

Research Article



Effect of Drought on Morpho-Physiological Traits of Sunflower (*Helianthus Annuus* L) Hybrids and their Parental Inbred Lines

Rizwana Qamar¹, Maria Ghias¹, Fida Hussain¹, Sajida Habib¹, Muhammad Khuram Razzaq^{2*}, Muhammad Aslam¹ and Imran Habib³

¹Directorate of Oilseeds, Ayub Agricultural Research Institute Faisalabad, Pakistan; ²Soybean Research Institute, National Center for Soybean Improvement, Nanjing Agricultural University, China; ³Directorate of Biotechnology, Ayub Agricultural Research Institute Faisalabad, Pakistan.

Abstract | Water stress is the most significant restrictive factor for agricultural production in Pakistan. Sunflower productivity is sturdily delimited by the inaccessibility of water. Developments of sunflower hybrids having greater drought tolerance are the effective and inexpensive approach by plant breeders to manage with drought. A wire house experiment was conducted in Oilseeds Research Institute, Ayub Agricultural Research Faisalabad for the screening of genotypes against drought. Ten hybrids along with their 13 parental inbred lines were grown in polythene bags following factorial structured completely randomized design by three replications against three levels of water stress. Data was recorded on root length (cm), shoot length (cm), plant fresh weight (g), plant dry weight (g), chlorophyll fluorescence, leaf temperature (°C) and relative water contents (%). Results indicated the presence of significant genetic variability among hybrids and inbred lines. Hybrids had more tolerance to drought conditions on both levels of water stress treatments. With the increase in stress, root length and leaf temperature increased, while shoot length, plant fresh and dry weight, chlorophyll fluorescence and relative water content decreased at seedling stage. Hybrid FH-630 and Hysun-33 are identified as the most drought tolerant genotypes used in present study.

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***Correspondence** | Muhammad Khuram Razzaq, Soybean Research Institute, National Center for Soybean Improvement, Nanjing Agricultural University, China; **Email:** khuram.uos@gmail.com

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Introduction

Sunflower (*Helianthus annuus* L.) is grown all over the world for its finest quality premium oil. Its oil is considered best for heart patients because of higher proportion of unsaturated fatty acids. Sunflower is a major oilseed crop of Pakistan ranked third for the production of oil after cotton, rapeseed and mustard (Amjad, 2014; Razzaq, 2017). Pakistan is far behind in the domestic production of edible oil and government has to compromise a major share of its income (284.5 billion rupees) to purchase the oil seeds/ edible

oil, from other countries (ESP, 2016-17). Pakistan is mainly semiarid with less rainfall and greater solar radiation over most parts of the country. Water stress is the temporary deficiency of water but if shortage increases then it goes to the permanent wilting of plant in drought; that are prolonged climatic events which frequently affect crop growth and productivity (Riaz et al., 2010, Hamayun et al., 2010). Water scarcity is prevailing and it supposed to become a main restrictive factor against crop performance in the coming years due to global warming (Cook et al., 2007) and ever-increasing population pressure (Somerville and

Briscoe, 2001). Pakistan provinces especially main parts of Baluchistan and Sindh are facing the water shortage since last few decades due to erratic rain-falls. Less rain falls and lower level of river water becomes the chief factor for changing huge zones into deserts (Ashraf, 2006). The water storage capacities of reservoirs have been reduced because of siltation and water level continuously lowering down towards dead level. It is estimated that $\frac{1}{4}$ of total cultivated land of Pakistan (4.9 million hectares) is under water scarcity (Khan and Qayyum, 2015). Drought is a serious environmental factor which interferes with growth, nutrients and water relations, photosynthesis, assimilate partitioning and ultimately cause a significant reduction in crop yield (Praba et al., 2009).

In Pakistan, sunflower can grow 2 times (spring and autumn season) in a year, and has great potential to fulfill the requirements of edible oil of the country. Keeping in sight the importance of sunflower as a major oilseed crop in Pakistan, this study aimed to estimate the effect of water shortage on some morpho-physiological attributes of sunflower. This will also give vision to choice the tolerant inbreds and finally help in breeding of the better hybrids. Therefore, the objective of this research work is to select drought tolerant sunflower hybrids and inbreds that can acclimatize to the predictable effects of climate change.

