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Full Length Research Paper

Evaluation of drought tolerance in different growth stages of maize (*Zea mays* L.) inbred lines using tolerance indices

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In order to find the best drought tolerant inbred lines, experiment was performed at the Agricultural College of Islamic Azad University, Shoushtar Branch, Iran during 2010. Experiment treatments were compared in a split-plot design by a randomized complete block design with 3 replications. Main factors included non-stress, drought stress in 6 to 7 leaves (vegetative) stage, drought stress in pollination stage and drought stress in grain filling stage. Sub factors were 7 inbred lines. Five stress tolerance indices, including mean productivity (MP), stress tolerance (TOL), stress susceptibility (SSI), stress tolerance index (STI) and geometric mean productivity (GMP) were used in this study. Drought stress in grain filling stage had the minimum grain yield that showed severe effects of drought stress at this stage of growth. Data analysis revealed that the MP, GMP and STI indices were the more accurate criteria for selection of drought tolerant and high yielding inbred lines. The positive and significant correlation of STI and grain yield under all conditions revealed that this index is more applicable and efficient for selection of parental inbred lines in producing hybrids tolerant to drought in vegetative, pollination and grain filling stages and high yielding under non-stress and stress conditions. Based on the STI, GMP and MP indices, K166B proved to be the most drought tolerant line. Biplot analysis allowed us to distinguish groups of tolerant and sensitive inbred lines. Based on the results of this study, the inbred line K166B can be recommended in future breeding programs for production of drought tolerant hybrids.

Key words: Biplot, drought stress, maize, tolerance indices.

INTRODUCTION

Among various abiotic and biotic stress factors, drought is an important cause of genotype and environmental interactions in maize across years, locations (Löffler et al., 2005; Setimela et al., 2005) and most likely within individual fields (Bruce et al., 2002). Drought is one of the most important abiotic stress factor (Bruce et al., 2002), which affects almost every aspect of plant growth (Sadras and Milroy, 1996; Aslam et al., 2006). Drought, or more generally, limited water availability is the main factor limiting crop production (Golbashy et al., 2010). Drought is a permanent constraint to agricultural production in many developing countries, and an occasional cause of losses of agricultural products in developed ones (Ceccarelli and Grando, 1996). The best option for crop production, yield improvement and yield stability under drought stress conditions is to develop drought tolerant crop varieties. One of the main goals in breeding programs is selection of the best genotypes under drought stress conditions (Richards et al., 2002). No exact figures on yield and economic losses in maize due to drought are available. In maize, grain yield reduction caused by drought ranges from 10 to 76%, depending on the severity and stage of occurrence (Bolaòos et al., 1993). Leta et al. (2001) reported that drought stress was at vegetative growth stage, the minimal effect at the grain filling stage caused the greatest decrease in grain yield.

To evaluate response of plant genotypes to drought

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stress, some selection indices based on a mathematical relation between stress and optimum conditions has been proposed (Rosielle and Hamblin, 1981; Clarke et al., 1992; Fernandez, 1992). Fernandez (1992) classified plants according to their performance in stressful and stress free environments into four groups: genotypes with similar good performance in both environments (Group A); genotypes with good performance only in non-stress environments (Group B) or stressful environments (Group C); and genotypes with weak performance in both environments (Group D).

Moghaddam and Hadi-Zadeh (2002) found that stress tolerant index (STI) was more useful in order to select favorable corn cultivars under stressful and stress-free conditions. Khalili et al. (2004) showed that based on geometric mean productivity (GMP) and STI indices, corn hybrids with high yield in both stress and non-stress environments can be selected. Khodarahmpour et al. (2011) reported that the SSI, STI and GMP indices were the more accurate criteria for selection of heat tolerant and high yielding maize genotypes. Biplot is an exploratory data visualization technique that displays the multivariate data into a two dimensional scatter plot. The concept of biplot was first developed by Gabriel (1971). This technique has extensively been used in the analysis of multi-environmental traits (Ahmadzadeh, 1997; Shiri et al., 2010; Fernandez, 1992; Souri et al., 2005; Karami et al., 2006; Khodarahmpour et al., 2011).

