## **EVALUATION OF DROUGHT STRESS TOLERANCE IN SOME FABA BEAN GENOTYPES USING DROUGHT TOLERANCE INDICES**

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#### ABSTRACT

To evaluate drought tolerance in faba bean genotypes, 21 genotypes were studied in a randomized complete block design with three replications under normal irrigation and drought stress conditions. The experiment was conducted in 2017-2018 at research station of Gorgan, Iran. Indices such as stress tolerance index (STI), stress susceptibility index (SSI), tolerance (TOL), mean productivity (MP), geometric mean productivity (GMP), yield index (YI) and yield sustainability index (YSI) were calculated for all genotypes according to grain yield in normal irrigation and stress conditions. Based on analysis of variance results, there was a significant difference among the genotypes in terms of grain yield in both environments (normal irrigation and drought stress). The results of correlation showed that STI, MP, GMP and TOL are suitable for selection of high-yielding genotypes in both environments, among which STI and MP were the most suitable indices to select drought-tolerant genotypes. Based on STI, MP and GMP indices, GF-65 genotype was classified drought-tolerant, and GF-525 and GF-21 were susceptible according to TOL and SSI indices. Results of cluster analysis showed that GF-66, GF-62, GF-75 and GF-67 were superior compared to other genotypes based on YSI, YI, GMP, MP and STI indices, and utilization of these genotypes may be recommended to achieve high yields in drought stress conditions.

Keywords: drought stress, genotype, legumes, tolerance index, yield.

#### **INTRODUCTION**

Faba bean is a major legume used widely all over the world as a rich source of protein, cellulose and minerals in developing and developed countries for human and livestock, respectively (Haciseferogullari et al., 2003). Containing 20-38% protein gives faba bean a high nutritional value like the rest of legumes (Jelenic et al., 2000).

Drought is considered an important environmental stresses limiting crop production in the world, and adversely affects plant growth and development (Khan et al., 2010; Akram et al., 2013; Lum et al., 2014).

Extent of yield under stress condition is not a suitable indicator to study stress-tolerance of genotypes, whereas yield sustainability and comparison of yield under stress and non-stress conditions may be more suitable indices to investigate the response of genotypes to drought stress. The faba bean (Vicia faba L.) crop often experiences drought during its growth and development such that soil moisture deficits constrain its production (Khan et al., 2010). Hence, various methods have been utilized by researchers to evaluate crop response to environmental stresses and screening of tolerant and susceptible genotypes such as susceptibility index (SSI) (Fisher and Maurer, 1978), tolerance index (TOL) and mean productivity (MP) (Rosielle and Hamblin, 1981), stress tolerance index (STI) and geometric mean productivity (GMP) (Fernandez, 1992).

Genotypes may be categorized in 4 groups based on their response to stress and non-stress conditions: 1 - genotypes producing high yields under both conditions (Group 1); 2 - genotypes producing high yields only under non-stress conditions (Groups 2); 3 - genotypes producing higher yields under stress conditions. These genotypes are susceptible to optimal

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environmental conditions and produce lower yields due to lodging, high vegetative growth or being infested with pathogens and deceases (Group 3); 4 - genotypes which have low yields under optimal and stress conditions (Group 4) (Fisher and Wood, 1976).

Utilization of stress tolerance indices in chickpea showed that MP, GMP, STI and MH were the most suitable indices to screen chickpea lines in terms of drought tolerance (Farshadfar et al., 2011). Indices of GMP, STI and MP had the highest correlation with grain yield in wheat genotypes under stress and normal conditions and identifying group 1 genotypes was possible using these indices (Golparvar, 2000). Naderi (2000) reported that STI and GMP are more important compared to other indices for selection of high yielding and stress-tolerant genotypes. In a study, STI, GMP and MP had high correlations with yield under stress and non-stress conditions, and were identified as the best indices for selection of drought-tolerant soybean genotypes (Zeinaly Khanghah et al., 2004). In another report, the effect of normal irrigation and drought stress was investigated in maize hybrids. Drought stress resulted in lower grain yield and 1000 grain weight compared to normal irrigation. Among maize hybrids, Single-crosses 704, KLM and 700 were identified as tolerant, semi-tolerant and susceptible to drought stress. Single-cross 704 had the highest STI as well as grain yield and 1000 grain weight under drought stress conditions. Thus, STI may be used for identification of stress-tolerant genotypes (Nasrollahzadeh Asl et al., 2017). Faba bean is an important crop in Golestan province, Iran, so the following study was conducted to evaluate the response of faba bean genotypes to drought stress using grain yield and drought tolerance indices.

