ABSCISIC ACID, A STRESS HORMONE HELPS IN IMPROVING WATER RELATIONS AND YIELD OF SUNFLOWER (HELIANTHUS ANNUUS L.) HYBRIDS UNDER DROUGHT

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Abstract

Genotypic variation in water relations under drought is an important index of studying drought tolerance of crops. Abscisic acid (ABA) application helped in mitigating drought stress by improving water relations and yield. Three sunflower hybrids viz., DK-4040 (tall stature), S-278 (medium stature) and SF-187 (short stature) were subjected to different irrigation and ABA application regimes i.e., four irrigations (25 days after sowing (DAS), at bud initiation, at flower initiation and at achene formation) and with no ABA spray, three irrigations (25 days after sowing, at flower initiation and at achene formation) and with no ABA spray, three irrigations (25 days after sowing, at flower initiation and at achene formation) and with ABA spray at bud initiation, three irrigations (25 days after sowing), at bud initiation and at achene formation) and with no ABA spray, three irrigations (25 days after sowing), at bud initiation and at achene formation) and with ABA spray at flower initiation. The experiment was laid out in randomized complete block design with split plot arrangement and had three replications. Exogenous application of ABA under drought at either stage (bud or flower initiation) was helpful in ameliorating drought stress by improving water relations and yield of sunflower hybrids; however response was better when ABA was applied under drought at bud initiation than at flower initiation stage. Sunflower hybrid DK-4040 showed better enhancement of drought tolerance by exogenous application of ABA under drought than SF-187 and S-278 because it showed more improvement in water potential, osmotic potential, turgor pressure, relative leaf water contents and achene yield.

Introduction

Sunflower is successfully cultivated over a widely scattered geographical area in the world. It has ability to adopt in a variety of environmental conditions (Beard & Geng, 1982). Sunflower is a high yielding oilseed crop and has the potential to bridge up the gap existing between consumption and domestic production of edible oil. Furthermore, it is a short duration crop (90-120 days) and can be grown twice a year. It fits well in existing cropping system and can be grown without replacing any major crop.

Water is very essential for plant growth and makes up 75 to 95 % of plant tissue. A vast amount of water moves throughout the plant daily. Plants use water and carbon dioxide to form sugars and complex carbohydrates. Water acts as a carrier of nutrients and also a cooling agent. It also provides an element of support through turgor and as an intercellular reaction medium (Ashraf & Harris, 2005).

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Environmental stresses represent a major constraint to meet the world food demand. There are relatively few stress free areas where crops may approach their potential yields. For instance, up to 45% of the world agricultural lands are subjected to continuous or frequent drought, wherein 38% of the world human population resides (Bot *et al.*, 2000). In the face of a global scarcity of water resources, drought has already become a primary factor in limiting crop production worldwide. At present, around 18% of the global farmland only is irrigated (more than 240 million hectares) and up to 40% of the global food supply is produced on this land (Somerville & Briscoe, 2001). It is, therefore imperative to improve the drought tolerance of crops to increase crop production in drought hit areas. Water-limited crop production depends on the intensity and the pattern of drought which vary from year to year. In some sub-tropical countries like Pakistan however, there is a high probability that crop water deficits increase in severity as the season progresses, due to lack of rainfall and also due to the high evaporative demand (Ashraf & Foolad, 2006).

Sunflower genotypes have performed differently in various environmental conditions. Different environmental conditions are required to evaluate the good yielding, better adaptive and stable crops varieties. (Luquez *et al.*, 2000; Azhar *et al.*, 2009). The severity of water shortage imposed on field crops also depends on the susceptibility of crops during different stages of their development (Shamim *et al.*, 2009). The general effects of drought on plant growth are well known (Manivannan *et al.*, 2007; Ahmad Sankar *et al.*, 2007). When soil moisture is deficient, crop establishment may be reduced. Plant growth is limited, normal development patterns disrupted and eventually final yield lowered.

Varietal differences in drought tolerance may be associated with change in ABA in response to various environmental stresses. This includes drought tolerance of maize (Pekic & Quarrie, 1987), chilling tolerance of rice seedlings (Lee et al., 1993) and salt tolerance of rice (Moons et al., 1995). ABA, a well-known stress reducing/tolerance promoting plant hormone has long been studied as potential mediator for induction of drought tolerance in plants (Davies & Jones, 1991; Zeevaart, 1999). It helps in promoting drought tolerance, both from the use of exogenous application to intact plants (Gibson et al., 1991; Heschel & Hausmann, 2001; Xu et al., 2002; Wang et al., 2003) and from the measurement of the endogenous ABA concentration (Van Rensburg & Kruger, 1994; Li & Wang, 2003). Exogenous application of 5 µM ABA to cotton under drought significantly increased seed number and lint mass per plant (Pandey et al., 2003). Jia & Zhang (1999) have also shown 50 % decrease in leaf conductance in sunflower detached shoot fed with 5µM ABA solution for 30 minutes. In soybean, reproductive potential reduced considerably due to less pod set under drought stress. This problem has been alleviated by exogenous application of 0.1mM ABA (Liu et al., 2004). Population variation due to exogenous application of ABA have been observed in well watered and stressed plants (Amdt et al., 2001; Tsialtas et al., 2001; Ponton et al., 2001). Exogenous application of ABA affected water use efficiency more in droughted plant than wellwatered plants of jewelweed (Heschel & Hausmann, 2001). Plants would grow more rapidly by using a finite water supply more efficiently when water is limited, in this case high water use efficiency would positively affect plant productivity (Li, 1999).