To improve maize yield and stability in stressful environments, there is a necessity to identify selection indices that are able to distinguish high yielding maize cultivars in these situations. Thus, the aim of this study was to evaluate efficiency and profitability of different selection indices in identification of cultivars which are compatible with stressful and optimal conditions, to achieve cultivars that can tolerate long irrigation intervals or are likely not to tolerate irrigation at sensitive growth stages.

MATERIALS AND METHODS

The present study was conducted at the Agricultural College of Islamic Azad University, Shoushtar Branch, Iran during 2010. Experiment treatments were compared in a split-plot design by a randomized completely block design with 3 replication. Main factors included: normal irrigation (non-stress), drought stress in 6 to 7 leaves (vegetative) stage, drought stress in pollination stage and drought stress in grain filling stage. Sub factors were 7 inbred lines (Table 1). The inbred lines were grown in three-row plots with 10 m length and 75 cm spacing between rows. Fertilizer was used based on soil test. Irrigation was applied once every 7 days for non-stress and stress conditions, respectively. Irrigation in stress conditions was separated into two rounds of irrigation in the stage applied to the drought stress. Drought tolerance indices were calculated using the following equations:

Tolerance index $(TOL) = Yp - Ys$ (Rosielle and Hamblin, 1981)

Mean productivity $(MP) = \frac{Yp + Ys}{2}$ (Rosielle and Hamblin, 1981)

Stress tolerance index $(STI) = \frac{Ys.Yp}{(\bar{Y}p)^2}$ (Fernandez, 1992)

Geometric mean productivity $(GMP) = \sqrt{(Yp)(Ys)}$ (Fernandez, 1992)

Stress susceptibility index $(SSI) = \frac{1 - \frac{Ys}{Yp}}{SI}$ $SI = 1 - \left(\frac{\bar{Y}s}{\bar{Y}p}\right)$ (Fischer and Maurer, 1978)

In all the equations, Ys and Yp are stress and non-stress (potential) yield of a given genotype, respectively. $\bar{Y}s$ and $\bar{Y}p$ are average yield of all genotypes under stress and non-stress conditions, respectively. Analysis of variance was performed using the SPSS ver. 16 computer program as well as mean comparison and correlation coefficients. The biplot display was used, which provides a useful tool for data analysis. To display the genotypes in biplot, a principal component analysis was performed.

RESULTS AND DISCUSSION

Results of ANOVA showed significant differences among different levels of drought stress, inbred lines and interaction between drought stress and inbred lines for grain yield under studied conditions ($P \leq 0.01$) (Table 2), which demonstrated existence of high diversity among inbred lines studied for drought tolerance and difference between time of applied stress.

Among all inbred lines, K166B (3254.2 kg/ha) had the maximum grain yield, but MO17 (750.63 kg/ha) had the minimum grain yield produced in all conditions, respectively (Tables 3 and 4). Drought stress in grain filling stage had the minimum grain yield (Tables 3 and 4) that showed severe effects of drought stress at this stage of growth. Drought stress reduces grain yield by 15, 40 and 60% at vegetative growth, pollination and grain filling stages than non-stress condition, respectively (Table 3). Leta et al. (2001) reported that drought stress at vegetative growth stage had minimal effect and at the grain filling stage, caused the greatest decrease in grain yield. Other researchers showed that drought stress decreased grain yield (Shiri et al., 2010, Golbashy et al., 2010).Drought tolerance indices were calculated to identify the tolerant inbred lines (Table 4). Based on the MP, GMP and STI indices, the K166B line was identified as tolerant in vegetative, pollination and grain filling stages of drought stress (Table 4).Therefore, according to these results, selection based on MP, GMP and STI will improve mean yield under non-stress and drought

Table 1. Pedigree/origin of studied inbred lines of maize

Inbred lines	Pedigree sources/origin
Lancaster Sure Crop (LSC)	
MO17	Cl. 187–2 × C103
K18	Derived from MO17 changes in Iran
K19	Derived from MO17 changes in Iran
Reid Yellow Dent (RYD)	
A679	A B73 back-cross derived line [(A662 × B73)(3)]
Extracted from late synthetic (created in Iran)	
K3651/1	SYN-Late(Iran)
Lines extracted from CIMMYT originated materials in Iran	
K166A	
K166B	

Table 2. Analysis of variance of mean squares of grain yield trait.

