Assessment of Yield-based Drought Tolerance Indices and Physiological Traits for Screening Pomegranate (*Punica granatum* L.) Genotypes

Soraya Karami[1]*, Sakine Faraji[2], Tayebeh Basaki[1]*, Sedighe Ghanaei[3]

1 Department of Agriculture, Payame Noor University (PNU), Tehran, Iran

2 Crop and Horticulture Science Research Department, Markazi Agricultural and Natural Resources Research and Education Center (AREEO), Arak, Iran

3 Department of Agricultural Science, Technical and Vocational University (TVU), Tehran, Iran

ARTICLE INFO

COPYRIGHT

ABSTRACT

The present research considered identifying drought-tolerant genotypes through an index-based analysis of stress tolerance and physiological traits in pomegranates. Ten local pomegranate genotypes in Iran were evaluated for drought stress tolerance in two different irrigation regimes, i.e., normal conditions and severe drought stress (SDS). The experiment was arranged in a randomized block design with three replications and took two consecutive years to accomplish (2019-2020). Indices for stress tolerance were estimated from the fruit yield of a single tree under each irrigation regime. There were significantly positive correlations among several parameters, including Yp (fruit yield under normal conditions), Ys (fruit yield under SDS), mean productivity (MP), geometric mean productivity (GMP), stress tolerance index (STI), and proline (Pro) content. Therefore, these indices can be applied when identifying drought-tolerant genotypes under SDS and normal conditions. The Pro content was introduced as the best physiological index to assist in indirectly selecting drought-tolerant genotypes. A negative correlation occurred between Ys and stress susceptibility index (SSI). This index may assist in identifying susceptible genotypes. Based on principal component analysis (PCA), two genotypes (MTS and PSS) performed best in fruit yield and showed a high Pro content under the SDS condition.

Introduction

Pomegranate (*Punica granatum* L.) is a valuable fruit crop, not only because of its broad medicinal effects but also for its nutritional properties (Johanningsmeier and Harris, 2011). Pomegranates generate high economic value, especially in arid and semi-arid regions. However, drought stress is still a significant environmental constraint that limits plant growth, fruit yield, quality, and quantity (Faraji et al., 2020). Therefore, selecting effective criteria for screening and identifying tolerant genotypes can

*Corresponding author's email: soraya.karami@pnu.ac.ir; tbasaki2@pnu.ac.ir

be essential to alleviate the adverse effects of unusual environmental conditions such as drought.

Decreased economic yield under environmental stress conditions compared to yield under normal conditions is usually a fundamental criterion for assessing drought susceptibility in plants (Fernandez, 1992; Blum, 2011). In line with this criterion, extensive efforts have led to new variables and cost-effective, yield-based indicators under different environments to help breeders identify drought-resistant genotypes reliably. Stress tolerance index (STI) (Fernandez, 1992), yield stability index (YSI) (Bouslama and Schapaugh, 1984), stress tolerance (TOL) (Rosielle and Hamblin, 1981), mean productivity (MP) (Rosielle and Hamblin, 1981), geometric mean productivity (GMP) (Fernandez, 1992), and stress susceptibility index (SSI) (Fischer and Maurer, 1978) are examples of these selection indices.

Several indices for selecting drought-tolerant genotypes were reportedly applicable to many annual plants such as safflower, cumin, and barely (Golkar et al., 2021; Karimi Afshar et al., 2021; Barati et al., 2020). For perennial plants, Ebrahimiyan et al. (2013), Irani et al. (2015), Saeidnia et al. (2017), and Quevedo et al. (2022) evaluated perennial plants such as *Festuca arundinacea, Onobrychis viciifolia, Bromus inermis Leyss,* and *Gossypium hirsutum*. They reported STI as a superior criterion in selecting genotypes with higher levels of yield potential and stress tolerance under drought stress and normal conditions. In addition to yield indices, one of the effective strategies for genotype selection (though not very economical) and a reliable selection criterion for drought resistance is to monitor changes in various physiological activities during drought stress (Pour-Aboughadareh et al., 2017). For example, the CAT enzyme is a defensive agent against oxidative stress. It is a peroxisome compound that decomposes H_2O_2 into molecular oxygen and water (Kumar et al., 2011) and remains a candidate index when identifying dehydration tolerance in genotypes (Faraji et al., 2020). Indeed, cell membranes that mediate the effects of different environmental stresses are the central site of physiological responses (Nandhini et al., 2022).

It is worth noting that screening genotypes based on physiological indices reportedly led to valuable results in many horticultural crops under drought stress, such as *Juglans regia* L. (walnut) (Lotfi et al., 2010), *Vitis vinifera* L.; grape (Król et al., 2014), *Olea europaea*; olive (Sofo et al., 2005), *Malus domestica*; apple (Wang et al., 2012), and *Punica granatum* L.; pomegranate (Faraji et al., 2020). The potential of yield indices to assist in selecting drought-tolerant genotypes in woody horticultural crops, especially pomegranate, is poorly understood.

Since drought-tolerant genotype differentiation based on one criterion can be inconsistent, a joint analysis of variables can provide deeper and broader information about genotypes for gauging their genetic diversity. Thus, this study aimed to identify and introduce the most efficient indices for selecting high-yielding and drought-tolerant pomegranate genotypes using principal component analysis as one of the most efficient multivariate analysis techniques. The current research aimed to compare the screening potential of some physiological characteristics and indices in studying drought tolerance/susceptibility in two consecutive years.

Materials and Methods
Plant materials
Ten native and commercial pomegranate genotypes were used in this study. These genotypes were previously grouped by Faraji et al. (2020) using physiological indices that grouped the genotypes as drought-stress tolerant or susceptible (Table 1).

Field establishment
A field experiment was designed as a randomized complete block, with three replications per irrigation regime per year (normal conditions and severe drought stress: SDS). The study was conducted at the Research Orchard of the Pomegranate Research Station, Saveh, Iran (35°1′N, 50°21′E, 960 m a.s.l.) on 9-year-old pomegranate trees during two growing seasons (2019 and 2020). The soil was sandy loam (pH 7.7) with an average bulk density of 1.48 g cm^{-3}. The average annual temperature and precipitation were 20.5 °C and 151 mm, respectively. The irrigation water had electrical conductivity (EC) measuring 1.3 ds m^{-1}. The plants received water uniformly until bud-burst, and then the irrigation regimes were established based on 50 and 90% depletion levels of field capacity in the root zone (at a depth of approximately 40 cm). Under normal conditions, plants were irrigated when 50% of the available soil water was depleted from the root zone. Under severe drought stress, irrigation was performed when 90% of the total available soil water was depleted from the root zone.

Table 1. Information about the pomegranate germplasm in the present study.

