a result of higher translocation rate of photo-assimilates into the spike during the pre-anthesis critical phase (Miralles et al. 2002). The yield increase in the present study is very similar to the study with durum wheat conducted by (De Vita et al. 2010; Mohammadi and Amri 2013; Rharrabti et al. 2010; Subira et al. 2015). Analogous results were also reported in studies conducted on Moroccan durum wheat by (Boussakouran et al. 2018; Rharrabti and Elhani 2014; Taghouti et al. 2018).

Table 5. Stepwise regression analysis considering stress susceptibility index (SSI) as dependent variable and flag leaf length (FLL), flag leaf area (FLA), peduncle length (PL), spike length (SL), spike area (SA), green leaf area (GLA) as independent variables of six durum wheat varieties released in different periods (old, intermediate, and modern) grown under two water regimes (irrigated and rainfed) during 2015-2016.

Traits included	Partial R^2	Model R^2
FLL	0.835**	0.835
PL	0.117*	0.952
FLA	0.045*	0.997
Final equation	SSI = 2.921 - 0.091×FLL + 0.036×FLA - 0.052×PL	

* Significant at 0.05 probability level; ** Significant at 0.01 probability level,

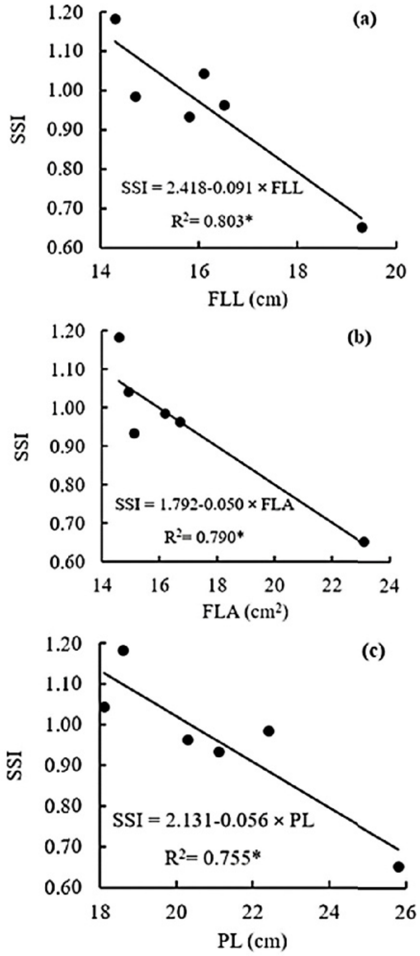

Fig. 3. Relationships between stress susceptibility index and (a) FLL (b) FLA and (c) PL. Each point represents the mean value of one variety under two water regimes.

Our results indicated that breeding during the 30 years increased the DSA and GFP. These results are in agreement with Rharrabti and Elhani (2014) who compared old and modern Moroccan durum wheat. A different scenario was observed by Motzo and Giunta (2007) and Isidro et al. (2011) who found the duration of the following phases were significantly reduced throughout the breeding process. This reduction could be a consequence of breeding modern cultivars under favorable conditions as compared to old cultivars, which attenuate the negative effect of water shortage during the grain-filling period, as usually occurs in the Mediterranean region (Royo et al. 2006). In comparison to old varieties, modern ones had shorter FLL, FLA, and PL. These results agree with those reported by Giunta et al. (2008) for durum wheat and by Foulkes et al. (2007) for bread wheat.

Concerning the relationships among studied traits, a significant positive correlation was found between GYP and FLL under rainfed conditions, but not under irrigated conditions. A number of studies showed a positive association between flag leaf size and grain yield per plant in cereals such as wheat and rice (Khaliq et al. 2008; Wang et al. 2011). In fact, flag leaf is considered to be the first source of translocated assimilates to fill the grains and determine grain yield (Ahmed et al. 2004; García et al. 2010; Sanchez-Bragado et al. 2014; Sharma et al. 2003) because to its proximity to the spike and the fact that it remains green for longer time as compared to the rest of leaves (Christopher et al. 2008; Kipp et al. 2014). In addition, in this study, grain yield per plant increased when green leaf area increased, similar results were obtained by Dalirie et al. (2010) and recently by Chen et al. (2019) who demonstrated that the area of photosynthetic organs above the flag leaf node is an important factor determining wheat grain yield.

Under irrigated conditions, GYP was positively correlated with GFP. These results agree with those reported by García del Moral et al. (2003). Also, Rharrabti et al. (2001) previously observed a strong correlation between GFP and grain weight. The positive relationship between GYP and GFP found under favorable conditions indicates that when grain maturation is prolonged, grain growth continues under conditions of high radiation and temperature, which tend to accelerate photosynthesis and translocation of assimilates to the grains (García del Moral et al. 2005).

Although the correlation coefficients were non-significant, the DSA was negatively correlated with GYP under both water regimes. These results are in agreement with those of Maccaferri et al. (2010) who compared 189 durum wheat accessions in 15 Mediterranean environments and demonstrated a negative correlation between DSA and GYP. The negative association between GYP and DSA indicates the importance of earliness under water deficit condition, it increases the likelihood to escape drought, and it improves the partitioning of the water used by the crop and transpired after anthesis (González et al. 2007; Slafer et al. 2005).

In the present study, the results of the principal component analyses (PCA) supported those of ANOVA and correlation analyses, PCA allowed a good separation between irrigated and rainfed trials. Moreover, values of all morphological traits were higher under favorable conditions. PCA analyses also showed the strongest relationship between GYP and GLA, and the negative association between GYP and DSA; this confirmed what was previously illustrated by correlation studies.

FLL was the most consistent trait entered in SSI regression model explaining 83.5% of SSI variations followed by PL (12%) and FLA (4.5%), respectively. The morphological traits of flag leaf, including length and area, and peduncle length are highly influenced by water stress conditions (Coleman et al. 2001; Villegas et al. 2007). Moreover, the peduncle is more advantageous in performing photosynthesis during later stages of grain filling as compared to the flag leaf (Kong et al. 2010). Our results suggest the suitability of FLA, FLL, and PL as proper estimators of drought tolerance. When SSI was plotted against FLL, FLA, and PL negative correlations were revealed indicating that varieties with longer flag leaf and peduncle would have more drought resistance. Similarly, Bogale et al. (2011) previously observed a negative correlation of PL with SSI. In addition, a strong relationship was found between peduncle weight and SSI in the more stressed environments (Villegas et al. 2007).

Conclusion

The results reported in the present study indicated that water regime significantly decreased grain yield per plant and all morphological traits above flag leaf. In addition, our results demonstrated that breeding during the last 30 years in Morocco increased the grain yield per plant, days from sowing to anthesis, and grain filling period. In contrast, a decrease in flag leaf length, flag leaf area, and peduncle length were observed from old to modern varieties. Moreover, varieties with longer flag leaf and peduncle would have more drought resistance. Therefore, these traits merit attention in breeding programs and could be used as selection criteria for higher grain yield under rainfed conditions.

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