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Screening of Iranian Cumin (*Cuminum cyminum* L.) Ecotypes under Normal Moisture and Drought Conditions using Tolerance Indices

Azadeh Karimi Afshar[1]*, Amin Baghizadeh[2], Ghasem Mohammadi-Nejad[3]

[1]*Department of Plant Breeding, Graduate University of Advanced Technology, Kerman, Iran.*
[2]*Department of Biotechnology, Institute of Science and High Technology and Environmental Sciences, Graduate University of Advanced Technology, Kerman, Iran.*
[3]*Horticultural Research Institute, Shahid Bahonar University of Kerman, Kerman, Iran.*

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Abstract

Cumin is one of the most agriculturally valuable plants, in the semi-arid tropical regions of Iran. In this research, drought tolerance of 49 cumin ecotypes were evaluated under irrigated, and rained conditions in the field during two years (2010 and 2011). Five drought tolerance/susceptibility indices including mean productivity (MP), geometric mean productivity (GMP), stress tolerance index (STI), tolerance (TOL) and susceptible stress index (SSI) were applied. Results of combined analysis based on the experiments showed a significant variation among ecotypes for grain yield and, it was decreased due to drought stress. The mean grain yield of Ardestan and Ravar in normal and drought stress conditions possessed the highest values respectively. According to the results derived from principal component analysis, bi-plot display and STS equation, Ravar was identified as the most drought tolerant ecotype. In conclusion, this suitable ecotype could be recommended for cropping in regions with limited water resources, also MP, GMP and STI indices were found to be more effective in identifying drought-tolerant and high yielding ecotypes in both conditions.

1. Introduction

Cumin (*Cuminum cyminum* L.) as a herbaceous, annual medicinal plant, is one of the most important export crops for countries such as India, Iran and some other Asian countries (Kafi., 2002). In arid and semi-arid area such as Iran, among the different environmental constraints, the drought of an area is the most limiting factor for farming. Basic risk management in agriculture includes choosing plant against adverse weather events. Cumin has a potential to be a rainfed crop, but supplemental irrigation is needed to produce more (Rezaei Nejad., 2011). Yield under drought-prone environment may be considered to be affected by three components including yield potential, appropriate phenology and, drought tolerance (Ouk *et al*., 2006). Drought tolerance is defined as the ability of plants to live, grow and reproduce satisfactory with limited water supply or under periodic conditions of water deficit. Drought susceptibility of a genotype is often measured by reduction in yield under drought stress (Turner., 1979; Blum., 1998). However, breeding for drought tolerance is particularly challenging because of the genetic complexity of this trait (Cattivelli *et al*., 2008). To have high and durable yield under drought-prone environments, drought tolerant genotypes are needed (Abdoshahi *et al*., 2013). Different genotypes may have different responses to drought stress. Several indices have been utilized to evaluate genotypes for drought tolerance on the basic of a mathematical relationship between different grain yield in different environments (Mohammadi *et al*., 2011). Rosielle and Hamblin (1981) defined stress tolerance (TOL) as a difference in yield, between the stress (Ys) and non-stress (Yp) environments and mean productivity (MP) as the average yield of Ys and Yp. The geometric mean is often used by breeders interested in relative performance, since drought stress can vary in severity in field environments over years (Ramirez & Kelly., 1998). Fisher and Maurer (1978) proposed a stress susceptibility index (SSI) for genotypes. Fernandez (1992) defined an advanced index (STI= stress tolerance index), which can be used to identify genotypes

*Corresponding Author: Department of Plant Breeding, Graduate University of Technology, Kerman, Iran
E-mail Address: Azadehkarimi.a@gmail.com.

that produce high yield under both stress and non-stress conditions. The optimal selection criterion should distinguish genotypes that express uniform superiority in both stressed and non-stressed environments from the genotypes that are favorable only in one environment (Fernandez., 1992). The main objects of this study were to identify drought tolerant genotypes, suitable for dry land regions of Iran and also to discover high-yielding genotypes in drought stress and normal conditions.