The success of semi-dwarf genes in cereals has prompted scientists to consider height reduction in sunflower. Agronomic benefits of dwarf sunflowers have been reported (Miller, 1992; Johnston *et al.*, 1995). In sunflowers, differential water relations response has been demonstrated by different stature genotypes. Dwarf cultivars of

sunflower have higher water potential i.e., least water stress as compared to intermediate and long stature cultivars. This is due to difference in canopy architecture and root penetration depth among different stature sunflower cultivars (Angadi & Entz, 2002a). Tall stature cultivars produced more leaf area, aerial biomass and deeper root system so they transpired more water (Angadi & Entz, 2002b). Giminez & Fererez (1986) have reported a negative correlation between leaf area and leaf water potential. Greater leaf overlap due to compact canopy in dwarf versus standard height sunflower has also been reported by Sadras *et al.*, (1991). Compact crop canopies affect microclimate by reducing wind speed and light penetration by increasing relative humidity (Nobel, 1999), which in turn reduces transpiration loss from the canopy (Pataki *et al.*, 1998). An open canopy, with greater light penetration and better air circulation has potential to increase the plant water status (Nobel, 1999).

So the present study was conducted to determine that how exogenous application of ABA helps in mitigating the effect of drought stress on water relations and yield of sunflower. Other objective of this study was to find out the appropriate stage at which ABA application could be more beneficial to ameliorate the water deficit effect on sunflower hybrids.

Materials and Methods

Three sunflower hybrids viz., DK-4040 (tall stature), S-278 (medium stature) and SF-187 (short stature) were used in the current study. Their seed was obtained from the Regional Office of the Pakistan Oil Development Board, Faisalabad. The experiment was conducted during spring at Agronomic Research Farm, University of Agriculture, Faisalabad, Pakistan for two consecutive years 2008 and 2009. The meteorological data for the growing period of the crop were collected from the Agro-meteorological cell, Department of Crop Physiology, University of Agriculture, Faisalabad, Pakistan. The data on average temprature, relative humidity and rainfall are presented in Fig. 1 while physiochemical characteristics of soil in Table 1. The experiment was laid out in randomized complete block design (RCBD) with split plot arrangement and replicated thrice. Net plot size was 3.0 x 5.0 m.

Prior to seedbed preparation, pre-soaking irrigation of 10 cm was applied. The seedbed was prepared by cultivating the field for 2-3 times with tractor-mounted cultivator. Sunflower hybrids (DK-4040, S-278 and SF-187) were sown on 17^{th} and 12^{th} of February, 2008 and 2009, respectively on ridges with the help of dibbler using seed rate of 8 kg ha⁻¹ Ridges were made 75 cm apart and plant-to-plant distance of 25 cm was maintained. Fertilizers were applied at the rate of 150 kg N and 100 kg P₂O₅ ha⁻¹ in the form of urea and diammonium phosphate (DAP). Half of N and whole of P were applied at sowing, while the remaining N with 1st irrigation. Field hoeing was done in order to keep the field weed free. Plant protection measures were carried out to keep crop free of insect pests, diseases and parrots. The first and second year crops were harvested on May 29, 2008 and June 5, 2009, respectively.

First irrigation was applied at 4-6 leaf stage [25 days after sowing (DAS)], 2nd irrigation at bud initiation stage (45 DAS) except the plots which were subjected to water stress at this stage, 3rd irrigation at flower initiation stage (67 DAS) except the plots which were subjected to water stress at this stage and 4th irrigation to all plots at achene formation stage (90 DAS).



Fig. 1. Mean temperature (°C), relative humidity (%) and rainfall (mm) during experimental period (February-June).

Data	rmination	Unit	Value	
Dete	rinnation	Umt	2008	2009
A.	Physical analysis			
	Sand	%	66.6	64.5
	Silt	%	16.6	18.5
	Clay	%	16.8	17
	Textural class	Sandy clay loam		
В.	Chemical analysis			
	рН		8.2	8.00
	EC	dSm ⁻¹	1.37	1.42
	Organic matter	%	0.74	0.70
	Total nitrogen	%	.046	.047
	Available phosphorus	mg kg ⁻¹	6.52	6.58
	Available potassium	mg kg ⁻¹	171	170

Table 1. Pre-sowing field soil analysis.

