International Journal of Farming and Allied Sciences
Available online at www.ijfas.com
©2015 IJFAS Journal-2015-4-3/177-184/ 31 March, 2015
ISSN 2322-4134 ©2015 IJFAS

EVALUATION OF GENETIC VARIATIONS FOR DROUGHT TOLERANCE IN SOME ADVANCE LINES OF BARLEY (*HORDEUM VULGAR*)

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ABSTRACT: In order to evaluation of genetic variation for drought tolerance, seven advanced barley lines (*Hordeum vulgar*) and two control cultivars planted in the field of the Agricultural and Natural Resource Station of Kermanshah, Iran, using randomized complete block design with three replications in 2009 - 2011. The Results of variance analysis showed a significant differences between genotypes for yield potential (Yp), stress yield (Ys), mean productivity (MP), geometric mean productivity (GMP), harmonic mean (MH), stress tolerance index (STI), yield index (YI) and abiotic tolerance index (ATI) at ٪1 level and for tolerance index (TOL), stress susceptibility index (SSI), Yield stability index (YSI), drought resistance index (DI), stress susceptibility percentage index (SSPI) and stress non-stress production index (SNPI) at ٪5 level. Screening drought tolerance indicators using correlation analysis displayed that the most suitable drought tolerance criteria for screening advanced barley lines were MP, GMP, MH and STI. The results of compare means, cluster analysis and principal component analysis showed that the genotypes of MBD-85-8, MB-85-3 and Nosrat had the most resistance to drought and the highest seed yield; on the other side, the genotypes of MBD-85-6, MBD-85-3 and MBD-85-14 had the lowest resistance to drought and seed yield. The genotypes of MB-85-18, MB-85-5 and Yosef had the moderate resistance to drought and seed yield.

***Keywords*:** drought tolerance indices; Kermanshah; Multivariable analysis; Stress conditions; variance analysis

INTRODUCTION

Barley is one of the most important cereal crops grown in many countries such as IRAN, where it is often subject to extreme drought stress that significantly affects production (Ceccarelli, 2007). Drought is an important factor limiting crop production in arid and semi-arid conditions. Breeding for drought tolerance by selecting solely for grain yield is difficult, because the heritability of yield under drought conditions is low, due to small genotypic variance or large genotype-environment interaction variances (Blum, 1988.). The genetic structure and phenotypic expression of a quantitative trait are highly influenced by environmental factors, thus, one barrier for understanding the inheritance of a quantitative trait is genotype-environment interactions (Breese, 1969). Drought tolerance is not a simple response, but is mostly conditioned by many component responses, which interact and may different for crops, in relation to types, intensity and duration of water deficit. Moreover, most agronomical characters are expressed differently in normal and stress conditions and are known to be affected by environmental factors. Therefore, selection based on the phenotype would be difficult for such traits (Hittalmani, 2003). Many methods have been employed to identify crop lines that are productive in dry environments (Reynolds, 2007) Some use mathematical models to compare the change in seed yield between stressed and non-stressed environments (Rosielle and Hamblin, 1981). Loss of yield is the main concern of plant breeders and they hence, emphasize on yield performance under moisture-stress conditions. But variation in yield potential could arise from factors related to adaptation rather than to drought tolerance. Thus, drought indices providing a measure of drought based on yield loss under drought-conditions compared to normal conditions are being used in screening drought-tolerant genotypes (Mitra, 2001). Genotype can be categorized into four groups based on their performance in stress and non-stress environments: genotypes express uniform superiority in both stress and non-stress environments (Group A); genotypes perform favorably only in non-stress environments (Group B); genotypes yield relatively higher only in stress environments (Group C); and genotypes perform poorly in both stress and non-stress environments (Group D). The optimal selection criterion should distinguish Group A from the other three groups (Fernandes, 1992).