#### MATERIAL AND METHODS

This study was conducted in 2017-2018 at agricultural research station of Gorgan, Iran. The station is located in 5 km from north of Gorgan (36°54' N, 54°25' E). Altitude from sea level is 5 m, average annual temperature and rainfall are 17°C and 450 mm, respectively. In this investigation, 21 faba bean genotypes were evaluated and compared under drought stress and normal conditions. Yield and yield components of faba bean genotypes were investigated under two mentioned conditions. These genotypes were sown in a randomized complete blocks design with three replications with 65 and 10 cm distances between and on the rows, respectively. The events were recorded at various growth stages.

In normal irrigation treatment, the plots were irrigated when necessary based on climatic conditions and common local practices (before flowering, start of flowering and grain filling period) and the plant was not exposed to drought stress, whereas no irrigation was performed in drought stress treatment. Metal bars with 1.5 m height were placed around the plots associated with drought stress treatment to cover the plots with plastic in case of rainfall. The plastic cover was removed in non-rainy days (similar to greenhouse, but performed in the open air).

The following drought tolerance indices were investigating for evaluation of genotypes (Table 1).

Table 1. Drought tolerance indices used for evaluation of the reaction of Faba bean cultivars to water stress conditions

Drought tolerance indices	Equation	Reference		
Tolerance index	TOL = YP - YS	Rosielle and Hamblin (1981)		
Mean productivity	MP = YP + Ys/2	Rosielle and Hamblin (1981)		
Geometric mean productivity	$GMP = \sqrt{Yp \times YS}$	Fernandez (1992)		
Yield stability index	YSI = YS / YP	Bouslama and Schapaugh (1984)		
Yield index	$YI = YS / \ddot{Y}S$	Gavuzzi et al. (1997)		
Stress tolerance index	$STI = (Y_p \times Y_s) / \ddot{Y}_p^2$	Fernandez (1992)		
Stress susceptibility index	$SSI = [1 - (Ys / Yp)] / [1(\ddot{Y}s / \ddot{Y}p)]$	Fisher and Maurer (1978)		

Yp and Ys = mean grain yield/plant of genotype under non-stress and water stress conditions, respectively; YP and YS = grain yield/plant of all genotypes under non-stress and water stress conditions, respectively. Combined analysis of the data in year was performed using SAS software and Excel software was utilized to draw graphs.

#### **RESULTS AND DISCUSSION**

The results of combined analysis of variance demonstrated that there was a significant difference between genotypes for grain yield (under non-stress and water stress conditions), 100 grain weight, STI, SSI, MP, GMP, YI and YSI (Table 2).

# Grain yield under non-stress and water stress conditions

The highest grain yield (GF-65: 6410.3 kgha<sup>-1</sup>) was obtained in non-stress condition whereas minimum grain yield (GF-21: 1402.7 kgha<sup>-1</sup>) obtained in water stress condition (Table 3). High phenotypic variation was observed for grain yield occurred under non-stress and water stress conditions in both years. Water stress conditions reduced mean grain yield by 59% in average. There was a no significant relationship between grain yield under well-watered and drought stress conditions  $(r = 0.31^{ns})$  (Table 5). The majority of lines performed well only under favorable condition (Table 3), few lines showed high grain yield under both conditions (GF-65, GF-66 and GF-67),

#### **Stress Tolerance Index**

Since the highest and lowest yield under normal irrigation and drought stress conditions was not associated with only one genotype, calculation of stress susceptibility and tolerance indices (SSI and STI) was necessary to evaluate and identify superior genotypes. Significant difference among genotypes indicates genetic diversity and thus, possibility of selection for drought tolerance and cross breeding in genetic studies (Babaei et al., 2017).

Higher values for STI indicate higher drought tolerance. Among faba bean genotypes, GF-65 with 0.73 and GF-67, GF-66, GF-62 and GF-72 with respectively 0.57, 0.56, 0.55 and 0.50 had the highest STI indices, whereas the lowest STI was related to GF-293, GF-525, GF-296 and GF-21 with 0.28, 0.28, 0.27 and 0.29 STI values, respectively (Table 3). Also, these genotypes had lower yields under drought stress. According to Blum (1988), the best index should have a significant correlation with grain yield under both normal irrigation and drought stress conditions. Thus, STI may be utilized for identification of stress-tolerant genotypes due to high correlation of this index with grain yield under drought stress condition  $(r = 0.95^{**})$  and normal irrigation  $(r = 0.57^{**})$  (Table 5).