In this study, Abscisic acid of Sigma Aldrich, Japan was applied to the crop as foliar spray. The effect of application of two ABA levels (0 and 8μ M) at two growth stages (bud initiation and flower initiation) under three irrigation regimes (no drought stress, drought stress at bud initiation and drought stress at flower initiation) was studied.

Data regarding water relations including water potential, osmotic potential, turgor pressure and relative leaf water contents were recorded ten days after exogenous spray of abscisic acid. Achene yield was recorded at maturity. The third leaf from top (fully expanded youngest leaf) of two plants from each treatment was used to determine the leaf water potential. The water potential was measured from 8.00 to 10.00 A.M. by using Scholander pressure chamber according to the technique given by Scholander *et al.*, (1965). The same leaf, as used for water potential, was frozen in a freezer at below -20°C for 7 days. Then the frozen leaf material was thawed and cell sap extracted with the help of a disposable syringe. The extracted sap was directly used for the determination of osmotic potential by means of an osmometer (Wescor 5500). Turgor pressure was calculated as the difference between the values of osmotic potential (Ψ_s) and water potential (Ψ_w).

$$(\Psi_{\rm p}) = (\Psi_{\rm w}) - (\Psi_{\rm s})$$

The third leaf from top (fully expanded youngest leaf) of two plants of each treatment was taken to determine the relative leaf water contents. Soon after cutting at the base of lamina, leaves were sealed within plastic bags and quickly transferred to the laboratory. Fresh weight (FW) was taken within two hours after excision of leaves. Then turgid weight (TW) was determined after soaking leaves in distilled water for 16-18 h at room temperature. After soaking, leaves were quickly and carefully blotted dry with tissue paper to compute turgid weight. Dry weight (DW) was obtained after drying the leaf samples in oven for 72 h at 70°C. Relative leaf water content (RLWC) was calculated from the formula (Schonfeld *et al.*, 1988) and then averaged.

RLWC (%) = (FW-DW)/ (TW-DW)
$$x100$$

where FW, DW and TW are fresh weight, dry weight and turgid weight of leaf, respectively.

Data collected were analyzed by using Fisher's analysis of variance technique. LSD test at 5% probability was used to compare the differences among treatments means (Steel *et al.*, 1997).

Results

Different irrigation and ABA application schedules for different hybrids of sunflower showed significant effect on leaf water potential during both years of study. Sunflower hybrid DK-4040 had maximum leaf water potential than S-278 and SF-187. Drought stress significantly decreased leaf water potential and more decrease in leaf water potential was observed when drought stress was imposed at flower initiation than at bud initiation. Exogenous application of ABA at bud initiation statistically increased leaf water potential over no ABA application at same stage (Fig. 2a). This increase was 32.33% in DK-4040, 15.96% in S-278 and 27.66% in SF-187 during 2008. Similar trend was observed during both years of study. ABA spray at flower initiation also significantly increased leaf water potential than no ABA spray at same stage (Fig. 2b). This improvement in leaf water potential was 27.10% in DK-4040, 15.25% in S-278 and 13.89% in SF-187 during first year of study. Similar observations were recorded during both years of study.

Drought stress significantly increased leaf osmotic potential and more increase in leaf osmotic potential was observed when drought stress was imposed at flower initiation than at bud initiation. Applying ABA under stress at bud initiation significantly decreased leaf osmotic potential in sunflower hybrids (Fig. 3a). This decrease was 4.01% in DK-4040, 2.51% in S-278 and 4.95% in SF-187 during 2008. Similar trend was observed during both years of study. Foliar application of ABA at flower initiation also significantly decreased leaf osmotic potential than no ABA application at this stage (Fig. 3b). This reduction in leaf osmotic potential was 3.61% in DK-4040, 4.16% in S-278 and 3.21% in SF-187 during first year of study. Similar observations were recorded during both years of study.



Fig. 2. Effect of irrigation and exogenous application of abscisic acid on leaf water potential of sunflower hybrids; a) Budding b) Flowering; T_1 = 4 irrigations and no ABA; T_2 = Irrigation skip at budding and no ABA; T_3 = Irrigation skip at budding and ABA; T_4 =Irrigation skip at flowering and no ABA; T_5 =Irrigation skip at flowering and ABA.