2. Material and Methods

The field experiments were carried out in the private research field of Kerman in two years (2010 and 2011). The average annual rainfall is 245.9 mm in the region. During the period of this research, climatic conditions were characterized by an annual average temperature of 15.1 ˚C and 16.3 ˚C and average annual rainfall of 260.9 and 177.9 mm in 2010 and 2011, respectively (Table 1). In split plot arrangement based on a randomized complete blocks design, two separate experiments including rained (drought condition) and supplemental irrigation were considered as the main factor levels, while forty-nine sub-population cumin ecotypes belonged to nine populations from different provinces of Iran, were arranged to sub plots (Table 2).

Table 1 Mean daily temperature and total rainfall during the experiments.

	Jan	Feb	Mar	Apr	May
2010					
Mean air Temperature (˚C)	3.3	6.5	10.5	11.5	17.5
Rain(mm)	16	19.5	47.3	100	27.6
2011					
Mean air temperature(˚C)	5.5	4.4	9.2	15.3	20.2
Rain(mm)	13.11	47.7	41.2	20.5	39.3

Planting time was in 16th January 2010 and 2011 in first and second years, respectively. The genotypes were planted in plots of 2 m long. There was 40 cm row spacing and the distance between plants was 4cm.

Table 2 List of 49 studied cumin ecotypes/sub-populations from 9 different provinces of Iran

NO.	Populations	Ecotype	NO.	Populations	Ecotype
1	Fars	Sarvestan	26	South khorasan	Birjand
2	Fars	Sepidan	27	South khorasan	Sarayan
3	Fars	Sivand	28	South khorasan	Darmian
4	Fars	Estahban	29	Esfahan	Feridan
5	Yazd	Ardekan	30	Esfahan	Semirom
6	Yazd	Bafq	31	Esfahan	Ardestan
7	Yazd	Sadoq	32	Esfahan	Naien
8	Yazd	Khatam	33	Esfahan	Khansar
9	Yazd	Sadroea	34	Esfahan	Natanz
10	Golestan	Maraveh-Tapeh	35	Semnan	Shahmirzad
11	Golestan	Aq-Qala	36	Semnan	Sorkheh
12	Golestan	Jat	37	Semnan	Ivanaki
13	Golestan	Gonbad	38	Semnan	Kalateh
14	Kerman	Baft	39	North Khorasan	Esfarayen
15	Kerman	Bardsir	40	North Khorasan	Shirvan
16	Kerman	Chatrood	41	North Khorasan	Bojnord
17	Kerman	Joopar	42	North Khorasan	Baneh
18	Kerman	Kooh-banan	43	Razavi Khorasan	Gonabad
19	Kerman	Mahan	44	Razavi Khorasan	Ferdows
20	Kerman	Ravar	45	Razavi Khorasan	Torbat- Heidareh
21	Kerman	Rafsanjan	46	Razavi Khorasan	Torbat-Jam
22	Kerman	Sirjan	47	Razavi Khorasan	Kashmar
23	Kerman	Zarand	48	Razavi Khorasan	Taybad
24	South khorasan	Qaen	49	Razavi Khorasan	Bardsekan
25	South khorasan	Nahbandan			

Both normal and stressed experiments were watered at sowing time. Plants in normal condition were irrigated until, they reached physiological maturity. Irrigation was terminated for plants in drought stress condition, before 50% of plants in each plot reached flowering stage. Therefore, plants in drought stress condition received less water in both years. Plants were harvested after removal of edge effect and grain yield were measured. Drought tolerance/susceptibility indices were calculated for each genotype using the following relationships:

- Stress Susceptibility Index (SSI)= $\frac{1-\frac{Ys}{\bar{Y}s}}{1-\frac{Ys}{\bar{Y}p}}$ (Eq. 1)
- Mean Productivity (MP)= $\frac{Ys+Yp}{2}$ (Eq. 2)
- Tolerance (TOL)= $Yp - Ys$ (Eq. 3)
- Stress Tolerance Index (STI)= $\frac{Yp \times Ys}{\bar{Y}_p^2}$ (Eq. 4)
- Geometric Mean Productivity (GMP)= $\sqrt{Yp \times Ys}$ (Eq.5)
- Stress Tolerance Score (STS)=MP+STI+GMP - SSI-TOL (Eq. 6)

Where Ys, $\bar{Y}s$, Yp, and $\bar{Y}p$ are grain yield and the mean yield of all genotypes under drought stress and normal conditions, respectively. Analysis of variance, correlation between different indices, Ys, Yp and principal component analysis based on correlation matrix of genotypes was computed by SAS ver9.1.

