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Evaluation of a Hexaploid Wheat Collection (*Triticum aestivum* L.) under Drought Stress Conditions Using Stress Tolerance Indices

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Authors' contributions

This work was carried out in collaboration among all authors. Author ADH conducted the study, performed the statistical analysis and wrote the first draft of the manuscript. Author AD designed the experiment, provided the funds and revised the manuscript. Author BH managed the analyses of the study and revised the manuscript. Author SAK assisted with the technical aspects of the experiment. All authors read and approved the final manuscript.

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ABSTRACT

Wheat is the most important crop in the world which faces the global problem of drought. Its production is affected by water deficit after pollination in arid and semi-arid regions. An experiment was conducted to assess tolerance of 39 bread wheat genotypes to end-season drought. The experimental design was Randomized Complete Block in three replications and the drought tolerance indices (SSI, STI, TOL, MP and GMP) were calculated for grain yield. The cultivar Cambin produced the highest grain yield under normal irrigation by 369.19 g m^{-2} while Arina had the highest yield (223.35 g m^{-2}) under drought stress conditions. Stress tolerance (TOL) introduced Hindukesh, Iran2355 and Iran6476 as drought tolerant genotypes. Also, results showed that grain yield under stress and non-stress environments were highly correlated with the mean productivity (MP), the geometric mean productivity (GMP) and tolerance index (TOL). These genotypes could be further used in crosses for genetic studies and breeding programs for improvement tolerance to drought.

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1. INTRODUCTION

Wheat is a strategic crop being cultivated under all environmental conditions and provides more than a quarter of the total world's cereal products. Its grains are the main sources of calories for more than 1.5 billion people (CIMMYT, 2000). However, drought stress is one of the limiting factors of wheat production in the arid and semi-arid regions. Insufficient precipitation and water deficit at various growth and development stages (vegetative, reproductive, and grain development) have negative influence on the physiological processes of wheat that subsequently affect yield [1]. The problem of drought is acute in many developing countries, where about 37% of growing areas are semi-arid and have low soil moisture content, thus presenting a limiting factor for higher yields [2]. Drought occurs in many parts of Fars Province located in southern Iran where it affects the potential yield to as low as 800 kg ha^{-1} [3].

The impact of drought stress on potential yield and its components depends on the plant growth stage. An average loss of 17% to 70% in grain has been estimated due to drought stress [4]. Response to this stress differs in various crop species and hence, drought tolerance is an important aim in breeding programs [5]. With dwindling water resources and increasing drought intensity, grain yield loss is a great concern to breeders in drought-affected areas [6]. Therefore, understanding the compensating strategies in a particular environment is key for successful breeding programs. Although drought stress affects morphological traits, grain yield-associated traits are highly used for selection of superior varieties under water deficit conditions [7]. Several statistical equations have been defined as drought indices which represent a measure of drought stress based on the reduction of grain yield under drought stress conditions in comparison to irrigated conditions [6]. These indices are able to discriminate genotypes that are productive in dry environments. Rosielle and Hamblin [8] defined stress tolerance index (TOL) as the differences in yield under stress (Ys) and nonstress (Yp) environments, and mean productivity (MP) as the average of Ys and Yp. Fischer and Maurer [9] proposed stress susceptibility index (SSI). Fernandez [10] defined the stress tolerance index (STI), which can be used to identify high-yielding genotypes under both stress and non-stress conditions. Another yield-based estimate of drought tolerance is the geometric mean (GMP) [11].

Iran is the center of wheat origin representing a rich germplasm of landraces that could harbor genes for abiotic and biotic stresses with possible use in breeding wheat. Landraces, which have arisen through a combination of natural selection and the selection performed by farmers [12], usually have a broad genetic base and can therefore provide valuable characteristics for wheat [13,14]. Tolerance to environmental stresses [15] and the resulting good yield stability are also often referred to in landraces [14]. Therefore, these genetic resources can be considered as a valuable portion of the gene pool [16,17], because they represent the broad intra-specific genetic diversity of crops, from which new cultivars could arise. Several studies have employed commercial genotypes to assess and improve wheat characteristics, but little is known regarding the drought tolerance of landraces. Therefore, the aims of this study were to screen a collection of Iranian, Afghan and Swiss wheat genotypes under rain-fed and irrigated conditions using drought indices and to identify drought-tolerant genotypes for further use in breeding programs.