Several drought resistance indices were proposed based on genotypes performance in stress (Y_s) and non-stress (Y_p) conditions. Rosielle and Hamblin (1981) defined stress tolerance (TOL) as the differences in yield under stress (Ys) and non-stress (Yp) environments and Mean Productivity (MP) as the average of Ys and Yp. Fischer and Maurer (1978) proposed a Stress Susceptibility Index (SSI) of the cultivar. Fernandez (1992) defined stress tolerance index (STI), which can be used to identify genotypes that performance high yield in two environments conditions. The geometric mean productivity index (GMP), which often used by breeders interested in relative performance since drought stress can vary in severity in field environment over years (Ramirez and Kelly, 1998). Gavuzzi (1997) defined yield index (YI), by genotype yield on average yield of stress condition. Other yield based estimates of drought resistance are drought resistance index (DI) and yield stability index (YSI), which introduced by Lan (1998) and Bouslama & Schapaugh (1984), respectively. In recently years several drought resistance indices were suggested based on genotypes performance in stress (Y_s) and non-stress (Y_p) conditions, consisted of abiotic tolerance index (ATI), stress susceptibility percentage index (SSPI) and stress non-stress production index (SNPI) (Moosavi, 2008). Among the stress tolerance indicators, a larger value of TOL and SSI represent relatively more sensitivity to stress, thus a smaller value of TOL and SSI are favored. Selection based on these two indices favors genotypes with low yield under non-stress conditions and high yield under stress conditions (Golabadi, 2006). To evaluate quantitative drought resistance criteria of barley genotypes in stressed and non-stressed conditions reported that genotypes were significantly different for their yield under stress and non-stress conditions. (Giancarla, 2010). Drought stress reduced the yield of some genotypes while others were tolerant to drought, suggesting genetic variability in this material for drought tolerance. The results of a correlation matrix revealed highly significant associations between grain yield (Yp) and mean productivity (MP), stress tolerance index (STI), geometric mean productivity (GMP) and yield index (YI) under irrigated conditions while, the mean productivity (MP), yield stability index (YSI), stress tolerance index (STI), geometric mean productivity (GMP) and yield index (YI) had a high response under stressed condition. Based on a principal component analysis, GMP, MP and STI were considered to be the best parameters for the selection of drought-tolerant genotypes. Nazari and Pakniat (2010) indicated that STI, MP and GMP are the best criteria for the selection of high yielding barley genotypes both under stress and non-stress conditions. Results of calculated gain from indirect selection indicated that selection under moisture stress would be efficient in yield improvement compared to non-stress condition. Genotypes were significantly different for their yield under stress and non-stress conditions (Zare, 2012). Ahmadizadeh (2012) showed that MP, STI, GMP and HM were the most suitable indices to screen durum wheat genotypes in drought stress condition. Based on geometric mean productivity (GMP) and STI indices, corn hybrids with high yield in both stress and non-stress environments can be selected (Khalili, 2004). Mean productivity (MP), geometrical mean productivity (GMP) and stress tolerance index (STI) were suitable resistance indices for the identification of bread wheat genotypes to drought stress (Abdi, 2012). Talebi (2009) reported that MP, GMP and STI were more effective in identifying high yielding cultivars under different moisture conditions. Akcura (2011) revealed that SSI was suggested as useful indicator for wheat breeding where the stress was severe while MP, GMP, TOL, HM and STI were suggested if the stress was less severe.

In agronomic and breeding studies, correlation coefficients are generally employed to determine the relation of grain yield with yield components. Correlation coefficients mostly bring forth the interrelations of independent components. However, in plant production, the Barley cultivars grain yield is a function of many parameters which have interrelations among themselves and affect the grain yield directly or indirectly. For this reason, the correlation coefficients become insufficient in using yield components for selection criteria to improve grain yield. It is reasonable to know whether any yield component has a direct or an indirect effect on grain yield, so that the selection studies can be carried out successfully. Here, the PCA analysis clear relations between genotypes and drought tolerance indices and is used to determine the direct or indirect effect of any drought indices on yield under normal and stress conditions. Many researches were done correlation and PCA analysis methods simultaneously on barely drought indices. PCA methods clear relationships between genotypes, drought tolerant indices and YP and YS and can to help plant breeding researchers to selection genotypes with high yield under normal and stress conditions.

The objectives of this study were to, identify drought tolerant barley varieties under different conditions in Kermanshah at west of Iran, determine the efficiency of tolerance indices to classify barley varieties into sensitive and tolerant and interpret interrelationships among the tolerance indices by biplot analysis.

