Evaluation of Genetic Variation for Drought Tolerance and Determination of the Best Selection Criteria in Safflower Genotypes (Carthamus tinctorius L.)

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ABSTRACT

In order to evaluate genetic variation and drought tolerance of safflower cultivars (Carthamus tinctorius L.), an experiment was conducted using fifteen cultivars in a randomized complete block design with three replications under drought and normal conditions during 2014-2015 growing season. Drought tolerance indices, such as tolerance (TOL), stress tolerance index (STI), stress susceptibility index (SSI), mean productivity (MP) and geometric mean productivity (GMP) were calculated to distinguish cultivars based on seed yield under both moisture regimes. The correlation coefficients illustrated that STI and GMP were the best and efficient selection criteria to distinguish drought tolerant and high-yielding cultivars. Significant and positive correlation was found between yield in both stress and normal conditions with GMP, MP and STI. Principal component analysis (PCA) showed that first and second PC accounted for 97.1% of the total variation. Biplot graphical display represented that lines 2, 11, 14 and 15 were highly adapted to the both normal, stress conditions, and classified them in high-yielding and drought tolerant groups, while genotypes numbered as 10, 12 and 13 were potential and stable under normal condition. Based on data analysis, cultivars numbered as 1, 5, 6 and 9 had lowest yield under both moisture regimes, lines 3, 4, 7 and 8 showed high-yielding under stress regimes. Cluster analysis ordered the genotypes into six groups with 5, 3, 2, 2, 2 and 1 genotypes, respectively. In conclusion, present investigation revealed that drought conditions induced reduction of yield of some cultivars, while others were tolerant to drought stress. Hence, breeders can select drought tolerant safflower lines based on the GMP and STI indices.

Key words: Safflower, Drought tolerance indices, Genetic improvement, Biplot, Cluster analysis.

INTRODUCTION

Safflower (Carthamus tinctorius L.) is one of the seed oil crops grown in Iran. It is one of the plants, which have a high conformity to various conditions such as resistance to drought, and it is appropriated to be grown in arid and semi-arid areas. Due to the growing request for edible oils, improvement of oilseed crop is very important (Safavi et al., 2013; Rameshknia et
In normal conditions, plants are subjected to various stress factors with harmful influence on growth and crop production (Roudbari et al., 2012). Drought as an environmental stress is the limitation that induces a highly negative effect on yield (Khalili et al., 2014). Drought tolerant is an important characteristic for increasing and enhancing crop efficiency in dry regions (Guo et al., 2009). Recognition of the important yield component is very efficient in genetic programs of these traits via indirect selection (Golparvar, 2011). The identified genes from wild plant species provide a mean for sustaining genetic improvement in plants cultivated in dry regions. The cultivated lines tolerated less drought stress than wild plants and fluctuating water stress levels caused meaningful more declines in the seed yield of cropping genotypes as compared with wild genotypes (Majidi et al., 2011). The inheritance of agronomic traits was studied in safflower under drought stress condition. In order to improve seed yield and seed yield of safflower under drought regimes, obtained outcomes could be suitable for designing of breeding programs (Mirzahashemi et al., 2014). Detection of the drought tolerance genes in barley (Hordeum Vulgare L.) will facilitate the molecular mechanisms conception of drought tolerance, and also facilitate the genetic breeding of barley via marker-assisted selection or transformation of genes. These results showed that new understanding into further comprehension of drought tolerance procedures in barley plants could be provided (Guo et al., 2009). Various plants react differently to drought stress. Drought condition induced varied molecular and physiological responses such as changes in gene expression in plants (Savitri et al., 2013). Shiranirad et al. (2011) announced that drought is a common obstacle seriously influencing rapeseed (Brassica napus L.) production, mostly in arid region in the world. They observed that MP, GMP and YI parameters were the best for screening high seed yield genotypes under stress conditions. Farshadfar et al. (2013) cleared that grain yield of bread wheat in normal and stress regimes were significantly and negatively correlated with SSI. Their findings indicated that some indices such as RDI, ATI, SNPI and DI can be used as the most favorable indicators for identifying drought tolerant genotypes. Cluster analysis of mentioned investigation classified the cultivars into three groups including; tolerant, susceptible and semi tolerant or semi-sensitive to drought regimes. In order to assess of drought tolerance indices under different environmental conditions for screening of Turkish oat (Avena sativa L.), fourteen landraces and cultivars were used. The experiments were applied both under rain-fed and irrigated regimes for three cropping seasons. Correlation coefficient matrix showed that the drought parameters were significantly inter-correlated with each other and can be classified into four groups. Their results demonstrated that the STI, GMP, MP, YI and HM indices under dry and irrigated conditions can be suggested to screen drought tolerant cultivars with high-yielding potential (Akcura et al., 2011).