Fig. 3. Effect of irrigation and exogenous application of abscisic acid on leaf osmotic potential of sunflower hybrids; a) Budding b) Flowering; T_1 = 4 irrigations and no ABA; T_2 = Irrigation skip at budding and no ABA; T_3 = Irrigation skip at budding and ABA; T_4 =Irrigation skip at flowering and no ABA; T_5 =Irrigation skip at flowering and ABA.

achene yield of sunflower hybrids.						
Hybrids (H)	Irrigation and ABA application schedules	Achene yield (kg ha ⁻¹)				
Hybrids (H)	In rigation and ADA application schedules	2008	2009			
H ₁ (DK-4040)	T ₁ =Control (no ABA application)	3226.64a	3452.17a			
	T ₂ =Stress at bud initiation and no ABA	1824.34c	1954.08d			
	T ₃ =Stress at bud initiation and ABA	2702.35b	2893.87b			
	T ₄ =Stress at flower initiation and no ABA	1210.67d	1296.12e			
	T ₅ =Stress at flower initiation and ABA	2454.65b	2628.59c			
H ₂ (S-278)						
	T ₁ =Control (no ABA application)	2728.97a	2922.55a			
	T ₂ =Stress at bud initiation and no ABA	1585.96d	1697.69d			
	T ₃ =Stress at bud initiation and ABA	2316.08b	2479.96b			
	T ₄ =Stress at flower initiation and no ABA	1124.25e	1203.66e			
	T ₅ =Stress at flower initiation and ABA	1814.19c	1942.92c			
H ₃ (SF-187)						
	T ₁ =Control (no ABA application)	3122.58a	3344.23a			
	T ₂ =Stress at bud initiation and no ABA	1939.34d	2076.73d			
	T ₃ =Stress at bud initiation and ABA	2745.32b	2923.76b			
	T ₄ =Stress at flower initiation and no ABA	1432.07e	1533.75e			
	T ₅ =Stress at flower initiation and ABA	2389.98c	2514.89c			
LSD (5%)		49.051	63.369			
Year mean		2139.51b	2303.36a			
LSD (5 %)	51.029					

 Table 2. Interactive effect of irrigation and ABA application scheduling on achene vield of sunflower hybrids.

Means sharing the same letter within a column don't differ significantly from each other at 5% probability level.

Sunflower hybrid DK-4040 had maximum leaf turgor pressure as compared to S-278 and SF-187. Drought stress significantly decreased leaf turgor pressure and more decrease in leaf turgor pressure was observed when stress was imposed at flower initiation than at bud initiation. Turgor pressure in sunflower hybrids was significantly improved by the application of ABA under drought stress at bud initiation than no ABA under similar condition (Fig. 4a). This increase was 12.94% in DK-4040, 10.59% in S-278 and 11.95% in SF-187 during 2008 and 12.95% in DK-4040, 10.43% in S-278 and 11.98% in SF-187 during 2009. Spray of ABA at flower initiation also significantly improved leaf turgor pressure than no ABA spray at same stage (Fig. 4b). This improvement was 13.48% in DK-4040, 7.93% for S-278 and 6.95% for SF-187 during 2008 and 13.50% in DK-4040, 7.81% in S-278 and 6.85% in SF-187 during 2009.

Sunflower hybrid DK-4040 had maximum relative leaf water contents as compared to S-278 and SF-187. Drought stress significantly decreased relative leaf water contents and more decrease in relative leaf water contents was observed when drought stress was imposed at flower initiation than at bud initiation. Applying ABA under stress at bud initiation significantly increased relative leaf water contents in sunflower hybrids (Fig. 5a). This improvement was 6.16% in DK-4040, 3.62% in S-278 and 4.68% in SF-187 during 2008. Similar trend was observed during both years of study. ABA application at flower initiation also significantly increased relative leaf water contents than no ABA application at same stage (Fig. 5b). This increase in relative leaf water contents was 5.19% in DK-4040, 3.10% for S-278 and 4.51% in SF-187 during first year of study. Same trend was observed during both years of study.



Fig. 4. Effect of irrigation and exogenous application of abscisic acid on leaf turgor pressure of sunflower hybrids; a) Budding b) Flowering; T_1 = 4 irrigations and no ABA; T_2 = Irrigation skip at budding and no ABA; T_3 = Irrigation skip at budding and ABA; T_4 =Irrigation skip at flowering and no ABA; T_5 =Irrigation skip at flowering and ABA.



Fig. 5. Effect of irrigation and exogenous application of abscisic acid on relative leaf water contents of sunflower hybrids; a) Budding b) Flowering; T_1 = 4 irrigations and no ABA; T_2 = Irrigation skip at budding and no ABA; T_3 = Irrigation skip at budding and ABA; T_4 =Irrigation skip at flowering and no ABA; T_3 =Irrigation skip at flowering and ABA.

Different irrigation cum ABA application schedules imposed on different hybrids of sunflower showed significant effect on achene yield during both years of study. Drought stress significantly decreased achene yield and more decrease in achene yield was observed when stress was imposed at flower initiation than at bud initiation. Achene yield in sunflower hybrids was significantly increased with the application of ABA under drought stress at bud initiation than no ABA under similar condition. This increase was 48.12% in DK-4040, 46.03% in S-278 and 41.55% in SF-187 during 2008 and 48.09% for DK-4040, 46.07% for S-278 and 40.78% for SF-187 during 2009. Foliar application of ABA at flower initiation also significantly increased achene yield than no ABA application at same stage. This improvement in achene yield was 102.75% for DK-4040, 61.36% for S-278 and 66.68% for SF-187 during 2008 and 102.79% in DK-4040, 61.41% in S-278 and 63.97% in SF-187 during 2009.