Genotype	Origin
Malase Torshe Saveh (MTS)	Markazi, Iran
Post Sefide Shirin (PSS)	Markazi, Iran
Tabestaniye Torsh (TT)	Markazi, Iran
Malas Shirin Saveh (MSS)	Markazi, Iran
Post Sefide Torsh (PST)	Markazi, Iran
Post Siyah (PS)	Yazd, Iran
Agha Mohamad Ali (AMA)	Markazi, Iran
Alake Torsh (AT)	Markazi, Iran
Post Sefide Bihaste Shomal (PSBS)	Markazi, Iran
Alake Shirin (AS)	Markazi, Iran

Irrigation planning

Basins with dimensions of 1.5 × 1.5 m were prepared around the experimental genotypes and were properly leveled at the beginning of the experiment. The first irrigation for the genotypes under each treatment was done to bring the soil to field capacity by flooding irrigation. The soil moisture in the respective treatments was depleted to 50 and 90% of field capacity, and then it reached the field capacity again by applying the measured amount of water. The soil moisture characteristic curve assisted in determining the water quantity for bringing the soil moisture to field capacity in the prepared basins during each irrigation.

Then, to determine the characteristic curve of soil moisture, soil samples were taken from the prepared basins (0-40 cm). These samples were saturated with water for 24 h and then subjected to 6 atmospheric pressures, including -0.3, -0.5, -1.0, -5.0, -10.0, and 15.0 via an atmospheric pressure device. The amount of soil moisture retained in different atmospheric pressures was determined by the standard gravimetric method and was expressed as a percentage of dry weight basis (Gregorich and Carter, 2007). Based on these observations, the soil moisture characteristics curve determined the amount of water retained by the soil at different soil moisture contents, which served as a guide to calculate the amount of water that should bring the soil moisture to field capacity.

Subsequently, we calculated the amount of water required to bring the soil moisture to field capacity from 50 and 90% depletion levels according to the following equation (Singh, 2008):

Total quantity of water applied per tree (liters) = A × d

Where, A: basin area to be irrigated (m2); d: depth of irrigation water (cm).

The depth of irrigation water for each application was calculated by the following formula:

$$d = \frac{Pw \times Bd \times D}{100}$$

Where, Pw: moisture percentage to be raised based on the soil moisture characteristics curve; Bd: bulk density of the soil (1.48 g cm^{-3}); D = depth of root- zone to be moistened (40 cm).

The quantity of water applied under each treatment on the basis of the above calculation was 398.1, 495.15, and 693.22 L at 50 and 90% depletion levels of field capacity, respectively.

To supply the amount of water consumed (calculated according to the above formula) during the growing season and under each treatment, irrigation was applied with a drip irrigation system using two side pipes per tree row and four emitters (4 L h^{-1}) per tree.

Recording the yield and physiological characteristics

At fruit maturity stage, based on commercial harvesting times for each genotype (Table 1), the fruits were harvested and the fruit yield was recorded from five random plants per genotype in each plot per replication. There were fifteen trees per genotypes in each irrigation regime. For each genotype, there were 30 trees per genotype and 10 trees per replication. They were planted in a plot of 3 rows in each replication, 4 m wide and 2 m apart.

Six different selection indices, including YSI, SSI, TOL, MP, GMP, and STI, were calculated based on fruit yield under normal conditions and SDS (Table 2).

Fruit yield (Y_p and Y_s) was recorded for each genotype at the fruit maturity stage based on commercial harvesting times. For physiological assessments, leaf samples comprised 10 healthy leaves per plot from each genotype at the mid-fruit growth stage. Leaf water status was determined by estimating the RWC according to Ritchie et al. (1990). Also, the Pro content, water-

soluble carbohydrates content (WSCs), superoxide dismutase (SOD) activity, and CAT activity were measured according to Bates et al. (1973), Zhang et al. (2006), Sairam et al. (2002), and Aebi (1984), respectively. Protein content was determined using bovine serum albumin as a Bradford standard (1976).

Table 2. Drought tolerance/susceptibility indices calculated for each genotype in the present study.

Equation	Designation	Formula	Reference adapted from
Eq. 1	Tolerance	$TOL = Yp - Ys$	Rosielle and Hamblin, 1981
Eq. 2	Mean Productivity	$MP = \frac{Yp+Ys}{2}$	Rosielle and Hamblin, 1981
Eq. 3	Geometric Mean Productivity	$GMP = \sqrt{Yp \times Ys}$	Fernandez, 1992
Eq. 4	Stress Susceptibility Index	$SSI = \frac{1-\frac{Ys}{Yp}}{1-\frac{\bar{Y}s}{\bar{Y}p}}$	Fischer and Maurer, 1978
Eq. 5	Stress Tolerance Index	$STI = \frac{(Yp \times Ys)}{\bar{Y}p^2}$	Fernandez, 1992
Eq. 6	Yield Stability Index	$YSI = \frac{Ys}{Yp}$	Bouslama and Schapaugh, 1984

Ys and Yp: fruit yield of genotypes under severe drought stress and normal conditions, respectively, $\bar{Y}$s and $\bar{Y}$p: mean fruit yields of all genotypes under severe drought stress and normal conditions, respectively.

Statistical analysis
Before conducting analyses of variance (ANOVA), SAS software (9.3.1) executed a normality test of performance (SAS Institute, Cary, NC, USA). The data were then subjected to ANOVA via SAS software to determine differences among treatments and genotypes per trait. The experiment was conducted using two irrigation regimes for two years. Thus, regarding fruit yield and yield-related indices, a combined analysis of variance was performed. Regarding physiological traits, a split-plot in time (year) was used for combined analysis as described by Steel and Torrie (1980) and Nguyen and Sleper (1983). Before combining the two-year data, the Bartlett test for homogeneity in environmental variance was performed. To determine differences among treatments and genotypes for each trait, the LSD test enabled a comparison of mean values (least significant difference) ($P \leq 0.05$) (Steel and Torrie, 1980). The Statgraphics Centurion software XVI (www.statgraphics.com) enabled multivariate statistical analysis, including correlation analysis (Pearson coefficients), principal component analysis, and biplot preparation.

Results
Yield, tolerance indices, and physiological traits
The results of combined ANOVA indicated a significant variation in fruit yield (Y_p and Y_s) and tolerance indices among the genotypes ($P \leq 0.01$) (Table 3). In contrast, there was no significant interaction effect in genotype × year, regarding fruit yield and tolerance indices (Table 3). Thus, data were averaged over the two years and used for comparing mean values (data not shown).

Table 3. Combined analysis of variance for drought tolerance/susceptibility indices of 10 pomegranate genotypes tested across two irrigation regimes in 2019 and 2020.