**MATERIALS AND METHODS**

A field trial was carried out during 2014-2015 at the Isfahan (Khorasgan) Branch, Islamic Azad University, research station (50° 44’ N, 32° 40’ W and altitude of 1517 m above mean sea level). The study location is characterized by arid climate with an annual average rainfall of 120 mm, and the annual mean maximum and minimum temperatures of 25 °C and 1 °C, respectively. Soil type of the study site was silty loam and soil pH was 7.7 to 8. Generally, there is no precipitation during safflower growth cycle in this region.

Fifteen spring safflower cultivars including U.S.10, Kuseh landrace, Nebraska-10, Gila, S149, Bushehr landrace, Shiraz landrace, Arak-2811, Kerman landrace, Isfahan landrace,
C111, Lordegan landrace, S3110, A.C.Sterling and Semnan landrace were planted at first of March 2012. The plots comprising of three rows were 3 m long and 0.5 m apart. Interplants distance within rows was 5 cm, hence, seedling density was 400/000 plants ha$^{-1}$. The experiment was watered at planting and flowering stages. Irrigation regimes were started at emerging of seedling. Two irrigation programs were considered in this study: IR1, irrigation after 75 mm cumulative evaporation from class A evaporation pan (CE) during the whole growth cycle as optimum irrigation treatment. IR2, irrigation after 150 mm cumulative evaporation from class A evaporation pan (CE) during the whole growth cycle as stress treatment. Various drought tolerance indices were evaluated (Table 1).

<table>
<thead>
<tr>
<th>Code</th>
<th>Drought tolerance indices</th>
<th>Equation*</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Stress Susceptibility Index (SSI)</td>
<td>$SSI = \frac{1-(Y_s / Y_p)}{1-(Y_s / Y_p)}$</td>
<td>Fischer and Maurer, 1978</td>
</tr>
<tr>
<td>2</td>
<td>Geometric Mean Productivity (GMP)</td>
<td>$GMP = \sqrt{Y_s \times Y_p}$</td>
<td>Fernández et al., 1992</td>
</tr>
<tr>
<td>3</td>
<td>Stress Tolerance Index (STI)</td>
<td>$STI = \frac{Y_s \times Y_p}{Y_p}$</td>
<td>Fernández et al., 1992</td>
</tr>
<tr>
<td>4</td>
<td>Mean Productivity (MP)</td>
<td>$MP = \frac{Y_s + Y_p}{2}$</td>
<td>Rosielle and Hambling, 1981</td>
</tr>
<tr>
<td>5</td>
<td>Tolerance Index (TI)</td>
<td>$TOL = Y_p - Y_s$</td>
<td>Rosielle and Hambling, 1981</td>
</tr>
</tbody>
</table>

* $Y_s$ and $Y_p$ are seed yield in stress and normal conditions, respectively.

The research was conducted in two independent randomized complete block design (as stress and non-stress conditions) with three replications in each experiment. Analysis of variance and Duncan’s multiple range test was utilized for mean comparisons were applied using proc GLM procedure of SAS software (version 9.2, SAS institute Inc., NC, USA). Correlation analysis, principal component analysis (PCA) and biplot graphical display were done by using STATGRAPHICS PLUS software while cluster analysis based on Ward’s method was carried out by SAS9.2 software.