Discussion

In sunflower hybrids, differential water relations response was demonstrated by different statured genotypes. Present study categorized DK-4040 as tall statured hybrid, SF-187 as short stature hybrid and S-278 as intermediate stature hybrid. DK-4040 and SF-187 under drought either at budding or at flowering had higher water potential than S-278. This variation in water potential among sunflower hybrids was due to differences in their canopy architecture and root penetration (Angadi & Entz. 2002b). DK-4040 and SF-187 had compact crop canopy with larger leaves while sunflower hybrid S-278 had an open or lag canopy with smaller leaves. Number of leaves per plant was also more in S-278 than DK-4040 and SF-187. Short stature cultivar SF-187 had more leaf water potential under drought (Miller, 1992; Johnston et al., 1995) due to compact canopy (Sadras et al., 1991) and lesser transpiration rate (Pataki et al., 1998) as wind speed and light penetration reduced. Tall stature cultivar DK-4040 also showed more leaf water potential under drought as it has compact canopy and also deeper root system due to which not only reduced wind speed and light penetration reduced (Nobel, 1999) but also might has increased water extraction from deeper soil layer. Intermediate stature hybrid S-278 had lag canopy, smaller and more leaves which increased light penetration and dry air circulation (Giminez & Fereres, 1986) and finally rate of transpiration increased and its leaf water potential decreased under drought. Results further indicated that exogenous application of ABA under drought either at budding or at flowering significantly increased leaf water potential. This increase in leaf water potential was due to increase in water availability to plants. Water availability might be improved by conserving plant moisture due to partial closing of stomata, reduction in transpiration, increase in root penetration and inhibition of shoot growth (Salisburry & Marinos, 1985; Blum, 1997; Alfredo & Setter, 2000; Hoad et al., 2001).

Results of the present study indicated that sunflower hybrid DK-4040 (tall stature) under drought had more osmotic potential than SF-187 (short stature) and S-278 (intermediate stature). This more osmotic potential of DK-4040 and SF-187 than S-278 might categorize DK-4040 and SF-187 as drought tolerant and S-278 as drought sensitive sunflower hybrids. Genotypic variation for osmotic adjustment in sunflower hybrids was also reported (Chimenti & Hall, 1993; Iqbal & Ashraf, 2006). This variation was due to that SF-187 (short stature) initiated osmotic adjustment earlier (Angadi & Entz, 2002b) than medium stature sunflower hybrid (S-278) which indicated better response of SF-187 in osmotic adjustment and had more osmotic potential than S-278. Intermediate

sunflower hybrid (S-278) showed greater water depletion from soil as compared to short stature hybrid (SF-187) so they felt drought earlier and lowered its osmotic potential due to poor osmotic adjustment. The maintenance of higher osmotic potential of large stature sunflower hybrid (DK-4040) than intermediate stature hybrid (S-278) under drought was due to faster root penetration and more water extraction from deeper soil layers which delayed osmotic adjustment (Angadi & Entz, 2002b). So advantages of better drought tolerance in short and large stature sunflower hybrids were due to earlier osmotic adjustment by short stature (SF-187) and delay in osmotic adjustment by tall stature (DK-4040) hybrids. Exogenous application of ABA to sunflower hybrids under drought made osmotic potential less negative (increased) which was an indication that exogenous application of ABA improved drought tolerance in sunflower hybrids. ABA helped in mediating drought tolerance in plants (Zeevart, 1999) and its exogenous application to sunflower under drought converted tolerant cultivars to more tolerant and sensitive cultivars to tolerant cultivars (Quarrie, 1980; Cellier et al., 1998). Conversely exogenous application of ABA might increase the activity of certain enzymes like α -amylase and ribonuclease. These enzymes might breakdown starches and other materials to make osmotic potential more negative (Bradford & Hsiao, 1982).

Varietal differences in leaf turgor pressure were also observed. Sunflower hybrid DK-4040 (tall stature) under drought had maximum leaf turgor pressure as compared to S-278 (intermediate stature) and SF-187 (short stature). Genotypic variation for leaf turgor pressure under water deficits was also reported in field crops like maize (Saneoka et al., 1995), sunflower (Angadi & Entz, 2002b), tomato (Makela et al., 1998) and cotton (Meek et al., 2003). Drought stress significantly decreased leaf turgor pressure in sunflower hybrids and more decrease in leaf turgor pressure was observed when stress was given at flower initiation than at bud initiation. The maintenance of higher leaf turgor pressure of tall stature sunflower hybrid (DK-4040) than intermediate stature hybrid (S-278) under drought was due to faster root penetration and more water extraction from deeper soil layers (Angadi & Entz, 2002a) while in short stature hybrid (SF-187) it was due to earlier osmotic adjustment (Angadi & Entz, 2002a) and compact crop canopy which reduced rate of transpiration (Nobel, 1999) and finally increased water conservation in plants. Our findings further demonstrated that foliar application of ABA under drought significantly improved leaf turgor pressure. This increase in leaf turgor pressure was due to enhancement in water availability and conservation of water in plants which finally improved leaf water potential. Water availability might also be improved by conserving plant moisture due to partial closing of stomata, reduction in transpiration, increase in root penetration and inhibition of shoot growth (Salisburry & Marinos, 1985; Blum, 1997; Alfredo & Setter, 2000; Hoad et al., 2001).