3. Results

Grain yield under irrigated condition showed positive significant correlation with rained condition (r=0.3*) (Table 4).

Among all genotypes over two years, Ardestsn and Ravar with averages 1265.7 and 1152.2 kg/ha had the highest grain yield; also Khatam and Naien with averages 1036.6 and 1011.2 kg/ha produced the lowest in normal and stress conditions, respectively (Table 5). To identify tolerant genotypes some drought tolerance/susceptibility indices including STI, MP, GMP, SSI and TOL were calculated on the basis of grain yield in normal and stress conditions over two years. In this research GMP had the highest correlation with Yp and Ys (r= $0.9^{**}$and r=0.7^{**} respectively). There were high and significant correlations between TOL and SSI also MP, GMP and STI indices, that had positive significant correlation with each other, Y_S and Y_P. (Table 4).Therefore the results showed GMP, STI and MP indices will produce similar results (Table 5). Based on ranking of MP, GMP and STI indices, Ardestan and Ravar had the best performance and showed the highest value (Table 5).

Table 3 Mean squares for grain yield (kg.h^{-1}) based on combined analysis

SOV	DF	Grain yield (kg.h^{-1})
Year	1	24304.5^{ns}
Irrigation	1	779713.7*
Irrigation* Year	1	327603.8**
Genotype	48	63675.2*
Irrigation*Genotype	48	71207.8**
Year*Genotype	48	66985.9*
Year* Irrigation *Genotype	48	76192.1**

^{ns}, * and **: Non Significan , Significant at the probability levels of 5% and 1%

Table 4 Simple correlation coefficients between Yp, Ys and drought tolerance/susceptibility indices of 49 cumin ecotypes

	Ys	Yp	TOL	SSI	MP	GMP
Yp	0.3*	-	-	-	-	-
TOL	-0.3**	0.8**	-	-	-	-
SSI	-0.32*	0.7**	0.9**	-	-	-
MP	0.69**	0.89**	0.46**	0.43**	-	-
GMP	0.7**	0.9**	0.45**	0.42**	0.99**	-
STI	0.69**	0.89**	0.45**	0.43**	0.99**	1**

* and ** : significant at 0.05 and 0.01 probability level, respectively

Whereas, a larger value of TOL and SSI represent relatively more sensitivity to stress; thus a smaller value of TOL and SSI are favorable. The lowest value of SSI and TOL was assigned to Khatam and Sadoq. Principal component analysis (PCA) technique decreased five variations into two components. The first two components in total, explained 99.2 percent of the variation between the data (Table 6). Thus, bi-plot was drawn based on the first two components. The first component (PC_1) justified 69.5 percent of variation in the matrix of data and showed a high coordination with yield in both environments and MP, GMP and STI indices. Therefore, it was named as high yield and stress tolerance component.

Table 5 Estimation of stress tolerance indices from the potential yield and the stress yield data for 49 cumin ecotypes