2. MATERIALS AND METHODS

2.1 Plant Material and Experimental Design

Thirty-nine wheat genotypes including cultivars and landraces; were cultivated in two separate experiments (normal and stress) in a randomized complete block design (RCBD) with three replications at the research farm, School of Agriculture, Shiraz University (52° 32′ E 29° 36′ N) Iran (Table 1). The soil texture was silt loam with pH 7.0, EC 0.605 dS m^{-1}, CEC 0.512 dS m^{-1}, 0.089 % total N, 20.12 mg kg^{-1} available P, 589.82 mg kg^{-1} K, OC (0.98% at 0-15 cm soil depth) and FC 27.65%. The trial was repeated in 2014-2015; 2015-2016 and 2016-2017 growing seasons and standard agronomic practices were followed. A sample of 100 seeds was planted in each 1 m^2 plot on October 27th of each year. Sowing depth was 5-cm with 10 cm between seed space. Prior to sowing, the field was fertilized with 300 kg urea (46% N) ha^{-1} and 110 kg ha^{-1} triple superphosphate according to soil analysis results. During the growing season, 50 kg N ha^{-1} was applied at both the stem elongation and heading stages. Drought stress was applied by ceasing irrigation after the flowering stage.

2.2 Stress Index

Drought tolerance indices were calculated as mentioned in Table 2.

2.3 Statistical Analysis

The data were first subjected to combined analysis of variance using Proc GLM in SAS software V. 9.3 for estimating the variation source of traits among genotypes. Pearson's simple linear correlation coefficients and principal component analysis (PCA) were calculated on the basis of tolerant indices.

Table 1. Wheat genotypes studied for drought stress under normal and drought stress conditions in three growing seasons

Code	Genotype	Origin
1	Hindekush	Afghanistan
2	Butshak	Afghanistan
3	Kabul	Afghanistan
4	Kutschos	Afghanistan
5	Tschardeh	Afghanistan
6	Iran246	Iran
7	Iran 811	Iran
8	Iran 880	Iran
9	Iran 906	Iran
10	Iran 936	Iran
11	Iran 969	Iran
12	Iran 1307	Iran
13	Iran 2355	Iran
14	Iran 2588	Iran
15	Iran 5775	Iran
16	Iran 6476	Iran
17	Sadra-I-Safid Senzid	Afghanistan
18	Ghandozi-I-Wheat Paynda Village (population)	Afghanistan
19	Hazarjat (population)	Afghanistan
20	Lami-Surkh Paynda Village (population)	Afghanistan
21	Garma-I- Safid Bam (population)	Afghanistan
22	Sarda_I- Surkh Senzid (population)	Afghanistan
23	Lalmi-Khagi Dolana	Afghanistan
24	Sarda-I-Surkh Mamad Quli (Population)	Afghanistan
25	Lalmi surkh Dolana (population)	Afghanistan
26	Garma-I-Surkh Bam (population)	Afghanistan
27	Arina	Switzerland
28	Orzival	Switzerland
29	Forno	Switzerland
30	Camedo	Switzerland
31	Forel	Switzerland
32	Lorenzo	Switzerland
33	Cambin	Switzerland
34	Zinal	Switzerland
35	Shiraz	Iran
36	Cross Boolani	Iran
37	Havasi	Iran
38	Tonichi 81	CIMMYT
39	Pavon 76	CIMMYT

Table 2. Formula of indices used to evaluate drought tolerance in 39 wheat genotypes under water deficit in three growing seasons

Stress Tolerance Index	$\frac{Yp \times Ys}{\bar{Y}p^2}$	Fernandez [10]
Tolerance	Yp –Ys	Rosielle and Hamblin [8]
Geometric Mean Productivity	$\sqrt{(YS \times Yp)}$	Fernandez [10]
Mean Productivity	$\frac{Ys + Yp}{2}$	Rosielle and Hamblin [8]
Stress Susceptibility Index	$\frac{1 - \frac{Ys}{Yp}}{1 - \frac{\bar{Y}s}{\bar{Y}p}}$	Fischer and Maurer [9]

NB: Ys and Yp are the genotype's yield under stress and normal conditions respectively. $\bar{Y}s$ represents yield mean of all genotypes under stress conditions and $\bar{Y}p$ is yield mean under normal conditions