Genotypic variation in leaf relative water contents was observed and sunflower hybrid DK-4040 (tall stature) under drought had maximum leaf relative water contents as compared to S-278 (intermediate stature) and SF-187 (short statured). Water deficit to sunflower hybrids at flowering reduced leaf relative water contents more as compared to drought given at budding. Reduction in leaf relative water contents under drought drastically affected sunflower growth and development (Velue & Palanisami, 2001; Lawlor, 2002; Tezara *et al.*, 2002). Higher leaf relative water contents of sunflower hybrids DK-4040 and SF-187 than S-278 was due to better water potential, turgor pressure and osmotic adjustment in these hybrids. Compact crop canopy of DK-4040 and SF-187 reduced rate of transpiration under drought which also improved their growth by increasing leaf relative water contents. Liu & Li, (2005) reported that low rate of transpiration under water deficits improved leaf relative water contents which ultimately

increased growth and physiological activities of drought tolerant line of wheat which was advantageous over drought sensitive line. Exogenous application of stress hormone ABA under drought significantly increased leaf relative water contents of sunflower hybrids. This improvement in leaf relative water contents was due to conservation of moisture in plants. Conservation of water in plants due to foliar application of ABA occurred by partial closing of stomata, reduction in transpiration, increase in root penetration and inhibition of shoot growth (Salisburry & Marinos, 1985; Blum, 1997; Alfredo & Setter, 2000; Hoad *et al.*, 2001).

References

- Ahmad, S., R. Ahmad., M.Y. Ashraf, M. Ashraf and E.A. Waraich. 2009. Sunflower (*Helianthus annuus* L.) response to drought stress at germination and seedling growth stages. *Pak. J. Botany*, 41: 647-654.
- Alfredo, A.C.A. and T.L. Setter. 2000. Response of cassava to water deficit: Leaf area growth and abscisic acid. *Crop Sci.*, 40: 131-137.
- Amdt, S.K., S.C. Clifford, W. Wanek, H.G. Jones and M. Popp. 2001. Physiological and morphological adaptations of the fruit tree *Ziziphus rotundifolia* in response to progressive drought stress. *Tree Physiol.*, 21: 705-715.
- Angadi, S.V. and M.H. Entz. 2002a. Agronomic performance of different stature sunflower cultivars. *Can. J. Plant Sci.*, 202: 117-125.
- Angadi, S.V. and M.H. Entz. 2002b. Water relation of standard height and dwarf sunflower cultivars. Crop Sci., 42: 152-159.
- Ashraf, M. and M.R. Foolad. 2006. Roles of glycinebetaine and proline in improving plant abiotic stress resistance. *Environ. Exp. Bot.*, 59: 206-216.
- Ashraf, M. and P.J.C. Harris. 2005. Abiotic stresses: plant resistance through breeding and molecular approaches. Haworth Press, New York.
- Azhar, N., M.Y. Ashraf, M. Hussain, M. Ashraf and R. Ahmed. 2009. EDTA-induced improvement in growth and water relations of sunflower (*Helianthus annuus* L.) plants grown in lead contaminated medium. *Pak. J. Botany*, 41: 3065-3074.
- Beard, B.H. and S. Geng. 1982. Interrelationship of morphological and economic characters of sunflower. Crop Sci., 22: 817-822.
- Blum, A. 1997. Crop responses to drought and the interpretation of adaptation. In: Drought Tolerance in Higher Plants: Genetical, Physiological, and Molecular Biological Analysis. (Ed.): I. Belhassen. Kluwer Academic Publish, Dordrecht, pp. 57-70.
- Bot, A.J., F.O. Nachtergaele and A. Young. 2000. Land resource potential and constraints at regional and country levels. *World Soil Resources Reports 90*. Land and Water Development Division, Food and Agriculture Organisation, Rome.
- Bradford, K.J. and T.C. Hsiao. 1982. Physiological responses to moderate water stress. In: *Physiological Plant Ecology II, Water relations and carbon assimilation.* (Eds.): O.L. Lange, P.S. Nobel, C.B. Osmond and H. Ziegler. pp. 263-324. In: A. Person, M.H. Zimmermann (Eds.) Encyclopedia of plant physiology, new series. Vol. 12B. Springer-verlag, Berlin, Heidelberg, New York.
- Cellier, F., G. Conejero, J.C. Breitler and F. Casse. 1998. Molecular and physiological responses to water deficit in drought sensitive lines of sunflower. *Plant Physiol.*, 116: 319-328.
- Chimenti, C.A. and A.J. Hall. 1993. Genetic variation and changes with ontogeny of osmotic adjustment in sunflower. *Euphytica*, 71: 201-210.
- Davies, W.J. and H.G. Jones. 1991. Abscisic Acid: Physiology and Biochemistry. BIOS Scientific Publishers, Oxford, UK, pp. 266.
- Gibson, A., K.T. Hubick and E.P. Bachelard. 1991. Effects of abscisic acid on morphological and physiological responses to water stress in *Eucalyptus camaldulensis* seedlings. *Aust. J. Plant Physiol.*, 18: 153-163.