RESULTS AND DISCUSSION

Yield ranged from 3534 kg ha$^{-1}$ (cultivar S149) to 1366 kg ha$^{-1}$ (cultivar Arak-2811) in non stress treatment (Yp) (Table 2). The values of yield under stress conditions varied from 1326 to 1966 kg ha$^{-1}$ and the Semnan and Lordegan landraces had lower seed yields Ys. Gila, U.S.10 and Nebraska-10 showed higher yield (1966, 1946 and 1934 kg ha$^{-1}$, respectively) in Ys non-stress. Mean yields under and stress conditions, were 2253 and 1657 kg ha$^{-1}$,
respectively revealing a reduction of 27% compared to normal irrigation conditions (data not shown).

Table 2. Average values of drought tolerance indices in safflower cultivars

<table>
<thead>
<tr>
<th>Code</th>
<th>genotype</th>
<th>Yp</th>
<th>Ys</th>
<th>TOL</th>
<th>MP</th>
<th>SSI</th>
<th>GMP</th>
<th>STI</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Esfahan landrace</td>
<td>1714</td>
<td>1472</td>
<td>242</td>
<td>1593</td>
<td>0.52</td>
<td>1588.39</td>
<td>0.49</td>
</tr>
<tr>
<td>2</td>
<td>Kuseh landrace</td>
<td>2392</td>
<td>1740</td>
<td>652</td>
<td>2066</td>
<td>1.01</td>
<td>2040.11</td>
<td>0.81</td>
</tr>
<tr>
<td>3</td>
<td>Arak-2811</td>
<td>1366</td>
<td>1686</td>
<td>-320</td>
<td>1526</td>
<td>-0.87</td>
<td>1517.58</td>
<td>0.45</td>
</tr>
<tr>
<td>4</td>
<td>Nebraska-10</td>
<td>1838</td>
<td>1934</td>
<td>-96</td>
<td>1886</td>
<td>-0.19</td>
<td>1885.38</td>
<td>0.69</td>
</tr>
<tr>
<td>5</td>
<td>Semnan landrace</td>
<td>1566</td>
<td>1326</td>
<td>320</td>
<td>1526</td>
<td>0.57</td>
<td>1441.01</td>
<td>0.4</td>
</tr>
<tr>
<td>6</td>
<td>Lordegan landrace</td>
<td>1494</td>
<td>1326</td>
<td>168</td>
<td>1410</td>
<td>0.42</td>
<td>1407.49</td>
<td>0.39</td>
</tr>
<tr>
<td>7</td>
<td>Bushehr landrace</td>
<td>1496</td>
<td>1846</td>
<td>-350</td>
<td>1671</td>
<td>-0.87</td>
<td>1661.81</td>
<td>0.54</td>
</tr>
<tr>
<td>8</td>
<td>Shiraz landrace</td>
<td>2146</td>
<td>1680</td>
<td>466</td>
<td>1913</td>
<td>0.81</td>
<td>1898.75</td>
<td>0.7</td>
</tr>
<tr>
<td>9</td>
<td>Kerman landrace</td>
<td>2234</td>
<td>1360</td>
<td>874</td>
<td>1797</td>
<td>1.46</td>
<td>1743.05</td>
<td>0.59</td>
</tr>
<tr>
<td>10</td>
<td>A.C. Sterling</td>
<td>2600</td>
<td>1446</td>
<td>1154</td>
<td>2023</td>
<td>1.66</td>
<td>1938.96</td>
<td>0.74</td>
</tr>
<tr>
<td>11</td>
<td>S3110</td>
<td>3146</td>
<td>1820</td>
<td>1326</td>
<td>2483</td>
<td>1.57</td>
<td>2392.84</td>
<td>1.12</td>
</tr>
<tr>
<td>12</td>
<td>C111</td>
<td>3080</td>
<td>1466</td>
<td>1614</td>
<td>2273</td>
<td>1.96</td>
<td>2124.91</td>
<td>0.88</td>
</tr>
<tr>
<td>13</td>
<td>S149</td>
<td>3534</td>
<td>1754</td>
<td>1780</td>
<td>2644</td>
<td>1.88</td>
<td>2489.7</td>
<td>1.22</td>
</tr>
<tr>
<td>14</td>
<td>U.S.10</td>
<td>2794</td>
<td>1946</td>
<td>848</td>
<td>2370</td>
<td>1.13</td>
<td>2331.76</td>
<td>1.07</td>
</tr>
<tr>
<td>15</td>
<td>Gila</td>
<td>2406</td>
<td>1966</td>
<td>440</td>
<td>2186</td>
<td>0.68</td>
<td>2174.9</td>
<td>0.93</td>
</tr>
</tbody>
</table>