Materials and Methods

Polythene bags experiment was conducted in the Oil-seeds Research Institute, Ayub Agricultural Research Faisalabad, Pakistan to evaluate 10 hybrids along with their 13 parents (inbred lines) and a commercial check listed in the Table 1, for some morpho-physiological characters at seedling stage. Experiment was laid out in factorial structured completely randomized design following three replications. Two treatments of field capacity (FC) (T2 = 50% FC, T3 = 25% FC) were used. Furthermore, T1= 80% FC, was considered control in this study. For the determination of field capacity take 100 g air dried sample of soil. Place a soil on a filter paper in a glass funnel. Then add 100 ml of water in soil and let it drained for overnight. The water collected in a cylinder placed below the funnel will give the amount of extra water. The amount of water that retained in the soil will give the field capacity of soil. Data was recorded on root length (cm), shoot length (cm), plant fresh weight (g), plant dry weight (g), chlorophyll fluorescence, leaf tempera-

ture ($^{\circ}\text{C}$) and relative water contents (%). Root and shoot length of 30 days seedling were measured using measuring tape. Plant fresh weight was measured immediately after uprooting of seedlings whereas dry weight was measured after oven drying for 2 days at 80°C . Relative water contents (RWC) were studied according the Turner (1986), fresh leaves were taken from each genotype and weighed immediately to record fresh weight (FW). Afterwards they were put in distilled water for 4 h and weighed again to record turgid weight (TW). Dry weight (DW) was recorded after oven drying at 70°C for 24 hours. The RWC was calculated by using following equation according to method given by Taiz and Zaiger (1998):

$$RWC = \{(FW-DW) / (TW-DW)\} \times 100$$

Maximum quantum yield of PSII (Fv/Fm) of leaves were estimated using a chlorophyll fluorometer (PAM-2000, Walz, Germany) using a leaf-clip holder (2030-B, Walz, Germany). Leaf temperature of all genotypes was estimated by using infrared thermometer (AR320) at 10:00 am to 12.00 noon. Various researchers utilize stress tolerance indexes for various traits to select genotypes under Drought stress (Neghavi et al., 2013; Farshadfar et al., 2013). Stress tolerance indexes of all traits were measured according to the following formula.

$$STI1 (\%) = \text{Value at } T2 \times 100 / \text{Value at control condition } (T1)$$

$$STI2 (\%) = \text{Value at } T3 \times 100 / \text{Value at control condition } (T1)$$

Recorded data was subjected to analysis of variance according to Steel et al. (1997).

Results and Discussion

Mean square values of studied traits showed that significant genetic variability is present among sunflower genotypes under study. Drought treatments also had significant effect on all traits under study except leaf temperature. Analysis of variance showed that Genotype \times Treatment interaction also had significant effect on all traits except leaf temperature (Table 2). Mean values of root length, shoot length, plant fresh weight and plant dry weight reduces with the increase in drought stress (Table 3). Mean values for chlorophyll fluorescence and relative water content decreases whereas leaf temperature increases with the advancement of drought stress (Table 4). Effect of drought stress on sunflower at seedling stage has

been studied by many scientists (Razzaq et al., 2017; Ivanova et al., 2016; Javaid et al., 2015).

Table 1: List of hybrids, inbred lines used in the experiment.

Sr. No.	Hybrids	Sr. No.	Inbred lines
1	FH-583	1	ORI-20
2	FH-606	2	RL-37
3	FH-629	3	RL-39
4	FH-630	4	ORI-44
5	FH-631	5	ORI-45
6	FH-634	6	ORI-49
7	FH-639	7	RL-60
8	FH-649	8	RL-62
9	FH-331	9	ORI-65
10	Hysun 33	10	RL-67
		11	RL-68
		12	ORI-73
		13	RL-108

FH: Faisalabad hybrid; **ORI,** CMS line; **RL:** Restorer line.

Stress tolerance index of morphological parameters

Stress tolerance indexes of all traits are shown graphically which showed that genotypes had variable magnitude of root length stress tolerance index (Figure 1). FH-634 had maximum root length stress tolerance index at 50% field capacity (T2) whereas Hysun 33 showed highest root length stress tolerance index at 25% FC. Hybrids have more root length stress tolerance than their corresponding parents. All genotypes showed increase in tolerance with the increase in drought stress except two hybrids FH-583 and FH-634 which showed relative decrease in tolerance index with the increase in stress incidence. Variable magnitude of shoot length stress tolerance index was observed among genotypes (Figure 2). FH-630 showed highest shoot length stress tolerance index at 50 % FC whereas FH-639 showed highest tolerance at 25% FC. With the increase in drought incidence shoot length stress tolerance among the genotypes decreases. Overall performance showed that hybrids have more shoot length tolerance index than female and male parents.