- Gimenez, C. and E. Fereres. 1986. Genetic variability in sunflower cultivars under drought. II. Growth and water relations. *Aus. J. Agric. Res.*, 37: 583-597.
- Heschel, M.S. and N.J. Hausmann. 2001. Population differentiation for abscisic acid responsiveness in *Impatiens capensis* (Balsaminaceae). *Inter. J. Plant Sci.*, 162: 1253-1260.
- Hoad, S.P., G. Russell, M.E. Lucas and I.J. Bingham. 2001. The management of wheat, barley and oats root systems. *Adv. Agron.*, 74: 193-246.
- Iqbal, N. and M.Y. Ashraf. 2006. Does seed treatment with Glycinebetaine improve germination rate and seedling growth of sunflower (*Helianthus annuus* L.) under osmotic stress. *Pak. J. Bot.*, 38: 1641-1648.
- Jia, W. and J. Zhang. 1999. Stomatal closure is induced rather by prevailing xylem abscisic acid than by accumulated amount of xylem derived abscisic acid. *Physiol. Plant.*, 106: 268-275.
- Johnston, A.M., S.A. Brandt and D.A. Derksen. 1995. Relative performance of canola, sunola, and mustard. p. 208-215. In: Proc. Western Canada Agron. Workshop, 5-7 July, Red Deer, AB, Canada.
- Lawlor, D.W. 2002. Limitations to photosynthesis in water stressed leaves: stomata vs. metabolism and the role of ATP. *Ann. Bot.*, 89: 1-15.
- Lee, T.M, H.S. Lur and C. Chu .1993. Role of abscisic acid in chilling tolerance of rice (*Oryza sativa* L.) seedlings. I. Endogenous abscisic acid levels. *Plant Cell Environ.*, 16: 481-490.
- Li, C. 1999. Carbon isotope composition, water use efficiency and biomass productivity of *Eucalyptus microtheca* populations under different water supplies. *Plant Soil*, 214: 165-171.
- Li, C. and K. Wang. 2003. Differences in drought responses of three contrasting *Eucalyptus* microtheca F. Muell. Populations. For. Ecol. Manage., 179: 377-385.
- Liu, H.S. and F.M. Li. 2005. Root respiration, photosynthesis and grain yield of two spring wheat in response to soil drying. *Plant Growth Regul.*, 46: 233-240.
- Liu, H.S., F.M. Li. and H. Xu. 2004. Deficiency of water can enhance root respiration rate of drought sensitive but not drought tolerant spring water. Agri. Water Manage., 64: 41-48.
- Luquez, J.E., L.A.N. Aguirrezebal, M.E. Augero and V.R. Pereyra. 2002. Stability and adaptability of cultivars in non balanced yield trials, comparison of methods for selecting high oleic sunflower hybrids for grain yield and quality. *J. Agron. Crop Sci.*, 188(4): 225-234.
- Makela, P., K. Jokinen, M. Kontturi, P. Peltonen-Sainio, E. Pheu and S. Somersalo. 1998. Foliar application of glycinebetaine a novel product from sugar beet as an approach to increase tomato yield. *Indust. Crops Prod.*, 7: 139-148.
- Manivannan, P., C.A. Jaleel, A. Kishorekumar, B. Sankar, R. Somasundaram, R. Sridharan and R. Panneerselvam. 2007. Changes in antioxidant metabolism of *Vigna unguiculata* (L.) Walp., by propiconazole under water deficit stress. *Colloids Surf. B: Biointerfaces*, 57: 69-74.
- Meek, D., D. Oosterhuis and J. Gorhuam. 2003. Does foliar applied glycinebetaine affect endogenous betaine levels and yield in cotton. *Online Crop Manag. Doi.*, 10. 1094/CM-2003 O 804-02-Rs.
- Miller, J.F. 1992. Update on inheritance of sunflower characteristics. In: Proc. of 13th Int. Sunflower Conf., 7-11 Sept., Pisa, Italy, Vol. 1, Int. Sunflower Association, Toowoomba, Australia. p. 905-945.
- Moons, A., G. Bauw, E. Prinsen, M. Van, Montagu and D. Van Der Straeten. 1995. Molecular and physiological responses to abscisic acid and salts in roots of salt-sensitive and salt-tolerant indica rice varieties. *Plant Physiol.*, 107: 177-186.
- Nobel, P.S. 1999. *Physicochemical and environmental plant physiology*. p. 474. 2nd Edn., Academy Press, San Diego, CA, USA.
- Pandey, D.M., C.L. Goswami and B. Kumar. 2003. Physiological effects of plant hormones in cotton under drought *Biologia Plantarum*, 47(4): 535-540.
- Pataki, D.E., R. Orem and N. Phillips. 1998. Responses of sap flux and stomatal conductance of *Pinus taeda* L. trees to stepwise reductions in leaf area. J. Exp. Bot., 49: 871-878.
- Pekic, S. and S.A. Quarrie. 1987. Abscisic acid accumulation in maize differing in drought resistance: a comparison of intact and detached leaves. *J. Plant Physiol.*, 127: 203-217.