Source of variance	Block	Drought stress	Drought stress error	Inbred line	Drought stress × inbred line	Error
Degree of freedom	2	3	6	6	18	48
Mean of squares	2990**	545**	138	327.8**	23.5**	2.47

**Significant at 0.01 probability level.

Table 3. Comparison of means simple effects of drought stress levels and inbred lines.

Parameter	Grain yield (kg/ha)
Non-stress	2748.1[a]
Drought stress in vegetative stage	2344.6[ab]
Drought stress in pollination stage	1660[b]
Drought stress in grain filling stage	1092.7[c]
MO17	750.63[d]
K18	1828[bc]
K3651/1	1384.25[c]
A679	1508.13[c]
K166A	2304.75[b]
K166B	3254.2[a]
K19	2449.5[b]

*In each column, means with similar letters do not differ significantly at 0.05 probability level.

stress conditions. The ability to separate group A genotypes from others using the STI and GMP indices are consistent with the results reported by Ahmadzadeh (1997) and Khalili et al. (2004) in maize, Kristin et al. (1997) and Fernandez (1992) in common bean, Souri et al. (2005) in pea, Karami et al. (2006) in barley and Rezaeizad (2007) in sunflower in drought stress condition and Khodarahmpour et al. (2011) in maize in heat stress

Table 4. Average yields of maize inbred lines under non-stress (Yp) and stress (Ys) conditions, and different calculated drought tolerance indices.

Inbred line	Stress condition		Grain yield (Kg/ha)	MP	GMP	TOL	STI	SSI
MO17	Non-stress	Yp	1121.9^{efg}					
	Vegetative	Ys	1002^{efg}	1061.75	1060.07	119.5	0.15	0.71
	Pollination		586^{fg}	853.75	810.68	535.5	0.09	1.20
	Grain filling		293^{h}	1414.5	573.24	828.5	0.04	1.23
K18	Non-stress	Yp	2639^{bc}					
	Vegetative	Ys	2428^{bc}	3853	2531.30	211	0.85	0.53
	Pollination		1895de	2267	2236.27	744	0.07	0.7
	Grain filling		350^{gh}	1494.5	961.07	2289	0.12	1.44
K3651/1	Non-stress	Yp	2250^{c}					
	Vegetative	Ys	1878de	2064	2055.6	372	0.55	1.10
	Pollination		1081^{efg}	1665.5	1559.6	1169	0.32	1.30
	Grain filling		328^{gh}	1289	859.1	1922	0.10	1.42
A679	Non-stress	Yp	2541^{bc}					
	Vegetative	Ys	1914de	2227.5	2205.33	627	0.64	1.64
	Pollination		997.5^{cd}	1769.25	1592.06	1543.5	0.33	1.52
	Grain filling		580^{fg}	1560.5	1991.3	1961	0.19	1.28
K166A	Non-stress	Yp	2887^{b}					
	Vegetative	Ys	2379^{c}	2633	2620.72	508	0.91	1.17
	Pollination		2059^{d}	2473	2438.1	828	0.80	0.72
	Grain filling		1894de	2390.5	405.93	993	0.72	0.57
K166B	Non-stress	Yp	4026^{a}					
	Vegetative	Ys	3793^{ab}	3909.5	3907.8	233	2.02	0.38
	Pollination		3005.8^{b}	3515.9	3478.7	1024.2	1.60	0.63
	Grain filling		2192^{cd}	3109	2970.7	1834	1.17	0.76
K19	Non-stress	Yp	3772^{ab}					
	Vegetative	Ys	3018^{b}	3395	3374	754	1.51	1.33
	Pollination		1996^{d}	2884	2743.88	888	0.99	1.17
	Grain filling		1012^{efg}	2392	1953.78	1380	0.51	1.22

*In each column, means with similar letters do not differ significantly at 0.05 probability level. Yp: Potential yield; Ys: stress yield; TOL: tolerance index; MP: mean productivity; GMP: geometric mean productivity; SSI: stress susceptibility index; STI: stress tolerance index.

condition.

