



Effectiveness Of Different Methods For Screening Of Sunflower (*Helianthus Annuus L.*) Drought Tolerant Cultivars

Mehdi Ghaffari

Agricultural and Natural Resources Research Station, Khoy, Iran. P.O.Box: 383

Corresponding author: ghaffari@areo.ir

Abstract

Effectiveness of index versus multivariate based methods for screening of sunflower genotypes were compared under drought stress in vegetative, flowering and grain filling stages. Field evaluation was carried out as a strip plot design with three replications at Khoy Agricultural and Natural Resources Research Station in Iran. Flowering stage identified as the most sensitive stage to water deficit with 38% reduction in grain yield compared with normal irrigation. Hybrid Farrokh had the highest seed yield in all irrigation treatments (3686, 2856, 2256 and 2506 Kg/ha in control and water deficit in vegetative, flowering and grain filling stages respectively). The lowest and highest reduction in seed yield was observed in Lakomka and Hysun33 respectively in all drought treatments. According to the stress tolerance and sensitivity indices Farrokh and Hysun33 were the most drought tolerant and sensitive cultivars under all drought treatments respectively. After drought stress in vegetative stage cluster analysis based on all agronomic measurements differentiated Farrokh from others, however there was no singly differentiated cultivar in flowering and seed filling stages. Principle component analysis identified Farrokh as the most drought tolerant while Record as the sensitive cultivars under all drought regimes. Relative water content, head diameter and SPAD value were the main determinant of seed yield under drought stress in vegetative, flowering and seed filling stages respectively. All the methods confirmed Farrokh as the most drought tolerant cultivar, however there were no unique results for identifying of drought sensitive cultivar. Regarding seed yield as the final target it is concluded that principle component analysis merging all plant characteristics can be used as an effective differentiator of genotypes under different water regimes.

Keywords: Cluster analysis, Drought stress, Phenological stages, Principle component analysis

Introduction

Drought stress is a major limitation to agricultural productivity worldwide. About one quarter of world's arable areas is under drought stress (Singh 2000). Sunflower following oil palm, soybean, and rapeseed constitute over 87% of global production of vegetable oils (Murphy 2010). Sunflower is considered moderately tolerant to drought stress however productivity of that is greatly affected by drought (Tahir et al. 2002; Chimenti *et al.* 2002). It is well known that sunflower yield decreases under drought stress (Erdem et al. 2006) but this is dependant to level of water deficit and cultivar (Rodriguez et al. 2002). Drought stress has unfavorable effects on productivity of sunflower (Razi and Assad 1999), however flowering stage is the most sensitive stage to water deficiency (Rauf 2008).

Plant breeders have used different criteria for screening of sunflower genotypes for drought tolerance. The most important of them are stress

susceptibility index (SSI) (Fischer and Maurer 1978) and stress tolerance index (STI) (Fernandez 1992). These indices identify sensitive and resistant genotypes based on only seed yield in both stressed and non stressed condition. Drought tolerance as a complex trait is a function of many plant attributes and there is no single trait conferring drought tolerance to a given genotype. Hence screening method should consider bundle of plant characteristics which confer drought tolerance to plant. In other hand effective selection of desirable genotype needs to understanding of associations among plant characteristics. Multivariate methods such as cluster, path, discriminant and principle component analysis (PCA) provide a base to recognizing intrinsic structures between genotypes based on multivariate traits. Among them PCA biplots provide a definite view of associations among plant characteristics and could differentiate genotypes effectively according to a series of characteristics. Tersac et al. (1993) used this

method to classification of sunflower populations according to the country of origin. De la Vega et al. (2001) reported effectiveness of PCA for revealing genotype - environment interactions. Ghaffari and Farrokhi (2008) used the PCA to reveal two dimensional structures based on general and specific combining abilities in sunflower. In the current study efficiency of different methods was evaluated in screening of sunflower drought tolerant cultivars.