Source of variation		Mean square of traits							
	df	Y_P	Y_S	TOL	MP	GMP	SSI	STI	YSI
Year (Y)	1	0.21^{ns}	0.47^{ns}	0.05^{ns}	0.01^{ns}	0.25^{ns}	0.001^{ns}	0.18^{ns}	0.002^{ns}
Block/Y	4	3.00	1.55	0.16	0.16	1.19	0.02	1.14	0.002
Genotype (G)	9	1057.97^{**}	548.66^{**}	70.33^{**}	70.30^{**}	363.88^{**}	1.95^{**}	382.65^{**}	0.22^{**}
G × Y	9	$.04^{ns}$	0.02^{ns}	0.01^{ns}	0.01^{ns}	0.01^{ns}	0.007^{ns}	0.01^{ns}	0.00^{ns}
Residual	36	6.07	1.97	8.80	2.20	1.65	0.07	0.004	0.008

df: Degree of freedom, Yp: Fruit yield under normal conditions, Ys: Fruit yield under severe drought stress, TOL: Stress tolerance, MP: Mean productivity, GMP: Geometric mean productivity, SSI: Stress susceptibility index, STI: Stress tolerance index, YSI: Yield stability index. [ns]: non-significant; [*] and [**], significant at 5% and 1% levels of probability.

Mean comparisons among genotypes (Table 4) showed that in the normal treatment, the highest Y_p values occurred in genotypes TT and MTS (55.21 and 52.33 kg tree^{-1}, respectively), followed by PSS (40.67 kg tree^{-1}). In this respect, the genotypes TT and MTS were not significantly different in fruit yield. However, the lowest fruit yield occurred under normal conditions in genotypes PSBS and AS (17 kg tree^{-1}). The genotype MTS also had the highest Ys (50.33 kg tree^{-1}), followed by genotype PSS (37.77 kg tree^{-1}) and TT (36.87 kg tree^{-1}).

Regarding tolerance indices, MTS and PSS genotypes had the lowest values of TOL and SSI, but the highest values of YSI. On the other hand, the highest values of STI, MP, and GMP were recorded in MTS, TT and PSS genotypes (Table 4). In contrast, TT genotype showed the highest values of relative tolerance index and stress susceptibility index, but the lowest value of yield stability index (Table 4).

Table 4. Values obtained for fruit yield and different drought tolerance/susceptibility indices in 2019 and 2020.

Genotype	Yp (kg tree^{-1})	Ys (kg tree^{-1})	TOL	MP	GMP	SSI	STI	YSI	STI
MTS	52.33[a]	50.33[a]	2.00[d]	51.33[a]	51.31[a]	0.19[c]	2.43[a]	0.96[a]	2.43[a]
PSS	40.67[b]	37.77[b]	2.90[cd]	39.21[c]	39.19[c]	0.38[c]	1.41[c]	0.92[a]	1.41[c]
TT	55.21[a]	36.87[b]	18.350[a]	46.04[b]	45.09[b]	1.74[a]	1.88[b]	0.677[c]	1.88[b]
MSS	32.61[cd]	29.47[c]	3.15[cd]	31.04[d]	30.99[d]	0.48[c]	0.89[d]	0.90[a]	0.89[d]
PST	36.43[bc]	24.67[d]	11.76[b]	30.55[d]	29.90[d]	1.64[ab]	0.82[d]	0.69[bc]	0.82[d]
PS	28.57[de]	23.70[d]	4.87[cd]	26.13[e]	26.01[e]	0.90[bc]	0.63[e]	0.82[ab]	0.63[e]
AMA	25.18[e]	22.68[d]	2.49[d]	23.93[e]	23.89[e]	0.51[c]	0.52[ef]	0.90[a]	0.52[ef]
AT	23.93[e]	16.43[e]	7.50[bc]	20.18[f]	19.82[f]	1.66[ab]	0.36[fg]	0.68[bc]	0.36[fg]
PSBS	17.00[f]	12.33[f]	4.67[cd]	14.67[g]	14.40[g]	1.39[ab]	0.19[g]	0.73[bc]	0.19[g]
AS	17.00[f]	12.33[f]	6.67[cd]	14.66[g]	14.46[g]	1.44[ab]	0.19[g]	0.72[bc]	0.19[g]

Yp: Fruit yield under normal conditions, Ys: Fruit yield under severe drought stress, TOL: Stress tolerance, MP: Mean productivity, GMP: Geometric mean productivity, SSI: Stress susceptibility index, STI: Stress tolerance index, YSI: Yield stability index. The same letter in each column indicates no significant difference among the genotypes.

Regarding physiological traits, there were significant differences (P<0.01) among irrigation regimes (E) in all of the measured physiological traits (Table 9). The effect of genotype (G) was also significant on all traits, which indicated significant variations among the genotypes. The effects of G × E were significant as well. However, the effects of the year (Y), Y × E, and G × Y were not significant on the measured traits (Table 5). Meanwhile, drought stress significantly decreased the RWC, but significantly increased the WSC and proline content, as well as CAT and SOD activities (Table 6).

Multivariate analysis of yield, indices, and physiological traits

Severe drought stress caused correlations among tolerance-related indices and yield (Table 6). Accordingly, MP, GMP, and STI correlated positively and significantly with both Y_p and Y_s. However, SSI and YSI correlated moderately with Ys (r = -0.566 and r = 0.566, respectively) and did not correlate with Y_p. In addition, a perfect correlation was established between SSI and YSI (r = -1**), indicating that these two elements behaved similarly in amount but inversely in value. Likewise, such a correlation was detected between GMP and MP (r = 1**). The TOL index showed that Ys did not correlate significantly with Y_p (Table 7).

Table 5. Combined analysis ANOVA for measured physiological traits in 10 pomegranate genotypes evaluated at two irrigation regimes (normal and severe drought stress conditions) during 2019 and 2020.