MATERIALS AND METHODS

This experiment was carried out in (2009 -2011) in Research Center of Kermanshah Agricultural and Natural Resources of Iran. This filed placed in (34°/08´) latitude, (26°/47´) longitude with 1346 meter altitude and. average rainfall 538 mm in year. Two separate experiments carried out in a Randomized Complete Block Design (RCBD) with three replications using seven advance barley lines (*Hordeum vulgar*): MBD-85-3, MBD-85-6, MBD-85-8, MBD-85-14, MB-85-3, MB-85-5, MB-85-18 and two control genotype (Nosrat and Yosef) in stress and non-stress conditions. Experiments carried out in the same conditions and stopped the irrigation at Early Heading stage in one of them. Seeds were planted in 3 to 5 cm deep on October 28 in 2010. Individual plot consisted of 6 rows with 240 cm long, 20 cm distances between rows and 5 cm distances between plants (400 seeds in 1 m^2). Used fertilizers include 200 kg/ ha phosphate and 150 kg/ha ammonium (the hole of phosphate and 1/3 of ammonium applied prior to planting and 2/3 of ammonium at the two stage of growth, Raw Rating and Early Heading). Yield was measured at end of growth season by harvesting 1 m of the central part of each plot at physiological maturity.

Drought resistance indices were calculated by grain yield per plot for stress (Ys), non-stress (Yp) and total mean of grain yield for stress ($\bar{Y}_s$) and non-stress ($\bar{Y}_P$) conditions consisted of:

Stress susceptibility index (SSI) were suggested by Fischer and Maurer (1978) as following:

$$SSI = \frac{1-(Y_S/Y_P)}{1-(\bar{Y}_S/\bar{Y}_P)}$$

Tolerance (TOL) and mean productivity (MP) indices defined by Rosielle and Hamblin (1981).

$$TOL = Y_P - Y_S \qquad MP = \frac{Y_S + Y_P}{2}$$

Fernandez (1992) suggested stress tolerance index (STI), to use for identification of high yield genotypes in both conditions and geometric mean productivity (GMP) as well:

$$STI = \frac{Y_S \times Y_P}{\bar{Y}_P^{\,2}} \qquad GMP = \sqrt{(Y_S \times Y_P)}$$

Yield index (YI) proposed by Gavuzzi (1997) using yield of stress condition.

$$YI = \frac{Y_S}{\bar{Y}_S}$$

Yield stability index (YSI) introduced by Bouslama and Schapaugh (1984) as following:

$$YSI = \frac{Y_S}{Y_P}$$

Drought resistance index (DI) = Ys × (Ys/Yp)/ $\bar{Y}_S$ (Lan, 1998).

Moosavi (2008) suggested abiotic tolerance index (ATI), Stress susceptibility percentage index (SSPI) and Stress non-stress production index (SNPI) to identification resistant genotypes as:

ATI= [(Yp-Ys) / ($\bar{Y}_P$/ $\bar{Y}_S$)] × [√Yp × Ys]

SSPI= [Yp-Ys /2($\bar{Y}_P$)] × 100

SNPI= [$\sqrt[3]{}$ (Yp+Ys) / (Yp - Ys)] × [$\sqrt[3]{}$ Yp × Ys × Ys]

Statistical analysis

Analysis of variance, were done for drought resistance indices and yield of stress and non-stress conditions to determine significant variation among genotypes in randomized complete block design (RCBD) by three replications and mean comparison were performed to determine resistant genotypes (*Farshadfar and Sutka, 2003).* Ward's hierarchical clustering procedure was determined for grouping genotypes. Spearman's rank correlation between each pair of drought resistance indices were measured to distinction relationships between indices. Principal component analysis was performed for two-way tables of genotypes ranks for drought resistance indices and yield of two growth conditions (*Nouri, 2011)*