3. RESULTS AND DISSCUSION

3.1 Drought Tolerance Indices

Tolerance index (TOL), identified Hindukesh and Iran 6476 (with TOL values of 15.91 and 15.46 g.m^{-2} respectively) as drought tolerant and Garma-I-Sefid Bam and Cambin (with yields of 252.14 and 369.19 g m^{-2}, respectively) as drought sensitive. Rosielle and Hamblin (1981) [8] proposed that low TOL is associated with low sensitivity to stress and as a consequence, high-yielding genotypes in stress conditions are selected. This index shows difference between Yp and Ys and therefore, high TOL indicates low value of Yp or high value of Ys, and as a consequence, higher TOL means higher sensitivity to drought stress. Obviously, TOL points out the lowest yielding genotypes under normal stress conditions only.

The MP index represents relatively high and low-yielding genotypes in non-stress and stress conditions, respectively. Yet, this index is associated with STI, GMP and HM indices and cannot be appropriate *per se*. The highest and lowest MPs were recorded in Cambin and Hindukesh (301.9 and 59.11 mg^{-2}), respectively. The highest GMP was observed in Cambin (GMP =293.7 g m^{-2}). The data for GMP and STI suggested Cambin and Iran2355 could be supposed relatively drought tolerant.

The SSI divides materials to tolerant and sensitive genotypes regardless of yield potential [18]. Genotypes with SSI less than unit represent higher grain yield under drought [19]. Stress sensitive index values calculated for the tested genotypes were very close and hence, no selection could be made based on this index. Naeimi et al. [20] stated that selecting two varieties based on stress susceptibility index with equal amount, is not acceptable. Guttieri et al. [21] stated that SSIs higher and less than unit mean above-average and below-average susceptibility to drought stress, respectively. Compared with MP and TOL, SSI is a better index to select genotypes under drought stress conditions. Clarke et al. [22] used SSI to evaluate drought tolerance in wheat genotypes and found a year to year variation in SSI for genotypes and their ranking pattern.

The highest STIs were identified in Iran2355 and Iran6476 (Table 3). Since drought stress severity varies in field over seasons, genotypes do not show a stable reaction to water deficit condition [9]. Genotypes with high STI also showed high GMPs and low TOLs, whilst genotypes representing low STI presented low GMPs and MPs but high SSIs and TOLs. The STI index was used to discriminate between drought tolerant and sensitive wheat varieties in several studies [23,24,25]. The optimal selection criterion should distinguish genotypes that express uniform superiority in both stress and non-stress environments.

A two-phase screening strategy has been suggested when breeders face a large number of genotypes in breeding programs for drought tolerance [3]. First, genotypes with high STI are to be selected and then genotypes from previous stage must be screened for SSI. This method of selection leads to high-yielding genotypes well adapted to both stress and non-stress conditions [11]. In the study by Khakwani et al. [26], they screened drought-tolerant wheat varieties on the basis of higher MP, GMP, STI and low SSI which ultimately resulted in selection of Hashim-8 as drought tolerant. Likewise, Ramirez and Kelly [11] noticed that combination of SSI and GMP

Table 3. Drought stress indices and yield of thirty-nine wheat genotypes under drought stress and normal conditions