MO17 was a tolerant inbred line based on TOL and its low quantity indicates tolerant inbred lines (Table 4). MO17 was low yielding under all conditions. It seems that TOL had succeeded in selecting genotypes with high yield under stress, but had failed to select genotypes with proper yield under both non-stress and stress environments (Rosielle and Hamblin, 1981). This is due to low yield differences between the two conditions, which decreased the value of the TOL index. Therefore, low TOL does not mean high yielding, and genotype yield should be taken into consideration in addition to this criterion. Similar results were reported by Ahmadzadeh (1997) for maize hybrids in drought stress and Khodarahmpour et al. (2011) for maize inbred lines and hybrids in heat stress condition. Limitations of using the TOL index have also been discussed in relation to wheat (Clarke et al., 1992) and common bean (Ramirez and Kelly, 1998). Although, low TOL has been used for selecting genotypes with tolerance to stress, the

Table 5. Correlations between different selection indices and mean yield of maize hybrids under non-stress and stress conditions.

Stress		Yp	Ys	MP	TOL	GMP	STI	SSI
	Yp	1						
	Ys	0.97**	1					
	MP	0.85*	0.89**	1				
Vegetative	TOL	0.44	0.22	0.13	1			
	GMP	0.99**	0.99**	0.88**	0.33	1		
	STI	0.96**	0.99**	0.83*	0.23	0.98**	1	
	SSI	-0.34	-0.26	-0.34	0.86*	-0.15	-0.26	1
	Yp	1						
	Ys	0.89**	1					
	MP	0.98**	0.97**	1				
Pollination	TOL	0.25	-0.06	0.11	1			
	GMP	0.96**	0.98**	0.99**	0.06	1		
	STI	0.85*	0.84*	0.87*	0.11	0.87*	1	
	SSI	-0.44	-0.79*	-0.62	0.51	-0.67	-0.451	1
	Yp	1						
	Ys	0.72	1					
	MP	0.76*	0.53	1				
Grain filling	TOL	0.30	-0.21	-0.21	1			
	GMP	0.73	0.44	0.52	0.40	1		
	STI	0.80*	0.98**	0.60	-0.09	0.60	1	
	SSI	-0.43	-0.93**	-0.34	0.47	-0.14	-0.83*	1

* and**Significant at 0.05 and 0.01 probability levels, respectively. Yp: Potential yield; Ys: stress yield; TOL: tolerance index; MP: mean productivity; GMP: geometric mean. Productivity, SSI: stress susceptibility index; STI: stress tolerance index.

likelihood of selecting low yielding genotypes can be anticipated (Ramirez and Kelly, 1998).

Using SSI, K166B was selected as tolerant inbred line in vegetative and pollination stages of stress, but K166A was selected as tolerant inbred line in grain filling stage of drought stress (Table 4). K166A yield was relatively high in all conditions, also, K166B had high yield under all conditions (Tables 3 and 4). Therefore, this index discriminated Group A genotypes from others. This finding is consistent with that reported by Moghaddam and Hadizadeh (2000) and Khodarahmpour et al. (2011) in maize.

To determine the most desirable stress tolerant criterion, the correlation coefficient between Yp, Ys and quantitative indices of stress tolerance were calculated (Table 5). There were high and significant correlations between MP, GMP and STI in vegetative and pollination stages of drought stress (Table 5). There were significant and positive correlations between Ys and Yp with MP, GMP and STI in vegetative and pollination stages of stress, but there were significant and positive correlations between Yp and Ys with STI and Yp with MP in grain filling stage of stress (Table 5). Fernandez (1992) proposed STI as an index which discriminates genotypes with high yield and stress tolerance potentials. In this study, we found positive and high correlation between grain yield under drought stress and STI at all stages under study. The correlation coefficients between STI and yield in stress and non-stress conditions were highly positive and significant (Table 5). Hence, selection for high STI should give positive responses in all stages of drought stress.