Shibanirad et al. (2015) confirmed the suitability of STI in selection of drought-tolerant wheat genotypes. Various researchers also have suggested STI as the most suitable index (Bokaie et al., 2008; Peighambri et al., 2017). High values for STI denote higher tolerance to stress as well as higher yield potential (Naser Gadimi et al., 2017).

Source of variance	d.f	Yn	Ys	100 grain weight	STI	SSI	MP	GMP	YI	YSI	TOL
Block	2	1106995.0 <sup>ns</sup>	13377.46 <sup>ns</sup>	63.8 <sup>ns</sup>	0.004 <sup>ns</sup>	0.036 <sup>ns</sup>	996674.40*	102263.02 <sup>ns</sup>	0.003 <sup>ns</sup>	0.012 <sup>ns</sup>	1347702.38 <sup>ns</sup>
Genotypes	20	699426.51**	846957.30**	773.88**	0.044**	0.090**	1151459.92**	665630.45**	0.196**	0.03**	1065795.65 <sup>ns</sup>
Error	40	314257.39	145635.13	69.87	0.003	0.035	256020.60	65416.74	0.033	0.011	649183.6
Coefficient of variation (CV)	-	10.86	18.35	6.8	15.76	19.26	8.16	7.89	18.35	25.72	26.14
Coefficient of determination (R <sup>2</sup> )	-	56.31	74.44	84.80	85.33	56.84	70.95	83.78	74.44	59.10	48.04

Table 2. Analysis of variance (Mean squares) for stress tolerance index of studied Faba bean genotyps

Yn and Ys: Grain yield under non-stress and water stress conditions, respectively, STI: Stress tolerance index, SSI: stress susceptibility index, MP: mean productivity, GMP: geometric mean productivity, YI: yield index, YSI: yield sustainability index, TOL: tolerance index. ns, \* and \*\*: non-significant and significant at p<0.05 and p<0.01, respectively.

#### Stress susceptibility index SSI

Among the studied genotypes (average of 2 years), GF-525 and GF-21 were the most drought stress-susceptible genotypes with SSI values of 1.24 and 1.25, respectively. The genotypes with value of SSI more than 1 (GF-398, GF-335, GF-293, GF-294, GF-523, GF-520 and GF-296) were categorized as stress-susceptible (Table 3). Negative relationship between drought-susceptibility and grain yield under drought stress conditions confirms that there is negative and significant correlation (r =  $-0.93^{**}$ ) between SSI and grain yield, so that the more a genotypes is susceptible to stress, the lower becomes its grain yield.

In accordance with the current study, it has been reported that lower SSI values lead to higher drought-tolerance (Shibanirad et al., 2015). Linford and NMS3129 soybean cultivars had lower SSI values and were identified drought-tolerant (Zeinaly Khanghah et al., 2004). Babaei et al. (2007) also investigated Stress susceptibility index in wheat genotypes. In general, cultivars possessing SSI values higher than 1 are regarded susceptible. Selection by this index may lead to identification of genotypes attributed with relatively low and high yields under normal and drought stress conditions, respectively (Fernandez, 1992; Fernandez and Reynolds, 2000). Atrak and Chamran wheat cultivars had the highest drought susceptibility with SSI values of 1.22 and 1.27, respectively (Babaei et al., 2007).