Genotype	Yp (gm^{-2})	Ys (gm^{-2})	SSI	STI	TOL (gm^{-2})	MP (gm^{2})	GMP (gm^{2})
Hindekush	67.467	51.556	1.000	0.768	15.911	59.511	58.932
Butshak	187.276	139.224	0.993	0.756	48.051	163.250	161.034
Kabul	139.937	105.751	0.990	0.763	34.186	122.844	121.146
Kutschos	178.076	137.326	0.997	0.781	40.750	157.701	155.990
Tschardeh	70.222	53.733	0.995	0.766	16.489	61.978	61.329
Iran246	163.542	124.498	1.000	0.784	39.044	144.020	142.339
Iran 811	80.644	64.078	1.000	0.800	16.567	72.361	71.832
Iran 880	204.438	167.063	0.990	0.812	37.374	185.751	184.345
Iran 906	208.214	146.869	0.996	0.675	61.346	177.542	174.446
Iran 936	183.714	91.969	0.999	0.528	91.746	137.842	128.847
Iran 969	158.611	127.628	0.989	0.812	30.983	143.119	141.635
Iran 1307	160.944	123.539	0.996	0.776	37.406	142.242	140.425
Iran 2355	169.538	150.431	0.978	0.900	19.107	159.984	159.605
Iran 2588	145.985	98.998	0.995	0.682	46.987	122.491	119.239
Iran 5775	176.969	133.669	0.998	0.723	43.300	155.319	152.682
Iran 6476	272.006	256.541	0.997	0.942	15.464	264.273	264.107
Sadra-I-Safid Senzid	295.834	212.251	0.999	0.722	83.583	254.043	250.431
Ghandozi-I-Wheat Paynda Village (population)	198.006	118.299	0.999	0.613	79.707	158.152	152.806
Hazarjat (population)	187.222	137.746	1.002	0.744	49.477	162.484	160.083
Lami-Surkh Paynda Village (population)	203.457	154.886	0.996	0.770	48.571	179.171	177.166
Garma-I- Safid Bam (population)	252.141	100.917	1.000	0.462	151.224	176.529	152.510
Sarda_I- Surkh Senzid (population)	178.476	141.29	0.986	0.803	37.186	159.883	158.620
Lalmi-Khagi Dolana	76.333	55.444	1.000	0.722	20.889	65.889	64.863
Sarda-I-Surkh Mamad Quli (Population)	157.53	99.391	0.998	0.693	58.139	128.461	124.065
Lalmi Surkh Dolana (population)	142.3	92.439	1.000	0.655	49.861	117.370	112.498
Garma-I-Surkh Bam (population)	177.537	120.421	0.997	0.682	57.116	148.979	146.004
Arina	317.164	223.359	0.999	0.704	93.805	270.261	265.995
Orzival	208.768	124.666	1.000	0.578	84.103	166.717	158.529
Forno	181.889	109.721	0.997	0.591	72.168	145.805	141.115
Camedo	180.607	127.97	1.000	0.721	52.637	154.288	151.795
Forel	180.614	117.047	0.998	0.606	63.567	148.831	142.799
Lorenzo	82.556	39.967	1.000	0.487	42.589	61.261	57.325
Cambin	369.19	234.666	1.000	0.645	134.524	301.928	293.746
Zinal	191.807	119.292	1.000	0.674	72.515	155.550	147.942
Shiraz	198.521	112.743	0.997	0.563	85.777	155.632	149.027
Cross Boolani (control)	170.072	139.024	0.997	0.814	31.048	154.548	153.286
Havasi	291.954	212.017	1.000	0.728	79.938	251.986	248.700
Tonichi 81	149.729	108.972	1.007	0.795	40.757	129.351	127.062
Pavon 76	157.106	75.557	0.996	0.489	81.549	116.331	107.167

NB: Y_P, yield of genotype under non stress conditions; Y_S, yield of genotype evaluated under stress conditions; TOL, tolerance index; MP, mean productivity; GMP, geometric mean productivity; SSI, susceptibility index; STI, stability tolerance index is more efficient for selecting common bean to improve drought tolerance. In another study, high- yielding corn hybrids were selected based on STI and GMP indices under both drought stressed and non-stressed conditions [27].

3.2 Correlation of Traits under Two Moisture Regims

The results of correlation analysis showed that Yp had direct and significant correlations with all drought tolerance indices except STI. Furthermore, GMP had a high correlation with MP (r=0.983). The lowest positive correlation was observed between SSI and Yp (r=0.05). The MP and GMP indices had high significant and direct correlations with grain yield in the normal irrigation (Yp) and stress (Ys) conditions. Cengiz and Ilhan [28] reported that the mean productivity was positively and significantly ($p< 0.01$) correlated with seed yield ($r = 0.885$) and tolerance to drought index, (STI) ($r = -0.426$) under drought stress and non-drought stress conditions for chickpea.

Ys had significant positive correlations with TOL ($r = 0.122$) which shows that genotypes with high TOL are not suitable under stress conditions because tolerant genotypes have low TOL. The TOL index was strongly correlated with STI (r=-0.795). Low TOL is desirable and selection for this parameter would tend to favor low yielding genotypes. Hence, TOL singly is not beneficial in screening drought tolerant genotypes. Positive significant correlations between STI, MP and GMP in both stress and non-stress conditions show that their effects are stronger than those of SSI and TOL [24,29]. Positive correlations between TOL, Yp and Ys show that selection based on TOL will result in increased yield under optimal conditions [30,31].