A higher STI, GMP and MP value is indicative of more drought stress tolerance (Fernandez, 1992). Based on these indices, K166B was identified as superlative (Table 4). Khodarahmpour et al. (2011) reported that the SSI, STI and GMP indices were the more accurate criteria for selection of heat tolerant and high yielding maize genotypes. The positive and significant correlation of GMP and grain yield under both conditions revealed that this index is more applicable and efficient for selection of parental inbred lines in producing hybrids that are tolerant

Table 6. Eigen values, cumulative proportion and Eigen vectors of tolerance indices and yield in two environmental conditions in maize inbred lines.

Stress condition	Component	Eigen values	Cumulative proportion (%)	Yp	Ys	MP	GMP	TOL	SSI	STI
Vegetative	1	4.87	69.53	0.978	0.996	0.917	0.995	0.261	-0.233	0.979
	2	1.20	96.94	0.200	-0.035	-0.133	0.078	0.959	0.968	-0.027
Pollination	1	5.12	73.1	0.974	0.957	0.994	0.99	0.177	-0.582	0.907
	2	1.46	93.94	0.122	-0.271	-0.600	-0.121	0.939	0.750	0.018
Grain filling	1	4.19	59.86	0.939	0.879	0.752	0.758	0.074	-0.649	0.944
	2	1.75	84.88	0.235	-0.412	-0.43	0.487	0.900	0.703	-0.250

Yp: Potential yield; Ys: stress yield; TOL: tolerance index; MP: mean productivity; GMP: geometric mean. Productivity, SSI: stress susceptibility index; STI: stress tolerance index.

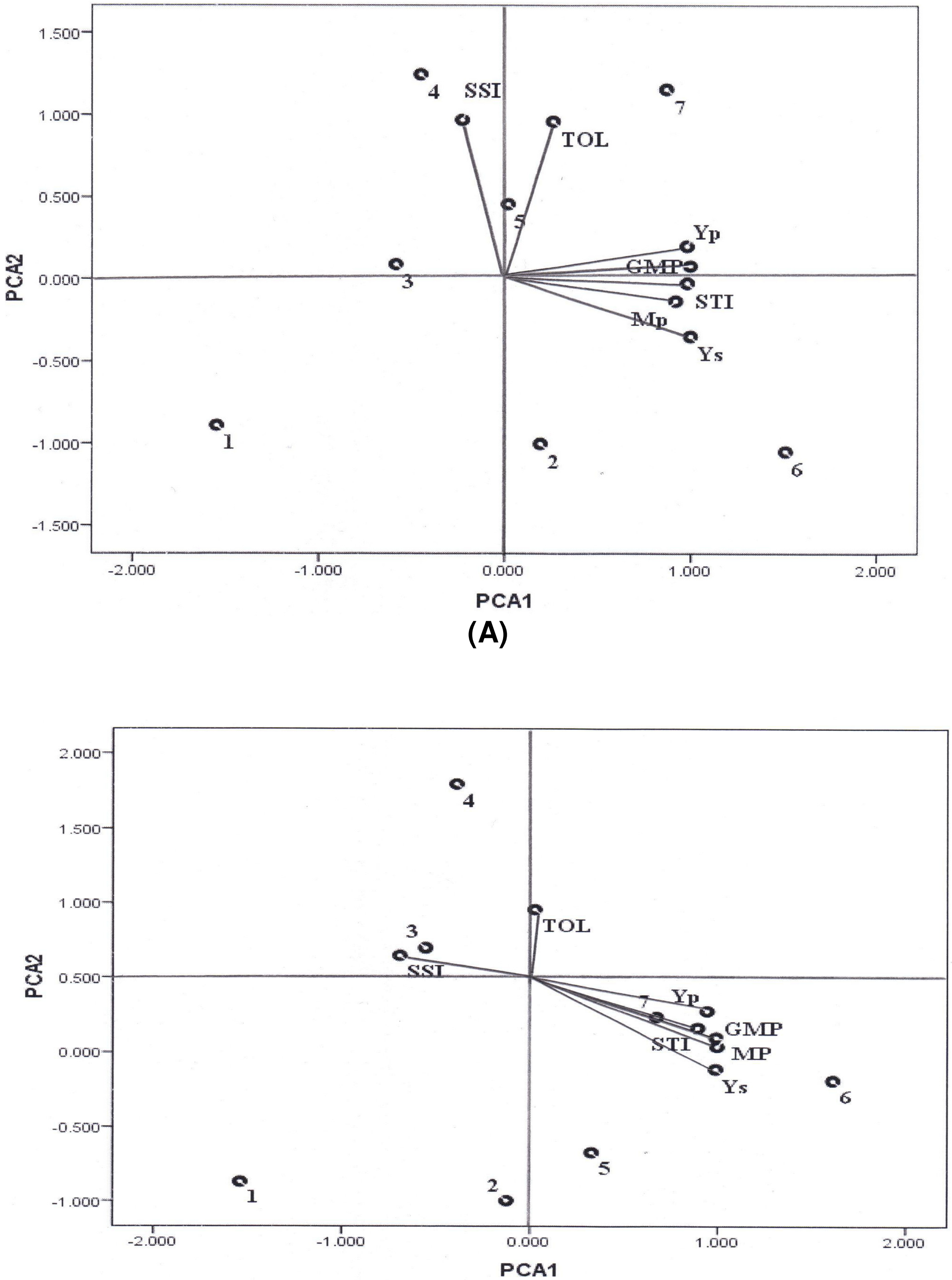
to high temperatures and high yielding under both conditions.