	Grain yield under	Grain yield under	100 grain		SSI	
Genotype	non-stress conditions	drought stress conditions	weight	STI		
	(kgha <sup>-1</sup> )	(kgha <sup>-1</sup> )	(gr)			
GF-67	4876.7cde	3193.8a	133.77bcd	0.57b	0.53e	
GF-66	5873.8ab	2734.4ab	121.60defg	0.56b	0.80de	
GF-75	5072.3bcde	2708.9ab	106.42hij	0.50bc	0.76de	
GF-72	4723.2cde	2313.1bc	104.94ij	0.40cd	0.85bcd	
GF-65	6410.3a	3136.3ab	113.24ghij	0.73a	0.817cde	
GF-62	5573.2abc	2754.2ab	111.78ghij	0.55b	0.815cde	
GF-61	5011.4bcde	2185.4bcd	106.47hij	0.39d	0.91bcd	
GF-398	5052.0bcde	1954.7cde	121.44defg	0.35de	1.0abcd	
GF-411	4618.6de	1921.4cde	101.73j	0.32de	0.96abcd	
GF-401	4251.4e	1871.3cde	99.46j	0.307de	0.92bcd	
GF-335	4954.9bcde	1812.2cde	116.45fghi	0.33de	1.04abcd	
GF-293	5057.5bcde	1537.7e	127.69cdef	0.28e	1.15ab	
GF-294	5548.6bcd	1880.9cde	130.71bcde	0.36de	1.08abc	
GF-290	5121.5bcde	1935.1cde	118.81efgh	0.36de	1.03abcd	
GF-292	5411.6bcd	1655.6de	133.86bcd	0.32de	1.15ab	
GF-523	5282.7bcd	1940.9cde	141.14ab	0.37de	1.05abcd	
GF-524	4647.5de	1847.7cde	142.24ab	0.31de	0.98abcd	
GF-525	5466.2bcd	1407.9e	142.10ab	0.28e	1.24a	
GF-520	5176.1bcde	1841.1cde	142.31ab	0.35de	1.07abc	
GF-296	4713.7cde	1618.0de	107.11hij	0.27e	1.09abc	
GF-21	5626.4abc	1402.7e	154.44a	0.29e	1.25a	
LSD	925.08	629.45	13.79	0.102	0.31	

Table 3. Mean comparison for faba bean genotypes for grain yield (average of 2 years), STI and SSI

<sup>†</sup> Means having common letters in each column have no significant difference at p<0.05 based on LSD test.

Although the difference among genotypes regarding tolerance index (TOL) was not significance, high yielding genotypes had proper tolerances. According to Table 4, genotypes GF-67, GF-75, GF-72 and GF-401 had the highest tolerance (lowest TOL) and highest yield under drought stress conditions,

whereas the lowest tolerance (highest TOL) was associated with GF-21 and GF-25 which were identified as the most susceptible genotypes. These genotypes produced the lowest yield under drought stress conditions which confirms their susceptibility to drought.

Genotypes GF-65 followed by GF-67 and GF-66 were considered as the most tolerant genotypes due to possessing the highest GMP, whereas GF-293, GF-525 and GF-296 were among the most susceptible genotypes (Table 4). Yield of the latter biotypes reduced severely under drought stress conditions so that they produced the lowest grain yield compared to other faba bean genotypes. Genotypes possessing high values of MP, GMP and STI produced higher yields as well. Noting the results of correlation and existence of a strong positive relationship between these indices and grain yield, it seems that there is a relationship among these indices which may lead to identification of stress-tolerant genotypes. This relationship has been reported by various researchers (Rosielle and Hamblin, 1981; Fernandez, 1992; Naderi, 2000; Babaei et al., 2007; Khan et al., 2010).

To choose proper drought tolerance indices, correlation between the indices and vield was calculated under normal irrigation and drought stress conditions (Table 4). According to results, correlation between grain yield under normal irrigation and drought stress was not significant, which was in accordance with the results of Babaei et al, (2007). Grain yield under normal irrigation had positive and significant correlation with STI, MP, GMP and TOL. On the other hand, there was a strong positive and significant relationship between grain yield under drought stress conditions with STI, MP, GMP, YI and YSI, whereas correlation between grain yield under drought stress conditions and SSI and TOL was negative and significant. Negativity indicates inverse relationship among these indices and grain yield. In other words, higher values of SSI and TOL indicate lower grain yield and more stress-susceptibility. According to Table 3, the lowest grain yield under stress conditions was obtained in GF-525 and GF-21 with values of 1407 and 1402.7 respectively.

These genotypes also had the highest TOL (4059.2 and 4222.6). Babaei et al. (2007) also reported that SSI and TOL had a negative and significant correlation. Researchers suggest that indices which possess high correlation with vield under stress and non-stress conditions are the best indices to identify drought-tolerant genotypes. Thus, STI, MP, GMP and TOL may be used for screening susceptible and tolerant faba bean genotypes under both conditions (normal irrigation and stress condition) due to high correlation with grain yield. Significant difference among genotypes regarding tolerance indices and grain yield indicates genetic diversity and possibility of selection for drought-tolerance cross-breeding in plant breeding and studies. In a study, STI, MP and GMP were identified as the most suitable indices for selection of stress-tolerant safflower (Carthamus tinctorius L.). Higher values for this indices indicate more sustainability and drought-tolerance (Mohammad, 2019).