With the increase in drought, stress tolerance index for plant fresh weight decreases (Figure 3). FH-639 had highest stress tolerance index followed by FH-630 and FH-606 at both levels of drought. Hybrids had more tolerance than parent inbred lines. ORI-73 is the only female parent which showed increase tolerance

with proceeding levels of stress. Variability in plant dry weight stress tolerance index among sunflower genotypes under drought stress is presented in Figure 4. FH-630 showed highest tolerance for plant dry weight followed by FH-583 at both levels of drought stress. With the increase in drought level, stress tolerance index for plant dry weight decreases. Hybrids showed more tolerance than parental inbred lines. Female line ORI-73 and Male line RL-37 showed increase in tolerance with the increase in stress level.

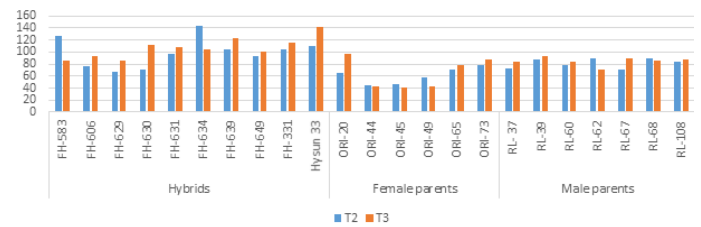


Figure 1: Root length stress tolerance index under drought condition.

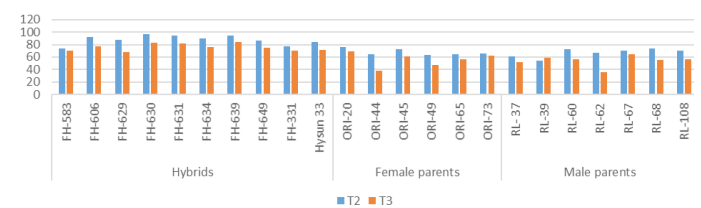


Figure 2: Shoot length stress tolerance index under drought condition.

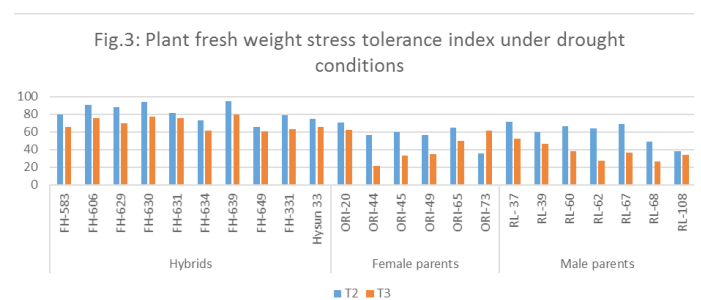


Figure 3: Plan fresh weight stress tolerance index under drought condition.

Stress tolerance index of physiological parameters

Chlorophyll fluorescence stress tolerance index under drought conditions are presented in Figure 5. Hybrids showed more tolerance than parental lines. FH-631 and FH-634 had highest stress tolerance for chlorophyll fluorescence at both levels. With the increase in drought, chlorophyll fluorescence in the studied genotypes decreased. Leaf temperature stress tolerance index also increases with increases stress as depicted in the Figure 6. The hybrid FH-649 and FH-630 showed more tolerance at 50% FC whereas the hybrid FH-606, Hysun 33 and FH-634 showed more tolerance at 25% FC. The hybrids FH-649 and FH-630 showed decrease in leaf temperature stress tolerance with increase in drought.

Table 2: ANOVA effects of treatments on different traits under drought stress.

SOV	Mean Squares									
	DF	RL	SL	RFW	SFW	RDW	SDW	Fv/Fm	LT	RWC
R	2	0.077	52.32	0.023	0.442	0.0004	0.004	0.004	17.90	18.34
G	22	9.649**	37.64**	0.629**	0.302**	0.0009**	0.005**	0.005**	11.48*	512.75**
T	2	26.17**	1464.6**	0.326**	37.79**	0.0140**	0.035**	0.035**	13.77	445.45**
G*T	4	6.228**	14.53**	0.012**	0.274**	0.0006**	0.004**	0.004**	7.7	531.54**

RL: Root Length; **SL:** Shoot Length; **RFW:** Root Fresh Weight; **SFW:** Shoot Fresh Weight; **RDW:** Root Dry Weight; **SDW:** Shoot Dry Weight; **Fv/Fm:** Chlorophyll Fluorescence Ratio; **LT:** Leaf Temperature; **RWC:** Relative Water Contents.