Characters	Moisture environment (E) (df = 1)	Replication/E (df = 4)	Genotype (G) (df = 9)	G × E (df = 9)	G × Replication (E) (df = 36)	Year (Y) (df = 1)	E × Y (df = 1)	G × Y (df = 9)	E × Y × G (df = 9)	Error (df = 40)
Fruit Yield	3100.52^{**}	4.63^{**}	1302.67^{**}	357.46^{**}	8.05^{**}	0.11^{ns}	0.006^{ns}	0.04^{ns}	0.016^{ns}	4.03
Proline content (Pro)	166.19^{**}	6.02^{**}	213.12^{**}	99.49^{**}	5.93^{**}	0.10^{ns}	0.001^{ns}	0.02^{ns}	0.02^{ns}	2.98
Water soluble carbohydrates (WSC)	76.27^{**}	0.30^{**}	0.84^{**}	0.79^{**}	0.43^{**}	0.0005^{ns}	0.0003^{ns}	0.0006^{ns}	0.001^{ns}	0.21
Superoxide dismutase (SOD) activity	3491.66^{**}	28.30^{**}	57.54^{**}	78.93^{**}	12.69^{**}	0.0001^{ns}	0.0003^{ns}	0.0002^{ns}	0.0001^{ns}	6.34
Catalase (CAT) activity	0.29^{**}	0.01^{**}	0.064^{**}	0.14^{**}	0.002^{**}	0.002^{ns}	0.0008^{ns}	0.0001^{ns}	0.0001^{ns}	0.002
Relative water content (RWC)	30.89^{*}	67.51^{**}	206.34^{**}	529.96^{**}	109.95^{**}	0.000^{ns}	0.001^{ns}	0.0002^{ns}	0.0005^{ns}	54.97

* significant at P<0.05; ** significant at P<0.01; ^{ns} not significant.

Table 6. Means of physiological traits of 10 pomegranate genotypes across two irrigation regimes in 2019-2020.

Moisture Environments	RWC (%)	WSC (mg ml^{-1})	CAT activity (U. min^{-1} mg^{-1} protein)	SOD activity (U. min^{-1} mg^{-1} protein)	Proline content (μmol g^{-1} FW)
Normal	82.63^{a}	10.97^{b}	0.09^{b}	10.27^{b}	9.58^{b}
Severe drought stress	69.52^{b}	12.04^{a}	0.21^{a}	19.77^{a}	11.52^{a}

Means followed by the same letter in each column are not significantly different according to the LSD test (P<0.05); RWC: relative water content, WSC: water soluble carbohydrates, CAT: catalase, SOD: superoxide dismutase.

Table 7. Coefficients of correlation between fruit yield of 10 pomegranate genotypes and the drought tolerance/susceptibility indices obtained in 2019-2020.

	Y_p	Y_s	TOL	MP	GMP	SSI	STI	YSI
YP	1							
YS	0.923^{**}	1						
TOL	0.413	0.031	1					
MP	0.982^{**}	0.979^{**}	0.235	1				
GMP	0.978^{**}	0.983^{**}	0.212	1.00^{**}	1			
SSI	-0.226	-0.566^{**}	0.755^{**}	-0.396	0.417-	1		
STI	0.959^{**}	0.973^{**}	0.189	0.985^{**}	0.985^{**}	-0.397	1	
YSI	0.226	0.566^{**}	-0.755^{**}	0.396	0.417	-1.00^{**}	0.397	1

* and ** significant at P<0.05 and P<0.01; respectively. Y_p: fruit yield under normal conditions, Y_s: fruit yield under severe drought stress, TOL: stress tolerance, MP: mean productivity, GMP: geometric mean productivity, SSI: stress susceptibility index, STI: stress tolerance index, YSI: yield stability index.

Regarding variable responses to irrigation regimes, correlation coefficients were calculated for physiological traits, yield, and SDS-related indices (Table 8). Under severe drought stress, correlation analysis (Table 8) revealed that only the proline content correlated significantly and positively with Yp, Ys, MP, GMP, STI, and CAT.

Table 8. Correlation coefficients between physiological traits, yield, and tolerance/susceptibility indices of 10 pomegranate genotypes under SDS in 2019 and 2020.

Traits	Y_p	Y_s	TOL	MP	GMP	SSI	STI	YSI
RWC	-0.255	-0.342	0.149	0.302	0.308	0.330	-0.328	-0.330
WSC	0.366	0.479	-0.185	0.428	0.435	-0.424	0.403	0.424
CAT	0.083	0.213	-0.290	0.148	0.155	-0.302	0.193	0.302
SOD	-0.176	-0.298	0.247	-0.239	-0.246	0.245	-0.259	0.245
Proline	0.786**	0.728**	-0.202	0.667**	0.674**	-0.459	0.730**	0.459

** Significant at P<0.01; Yp: fruit yield under normal conditions, Ys: fruit yield under severe drought stress, TOL: stress tolerance, MP: mean productivity, GMP: geometric mean productivity, SSI: stress susceptibility index, STI: stress tolerance index, YSI: yield stability index, RWC: relative water content, WSC: water soluble carbohydrates, CAT: catalase, SOD: superoxide dismutase.

Principal component analysis was performed on all genotypes and measured traits to show the weight of each trait (i.e., fruit yield, tolerant indices, and physiological traits) relative to the observed variations. Then, the analysis revealed interrelationships among genotypes and collinearity among the genotypes and traits. The PCA considered the mean values of traits obtained from both years and showed that the first and second principal components explained 83.85% of the total variation (Table 9). The first principal component (PC1) accounted for 70.34% of the total variation and had positive correlations with Y_S, STI, MP, GMP, Y_P, Pro, and YSI. The second component (PC2) explained 13.51% of the total variation and showed a highly positive correlation with TOL and SSI (Table 9). To classify the genotypes based on the PCs, the biplot of PC1 vs. PC2 was constructed (Fig. 1). The biplot classified the ten pomegranate genotypes into four distinct groups, comprising 1, 2, 3, and 4 genotypes. Clearly, two genotypes (MTS and PSS) formed a single group characterized by high values of Y_p, Y_s, Pro, STI, MP, and GMP, but low values of TOL and SSI indices (Group I). In contrast, the other four genotypes, namely, AS, PSBS, AT, and PST formed another distinct group characterized by higher values of TOL and SSI, but with lower values of Y_p, Y_s, and Pro (Group II). In addition, genotype TT was separated from the first group (Group I) because of its higher Y_p and TOL, thereby forming another distinct group (Group III). The fourth group (Group IV), including MSS, AMA, and PS genotypes, was characterized by moderate values of PC1 and low values of PC2. Evidently, the results showed that the pomegranate genotypes were successfully separated from each other through yield-based drought tolerance indices and physiological traits.

Table 9. Principal component analysis for fruit yield of pomegranate cultivars under non-stress (Yp) and drought stress (Ys) conditions, stress tolerance (TOL), mean productivity (MP), geometric mean productivity (GMP), stress susceptibility index (SSI), yield stability index (YSI), stress tolerance index (STI), relative water content (RWC), water soluble carbohydrates content (WSC), catalase activity (CAT), superoxide dismutase activity (SOD), and proline content (Pro).