In this study, there was a high correlation between grain yield under stress-conditions and YSI and YI, whereas no significant correlation was observed between these indices and grain yield under non-stress conditions (Table 5). Thus, these indices select genotypes which have high and low yields under stress and non-stress conditions, respectively. The lower the YSI and YI value, the higher the drought tolerance. According to YSI, GF-21 and GF-525 had the lowest yield. Hence, YI and YSI indices categorize the genotypes according to yield under stress conditions, and genotypes which possess higher YSI should result in high yields under both stress and non-stress conditions. However, this was not the case in some studies and genotypes having higher YSI indices produced the lowest and highest vields under stress and non-stress conditions, respectively (Sio-Se Mardeh et al., 2006). The same results were reported by Shibanirad et al. (2015).

#### ROMANIAN AGRICULTURAL RESEARCH

Genotype	MP	GMP	YSI	YI	TOL
GF-67	6473.6bcd	3919.3b	0.67a	1.53a	1682.8e
GF-66	7241.0ab	3928.3b	0.48bc	1.31ab	3139.4abcd
GF-75	6426.7bcd	3684.1bc	0.54ab	1.30ab	2363.4de
GF-72	5879.7defg	3296.2cd	0.49abc	1.11bc	2410.1de
GF-65	7978.4a	4440.4a	0.51abc	1.50a	3274.1abcd
GF-62	6950.3bc	3908.0b	0.49abc	1.32abc	2819.1bcde
GF-61	6104.1def	3298.0cd	0.44bcd	1.05bcd	2826.0bcde
GF-398	6029.4def	3138.0def	0.388bcdef	0.94cde	3097.4abcd
GF-411	5579.3efg	2946.2def	0.42bcde	0.92cde	2697.2cde
GF-401	5187.1g	2817.3ef	0.44bcd	0.90cde	2380.0de
GF-335	5861.0fg	2975.6def	0.37cdef	0.87cde	3142.7abcd
GF-293	5826.3defg	2769.6f	0.30def	0.73e	3519.9abcd
GF-294	6389.0cde	3199.9de	0.34cdef	0.90cde	3567.6abcd
GF-290	6089.1def	3144.4def	0.37bcdef	0.93cde	3186.5abcd
GF-292	6239.5cdef	2989.0def	0.30def	0.79de	3756.0abc
GF-523	6253.2cdef	3200.1de	0.36cdef	0.93cde	3341.9abcd
GF-524	5571.3efg	2925.4def	0.40bcdef	0.88cde	2799.9bcde
GF-525	6169.7cdef	2765.9f	0.25ef	0.67e	4059.2ab
GF-520	6096.7def	3084.2def	0.35cdef	0.88cde	3335.0abcd
GF-296	5522.7fg	2755.0f	0.34cdef	0.77de	3095.7abcd
GF-21	6326.7cdef	2808.3ef	0.25f	0.67e	4222.6a
LSD	834.98	422.07	0.173	0.303	1329.6

Table 4. Comparison of means for MP, GMP, YSI, YI, and TOL

 $\dagger$ Means having common letters in each column have no significant difference at p<0.05 based on LSD test.

Table 5. Linear correlation between faba bean yield traits and drought-tolerance indices

	Yn	YS	STI	SSI	MP	GMP	YI	YSI	TOL	100 grain weight
Yn	1									
Ys	0.31ns	1								
STI	0.57**	0.95**	1							
SSI	0.04ns	-0.93**	-0.77**	1						
MP	0.91**	0.67**	0.85**	-0.58ns	1					
GMP	0.57**	0.95**	0.99**	-0.78**	0.85**	1				
YI	0.31ns	1**	-0.95**	-0.93**	0.67**	0.95**	1			
YSI	-0.06ns	0.92**	0.76**	-0.99**	0.34ns	0.76**	0.92**	1		
TOL	0.53*	-0.63**	-0.38ns	0.86**	0.14ns	-0.38ns	-00.63**	-0.87**	1	
100 grain weight	0.32ns	-0.34ns	-0.22ns	0.45*	0.10ns	-0.22ns	-0.34ns	-0.46*	0.57**	1

ns, \* and \*\*: non-significant and significant at p<0.05 and p<0.01, respectively.