Materials and Methods

Field experiment was carried out at the Khoy Agricultural and Natural Resources Research Station located at 38°32' N and 44°58' E with altitudes of 1103m above sea level. The region is semi-arid with annual precipitation of 296mm. The experimental design was a strip plot design with three replications. Water regimes with 4 levels and 8 sunflower cultivars constitute the factors to be examined (table 1). Drought stress was imposed by water withholding during different growth stages according to the phenological description of Schneiter and Miller (1981). Each experimental plot consisted of 5 rows, 6 m length with 60cm spacing between rows and 25cm within rows. Fertilizers were applied at the rate of 100:70:90 NPK kg/ha. Field practices were followed according to the regional sunflower planting handbook (Ghaffari, 2006)

Morpho- physiological characteristics were measured at the end of drought treatment. Seed yield were measured after physiological maturity. The upper most fully expanded leaves were used for measurement of relative water content (RWC) and leaf temperature. Relative water content was calculated using $RWC = 100 \times (\text{fresh weight} - \text{dry weight}) / (\text{turgid weight} - \text{dry weight})$. Turgid weight was determined after 24 h rehydration at 4°C in a dark room with the leaf discs placed in a container with distilled water and dry weight determined after oven drying for 24 h at 72°C. Stress susceptibility index (SSI) (Fischer and Maurer, 1978) and stress tolerance index (STI) (Fernandez, 1992) were calculated using seed yield under controlled and drought stressed conditions. Cluster analysis was used to classification of varieties in different water regimes. Principle Component analysis (PCA) was used to ordination of entries in two dimensional biplots (Kroonenberg, 1997). Statistical analysis was performed with SPSS (version 21) and STATGRAPHICS (version 2.1) softwares.

Results

Hybrid Farrokh had the highest seed yield in all water regimes (3686, 2856, 2256 and 2506 Kg/ha

in control and water deficit in vegetative, flowering and grain filling stages respectively). In contrast in control and vegetative drought condition, Armavirski and in flowering and seed filling stages stress Record had the lowest seed yield (Fig.1). In all drought treatments the highest loss in seed yield was observed in Hysun33. The lowest loss in vegetative stage was in Master while in flowering and seed filling stages in Lakomka. In seed filling stage Armavirski had the same loss as Lakomka (Fig.1). The highest STI value for Farrokh indicated drought tolerance of that in all three drought treatments (table 2), however Farrokh had higher yield loss compared to Master and Lakomka. The STI index has been used to identification of drought tolerant genotypes. The high values of SSI for Hysun33 in all water regimes indicated the sensitivity of this hybrid to drought stress. The higher SSI for Hysun33 expressed that as drought sensitive variety however there were varieties with lower potential yield than Hysun33, i.e. Record, Master and Lakomka which expressed drought tolerant than Hysun33.

Cluster analysis based on all agronomic measurements using Ward linkage method differentiated varieties in different groups (Fig.2). Under drought stress in vegetative stage, Farrokh differentiated explicitly from others as a single group. Classification of varieties following drought treatment in flowering stage resulted in constitution of two groups one of them included SHF 81/90, Armavirski, Farrokh and Lakomka. Drought stress in seed filling stage resulted to the same grouping except moving Lakomka to other group. There was no singly differentiated variety in these stages.

Principle component analysis was used to differentiation of cultivars according to the measured characteristics under three drought treatments. When drought stress was imposed during vegetative stage Farrokh and Record differentiated in the reverse side of the PCA's biplot (Fig. 3). Farrokh was located at the same side of seed yield vector indicate higher seed yield of that. Whatever the genotype positioned closer to the tail end of a vector it has higher value for the corresponding trait and vice versa. Thus in Fig. 3 Farrokh at the same side of the seed yield's vector has the highest productivity while Record and Armavirski at the reverse side of that are low yielding varieties. In the same way drought stress in flowering and seed filling stages differentiated Farrokh and Record at the opposite sides of the PCA's biplot.

Table1. The factors and related levels in the experiment

Water regime		Variety	
C	Control	1- Hysun33	5-Record
V	Water withholding during (V4-R1)	2- Hysun25	6- Armavirski
F	Water withholding during (R5-R6)	3- Farrokh	7- Master
S	Water withholding during (R6-R9)	4- SHF81/90	8- Lakomka

C, V, F and S denote to control and vegetative, flowering and seed filling stages respectively.