The data indicated that drought stress could significantly reduce yield. The genotypes S149 and S3110 showed repop seed yield under both moisture regimes (Table 2). The values of mean productivity (MP) varied from 1410 kg ha⁻¹ (Lordegan landrace) to 2644 kg ha⁻¹ (line S149) and the genotypes S149, S3110, U.S.10, C111, Gila, Kuseh landrace, A.C. Sterling and Esfahan landrace were the most productive (1955 kg ha⁻¹). Based on geometric mean productivity (GMP), yield varied from 1407.5 kg ha⁻¹ (Lordegan landraces) to 2490 kg ha⁻¹ (line S149), proposing that the genotypes 13, 11, 14, 15, 12, 2 and 10 were the most productive. TOL index varied from -350 to 1780 kg ha⁻¹. Lower or negative TOL indices show tolerance to irrigation stress. Hence, Esfahan, Semnan and Lordegan landraces, Nebraska10, A.C. Sterling, S3110, C111 and S149 were more tolerant (297 kg ha⁻¹). Stability
tolerance index (STI) ranged from 0.39 (Lordegan landrace) to 1.22 (S149). The value of stress susceptibility index (SSI) varied from -0.87 (lines Arak and Bushehr landraces) to 1.96 (line C111). To detect the most desirable drought tolerance measures, correlation coefficient between yields under non-stress and stress conditions, and other quantitative indices of drought tolerance were estimated (Table 3). The outcomes indicated that the indices GMP, MP, STI and SSI were very similar for selection as Yp. This was supported by the high correlations among Yp and SSI (r= 0.84), TOL (r= 0.94), MP (r=0.95), GMP (r= 0.92) and STI (r=0.93). Correlation analysis demonstrated that the indices GMP and STI were similar for selection as Ys. Correlations between yields under stress regimes and GMP (r= 0.58) and STI (r= 0.57) confirmed this conclusion. The indices SSI, TOL and MP illustrated the lowest correlation with Ys (Table 3). Results of Safavi et al. (2013), investigations indicated that significant positive correlation was observed between grain yield in the drought regimes (Ys) with indicator stress tolerance index (STI), harmonic mean (HAR) and geometric mean productivity (GMP) and therefore these indices were suitable criteria for screening stress tolerant cultivars. Majidi et al. (2011), believed that GMP, STI and HM are superior criteria for identifying high yield genotypes under drought and normal regimes. The present results verified significant and positive correlation amongst Yp and Ys with GMP and STI; so these indices may be better predictors of Yp and Ys than MP, SSI and TOL indices. Our findings are in coincident with study of Rameshknia et al. (2013) who believed SSI and TOL indices were the best parameters for identification and screening of genotypes under normal and stress regimes in breeding programs. Safavi et al. (2013), also stated that tolerant index (TOL) and mean productivity (MP) can be regarded as desirable indices for detecting drought tolerant genotypes. Khalili et al. (2014) announced STI, MP, GMP and YI indices were the most appropriate criteria in safflower breeding plans and they revealed that these indices were used for screening high-yielding cultivars under both normal and stress conditions. In assessment of genetic properties of drought tolerance indices of durum wheat, the parameters such as; MP, GMP and STI had high positive genetic correlations with each other as well as with grain yield under stress regime (Ys) and normal condition (Yp). Hence, through these indices it is possible to select high-yielding cultivars in either conditions (Hussain Ali, 2015).