RESULTS AND DISCUSSION

Interrelationship among indices with seed yield

To determine the most desirable drought resistance criteria, the Spearman's rank correlation coefficient between yield of stress and non-stress conditions and indices of drought resistance were calculated (Table 1). The results indicated that MP, GMP, MH and STI had a significant ($P<0.01$) positive correlation with Yield of non-stress condition, beside TOL and SSPI had a significant ($P<0.01$) negative correlation. The indices of GMP, MH, STI, YI, DI and SNPI had a significant ($P<0.01$) positive correlation with Yield of stress condition and MP ($P<0.05$), therefore only four indices of MH, STI, MP and GMP had positive significant correlation with two environments. Some researchers believe in selection based on only favorable condition (*Betran, 2003*), and/or only stress condition (Giancarla*,* 2010), but others have chosen a mid-point and in selection based on both favorable and stress conditions (*Fernandes, 1992; Byrne, 2005*; Farshadfar*,* 2001) believe that most suitable indices for selection of drought resistance cultivars, is an indicator which has a relatively high correlation with grain yield in both conditions (*Fernandes, 1992;* Farshadfar*,* 2001; *Byrne, 2005)*. Fernandez (1992) referred that MP fails to distinguish between groups A and B, but STI is expected to distinguish group A from group B and group C and rank correlation between STI and GMP is equal to 1, also the higher value of STI for a genotype, the higher its stress tolerance and yield potential (*Fernandes, 1992).* Therefore, in this study we emphasis to using MP, STI, MH and GMP indices for genotype selection. This result were in close agreement with the findings of Fernandez (1992), Farshadfar (2001), Talebi (2009) and Nouri, (2011).

Table 1. Spearman's rank correlation between drought resistance indices and yield of stress and non-stress conditions

	YP	YS	TOL	MP	GMP	MH	SSI	STI	YI	YSI	DI	ATI	SSPI
YS	0.450												
TOL	-0.800**	0.117											
MP	0.833**	0.783*	-0.417										
GMP	0.800**	0.833**	-0.367	0.983**									
MH	0.800**	0.833**	-0.367	0.983**	1.000**								
SSI	-0.517	0.433	0.900**	-0.083	-0.017	-0.017							
STI	0.800**	0.833**	-0.367	0.983**	1.000**	1.000**	-0.017						
YI	0.450	1.000**	0.117	0.783*	0.833**	0.833**	0.433	0.833**					
YSI	-0.517	0.433	0.900**	-0.083	-0.017	-0.017	1.000**	-0.017	0.433				
DI	0.233	0.950**	0.367	0.617	0.683*	0.683*	0.650	0.683*	0.950**	0.650			
ATI	0.883**	-0.050	0.967**	-0.550	-0.500	-0.500	0.850**	-0.500	-0.050	0.850**	0.200		
SSPI	-0.800**	0.117	1.000**	-0.417	-0.367	-0.367	0.900**	-0.367	0.117	0.900**	0.367	0.967**	
SNPI	0.233	0.950**	0.367	0.617	0.683*	0.683*	0.650	0.683*	0.950**	0.650	1.000**	0.200	0.367

Assessment of resistant genotypes

The results of variance analysis showed significant differences between genotypes for all indices and indicated that genotypic differences were highly significant ($P<0.01$) for Y_P, Y_S, MP, STI, GMP MH, YI and ATI, also were observed for SSI, TOL, YSI, DI and SSPI a significant ($P<0.05$) genotypic differences (Table 2). Farshadfar (2011) reported significant differences for drought resistance indices in bread wheat.

Table 2. Mean squares for yield of stress and non-stress conditions and drought resistance indices

Source of Variation	DF	Mean of Square YP	Ys	SSI	TOL	MP	STI	GMP
Replication	2	0.322 *	0.093 ^{ns}	0.062 ^{ns}	0.258 ^{ns}	0.141 ^{ns}	0.012 ^{ns}	0.140 ^{ns}
Genotype	8	1.722 **	0.911 **	0.271 *	0.914 *	1.091 **	0.086 **	1.047 **
Error	16	0.073	0.156	0.103	0.289	0.042	0.005	0.055
Coefficient of Variation (%)		4.76	9.42	32.79	36.62	4.17	9.36	4.81
Source of Variation	DF	Mean of Square MH	YI	YSI	DI	ATI	SSPI	SNPI
Replication	2	0.144 ^{ns}	0.005 ^{ns}	0.004 ^{ns}	0.009 ^{ns}	6.221 ^{ns}	20.27 ^{ns}	1.302 ^{ns}
Genotype	8	1.023 **	0.053 **	0.018 *	0.063 *	14.880 **	71.26 *	10.274 *
Error	16	0.072	0.009	0.007	0.021	3.519	22.48	3.749
Coefficient of Variation (%)		5.59	9.41	11.11	19.31	35.1	36.60	20.92

*; ** significant at the 5% and 1% probability levels, respectively, ns; no significant