Characteristics	**PC1**	**PC2**
Yp	0.34	0.28
Ys	0.39	0.06
TOL	-0.03	0.60
MP	0.37	0.18
GMP	0.38	0.17
SSI	-0.24	0.45
STI	0.38	0.16
YSI	0.29	-0.45
RWC	-0.14	0.05
WSC	0.19	-0.11
CAT	0.09	-0.17
SOD	-0.13	0.10
Pro	0.32	-0.05
Eigenvalue	6.44	2.70
Percent of variation	70.34	13.51
Cumulative percentage	70.34	83.85
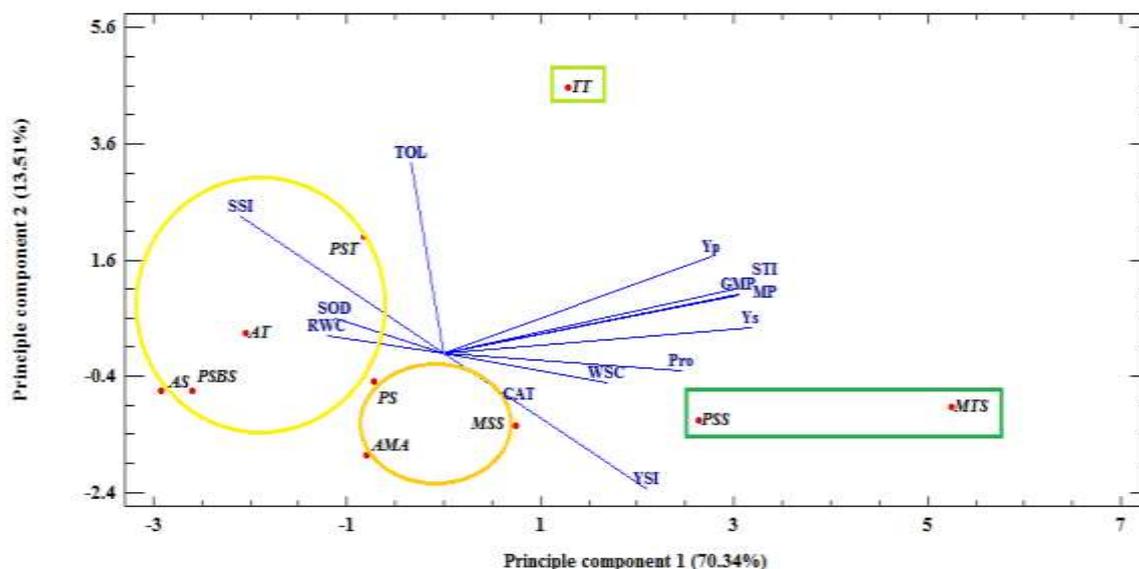
Fig. 1. Distribution of the first two principal components of physiological traits, and selection indices of 10 pomegranate genotypes under severe drought stress. Definition for abbreviations of the genotypes, traits and drought tolerance/susceptibility indices appear in Table 1 and footnotes under other tables (Table 2-8), respectively.

Discussion

Identifying effective strategies for selecting suitable genotypes is essential in breeding and genetic studies for drought tolerance. In this regard, the indicators of susceptibility and tolerance to drought were introduced as economical, efficient strategies to resolve the problems related to drought stress breeding (Vieira et al., 2016). Few studies exist in this regard, such as Faraji et al. (2020), Galindo et al. (2017), Laribi et al. (2013), and Selahvarzi et al. (2017), which dealt with the effects of drought stress on pomegranates. However, there is more potential to explore drought susceptibility and tolerance indices to identify and introduce tolerant pomegranate genotypes under both severe drought stress and normal conditions. The present study considered filling this knowledge gap in understanding drought susceptibility and tolerance indices regarding pomegranates. Several local Iranian pomegranate genotypes were evaluated for field drought tolerance during a two-year study using drought susceptibility criteria, tolerance indices, and physiological traits.

In the present investigation, a significant difference occurred among the fruit yield of the evaluated genotypes. The highest fruit yield was recorded in MTS, PSS, and TT genotypes in both irrigation regimes. Therefore, it seems that the above genotypes may be considered economical in performance when affected by drought. However, it is essential to note a significant difference between measuring drought sensitivity/tolerance in each genotype based on

the function of yield reduction under drought stress and the differential yield potential of each genotype (Blum, 2011). Thus, several indices were suggested for selecting and identifying resistant/sensitive genotypes. Therefore, in response to searching for pomegranate tolerant genotypes based on reliable indices, the present study evaluated six different selection indices (i.e., TOL, SSI, YSI, MP, GMP, and STI) to estimate fruit yield under severe drought stress.

We calculated phenotypic correlation coefficients among Y_p, Y_s, and quantitative indices for drought tolerance to find the most favorable criteria for drought tolerance. According to correlation analysis, a non-significant correlation occurred between TOL and Y_p (r = 0.413). It also appeared insignificantly between TOL and Ys (r = 0.031), thereby declaring the TOL index an inefficient TOL index to identify the most tolerant genotypes. The limited ability of the TOL index to identify drought-tolerant genotypes in some other crops was in agreement with earlier research (Golkar et al., 2021; Bahrami et al., 2014; Cabello et al., 2013; Mohammadi et al., 2011; Rizza et al., 2004). Regarding the SSI index, no significant correlation appeared between SSI and Y_p. A significantly negative correlation was between SSI and Ys (r = -0.566**), which implied that the superior genotypes recorded lower values of SSI under drought stress (Guttier et al., 2001). On the other hand, YSI did not correlate with Y_p but positively correlated with Y_s (r = 0.556**). Thus, it was considered a favorable index for selecting tolerant genotypes, mainly with lower yield potential under severe drought stress rather than in normal

conditions.

Generally, it is worth noting that indices with a high correlation in plant performance under both stressed and non-stressed conditions are introduced as the most efficient indices because they identify and separate genotypes with high performance in both environmental conditions (Golkar et al., 2021; Blum, 2018; Singh et al., 2016; Bahrami et al., 2014). Thus, the three indices, i.e., TOL, SSI, and YSI did not suitably distinguish superior and tolerant pomegranate genotypes in both conditions (normal and severe drought stress). The reason was a lack of significant correlation with Yp, Ys, or both. Similar to the TOL index, the limited ability of the SSI index to identify tolerant genotypes has also been reported in some other crops, such as wheat and potatoes (Mohammadi et al., 2011; Cabello et al., 2013).

According to the description of the most favorable indices in selecting drought-tolerant genotypes, the present study showed that fruit yield correlated positively and significantly with GMP, MP, and STI indicators in both conditions. Considering that the MTS genotype had the highest GMP, MP, and STI values, this genotype can be regarded as the most productive and stable pomegranate genotype among others under both irrigation regimes. The observed ability of the selection indices, GMP, MP, and STI, to satisfactorily identify pomegranate genotypes in both irrigation regimes is consistent with previous results on some other perennial plants. A relevant example is the case of GMP, STI, and MP in tall fescue (Ebrahymian et al., 2012), STI in smooth bromegrass (Saeidnia et al., 2017), and MP, GMP, and STI in cotton (Quevedo et al., 2022; Singh et al., 2016).