Table 3. Pearson’s correlation coefficients among drought tolerance indices

<table>
<thead>
<tr>
<th>index</th>
<th>Yp</th>
<th>Ys</th>
<th>SSI</th>
<th>TOL</th>
<th>MP</th>
<th>GMP</th>
<th>STI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Yp</td>
<td>1</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ys</td>
<td>0.249*</td>
<td>1</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>SSI</td>
<td>0.849**</td>
<td>0.231ns</td>
<td>1</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>TOL</td>
<td>0.94**</td>
<td>0.095ns</td>
<td>0.955**</td>
<td>1</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>MP</td>
<td>0.956**</td>
<td>0.521*</td>
<td>0.678*</td>
<td>0.799**</td>
<td>1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>GMP</td>
<td>0.927**</td>
<td>0.587*</td>
<td>0.634*</td>
<td>0.746**</td>
<td>0.994**</td>
<td>1</td>
<td></td>
</tr>
<tr>
<td>STI</td>
<td>0.93**</td>
<td>0.574*</td>
<td>0.635*</td>
<td>0.754**</td>
<td>0.993**</td>
<td>0.996**</td>
<td>1</td>
</tr>
</tbody>
</table>

For abbreviations, see Table1.
SSI values changed from -0.69 – 1.54, which were significantly and positively correlated with yield under non-stress and TOL index and negatively correlated with Ys. MP is the mean production under both moisture regimes, and was highly correlated with Yp and TOL indices. Results of Rameshknia et al. (2013) assessment illustrated that STI, GMP and MP indices could screen tolerant and sensitive genotypes under both environmental conditions, and mentioned indices that could be for selection of tolerant cultivars of spring safflower. TOL varied from -350- 1780 kg ha\(^{-1}\). A positive correlation between TOL and Yp (yield under non-stress conditions) and a negative correlation between TOL and yield under water stress (Ys) offered that selection based on TOL indices resulted in reduced yield under optimum irrigation regime. Hussain ali (2015) revealed that the genetic correlation of TOL and SSI indices with yield under stress conditions were high and negative, while correlation coefficient between TOL index and Yp was high and positive. Their findings cleared that selection can be based on TOL index to improve drought tolerance in durum wheat. Our correlations coefficient matrix illustrated that both GMP and STI indices were correlated with yield under both conditions. Moreover, a suitable index must be significantly correlated with yield in any of the two moisture regimes and show a low coefficient of variation. Therefore, these indices can be used to determine drought resistance cultivars with high yield in both moisture regimes. Selection based on a combination of indices may be more useful for improving drought resistance of safflower, but correlation coefficients are helpful for determining the degree of overall linear association between any two attributes (Safavi et al., 2013). Hence, a better approach than a correlation analysis such as biplot analysis is required to identify supreme cultivars for both moisture regimes. For further assessment of relation among drought tolerance indices, principle component analysis was applied. Accordingly, PC, two components accounted for 97.1% of the total variation (Table 4). The results of the principle component analysis of safflower cultivars indicated that the first PC accounted for 77.3% of the total variation, while the second PC justified 19.81% of the remaining variation (Table 4). Also reported that the first component with more than 68% of total variation is able to separate high-yielding and seed yield cultivars from other cultivars.

Table 4. Principal component analysis for drought tolerance indices in safflower cultivars