The results of mean comparison by LSD procedure at 5% and 1% probability levels and ranks of genotypes for indices were given in Table 3. These results indicated that the identification of drought-resistance genotypes based on a single index were antagonistic to other indices, therefore genotype selection were done considering correlation between indices and based on indices groups which positive and significantly correlated together, thus the genotypes MB-85-5, MBD-85-14 and YOSEF had the highest drought resistance based on SSI, TOL, DI, YSI, ATI, SSPI and SNPI, and the genotypes Nosrat, MB-85-3 and MB-85-8 had the most drought resistance based on MP, STI, GMP MH and YI. The genotypes MB-85-3, NOSRAT and MBD-85-8 had the highest yield in non stress condition, also the highest yield in stress condition were observed for Nosrat, MB-85-5, MB-85-3 and yousef.

Table 3. Mean comparison by LSD method at 5% and 1% probability levels for genotypes based on yield of stress and non-stress conditions and drought resistance indices and genotypes ranks for indices

Genotype	Yp	Ys	SSI	TOL	MP	STI	GMP
YOSEF	5.54(6)	4.48(4)	0.74(3)	1.07(3)	5.01(6)	0.77(6)	4.98(6)
MBD-85-3	4.86(8)	3.37(9)	1.18(7)	1.49(5)	4.12(8)	0.51(8)	4.05(8)
MBD-85-6	6.01(4)	3.80(7)	1.42(9)	2.21(8)	4.91(7)	0.71(7)	4.76(7)
MBD-85-8	6.48(1)	4.26(6)	1.31(8)	2.22(9)	5.37(3)	0.86(3)	5.25(3)
MBD-85-14	4.18(9)	3.38(8)	0.73(2)	0.80(2)	3.78(9)	0.44(9)	3.75(9)
MB-85-3	6.29(3)	4.55(3)	1.06(6)	1.73(7)	5.42(2)	0.89(2)	5.35(2)
MB-85-5	5.44(7)	4.76(2)	0.48(1)	0.67(1)	5.10(5)	0.81(4)	5.09(4)
MB-85-18	5.87(5)	4.38(5)	0.97(5)	1.49(4)	5.13(4)	0.80(5)	5.07(5)
NOSRAT	6.33(2)	4.79(1)	0.91(4)	1.54(6)	5.56(1)	0.94(1)	5.49(1)
Lsd (%5)	0.468	0.684	0.556	0.930	0.355	0.122	0.406
Lsd (%1)	0.644	0.942	0.765	1.282	0.488	0.169	0.559
Genotype	MH	YI	YSI	DI	ATI	SSPI	SNPI
YOSEF	4.95(6)	1.07(4)	0.81(3)	0.86(3)	3.94(3)	9.40(3)	10.18(3)
MBD-85-3	3.98(8)	0.80(9)	0.69(7)	0.56(9)	4.45(4)	13.11(5)	6.76(9)
MBD-85-6	4.62(7)	0.90(7)	0.63(9)	0.59(8)	7.57(8)	19.53(8)	7.57(8)
MBD-85-8	5.13(3)	1.01(6)	0.66(8)	0.67(6)	8.65(9)	19.58(9)	8.44(6)
MBD-85-14	3.73(9)	0.80(8)	0.81(2)	0.65(7)	2.21(1)	7.05(2)	7.82(7)
MB-85-3	5.28(2)	1.08(3)	0.72(6)	0.79(4)	6.86(7)	15.28(7)	9.34(4)
MB-85-5	5.08(4)	1.13(2)	0.88(1)	0.99(1)	2.53(2)	5.92(1)	12.36(1)
MB-85-18	5.01(5)	1.04(5)	0.75(5)	0.78(5)	5.58(5)	13.10(4)	9.33(5)
NOSRAT	5.43(1)	1.14(1)	0.76(4)	0.88(2)	6.31(6)	13.62(6)	11.52(2)
Lsd (%5)	0.464	0.164	0.145	0.251	3.247	8.207	3.351
Lsd (%1)	0.640	0.226	0.199	0.346	4.474	11.31	4.618

Clustering

Ward's hierarchical clustering for grouping genotypes based on ranks of drought resistance indices and yield of stress and non-stress conditions (Figure 1), were confirmed the results of mean comparison, consequently based on this groping three distinctive group was discussible. The first group consists of genotypes, MB-85-3, MBD-85-8 and Nosrat which had a desirable resistance to drought based on MP, STI, GMP MH and YI, also this group had the highest yield for both growth conditions. The second group including genotypes MB-85-18, MB-85-5, and Yosef, which had a desirable resistance to drought based on SSI, TOL, YSI, DI, ATI, SSPI and SNPI, also this group had the moderate yield in two environments. The lowest group for measured indices and grain yield in two environments was the third group, which consists of MBD-85-3, MBD-85-6 and MBD-85-14, which had a lowest resistance to drought based on most indices, also this group had the lowest yield in two environments.