Based on the current research, when drought stress differs in terms of severity in both years and field conditions, the GMP index is more potent in separating genotypes and calculating relative yields because this index shows less sensitivity to variations in yield values under normal and drought stress conditions (Schneider et al., 1997). As in the present study, the year factor did not significantly affect the yield and selection indices. The selected indices can effectively screen the germplasm when fruit yield is sufficiently heritable. Meanwhile, in this study, the GMP and MP indices correlated significantly ($r = 1^{**}$), confirming previous results reported by Quevedo et al. (2022). Furthermore, the STI correlated with both MP and GMP indices; thus, these indices can provide similar information.

Pomegranate reacts to different drought stress levels by modifying various biochemical and physiological processes (Faraji et al., 2020).

These changes, especially physiological ones, have been introduced as the main mechanisms of plant adaptation to resist water deficits (Goharrizi et al., 2021a). However, despite limited reports on the changes in physiological traits under severe drought stress, information related to the association between physiological traits and drought tolerance indices in pomegranates is less understood. In the present study, in agreement with previous results by Faraji et al. (2020), the occurrence of drought stress caused a significant increase in the content/activity of all physiological traits (Proline, WSC, CAT, SOD), except for the relative water content (RWC).

One of the common adaptation mechanisms to overcome stress conditions such as drought stress is the accumulation of compatible solutes such as proline and WSC in plants under stress (Kohli et al., 2020; Goharrizi et al., 2021a). Regarding proline, it seems that, in mitochondria, the simultaneous occurrence of two processes can lead to an increase in proline content under drought stress. These two processes are slow oxidation through decreasing the activity of proline oxidase and increased biosynthesis by increasing the activity of glutamate pathway enzymes such as γ-glutamyl kinase (Manivannan et al., 2007). This accumulation can improve stress tolerance in plants by suppressing the catabolic pathway to supply energy for growth and survival (Pirnajmedin et al., 2015), stabilizing and protecting enzymes and membranes, pre-venting electrolyte leakage and maintaining osmotic turgor during stress conditions (Hayat et al., 2012). In agreement with our findings, some studies reported elevated proline contents during drought stress in perennial plants such as *Pistacia vara*: pistachio (Goharrizi et al., 2021b), *Juglans regia:* walnut (Sheikh Beig Goharrizi et al., 2016; Lotfi et al., 2010), and *Gossypium hirsutum*: cotton (Quevedo et al., 2022). In the case of WSC, the reduction of water potential, hydrolysis of complex carbohydrates such as starch, and growth limitation can be seen as reasons for increasing WSC content in plants affected by drought stress (Yang et al., 2007). For antioxidant enzymes such as CAT, the effect of stress on the activity of these enzymes depends on the plant species, stress type, intensity, and duration (Pirnajmedin et al., 2015). In this study, an increase in CAT and SOD activity occurred, consistent with previous results on olive trees by Sofo et al. (2005). One of the possible reasons for the increase in enzymatic activity can be abscisic acid, which responds to drought (Boroomand et al., 2018). Indeed, plants must constantly adjust abscisic acid levels to respond to physiological changes caused by different environmental

conditions (El-Tayeb, 2006). The adjusted abscisic acid level induces an up-regulation in ROS scavenging enzyme activities such as CAT and SOD, which maintain plants cells against oxidative damage (Ye et al., 2011) by increasing the expression and activity of ROS network genes (Ma et al., 2014).

The leaf RWC level can be considered one of the important indexes of water balance in plant breeding programs under stress (Vaezi et al., 2010). A decrease in leaf RWC occurs when the osmotic potential decreases due to water limitation caused by drought stress (Bybordi, 2012). A decline in osmotic potential under drought stress may be related to stomatal conditions and leaf transpiration rate to cause better absorptions of moisture from the soil and prevent water loss through the stomata (Bybordi, 2012).

Among the mentioned physiological traits, Faraji et al. (2020) reported that three physiological variables, i.e., proline content, WSC, and CAT activity, can assist in identifying preferable and suitable genotypes with high fruit yield in severe drought stress. Thus, we evaluated relationships among the mentioned physiological traits with tolerance indices. In the present evaluation, no significant correlation occurred between the physiological traits and the tolerant indices, except for the proline content. The proline content positively correlated with STI ($r=$ 0.730**), GMP ($r=$ 0.674**), and MP (0.667**). Since genotypes with higher STI/MP/GMP are introduced as higher yield genotypes in both normal conditions and under drought stress conditions, based on higher proline, the result can be higher-yielding drought-tolerant genotypes. On the other hand, the lack or presence of a perfect similarity between the correlation results of tolerant indices with physiological traits could highlight the hypothesis that each index is a potential indicator for different or specific biological reactions under drought conditions.

Principal component analysis is a successful procedure in various fields of science for a detailed and comprehensive review of the relationship among variables (Johanson and Wichern, 2007). In this study, the effects of different yield-based drought tolerance indices in each PC showed that PC1 and PC2 could explain the suitable yield potential and stress susceptibility, respectively. Thus, based on the biplot, stable genotypes possessed greater PC1 but lower PC2 values and vice versa. Therefore, two genotypes, MTS and PSS, aligned closely with the best drought tolerance indices, high PC1 values, and low PC2 values. Thus, they may be superior genotypes for breeding in normal and

severe drought stress conditions. Genotypes with low PC1 and relatively high PC2 values were susceptible genotypes with high productivity (e.g., AS, PSBS, AT, and PST). However, MS, AMA, and PS showed low productivity and high susceptibility. Genotype TT was more suitable for normal than stress environments due to its high PC2 value. Genotypes MS, AMA, and PS proved more desirable in stress conditions than in normal ones. The lower the stress sensitivity index in a genotype, the more tolerant it would be to stress conditions. Regarding genotype TT, it is worth mentioning that despite its record of high STI, GMP, and MP indices, it was unsuitable for both irrigation regimes. The highly different yield of this genotype in the two environmental conditions was one of the reasons for not selecting this genotype. The more stable a genotype is under normal and stressful conditions, the greater the yield stability of the genotype. In agreement with this finding, Akcura and Ceri (2011) indicated that oat genotypes with a high STI are usually associated with very different performances under the two conditions. In this study, the proline content correlated positively with PC1, indicating that in selected pomegranate genotypes with high PC1 and low PC2, MTS and PSS were superior in fruit yield under both experimental conditions. Thus, they are stable genotypes. However, further evaluations of genotypes using drought tolerance indices across multiple locations are necessary before confirming their stability as a precursor to developing improved pomegranate genotypes.