Table 3: Mean values for various morphological traits under three treatments of drought stress.

Sr. No.	Name of geno- types	Root Length(cm)			Shoot Length (cm)			Plant Fresh Weight (g)			Plant Dry Weight (g)		
		T1	T2	T3	T1	T2	T3	T1	T2	T3	T1	T2	T3
1	FH-583	5.31	7.76	4.52	19.69	14.53	13.79	2.46	1.97	0.83	0.38	0.38	0.20
2	FH-606	6.66	5.08	6.21	20.77	12.91	13.92	2.14	1.51	0.87	0.44	0.32	0.18
3	FH-629	6.50	4.38	5.56	20.96	18.25	14.28	2.69	1.57	0.81	0.42	0.27	0.18
4	FH-630	5.27	3.77	5.87	24.80	18.97	15.73	2.98	1.90	0.82	0.35	0.35	0.17
5	FH-631	5.27	4.58	4.68	25.56	16.43	15.80	2.94	1.52	1.06	0.38	0.29	0.17
6	FH-634	5.76	8.26	6.06	24.37	14.67	13.76	2.84	1.78	0.89	0.38	0.33	0.19
7	FH-639	4.58	4.78	5.62	20.63	13.70	13.30	3.30	1.14	0.99	0.42	0.25	0.22
8	FH-649	4.77	4.42	4.83	22.61	12.22	9.44	3.01	1.97	0.71	0.42	0.32	0.17
9	FH-331	6.57	6.80	6.32	29.09	13.75	14.30	3.07	1.26	0.86	0.40	0.20	0.13
10	Hysun 33	4.41	4.85	6.22	24.51	18.11	12.48	2.62	1.70	0.94	0.38	0.34	0.17
11	ORI-20	4.79	3.11	4.67	15.01	11.40	10.33	1.95	1.71	1.22	0.35	0.29	0.25
12	ORI-44	5.62	4.07	8.08	17.92	14.51	14.57	2.66	1.90	1.39	0.44	0.28	0.34
13	ORI-45	5.48	4.77	8.93	26.03	13.99	15.44	2.54	1.51	1.69	0.35	0.39	0.32
14	ORI-49	5.89	2.61	2.48	21.94	14.12	8.35	2.94	1.67	0.62	0.34	0.27	0.16
15	ORI-65	4.51	2.09	6.07	20.11	17.83	12.23	2.87	1.70	0.95	0.34	0.34	0.25
16	ORI-73	3.22	4.43	5.12	21.98	13.81	10.27	2.77	1.57	0.97	0.44	0.27	0.23
17	RL- 37	3.29	4.53	5.07	19.63	14.34	11.18	1.55	1.18	0.59	0.32	0.19	0.14
18	RL-39	5.07	4.57	3.63	22.20	14.83	7.99	2.28	1.46	0.62	0.37	0.26	0.14
19	RL-60	3.17	4.15	4.39	14.31	13.55	10.89	1.91	1.62	0.96	0.33	0.30	0.19
20	RL-62	6.16	4.35	5.47	15.43	12.15	11.47	1.84	1.64	0.67	0.33	0.25	0.15
21	RL-67	4.02	3.56	3.43	18.79	15.76	10.34	2.64	1.29	0.70	0.33	0.25	0.15
22	RL-68	3.71	2.92	6.23	20.49	13.50	12.75	2.25	0.80	1.39	0.37	0.18	0.23
23	RL-108	3.98	6.10	6.13	21.93	15.52	12.35	3.98	1.52	1.35	3.98	0.32	0.28

FH: Faisalabad Hybrid; **ORI,** CMS line; **RL:** Restorer Line.

Relative water content stress tolerance index also decreases with the increase in drought incidence as shown in Figure 7. The hybrid FH-639 followed by Hysun 33 showed highest tolerance at 50% drought stress whereas the hybrid FH-639 and Hysun 33 showed high tolerance at 25 % drought stress. Male parent RL-39 showed more tolerance for most of the studied traits at higher level of drought. Hybrids showed more tolerance than parental inbred lines.