Principal component analysis (PCA) of inbred lines revealed that the first PCA explained 69.53, 73.1 and 59.86% of the variation with Yp, Ys, MP, GMP, SSI, TOL and STI in vegetative, pollination and grain filling stages of drought stress, respectively (Table 6). Thus, the first axis (PCA1) can be identified as yield potential and drought tolerance. Considering the high and positive value of this PCA on biplot, selected genotypes will be high yielding under stress and non-stress conditions. The second PCA explained 27.41, 20.84 and 25.02% of the variation with different attributes in vegetative, pollination and grain filling stages of drought stress, respectively (Table 6). Therefore, the second component (PCA2) can be named as a stress susceptible component with low yield in a stressful condition. Thus, selection of genotypes that have high PCA1 and low PCA2 are suitable for both stress and non-stress conditions. Therefore, K18 and K166B inbred lines in vegetative stage (Figure 1A) of stress, K166A, K166B and K19 in pollination stage (Figure 1B) of stress and K166A inbred line in grain filling stage (Figure 1C) of stress are superior for both environments with high PCA1 and low PCA2 (Figure 1). Kaya et al. (2002) revealed that genotypes with larger PCA1 and lower PCA2 scores gave high yields (stable genotypes), and genotypes with lower PCA1 and larger PCA2 scores had low yields (unstable genotypes). The use of biplot display in selecting drought tolerant genotypes has already been used by Ahmadzadeh (1997) and Shiri et al. (2010) in maize, Fernandez (1992) in common bean, Souri et al. (2005) in pea and Karami et al. (2006) in barley.

The correlation coefficient among any two indices is given approximately by the cosinus of the angle between their vectors. Hence, r = cos 180º = -1, cos 0º = 1 and cos 90º = 0 (Yan and Rajcan, 2002). Thus, a strong positive association between GMP, MP and STI with Yp and Ys was revealed by the acute angles between the corresponding vectors in all stages of drought stress. A negative association between SSI and Ys was reflected by the larger obtuse angles between their vectors in pollination and grain filling stages of drought stress in a biplot display (Figure 1). The results obtained from the biplot graph, confirmed the correlation analysis. Results of this study are in agreement with Golabadi et al. (2006) in durum wheat for drought tolerance. Since selection of drought tolerant genotypes is done based on the combination of indices in the biplot, this method is better than one index alone, to identify superior genotypes for drought conditions.