Conclusion

Current findings showed that the simultaneous or the sole use of drought tolerance indices, i.e., MP, GMP, STI, and specific physiological traits, i.e., proline content, can be the most effective screening criteria to identify drought-tolerant pomegranate genotypes for breeding programs. MTS and PSS were the most tolerant genotypes among the studied ones for cultivation in arid and semi-arid regions. They also would suitably serve as parents in hybridization programs. Additionally, the genotypes identified as most tolerant (MTS and PSS) and most susceptible (AS and PSBS) over the two years of this study can generate mapping populations for drought tolerance in pomegranates.

and equipment for this research.

Conflict of Interest
The authors indicate no conflict of interest for this work.

References
Aebi H. 1984. Catalase in vitro Methods in Enzymology. Academic press, New York.

Akcura M, Ceri S. 2011. Evaluation of drought tolerance indices for selection of Turkish oat (*Avena sativa* L.) landraces under various environmental conditions. Zemdirbyste-Agriculture 98, 157-166.

Bahrami F, Arzani A, Karimi V. 2014. Evaluation of yield based drought tolerance indices for screening safflower genotypes. Agronomy Journal 106(4), 1219-1224.

Barati M, Majidi MM, Safari M, Mostafavi F, Mirlohi A, Karami Z. 2020. Comparative physiological attributes of cultivated and wild relatives of barley in response to different water environments. Agronomy Journal 112(1), 36-43.

Bates L.S, Waldren RP, Teare LD. 1973. Rapid determination of free proline for water-stress studies. Plant and Soil 39, 205–207.

Blum A. 2011. Plant breeding for water-limited environments. CRC, Boca Raton, FL, pp 163–178.

Blum A. 2018. Plant Breeding for Stress Environments. CRC press, 231 pp.

Boroomand N, Sadat-Hosseini M, Moghbeli M, Farajpour M. 2018. Phytochemical components, total phenol and mineral contents and antioxidant activity of six major medicinal plants from Rayen, Iran. Natural Product Research 32, 564–567.

Bouslama M, Schapaugh WT. 1984. Stress tolerance in soybean: 1. Evaluation of three screening techniques for heat and drought tolerance. Crop Science 24, 933–937.

Bradford M.M. 1976. A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. Analytical Biochemistry 72, 248–254.

Bybordi A. 2012. Study effect of salinity on some physiologic and morphologic properties of two grape cultivars. Life Science Journal 9, 1092–1101.

Cabello R, Mendiburu FD, Monneveux P, Bonierbale M. 2013. Comparison of yield based drought tolerance indices in improved varieties, genetic stocks and landraces of potato (*Solanum tuberosum* L.). Euphytica 193, 147–156.

Ebrahimiyan M, Majidi MM, Mirlohi A, Noroozi A. 2013. Physiological traits related to drought tolerance in tall fescue. Euphytica 190, 401–414.

El-Tayeb M. 2006. Differential responses of pigments, lipid per-oxidation, organic solutes, catalase and per-oxidase activity in the leaves of two *Vicia faba* L. cultivars to drought. International Journal of Agriculture and Biology 8, 116–122.

Faraji S, Hadadinejad M, Abdossi V, Basaki T, Karami S. 2020. Screening pomegranate (*Punica granatum* L.) genotypes for drought tolerance using physiological and phytochemical characteristics. Fruits 75(3), 130-140.

Fernandez GCJ. 1992. Effective selection criteria for assessing plant stress tolerance. In: CG. Kuo, editor, Adaptation of Food Crops to Temperature and Water Stress: Proceedings of an International Symposium, Taiwan. 13–18 Aug, Asian Vegetable Res. and Dev. Ctr., Shanhua, Taiwan. p. 257–270.

Fischer RA, Maurer R. 1978. Drought resistance in spring wheat cultivars: 1. Grain yield response. Australian Journal of Agricultural Research 29, 897–912.

Galindo A, Calín-Sánchez A, Griñán I, Rodríguez P, Cruz Z, Girón I, Carbonell-Barrachina A. 2017. Water stress at the end of the pomegranate fruit ripening stage produces earlier harvest and improves fruit quality. Scientia Horticulturae 226, 68–74.

Goharrizi KJ, Hamblin MR, Karami S, Nazari M. 2021a. Physiological, biochemical, and metabolic responses of abiotic plant stress: salinity and drought. Turkish Journal of Botany 45(7), 623-642.

Goharrizi KJ, Meru G, Kermani SG, Heidarinezhad A, Salehi F. 2021b. Short-term cold stress affects physiological and biochemical traits of pistachio rootstocks. South African Journal of Botany 141, 90-98.

Golkar P, Hamzeh E, Maibody SAMM. 2021. Discrimination of drought tolerance in a worldwide collection of safflower (*Carthamus tinctorius* L.) genotypes based on selection indices. Acta Agriculturae Slovenica 117, 1-11.

Gregorich EG, Carter MR. 2007. Soil Sampling and Methods of Analysis. CRC Press, 76 pp.

Guttieri MJ, Stark JC, Brien K, Souza E. 2001. Relative sensitivity of spring wheat grain yield and quality parameters to moisture deficit. Crop Science 41, 327-335.

Hayat S, Hayat Q, Alyemeni MN, Wani AS, Pichtel J, Ahmad A. 2012. Role of proline under changing environments: a review. Plant Signal Behavior 7, 1456–1466.

Irani S, Majidi MM, Mirlohi A, Zargar M, Karami M. 2015. Assessment of drought tolerance in sainfoin: physiological and drought tolerance indices. Agronomy Journal 107, 1771–1781.

Johanningsmeier SD, Harris GK. 2011. Pomegranate as a functional food and nutraceutical source. Annual Review of Food Science and Technology 2, 181–201.

Johanson RA, Wichern DW. 2007. Applied Multivariate Statistical Analysis. Prentice Hall Inter. Inc., New Jersey.

Karimi Afshar A, Baghizadeh A, Mohammadi-Nejad G. 2021. Screening of Iranian cumin (*Cuminum cyminum* L.) ecotypes under normal moisture and drought

conditions using tolerance indices. Journal of Ethno-Pharmaceutical Products 2, 17-22.

Kohli SK, Gautum V, Bali S, Kaur P, Sharma A, Bakshi P, Bhardwaj R. 2020. Emerging trends of proline metabolism in abiotic stress management. In: Improving Abiotic Stress Tolerance in Plants. PP. 177-202.

Król A, Amarowicz R, Weidner S. 2014. Changes in the composition of phenolic compounds and antioxidant properties of grapevine roots and leaves (*Vitis vinifera* L.) under continuous of long-term drought stress. Acta Physiologiae Plantarum 36, 1491–1499.