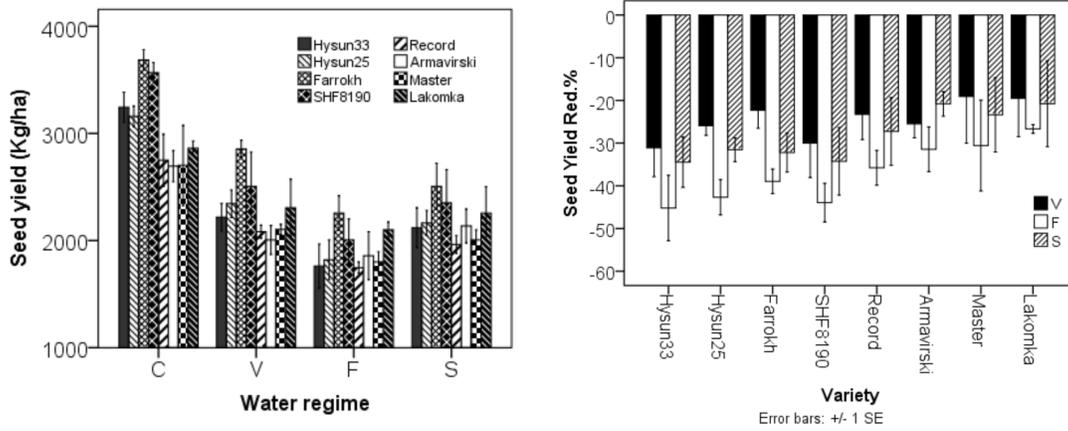


Fig 1. Seed yield of 8 sunflower varieties after drought treatment in different growth stages (Left). Reduction of seed yield under drought conditions in different varieties (Right). C, V, F and S denote to the control irrigation and water withholding in vegetative (V6-R1), flowering (R4-R6) and seed filling (R6-R9) stages respectively.

Table 2. Values of stress tolerance index (STI) and stress susceptibility index (SSI) for 8 sunflower varieties following drought stress during different growth stages.

Variety	STI			SSI		
	V	F	S	V	F	S
Hysun33	0.76	0.60	0.72	1.25	1.21	1.19
Hysun25	0.78	0.60	0.72	1.02	1.12	1.09
Farrokh	1.11	0.87	0.97	0.89	1.03	1.10
SHF81/92	0.94	0.75	0.88	1.18	1.16	1.18
Record	0.60	0.51	0.57	0.96	0.97	0.99
Armavirski	0.57	0.53	0.61	1.01	0.82	0.71
Master	0.60	0.51	0.57	0.87	0.88	0.89
Lakomka	0.69	0.63	0.68	0.77	0.70	0.73

V, F and S denote to the water withholding in vegetative (V6-R1), flowering (R4-R6) and seed filling (R6-R9) stages respectively.