- Dreyer. 2001. Comparison of water-use efficiency
- Ponton, S., J.L. Dupouey, N. Breda and E. Dreyer. 2001. Comparison of water-use efficiency of seedlings from two sympatric oak species: genotype environment interactions. *Tree Physiol.*, 22: 413-422.
- Quarrie, S.A. 1980. Genotypic difference in leaf water potential, abscisic acid and proline concentration in spring wheat during drought stress. *Annals Botany*, 46: 383-394.
- Sadras, V.O., D.M. Whitefield and D.J. Connor. 1991. Regulation of evapotranspiration and its partitioning between transpiration and soil evaporation by sunflower crops: a comparison between hybrids of different stature. *Field Crops Res.*, 28: 17-37.
- Salisbury, B.F. and N.G. Marinos. 1985. The ecological role of plant growth substances. In: *Encyclopedia of plant physiology*. Vol. 11. (Eds.): R.P. Pharis and D.M. Reid. Springer-Verlag, Berlin, Heidelberg, New York.
- Saneoka, H., C. Nagaska, D. Hahn, W.J. Yang, G.S. Premachandra, R.J. Joly and D. Rhodes. 1995. Salt tolerance of glycinebetaine deficient and containing maize lines. *Plant Physiol.*, 107: 361-368.
- Scholander, P., H. Hammel, E. Bradstreet and E. Hemmingsen. 1965. Sap pressure in vascular plants. Science, 148: 339-345.
- Schonfeld, M.A., R.C. Johnson, B.F. Carver and D.W. Mornhinweg. 1988. Water relations in winter wheat as drought resistance indicator. *Crop Sci.*, 28: 526-531.
- Shamim, A., R. Ahmad, R.Y. Ashraf, M. Ashraf and E.A. Wariach. 2009. Sunflower (*Helianthus annuus* L.) response to drought stress at germination and seedling growth stages. *Pak. J. Bot.*, 41(2): 647-654.
- Somerville, C. and J. Briscoe. 2001. Genetic engineering and water. Sci., 292: 2217.
- Steel, R.G.D., J.H. Torrie and D.A. Dicky. 1997. *Priciples and Procedures of Statistics: A biometrical approach*.3rd edn.McGraw Hill Book Co.Inc.New York: 400-428.
- Tezara, W., V. Mitchell, S.P. Driscoll and D.W. Lawlor. 2002. Effects of water deficit and its interaction with CO₂ supply on the biochemistry and physiology of photosynthesis in sunflower. *J. Exp. Bot.*, 53: 1781-1791.
- Tsialtas, J.T., L.L. Handley, M.T. Kassioumi, D.S. Veresoglou and A.A. Gagianas. 2001. Interspecific variation in potential water use efficiency and its relation to plant species abundance in water-limited grassland. *Funct. Ecol.*, 15: 605-614.
- Van Rensburg, L. and G.H.J. Kruger. 1994. Applicability of Abscisic acid and (or) proline accumulation as selection criteria for drought tolerance in *Nicotiana tabacum. Can. J. Bot.*, 72: 1535-1540.
- Velue, G. and K. Palanisami. 2001. Impact of moisture stress and ameliorants on growth and yield of sunflower. *Madras Agric. J.*, 88(10-12): 660-665.
- Wang, Z.L., B.R. Huang and Q.Z. Xu. 2003. Effects of abscisic acid on drought responses of Kentucky bluegrass. J. Am. Soc. Hort. Sci., 128(1): 36-41.
- Xu, X., G.Q. Zheng, X.P. Deng and H. Medrano. 2002. Effects of exogenous abscisic acid and water stress on the growth response of subterranean clover of different genotypes. *Acta Bot. Sinica.*, 44(12): 1425-1431.
- Zeevart, J.A.D. 1999. Abscisic acid metabolism and its regulation. In: *Biochemistry and Molecular Biology of Plant Hormones*. (Eds.): P.J.J. Hooykaas, M.A. Hall, K.R. Libbenga. Elsevier Science, Amsterdam, pp. 189-207.

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