ecotypes	Yp (kg.ha^{-1})	Ys (kg.ha^{-1})	STI	GMP	MP	SSI	TOL	STS
Sarvestan	1057.5	1081.8	0.93[40]	1069.56[40]	1069.63[4]	-0.77[4]	-24.3[4]	0.003[26]
Sepidan	1115.9	1032.0	0.93[38]	1073.12[38]	1073.94[37]	2.51[45]	84.0[43]	-4.417[45]
Sivand	1173.1	1105.5	1.05[3]	1138.77[3]	1139.27[3]	1.92[38]	67.6[39]	2.914[4]
Estahban	1051.1	1057.4	0.90[47]	1054.23[47]	1054.23[47]	-0.20[9]	-6.3[9]	-2.324[41]
Ardekan	1195.7	1069.7	1.03[6]	1130.93[6]	1132.68[6]	3.51[48]	125.9[48]	-0.284[28]
Bafq	1128.4	1074.9	0.98[23]	1101.33[23]	1101.66[23]	1.58[34]	53.5[34]	-0.280[27]
Sadoq	1037.6	1092.2	0.92[42]	1064.56[42]	1064.91[42]	-1.75[2]	-54.6[2]	0.909[21]
Khatam	1036.6	1093.8	0.92[41]	1064.82[41]	1065.20[41]	-1.84[1]	-57.2[1]	1.057[20]
Sadroea	1048.3	1029.3	0.87[49]	1038.75[49]	1038.80[49]	0.61[21]	19.1[20]	-4.992[47]
Maraveh-Tapeh	1132.6	1071.8	0.98[22]	1101.80[22]	1102.22[22]	1.79[35]	60.8[36]	-0.548[29]
Aq-Qala	1089.8	1030.9	0.91[44]	1059.93[44]	1060.34[44]	1.80[35]	58.8[35]	-4.655[46]
Jat	1104.0	1110.2	0.99[17]	1107.09[17]	1107.09[17]	-0.19[37]	-6.2[10]	2.897[5]
Gonbad	1099.2	1085.8	0.97[24]	1092.49[24]	1092.51[24]	0.40[17]	13.3[17]	0.577[23]
Baft	1104.5	1100.5	0.98[21]	1102.48[21]	1102.49[21]	0.12[15]	4.0[15]	1.986[11]
Bardsir	1098.4	1066.0	0.95[31]	1082.11[31]	1082.23[31]	0.98[27]	32.4[25]	-1.294[36]
Chatrood	1166.0	1103.2	1.04[4]	1134.17[4]	1134.61[4]	1.80[36]	62.9[37]	2.645[7]
Joopar	1044.4	1066.7	0.90[46]	1055.52[46]	1055.58[46]	-0.71[5]	-22.3[5]	-1.466[38]
Kooh-Banan	1117.5	1050.7	0.95[30]	1083.57[30]	1084.09[30]	1.99[39]	66.8[38]	-2.634[42]
Mahan	1112.0	1072.5	0.96[25]	1092.06[25]	1092.24[25]	1.19[28]	39.6[28]	-0.612[32]
Ravar	1173.3	1152.2	1.09[2]	1162.70[2]	1162.75[2]	0.60[20]	21.1[21]	7.315[1]
Rafsanjan	1141.4	1126.4	1.04[5]	1133.86[5]	1133.88[5]	0.44[18]	15.0[18]	4.654[3]
Sirjan	1174.9	1088.6	1.03[7]	1130.92[7]	1131.74[7]	2.45[43]	86.3[44]	1.338[16]
Zarand	1080.1	1095.2	0.96[26]	1087.61[26]	1087.64[27]	-0.47[6]	-15.1[6]	1.357[15]
Qaen	1072.7	1048.1	0.91[43]	1060.34[43]	1060.41[43]	0.76[23]	24.5[23]	-3.109[43]