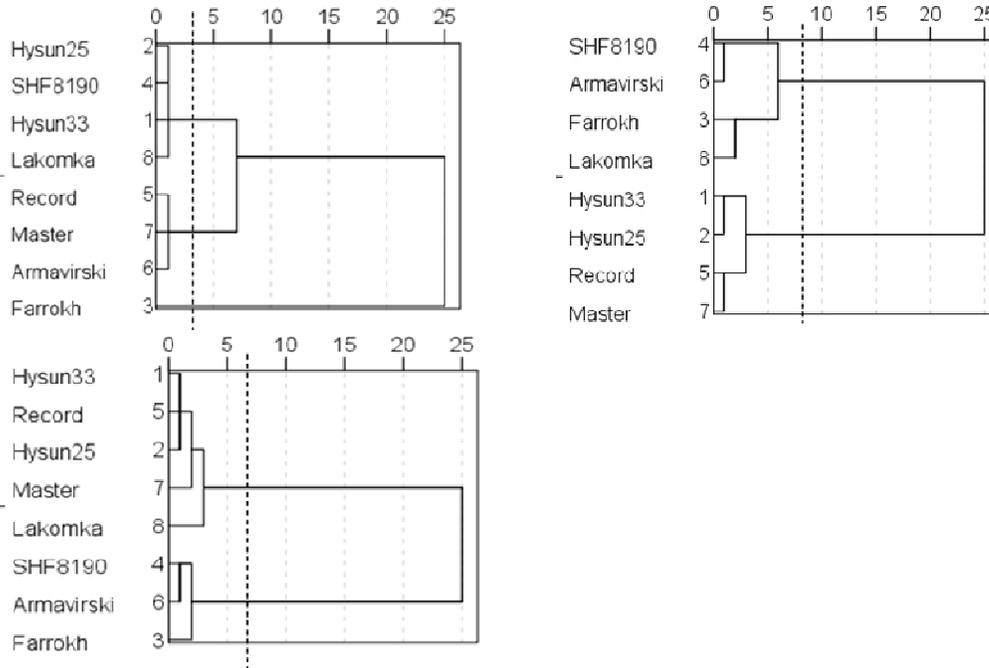


Fig.2 Cluster analysis using Ward linkage method to classification of sunflower varieties under drought stress in Vegetative (top left), Flowering (top right) and seed filling (down left) stages.

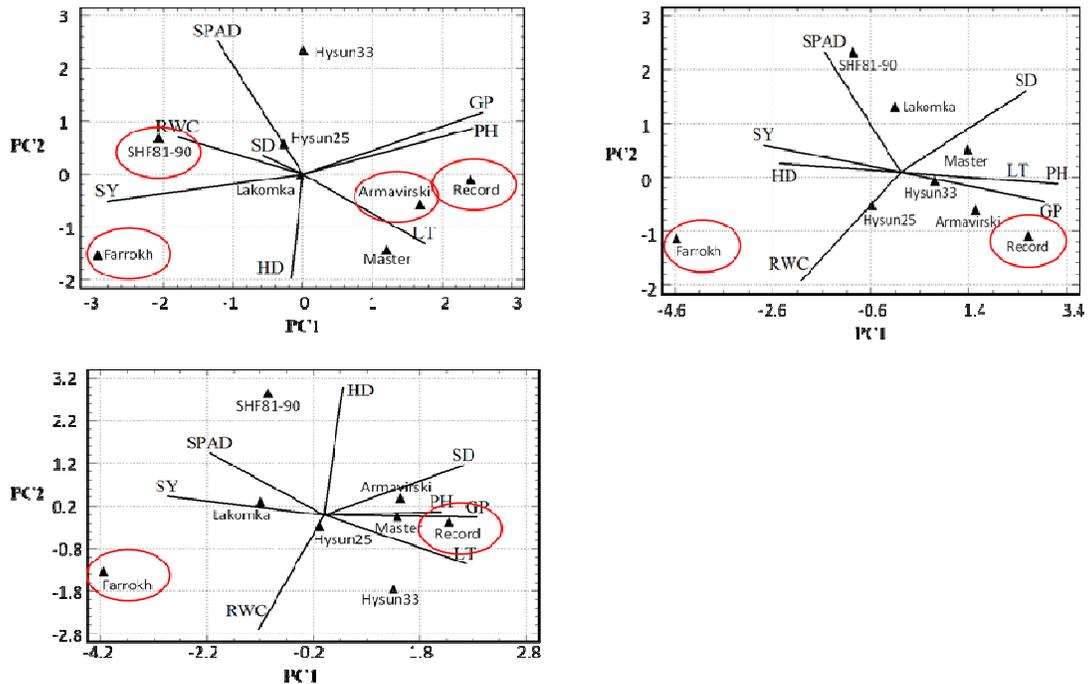


Figure 3. Principle component analysis for screening of sunflower varieties under drought stress in vegetative (top left), flowering (top right) and seed filling (down left) stages. Position of each variety represented by triangles. Straight lines show the vector of different traits. GP; Growth Period, PH, Plant Height, HD; Head Diameter, SD; Stem diameter, LT; Leaf Temperature, SPAD; Soil Plant Analysis Development (A measure for chlorophyll content), RWC; Relative Water Content and SY; Seed Yield.