Conclusions

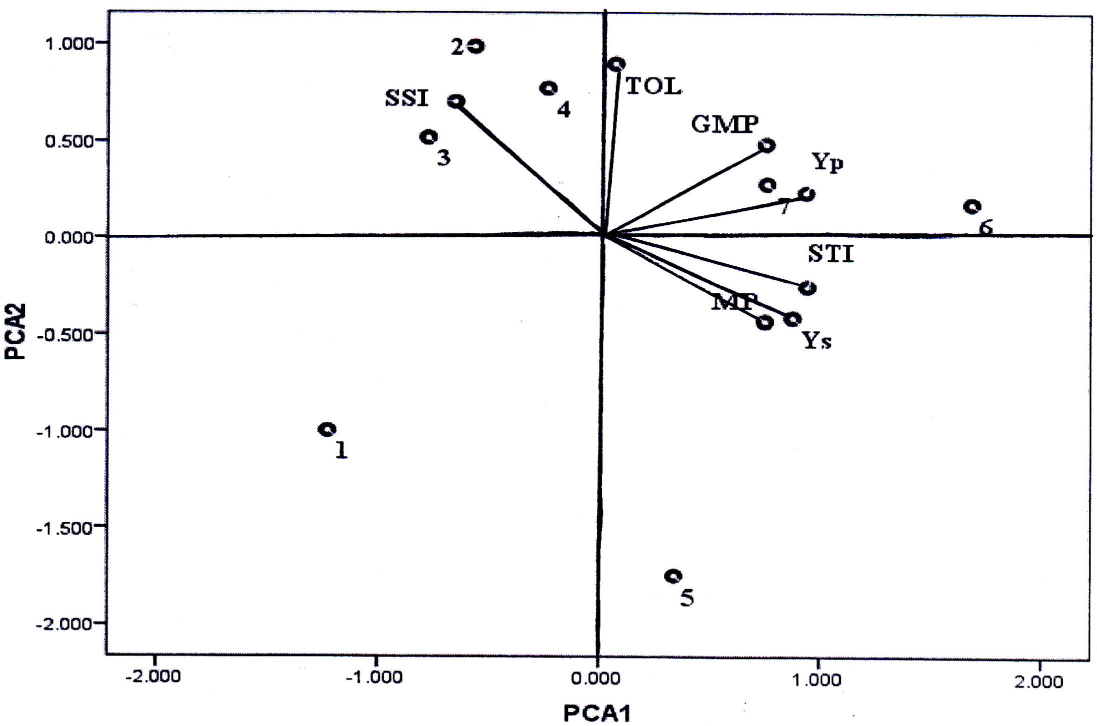
Drought stress in grain filling stage had the minimum grain yield that showed severe effects of drought stress at this stage of growth. The line K166B based on the MP, GMP and STI showed the highest tolerance to drought stress at all stages and produced the highest yield in all conditions. In this study, we found positive and high correlation between grain yield under drought stress and STI at all stages under study. Hence, selection for high STI should give positive responses in all stages of drought stress. Based on biplot display, the line K166B in vegetative and pollination stages of stress and K166A in grain filling stage of stress appeared to have high yield potential and low stress susceptibility. With reference to the results of grain yield in all conditions and tolerance indices, the line K166B can be recommended in future



(A)

(B)

Figure 1. The biplot display of maize inbred lines and drought tolerance indices on the first and second principal components in drought stress (A): vegetative stage, (B): pollination stage and (C): grain filling stage. 1: MO17, 2: K18, 3: K3651/1, 4: A679, 5: K166A, 6: K166B, 7: K19.



(C)

Figure 1. Contd.

breeding programs for production of drought tolerant hybrids.

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