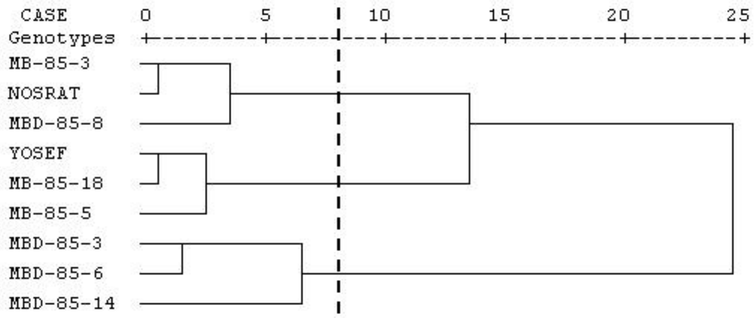

Figure 1. Dendrogeram of Ward's hierarchical clustering of genotypes ranks based on drought resistance indices and yield of stress and non-stress conditions

Biplot analysis

Principal component analysis for two way table of genotypes ranks for drought resistance indices and grain yield in two conditions showed that the first component explained 54.35% of the variation in the data matrix and had a high correlation among Y_S, Y_P with MP, GMP, STI, MH, DI and SNPI indices thus, the first component can be named the yield potential component which separates the high yielder from the low yielder (Fernandez, 1992). The second component explained 39.50% of total variability and had a high positive correlation among TOL, SSI, YSI, ATI and SSPI; therefore, the second component can be named as the stress resistant component, which separates the drought resistant genotypes (Fernandez, 1992). Biplot for the first two components were properly explained and confirmed the results of genotypes grouping based on cluster analysis and relationship among drought resistance attributes with Y_s and Y_p (Figure 2), thus the genotypes MBD-85-8, MB-85-3 and Nosrat had the most desirable performance for yield and drought resistance (group 1). The genotypes MB-85-18, Mb-85-5 and yousef had a desirable resistance to drought, but don't have a desirable production of grain yield (had a moderate grain yield). On the other hand, relationship among, MP, GMP, MH and STI with Y_s and Y_p were properly illustrated, considering the angles and the direction between vectors of these attributes, beside relation among ATI, TOL, SSPI, SNPI, YSI, SSI, YI and DI with Y_s were revealed by biplot. We finding that MP, GMP, MH and STI are desirable indices for selection drought resistant genotypes and had a strength correlation with two growth conditions and can are use for selection in barely genotypes with high drought resistance and high grain yield.

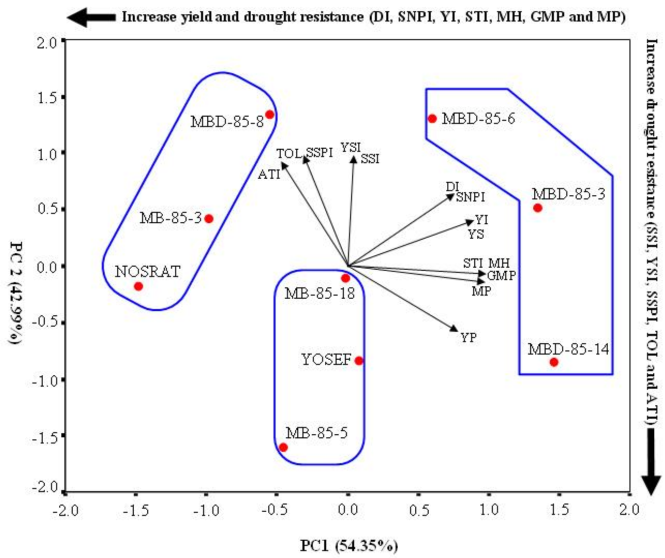

Figure 2. Biplot of drought resistance indices in barely lines based on two first components value and cluster grouping for genotypes

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