Kumar RR, Karajol K, Naik GR. 2011. Effect of polyethylene glycol induced water stress on physiological and biochemical responses in pigeonpea (*Cajanus cajan* L. Millsp.). Recent Research in Science and Technology 3(1), 148-152.

Laribi A, Palou L, Intrigliolo DS, Nortes P, Rojas-Argudo C, Taberner V, Pérez-Gago MB. 2013. Effect of sustained and regulated deficit irrigation on fruit quality of pomegranate cv. 'Mollar de Elche' at harvest and during cold storage. Agricultural Water Management 125, 61–70.

Lotfi N, Vahdati K, Kholdebarin B, Amiri R. 2010. Soluble sugars and proline accumulation play a role as effective indices for drought tolerance screening in Persian walnut (*Juglans regia* L.) during germination. Fruits 65, 97–112.

Ma D, Sun D, Wang C, Li Y, Guo T. 2014. Expression of flavonoid biosynthesis genes and accumulation of flavonoid in wheat leaves in response to drought stress. Plant Physiology and Biochemistry 80, 60– 66.

Manivannan P, Jaleel CA, Sankar B, Kishorekumar A, Somasunda-Ram R, Lakshmanan GA, Panneerselvam R. 2007. Growth, biochemical modifications and proline metabolism in *Helianthus annuus* L. as induced by drought stress. Colloids and Surfaces B: Biointerfaces 59, 141–149.

Mohammadi M, Karimizadeh R, Abdipour M. 2011. Evaluation of drought tolerance in bread wheat genotypes under dryland and supplemental irrigation conditions. Australian Journal of Crop Science 5, 487–493.

Nandhini K, Saraswathi R, Premalatha N. 2022. Identification of drought tolerant entries based on stress tolerant indices and physiological traits in RIL population of cotton (*Gossypium hirsutum*). Crop Design 1(2), 1-7.

Nguyen HT, Sleper DA. 1983. Theory and application of half-sib matings in forage grass breeding. Theoretical and Applied Genetics 64, 187–196.

Pirnajmedin F, Majidi MM, Gheysari M. 2015. Root and physiological characteristics associated with drought tolerance in Iranian tall fescue. Euphytica 202, 141–155.

Pour-Aboughadareh A, Ahmadi A, Mehrabi J, Etminan A, Moghaddam M, Siddique KHM. 2017. Physiological

responses to drought stress in wild relatives of wheat: implications for wheat improvement. Acta Physiologiae Plantarum 39, 106–115.

Quevedo YM, Moreno LP, Barragan E. 2022. Predictive models of drought tolerance indices based on physiological, morphological and biochemical markers for the selection of cotton (*Gossypium hirsutum* L.) varieties. Journal of Integrative Agriculture 21(5), 1310-1320.

Ritchie SW, Nguyen HT, Holaday AS. 1990. Leaf water content and gas-exchange parameters of two wheat genotypes differing in drought resistance. Crop Science 30, 105–111.

Rizza F, Badeckb FW, Cattivellia L, Lidestric O, Di Fonzoc N, Stancaa AM. 2004. Use of a water stress index to identify barley genotypes adapted to rainfed and irrigated conditions. Crop Science 44, 2127-2137.

Rosielle AA, Hamblin J. 1981. Theoretical aspects of selection for yield in stress and non-stress environment. Crop Science 21, 943–946.

Saeidnia F, Majidi MM, Mirlohi A, Soltan S. 2017. Physiological and tolerance indices useful for drought tolerance selection in smooth bromegrass. Crop Science 57(1), 282-289.

Sairam RK, Rao KV, Srivastava G. 2002. Differential response of wheat genotypes to long term salinity stress in relation to oxidative stress, antioxidant activity and osmolyte concentration. Plant Science 163, 1037–1046.

Schneider KA, Rosales R, Ibarra F, Cazares B, Acosta JA, Ramirez P, Wassimi N, Kelly JD. 1997. Improving common bean performance under drought stress. Crop Science 37, 43–50.

Selahvarzi Y, Zamani Z, Fatahi R, Talaei AR. 2017. Effect of deficit irrigation on flowering and fruit properties of pomegranate (*Punica granatum* cv. Shahvar). Agricultural Water Management 192, 189-197.

Sheikh Beig Goharrizi M, Dejahang A, Tohidfar M, Izadi Darbandi A, Carrillo NJ, Hajirezaei M, Vahdati K. 2016. Agrobacterium mediated transformation of somatic embryos of Persian walnut using *fld* gene for osmotic stress tolerance. Journal of Agricultural Science and Technology 18, 423–435.

Singh C, Kumar V, Prasad I, Patil VR, Rajkumar BK. 2016. Response of upland cotton (*G. hirsutum* L.) genotypes to drought stress using drought tolerance indices. Journal of Crop Science and Biotechnology 19(1), 53-59.

Singh G. 2008. Effect of irrigation, foliar fertilization and plant bio-regulators on growth, yield and quality of pomegranate cv. G-137. Ph.D. thesis, Nauni, Solan: Dr Y.S. Parmar University of Horticulture and Forestry.

Sofo A, Dichio B, Xiloyannis C, Masia A. 2005. Antioxidant defences in olive trees during drought stress: changes in activity of some antioxidant enzymes. Functional Plant Biology 32, 45–53.

Steel RGD, Torrie JG. 1980. Principles and Procedures of

Statistics. McGraw-Hill: New York.

Vaezi B, Bavei V, Shiran B. 2010. Screening of barley genotypes for drought tolerance by agro-physiological traits in field condition. African Journal of Agricultural Research 5, 881–892.

Vieira RA, Rocha R, Scapim CA, Amaral AT, Vivas M. 2016. Selection index based on the relative importance of traits and possibilities in breeding popcorn. Genetics and Molecular Research 15, 1-10.

Wang S, Liang D, Li C, Hao Y, Ma F, Shu H. 2012. Influence of drought stress on the cellular ultrastructure and antioxidant system in leaves of drought-tolerant and drought-sensitive apple rootstocks. Plant Physiology and Biochemistry 51, 81–89.

Yang DL, Jing RL, Chang XP, Li W. 2007. Identification of quantitative trait loci and environmental interactions for accumulation and remobilization of water-soluble carbohydrates in wheat (*Triticum aestivum* L.) stems. Genetics 176, 571–584.

Ye N, Zhu G, Liu Y, Li Y, Zhang J. 2011. ABA controls H2O2 accumulation through the induction of OsCATB in rice leaves under water stress. Plant and Cell Physiology 52, 689–698.

Zhang A, Jiang M, Zhang J, Tan M, Hu X. 2006. Mitogen-activated protein kinase is involved in abscisic acid-induced antioxidant defense and acts downstream of reactive oxygen species production in leaves of maize plants. Plant Physiology 141, 475–487.