Plant faces various biotic and abiotic stresses during their life cycle. As water is a fundamental constituent of plants for the maintenance of leaf structure and shape, photosynthesis, and thermal regulation (Sanaullah et al., 2013). Drought is the major stress that limits plant growth and crops production more than any other abiotic stress (Fahad et al., 2017). Damage index and stress tolerance index are better indicators to measure drought tolerance of plant

Table 4: Mean values for various physiological traits under three treatments of drought stress.

Sr. No.	Name of genotypes	Chlorophyll fluorescence (Fv/Fm)			Leaf temperature (°C)			Relative water content (%)		
		T1	T2	T3	T1	T2	T3	T1	T2	T3
1	FH-583	0.80	0.77	0.77	34.1	32.9	32.7	103.0	75.3	89.1
2	FH-606	0.81	0.75	0.76	32.2	33.9	30.2	97.2	67.9	73.8
3	FH-629	0.80	0.65	0.68	33.9	31.4	29.9	88.6	86.6	58.2
4	FH-630	0.78	0.74	0.69	32.2	33.4	29.6	82.6	78.0	48.7
5	FH-631	0.71	0.72	0.63	32.3	34.0	32.5	82.5	61.9	54.6
6	FH-634	0.79	0.79	0.69	32.0	35.2	33.4	75.3	71.5	78.4
7	FH-639	0.79	0.76	0.78	34.2	33.5	34.4	74.7	65.5	94.3
8	FH-649	0.77	0.66	0.75	33.4	35.5	33.8	89.4	70.2	61.5
9	FH-331	0.76	0.73	0.75	34.1	34.8	34.7	71.8	58.3	73.7
10	Hysun 33	0.79	0.74	0.69	32.6	33.6	36.7	73.5	81.4	86.4
11	ORI-20	0.76	0.65	0.75	34.0	35.9	36.1	64.5	80.0	76.6
12	ORI-44	0.78	0.71	0.74	33.1	36.0	34.6	96.5	80.4	68.9
13	ORI-45	0.73	0.78	0.77	33.3	36.1	33.4	83.2	84.9	57.8
14	ORI-49	0.78	0.73	0.72	33.6	33.7	33.5	84.2	92.1	79.0
15	ORI-65	0.73	0.71	0.76	30.3	32.6	35.5	74.6	82.8	102.2
16	ORI-73	0.78	0.71	0.79	32.9	31.9	34.5	83.4	98.2	90.0
17	RL- 37	0.74	0.62	0.71	31.8	33.4	37.4	94.3	123.7	72.4
18	RL-39	0.75	0.68	0.73	36.3	33.0	33.1	86.5	67.5	77.5
19	RL-60	0.71	0.79	0.74	33.3	31.8	30.2	81.4	70.3	101.7
20	RL-62	0.77	0.80	0.78	31.3	31.7	35.4	75.5	75.3	71.5
21	RL-67	0.75	0.74	0.73	34.4	35.5	36.6	72.6	103.2	87.3
22	RL-68	0.77	0.69	0.78	30.7	32.1	32.8	84.0	88.5	76.7
23	RL-108	0.78	0.75	0.73	32.9	35.5	32.5	69.5	79.5	92.9

FH: Faisalabad Hybrid; ORI; CMS line; RL: Restorer Line.

genotypes (Turhan and Baser, 2004; Onemali and Guccer, 2010). Genotypes used in the present studies have variable response on varying levels of drought stress.

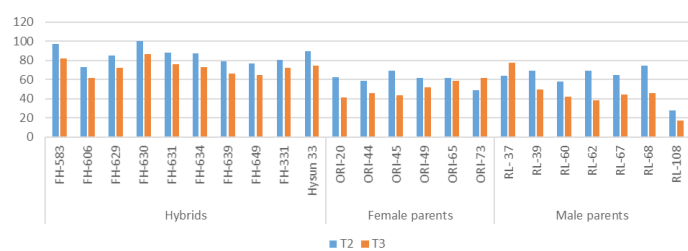


Figure 4: Plan dry weight stress tolerance index under drought stress.