<table>
<thead>
<tr>
<th>Indices</th>
<th>PC1</th>
<th>PC2</th>
</tr>
</thead>
<tbody>
<tr>
<td>GMP</td>
<td>0.416</td>
<td>0.198</td>
</tr>
<tr>
<td>MP</td>
<td>0.424</td>
<td>0.121</td>
</tr>
<tr>
<td>SSI</td>
<td>0.346</td>
<td>-0.442</td>
</tr>
<tr>
<td>STI</td>
<td>0.416</td>
<td>0.191</td>
</tr>
<tr>
<td>TOL</td>
<td>0.385</td>
<td>-0.361</td>
</tr>
<tr>
<td>Yp</td>
<td>0.42</td>
<td>-0.112</td>
</tr>
<tr>
<td>Ys</td>
<td>0.167</td>
<td>0.76</td>
</tr>
</tbody>
</table>
A biplot diagram from the first and second factor components is shown in Figure 1. The biplot is divided into four classes named A, B, C and D based on the two first principle components. Lines which were located in zone A (2, 14, 15 and 11) demonstrated high yield under both moisture conditions. Hence, these cultivars can be used as tolerant varieties into following breeding procedures for selection of drought tolerant and high-yielding cultivars under stress regime. Lines 13, 10 and 12 which were placed in region B, had suitable potential under both moisture conditions Safavi et al., (2013) believed that some indices such as STI, GMP, HAR and MP were more able to screening drought tolerant varieties and based on correlations between mentioned indices Yp and ys vectors (the angle between the vectors) in the biplot graph, STI was the favorable index for identifying drought tolerant cultivars in safflower.

On the other hand, genotypes that were located in zone D (genotypes 1, 5, 6 and 9) had the lowest yields under stress and normal conditions. Genotype number C area 3, 4, 7 and 8 were located in C area and had low and high yield under normal and stress regimes, respectively. Accordingly, genotypes of the area A was classified threfore high-yielding and drought resistance groups. Majidi et al. (2011) indicated that wild genotypes had a low yield but their seed yield was stable when the environment changed. As these landraces make a favorable genetic source for transferring drought tolerant genes to other genotypes. Cluster analysis of drought tolerance indices classified the mentioned 7 indices into three groups with 4, 2 and 1 indices, respectively (Figure 2). Group 1 consisted of indices with high positive values for first principle components (GMP, STI, MP and Yp indices). These results were verified by the biplot graph analysis which could locate genotypes 15, 14 and 11 with high GMP, STI and MP values into group A. Group 2 included indices with negative SSI and TOL values in second principle components. Ys index (yield under stress conditions) with lowest and positive high values for first and second principle components was located into group 3 (Figure 2).
Cluster analysis based on yield under both moisture regimes and drought tolerance indices classified the cultivars into six groups with 5, 3, 2, 2, 2 and 1 genotype, respectively (Figure 3). Group 3 included genotypes with high Ys, Yp, MP and GMP values, and is considered as a drought tolerant group with high-yielding under normal and stress conditions. Genotypes 14 and 15 (Gila) with high drought resistance and high GMP values were located in the same group. Roudbari et al. (2012) concluded that Gila genotype is more suitable genotype for drought stressed conditions. Grain yield, as a gross selection criterion for drought tolerance, is a complex characteristic that is defined by several metabolic, biochemical and physiological plant operations.

Group 4 which included genotypes 9 and 10 with low seed yield under drought regime. Genotypes 1, 3, 5, 6 and 7 were classified into group 5. These lines showed lower drought tolerance than the genotypes of group 4. The last group consisted of line 12 that had the lowest and high yield under drought and normal conditions respectively, and classified into susceptible group.

Figure 2. Dendrogram from cluster analysis of drought tolerance indices based on WARD’s method

Figure 3. Dendrogram from WARD cluster analysis of safflower cultivars based on drought tolerance indices (See Tables 2 for abbreviations and genotype codes).

noisulcnoc nl, the results of yduts tneserp, showed that moisture regimes had a clear impact on yield of safflower genotypes, so that drought conditions could decline yield up to
1657
kg ha⁻¹. This reduction is 27% compared to the normal treatment. Gila and S149 had higher yields in during stress and normal conditions, respectively. Genotypes Semnan and Lordegan landraces had the lowest seed yield under both moisture conditions. According to the results, GMP and STI were correlated with Yp and Ys, so they were determined as the best drought tolerance indices to select drought tolerant safflower cultivars. Selection based on these indices may be useful for determining a genotype with good seed yield under both stress and normal regimes. We can suggest that the genotypes Gila and U.S.10 can be recommended as candidate cultivars for drought resistance in arid area.

REFERENCES