#### **Cluster analysis**

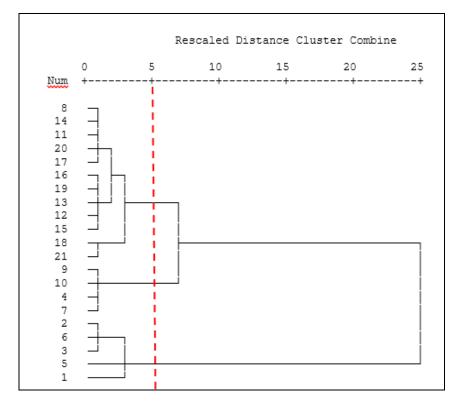
According to cluster analysis using ward and squared Euclidean distance, studied genotypes were classified in three groups. The first group included GF-398, GF-290, GF-335, GF-296, GF-524, GF-523, GF-520, GF-294, GF-293, GF-292, GF-525 and GF-21 genotypes. Four genotypes of GF-411, GF-401, GF-72 and GF-61 were placed in second group and GF-66, GF-62, GF-75, GF-65 and GF-67 belonged to the third group (Figure 1).

Groups 1 and 3 possessed higher yields compared to total average. The lowest yield under normal conditions was associated with group 2 which was 189.5 kg.ha<sup>-1</sup> (3.67%)

lower than total average, whereas under drought stress conditions. The extent of yield in the third group was 385.78 kg.ha<sup>-1</sup> (18.55%) higher than total average and average of other genotypes. Also, compared to total average and average of other groups, the third group had higher YSI, YI, GMP, MP and STI and lower SSI indices. Genotypes of this group were superior regarding yield and tolerance indices, so obtaining high yields is possible via cultivation of these genotypes under drought stress conditions.

Average of 100 grain weight in two environmental conditions (normal irrigation and drought stress) was utilized for cluster

analysis. This value was higher under normal conditions (126.14 g) compared to drought stress (119.35 g). There was a significant difference among genotypes regarding 100 grain weight. The highest 100 grain weight was associated with GF-21 with 154.44 g, followed by GF-520, GF-525, GF-524 and GF-523 with respectively 142.31, 142.10, 142.24 and 141.14 g. Genotypes GF-401 and GF-411 possessed the lowest value for 100 grain weight with respectively 99.46 and 101.73 g (Table 4). The difference among genotypes may be due to genetic properties of each genotype, so that some genotypes allocate assimilates to the seeds more efficiently.



*Figure 1.* Dendogram of faba bean genotypes using Ward's method based on grain yield under normal and stress conditions, 100 grain weight, STI, SSI, TOL, MP, GMP, YI and YSI

According to the results of cluster analysis for average of 100 grain weight, group 3 was placed between group 1 and 2 and was lower than total average. Average of 100 grain weight under drought stress in first, second and third was respectively 127.67, 102.08 and 113.21 g, whereas these values were 135.38, 104.22 and 121.52 g under normal irrigation. Third group may be regarded as a medium-sized grain genotype.

#### CONCLUSIONS

The following study was conducted to investigate the effect of drought stress on yield and stress tolerance indices of faba bean as well as selection of drought-tolerant genotypes. According to the results, a significant difference was observed among faba bean genotypes regarding all studied treatments which indicate genetic diversity in

faba bean genotypes. The highest and lowest STI was associated with GF-65 and GF-296, whereas GF-65 and GF-296 had the highest and lowest GMP values. Genotypes GF-65 and GF-401 showed the highest and lowest MP. Index of YI was the highest in GF-65, and the lowest value for this index was observed jointly in GF-525 and GF-21. The highest YSI value was observed in GF-67, whereas GF-525 and GF-21 genotypes had the lowest values. Genotypes GF-67 and GF-21 had the highest and lowest TOL values, respectively, whereas based on SSI index, GF-525 and GF-21 had the highest and GF-67 had the lowest values. Thus, noting the high correlation between stress-tolerance indices and grain yield, GF-65 may be regarded drought-resistant due to high GMP, MP and STI, whereas GF-525 and GF-21 had high TOL and SSI indices and were identified as drought- susceptible genotypes. According to cluster analysis, group 3 genotypes (GF-66, GF-62, GF-75, GF-65 and GF-67) were superior regarding tolerance indices and yield, so cultivation of these genotypes recommended under drought stress is conditions.

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# MORTEZA MEMARI ET AL.: EVALUATION OF DROUGHT STRESS TOLERANCE IN SOME FABA BEAN GENOTYPES USING DROUGHT TOLERANCE INDICES

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