Plant roots are primarily related to drought stress as it absorbs moisture from the soil. During water limited conditions roots proliferate to the deeper layers of soil to get moisture and other nutrients. So higher root length is a positive indicator of drought tolerance (Angadi and Entz, 2002). Moreover, root length along with root diameter, root density, root fresh and dry weight are also positively correlated with drought tolerance (Geetha, 2012; Rauf and Sadaqat, 2008;

Rauf et al., 2009). There are many reports which are in agreement with the present findings indicating that drought stress severely reduce the growth and biomass of the plant (Ivanova et al., 2016) due to reduction in cell division and elongation (Anjum et al., 2011). The varieties having genetic potential to maintain the higher growth under stress conditions are drought tolerant (Ahmad et al., 2009).

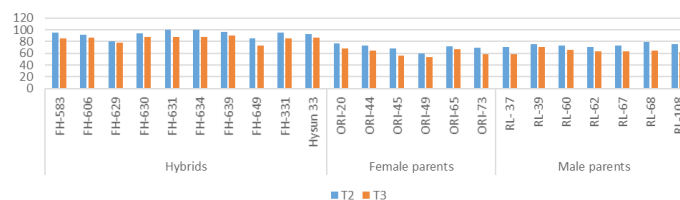


Figure 5: Chlorophyll fluorescence stress tolerance index under drought condition.

Water use efficiency decreases with increased drought stress (Zhang, 2018). It reduces plant transpiration which increases leaf temperature under drought stress (Shahenshah and Isoda, 2010). Stomatal conductance

becomes lower in limited water conditions which resulted in the increase in temperature so drought tolerant genotypes have warmer leaves whereas drought sensitive genotypes does not limit stomatal conductance resulting in cooler leaves (Khan et al., 2007).

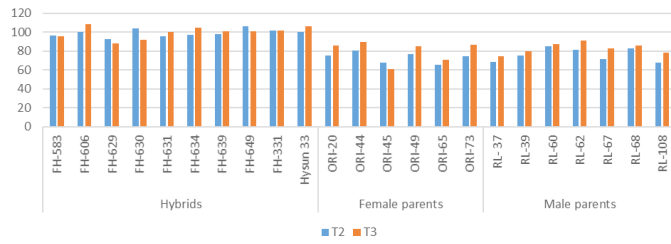


Figure 6: Leaf temperature relative tolerance index under drought condition.

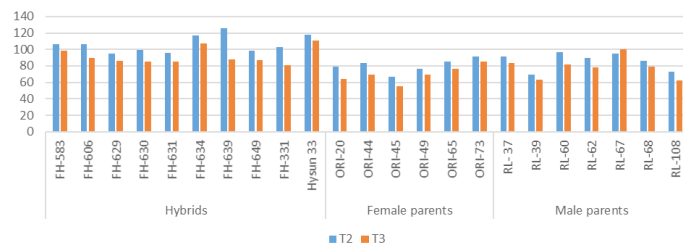


Figure 7: Relative water content stress tolerance index under drought condition.

Decrease in relative water content limits photosynthetic rate by closing down stomata (Cornic, 2000). High relative water contents are an indicator of plant drought tolerance (keyvan, 2010). Chlorophyll fluorescence measurements indicated the efficiency of photosystem II. Maximum chlorophyll fluorescence (Fm), Variable chlorophyll fluorescence (Fv) and their ration Fv/Fm decreases with the increase in drought stress (Arji et al., 2003). Khaleghi et al. (2012) also reported significant effect of drought stress on chlorophyll fluorescence in olive. Tolerant cultivars did not show decrease in Fv/Fm ratio whereas susceptible cultivars may decrease this ratio up to 90% (Faraloni et al., 2011).

Conclusions

Effect of drought stress have been extensively studied in different crop plants by different scientists and confirm the adverse effect on plant growth parameters and ultimately on yield. Present studies concluded that drought stress significantly affect the root shoot length, plant fresh and dry weight, Fv/Fm, leaf temperature and relative water contents. So, these traits may be used as an index of drought tolerance indicators and will be helpful in the screening of drought tolerant genotypes. Sunflower hybrids showed more

tolerance than female and male parents. FH-634 and Hysun-33 performed well for most of the traits. So, these hybrids can be used in the areas with scarcity of water.

Author's Contribution

Rizwana Qamar wrote the paper; Maria Ghias data collection, Fida Hussain conceived the idea, Sajida Habib Supervised the work, prepared methodology, Muhammad Khuram Razzaq improved the paper by critically review and correspondence, Muhammad Aslam analyzed the data and Dr. Imran Habib critically reviewed the paper at the end.

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