Nahbandan	1070.0	1070.3	0.93[39]	1070.17[39]	1070.17[39]	-0.01[13]	-0.3[13]	-1.028[35]
Birjand	1059.9	1019.1	0.87[48]	1039.28[48]	1039.48[48]	1.28[30]	40.8[29]	-5.911[48]
Sarayan	1166.0	1086.7	1.02[9]	1125.64[9]	1126.34[9]	2.27[41]	79.4[41]	1.086[18]
Darmian	1085.7	1087.1	0.95[28]	1086.41[28]	1086.41[28]	-0.04[12]	-1.4[12]	0.627[22]
Feridan	1125.6	1133.6	1.03[8]	1129.63[8]	1129.64[8]	-0.24[8]	-8.0[8]	5.227[2]
Semirom	1146.5	1066.3	0.99[18]	1105.69[18]	1106.42[18]	2.33[42]	80.2[42]	-0.972[34]
Ardestan	1265.7	1081.6	1.11[1]	1170.05[1]	1173.67[1]	3.85[49]	184.1[49]	1.475[13]
Naien	1102.4	1011.2	0.90[45]	1055.81[45]	1056.80[45]	2.76[47]	91.3[47]	-6.461[49]
Khansar	1054.6	1093.6	0.93[36]	1073.96[36]	1074.14[35]	-1.23[3]	-39.0[3]	1.104[17]
Natanz	1098.0	1071.3	0.95[29]	1084.54[29]	1084.62[29]	0.81[24]	26.7[24]	-0.803[33]
Shahmirzad	1146.2	1100.4	1.02[11]	1123.06[11]	1123.30[11]	1.33[31]	45.9[31]	2.235[10]
Sorkheh	1136.7	1103.9	1.01[12]	1120.17[12]	1120.29[12]	0.96[25]	32.8[27]	2.504[9]
Ivanaki	1102.0	1106.9	0.99[19]	1104.44[19]	1104.44[19]	-0.15[11]	-4.9[11]	2.572[8]
Kalateh	1081.0	1065.6	0.93[37]	1073.30[37]	1073.33[38]	0.48[19]	15.4[19]	-1.421[37]
Esfarayen	1080.3	1074.6	0.94[33]	1077.47[33]	1077.48[33]	0.18[16]	5.7[16]	-0.575[31]
Shirvan	1077.9	1075.0	0.94[34]	1076.45[34]	1076.45[34]	0.09[14]	2.9[14]	-0.553[30]
Bojnord	1126.6	1093.9	1.00[16]	1110.16[16]	1110.28[16]	0.97[26]	32.7[26]	1.501[12]
Baneh	1167.7	1080.3	1.02[10]	1123.14[10]	1123.99[10]	2.50[44]	87.4[45]	0.497[24]
Gonabad	1085.9	1062.4	0.93[35]	1074.05[35]	1074.11[36]	0.72[22]	23.5[22]	-1.706[40]
Ferdows	1124.5	1034.2	0.94[32]	1078.40[32]	1079.34[32]	2.68[46]	90.3[46]	-4.154[44]
Torbat- Heidareh	1114.0	1061.7	0.96[27]	1087.53[27]	1087.85[26]	1.57[33]	52.3[33]	-1.6[39]
Torbat-Jam	1133.8	1092.0	1.00[14]	1112.72[14]	1112.92[14]	1.23[29]	41.8[30]	1.365[14]
Kashmar	1095.5	1110.2	0.98[20]	1102.82[20]	1102.85[20]	-0.45[7]	-14.7[7]	2.848[6]
Taybad	1137.7	1088.7	1.00[13]	1112.92[13]	1113.19[13]	1.44[32]	49.0[32]	1.076[19]
Bardsekan	1149.2	1076.5	1.00[15]	1112.27[15]	1112.87[15]	2.11[40]	72.7[40]	0.01[25]