A positive significant correlation was observed between Yp and Ys ($r = 0.75$) which means that high-yielding genotypes can be selected under both stress and non-stress conditions. Our results were similar to those of Aghaei-Sarbarze et al. [3] on wheat cultivars and Nazari and Pakniyat [32] on barley cultivars since they also found positive significant correlations between Yp and Ys. It has been reported if correlation coefficient between Yp and Ys is between 0 and 0.5 under experimental conditions and genetic variance ratio is less than one [33], genotypic selection for yield might increase mean grain yield under non-stress condition.

Negative correlation was observed between Ys and SSI ($r = -0.20$) which means selection based on SSI results in yield loss under drought stress conditions. Hence, SSI is not a suitable index for screening drought-tolerant genotypes. Correlation analysis indicated that grain yield under both stress (Ys) and non-stress conditions (Yp) was correlated with STI, GMP and MP. Therefore, these indices are most appropriate in screening high-yielding wheat cultivars. Genotypes from such selection strategies could be useful not only for well-watered conditions, but also for drought-affected circumstances.

Some researchers believe that valid indices for screening have a good relation with yield in normal and stress condition [23,25]. In this context, Naroui Rad et al. [34] reported that the indices STI, GMP and MP had the highest positive correlation coefficient with yield under normal and drought stress conditions and they could be used for post anthesis water stress in sorghum. Also, Mohamadi [35] reported that correlations between the indices of STI, GMP and MP were highly significant ($P<0.01$) at all levels of drought stress and consequently, these indices could be used interchangeably for ranking genotypes. The stress susceptibility index (SSI) introduced by Fisher and Maurer [9] was significantly and negatively correlated with yield under stress and presented a positive correlation with TOL index.

Table 4. Correlation coefficients between different drought stress indices and yield under normal and drought stress conditions

	Y_P	Y_S	SSI	STI	TOL	MP
Y_S	0.750**					
SSI	0.054	-0.200				
STI	-0.199	0.388**	-0.304			
TOL	0.635**	0.122	0.299**	-0.795**		
MP	0.936**	0.901**	-0.072**	0.081	0.405**	
GMP	0.883**	0.954**	-0.114	0.193	0.311**	0.983**

*NB; **Indicates significant difference at 1% level of probability. Yp: yield under non-stress condition, Ys: yield under drought stress condition, TOL: tolerance index, MP: mean productivity, SSI: stress susceptibility index, GMP: geometric mean productivity and STI: stress tolerance index*

Table 5. Principal component loadings for the measured traits of wheat genotypes

Component	Total variation (%)	Cumulative (%)	Yp	Ys	TOL	MP	SSI	GMP	STI
PC1	59.8	59.8	0.48	0.44	0.30	0.47	0.04	0.47	-0.01
PC2	28.6	88.4	0.01	−0.25	0.43	-0.10	0.42	-0.14	-0.6

NB; Yp: yield under non-stress condition, Ys: yield under drought stress condition, TOL: tolerance index, MP: mean productivity, SSI: stress susceptibility index, GMP: geometric mean productivity and STI: stress tolerance index