Discussion

Flowering stage identified as the most sensitive stage to water deficit with 38% reduction in grain yield compared with normal irrigation. The results indicated that reduction in the seed number per head is the main cause for seed yield reduction under all water regimes. According to the STI and SSI indices which were used to identification of drought tolerant and drought sensitive cultivars, Farrokh was drought tolerant and Hysun33 was drought sensitive respectively. Clarke *et al.* (1992), Darvishzadeh *et al.* (2010) and Ghaffari *et al.* (2012) also suggested STI as the suitable criterion for screening of drought tolerant wheat and sunflower genotypes. It seems that potential seed yield and yield loss under drought condition was able to differentiate drought sensitive variety. So regarding these two criteria Hysun33 was drought sensitive in all water regimes. These results showed that potential yield or yield loss has the same results for screening of genotypes to some extent.

According to the results efficiency of SSI was under doubt when yield potential was considered as the main target for screening of genotypes. Although STI and SSI are simple criteria for identification of drought tolerant and sensitive genotypes however these are based on only seed yield and there is no information about relationship of other characteristics. So cluster analysis was used to finding any drought response structure among the cultivars. This method classified genotypes in different groups however without considering means of groups for related traits the analysis could not be able to clarify drought tolerant/sensitive genotype, so it is not a suitable method for identification of drought tolerant and sensitive varieties.

The results indicated that principle component analysis can eliminate these deficiencies and provide a straight view of plant attributes under drought condition. In the plots obtained by this analysis the angle between vectors represents the correlations among corresponding traits, the smaller the angle, the greater positive correlation among related traits and vice versa (Kroonenberg, 1995). Using PCA, Farrokh and Record were determined as the most drought tolerant and sensitive varieties respectively. All the methods confirmed Farrokh as the most drought tolerant cultivar however there were no unique results for identifying of drought sensitive cultivar. In spite of the SSI values which represented Hysun 33 as sensitive cultivar the PCA introduced Record as the most sensitive genotype. Because of involving many traits in PCA it is more reliable compared to the STI and SSI indices, although the same results was observed for drought tolerant

cultivar which expresses seed yield as a crucial factor under drought condition.

Higher correlation of RWC with seed yield was observed under drought stress in vegetative stage. It is concluded that water status during vegetative stage is the main determinant of seed yield. Following drought stress in flowering stage, Head diameter and RWC and after drought stress in seed filling stage, SPAD and RWC were the main determinants of seed yield. The results indicated unique effect of RWC in productivity of sunflower under drought condition. Crucial role of water status in productivity of sunflower under drought stress is reported also by Rauf and Sadaqat (2008) and Ghaffari *et al.* (2013). As RWC is related to cell volume, it may closely reflect the balance between water supply to the leaf and transpiration rate (Sinclair and Ludlow, 1985). Producing large heads as the exclusive seed bearers is an indicator of higher productivity under drought stress in flowering stage and maintaining chlorophyll content is a sign for higher seed yields when drought stress occurs after flowering stage. Higher correlation of SPAD with seed yield may have a positive impact on photosynthetic rate under drought condition. Because loss of chlorophyll contents under drought stress is considered as a main cause of inactivation of photosynthesis (Anjum *et al.* 2011).

Conclusions

Among the evaluated varieties Farrokh and Record were determined as the most drought tolerant and sensitive varieties respectively. Relative water content, head diameter and SPAD value were the main factors for establishment of seed yield under drought stress, among them RWC was expressed as the unique determinant of sunflower productivity under drought condition. Regarding seed yield as the final target it is concluded that PCA merging all plant characteristics can be used as an effective differentiator of genotypes under different water regimes.