Table 6 Results of principal component analysis for Yp, Ys and drought tolerance indices on 49 cumin ecotypes

Index	Component 1	Component 2
Yp	0.49	0.13
Ys	0.21	-0.60
TOL	0.30	0.50
SSI	0.29	0.50
MP	0.51	-0.17
GMP	0.50	-0.18
STI	0.50	-0.18
Eigenvalue	4.86	2.12
Percent of variation	69.5	29.7
Cumulative percentage	69.5	99.2

This component separated drought tolerant genotypes with high yield under stress and non-stress environments. The second component (PC_2) justified 29.7 percent of total variation. This component had high positive correlation with TOL, SSI indices and Yp. Thus, it was called as stress susceptibility component. This component separated genotypes with low and high difference yield in stress condition.

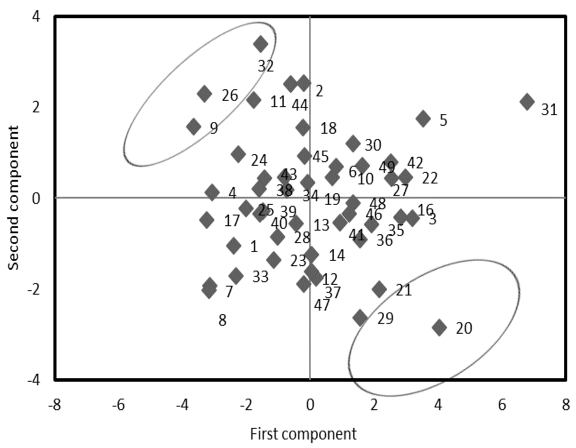

Fig. 1 Drawing bi-plot based on first and second components for 49 cumin genotypes.
(Numbers in the figures show the genotype position in the bi-plot)

Regarding the results of bi-plot display based on two first components (Fig. 1), G_{20} (Ravar), G_{21} (Rafsanjan), G_{29} (Feridan) were identified as tolerance genotypes due to located in yield potential and drought tolerance region (Fig. 1: bottom right) these genotypes had high value of PC_1 and low value of PC_2. G_9 (Sadroea), G_{32} (Naien), G_{26} (Birjand) were identified as drought sensitive genotypes, due to located in sensitive to drought stress and low yield region (Fig. 1: top left).
The ranking genotypes based on STS which is proposed by (Abdolshahi *et al.*, 2013) and results based on PCA were very similar (Table 4). According to bi-plot analysis, these genotypes (G_{20}, G_{21}, G_{29}) were located in the potential yield and drought tolerance region, have the highest and, (G_{32}, G_{26}, G_9) have the lowest value of STS index, respectively.

4. Discussion

A variety of approaches have been used to alleviate the problem of drought. Plant breeding, either conventional breeding or genetic engineering, seems to be an efficient and economic means of crops to enable them to grow successfully in drought-prone environments (Ashraf., 2010) .Breeders have made noticeable improvements in introduction of stress tolerance based on morphological traits on field experiments. Improvement in adaptation of cumin to drought stress environment has been largely achieved through field-based selection for stress tolerance. Significant difference between grain yield in normal and stress conditions demonstrated existence of high diversity between genotypes for drought tolerance and, possibility of selection for favorable genotypes in both environments (Table 5). Significant correlation between grain yield in normal and drought stress conditions show possibility of selection for favorable genotypes in both environments (Table 4). Fernandez (1992) believed that the most suitable index for selecting stress-tolerant genotypes is an index, which has a relatively strong correlation with the seed yield under stress and non-stress conditions. Therefore MP, GMP and STI are introduced as the best indices which highly correlated with grain yield in both environments. These indices are acceptable to screen drought-tolerant, high yielding genotypes in both drought-stressed and irrigated conditions. This result are in consistent Fernandez., 1992; Ramirez & Kelly., 1998, Sio Se- Mardeh *et al.*, 2006; Sanjari Pireivatlou & Yazdansepas., 2008; Jafari et al., 2009; Talebi *et al.*, 2009; Mohammadi *et al.*, 2010; Nouri *et al.*, 2011; Karimizadeh & Mohammadi., 2011 and Abdoshahi *et al.*, 2013. Based on STI, MP and GMP, Ardestan and Ravar were the most droughts tolerant which had the highest yield potential among genotypes in normal and stress conditions (Table 5). Whereas TOL and SSI had succeeded to screening genotypes with high and low yield under normal condition (Ardeatan and Khatam, respectively), but had failed to select genotypes with superior yield under both environments. This finding is in agreement with the results of Rosielle & Hambelen., 1981; Mohammadi *et al.*, 2011 and Jafari *et al.*, 2009. Considering the results of PCA and bi-plot display based on first two components G_{20} (Ravar), G_{21} (Rafsanjan), G_{29}

(Feridan) introduced as the most drought tolerant genotype, whereas G_9 (Sadroea), G_{32} (Naien), G_{26} (Birjand) were identified as drought sensitive genotypes. These extreme genotypes could be suitable parental in breeding program for improving stress tolerance and it could be possible to obtain drought tolerance lines, also they are valuable resources for identification of genes responsible for drought tolerance in molecular plant breeding. In conclusion, Ravar ecotype with high yield potential and ability to tolerate drought stress, were identified as a suitable genotype for introduction to farmers in dry land special in Kerman province.

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