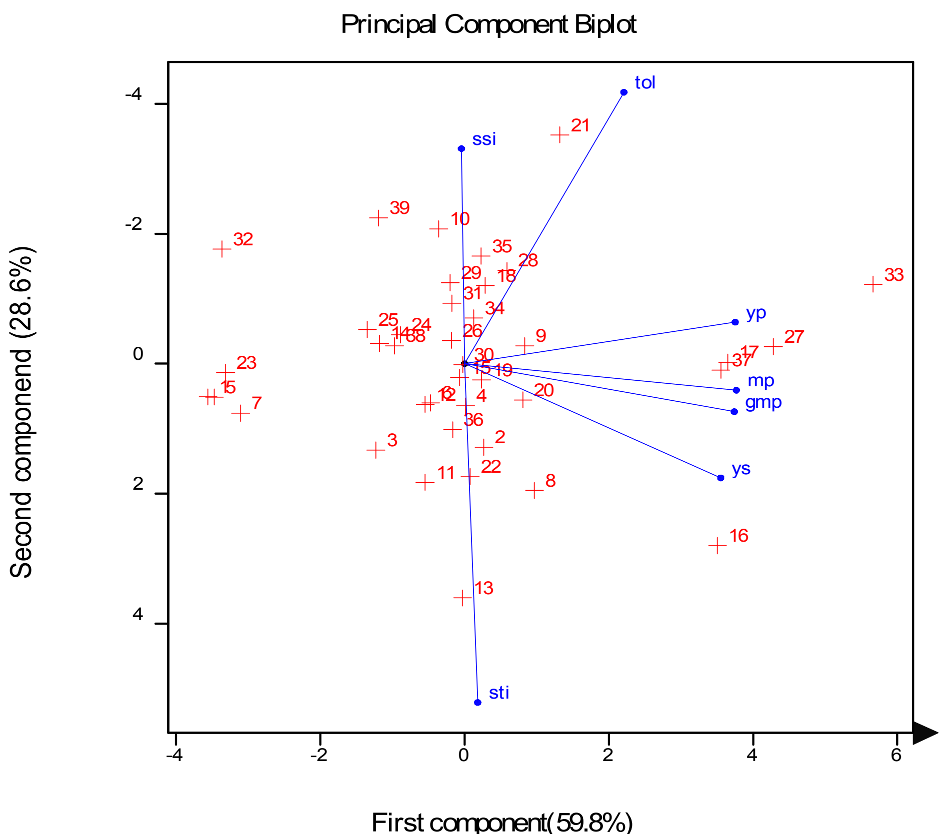

Fig. 1. Biplot for quantitative traits of wheat genotypes

NB: Yp: yield under non-stress condition, Ys: yield under stress condition and drought indices, TOL: tolerance index, MP: mean productivity, SSI: stress susceptibility index, GMP: geometric mean productivity and STI: stress tolerance index

3.3 Principal Component Analysis

Principal component analysis was performed to determine the relationship between genotypes and drought stress index.

The first PC indicated the significance of STI, MP and GMP and described 59.8% of total variation (Table 4). So, it was entitled as drought tolerance component. The second PC explained 28.6% of variations with higher importance being placed on SSI and TOL and consequently, was named as drought-sensitivity component that sieves sensitive genotypes.

These two components totally described 88.4% of drought indices variations which shows that selecting genotypes with high PC1 and low PC2 is appropriate for both conditions (Fig. 1). According to these results, genotypes number 2, 4, 8, 11, 15, 16, 17, 18, 20, 22, 36 and 37 with high PC1 and low PC2 (low sensitivity and high yield) probably perform better in both stressed and non-stressed conditions. These genotypes also presented high STI, MP and GMP. Genotypes 9, 18, 19, 21, 26, 27, 28, 30, 33, 34 and 35 with high PC1 and PC2 are suitable in non-stress conditions because they are sensitive to end- season drought.

Genotypes number 23, 1, 5, 6, 7, 12, 13, 23 and 36 with both low PC1 and PC2 had low sensitivity to stress conditions and can be used in breeding programs for drought tolerance. Genotypes number 10, 14, 24, 25, 32, 38 and 39 with low PC1 and high PC2 had low yields and were highly sensitive to end-season drought, and therefore, their cultivation cannot be not recommended (Fig. 1). Likewise, Dorostkar et al. [36] pointed out that selecting genotypes with high PC1 and low PC2 is suitable for both stress and non-stress conditions. Also, Kaya et al. [37] noticed that bread wheat genotypes with high PC1 (first genotype × environment) and lower PC2 (second genotype × environment) scores produced higher yields, whereas genotypes with higher PC2 and lower PC1 scores had low yields. Similarly, 25 accessions of meadow fescue collected from seven countries were discriminated using PCA [38].

4. CONCLUSION

Introducing drought tolerant cultivar(s) with early maturity is one of the efficient ways to minimize the effect of water deficit in combination with other water management methods. One suitable method to evaluate genotypes for their tolerance to drought is using morphological and physiological traits under both stress and non-stress conditions. In the current study, genotypes were evaluated under two irrigation regimes (stress and non-stress conditions) in three seasons and a high positive correlation was obtained between grain yield and some drought indices studied. The results showed that the genotypes Hindukesh, Iran2355 and Iran6476 had similar yields and low TOL under both normal and drought stress conditions, which indicates their stability under both conditions. In addition, we observed that mean productivity, geometric mean productivity and stress tolerance index are the best indices for selecting drought-tolerant lines. The findings of this study showed, breeders should choose the indices on the basis of stress severity in the target environment and the above-mentioned genotypes could be further used in crosses for genetic studies and breeding programs for improvement tolerance to drought in water-limited areas.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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