References

- Anjum, S.A., Xie, X.Y., Wang, L.C., Saleem, M.F., Man, C., Lei, W., 2011. Morphological, physiological and biochemical responses of plants to drought stress. *African J. Agri. Res.* 6(9), 2026-2032.
- Chimenti, C.A., Pearson, J., Hall, A.J., 2002. Osmotic adjustment and yield maintenance under drought in sunflower. *Field Crop Res* 75(2): 235-246.
- Clarke, J.M., Depauw, R.M., Townleysmith, T.F., 1992. Evaluation of methods for

- quantification of drought tolerance in wheat. *Crop Sci* 32:723-728.
- Darvishzadeh, R., Pirzad, A., Hatami-Maleki, H., Poormohammad Kiani, S., Sarrafi, A., 2010. Evaluation of the reaction of sunflower inbred lines and their F1 hybrids to drought conditions using various stress tolerance indices. *Span J Agric Res* 8:1037-1046.
- De la Vega, A., Chapman, S.C., Hall, A.J., 2001. Genotype by environment interaction and indirect selection for yield in sunflower. I. Two-mode pattern analysis of oil yield and biomass yield across environments in Argentina. *Field Crop Res* 27: 17-38.
- Erdem, T., Erdem, Y., Orta, A.H., Okursoy, H., 2006. Use of a crop water stress index for scheduling the irrigation of sunflower (*Helianthus annuus* L.). *Turk J Agric For* 30:11-20.
- Fernandez, G.C.J., 1992. Effective selection criteria for assessing plant stress tolerance. In: Proceedings of the Int. Symposium on Adaptation of Vegetables and Other Food Crops in Temperature and Water Stress, 13-18 August, p.257-270. Tainan, Taiwan.
- Fischer, R.A., Maurer, R., 1978. Drought resistance in spring wheat cultivars: I. Grain yield responses. *Aust J Agric Res* 29:897-912.
- Ghaffari, M., 2006. Sunflower production guide. Agricultural and Natural Resources Research Center of West Azerbaijan, Iran. pp.4. (In Persian).
- Ghaffari, M., Farrokhi, K. 2008. Principal component analysis as a reflector of combining abilities. Proc. 17th Int. Sunflower Conf. Cordoba, Spain, pp 499-504.
- Ghaffari, M., Toorchi, M., Valizadeh, M., Shakiba, M.R., 2012. Morpho-physiological screening of sunflower inbred lines under drought stress condition. *Turk J Field Crops* 17: 185-190.
- Kroonenberg, P.M. 1997. Introduction to biplots for G x E tables. Research report No. 51. Center for statistics. The University of Queensland, Brisbane, Qld, Australia.
- Murphy, D.J., 2010. Improvement of industrial oil crops. In: Singh, B (ed), Industrial crops and uses. CABI International, Cambridge, pp. 183-206.
- Razi, H., Assad, M.T., 1999. Comparison of selection criteria in normal and limited irrigation in sunflower. *Euphytica* 105:83-90.
- Rauf, S., 2008. Breeding sunflower (*Helianthus annuus* L.) for drought tolerance. *CBCS* 3(1):29-44.
- Rauf S. and Sadaqat H.A. 2008. Identification of physiological traits and genotypes combined to high achene yield in sunflower (*Helianthus annuus* L.) under contrasting water regimes. *Aust. J. Crop Sci.*, 1(1): 23-30.
- Rodriguez, D.J., Romero-Garcia, J., Rodriguez-Garcia, R., Angulo-Sanchez, J.L., 2002. Characterization of proteins from sunflower leaves and seeds: Relationship of biomass and seed yield. In: Janick J., Whipkey, A. (eds), Trends in new crops and new uses. ASHS Press, Alexandria, VA, pp. 143-149.
- Schneiter, A.A., Miller, J.F., 1981. Description of sunflower growth stages. *Crop Sci* 21: 901-903.
- Sinclair T.R. and Ludlow M.M. 1985. Who taught plants thermodynamics? the unfulfilled potential of plant water potential. *Funct. Plant Biol.*, 12: 213-217.
- Singh, B.D., 2000. Plant Breeding, Principles and Methods. New Delhi, India: Kalyani Publishers, 896 p.
- Tahir, M.H.N., Muhammad, I., Hussain, M.K., 2002. Evaluation of sunflower (*Helianthus annuus* L.) inbred lines for drought tolerance. *Int J Agric Biol* 3: 398-400.
- Tersac M., Vares, D., Vincourt, P., 1993. Combining groups in cultivated sunflower populations and their relationships with country of origin. *Theor Appl Genet* 87: 603-608.