

FOLIAR NITROGEN AND POTASSIUM APPLICATIONS IMPROVE PHOTOSYNTHETIC ACTIVITIES AND WATER RELATIONS IN SUNFLOWER UNDER MOISTURE DEFICIT CONDITION

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Abstract

This study investigated the influence of foliar supplementation of nitrogen (N) potassium (K) and their combination on photosynthetic activities, physiological indices and water relations of two sunflower (*Helianthus annuus* L.) hybrids Hysen-33 and LG-5551 under water deficit condition. Studies were conducted in a wire-house at Nuclear Institute for Agriculture and Biology, Faisalabad, Pakistan. Treatments were two water stress levels [100 (control) and 60% field capacity (water deficit)], six levels of foliar spray (no spray, water spray, 1% N, 1% K, 0.5% N + 0.5% K and 1% N + 1% K) and each treatment was replicated three times. Results showed that water stress reduced the photosynthetic activities: P_n (photosynthetic rate), E (rate of transpiration) and g_s (stomatal conductance) and water relations i.e., ψ_w (water potential), ψ_s (osmotic potential) and ψ_p (turgor potential). Soil moisture deficit also significantly reduced the plant height, root length, fresh and dry matter which consequently affected the plant height stress tolerance index (PHSI), root length stress tolerance index (RLSI) and dry matter stress tolerance index (DMSI) in both sunflower hybrids. However, foliar supplementation with N and K or N+K improved the photosynthetic activities, water relations and physiological indices of both the sunflower hybrids. The findings of present study suggest that application of N+K is necessary to have high plant productivity.

Key words: Gas exchange, Water relations, N and K foliar supplementation and Sunflower

Introduction

Sunflower (*Helianthus annuus* L.) is an important oilseed crop which ranks third after soybean and groundnut as a source of edible oil in the world. Sunflower being a non-traditional oil seed crop have enough role in reducing a wide gap between production and consumption of edible oil in Pakistan (Khan *et al.*, 2002; Ahmad *et al.*, 2014). Sunflower is a very sensitive crop to water stress at flowering, fertilization and grain filling stages however, it is not very susceptible to water shortage at the start and end of the growing period (Ahmad *et al.*, 2009).

Drought tolerance is a mechanism that is essentially linked to crop ability to acquire soil water and utilize it efficiently (Richard *et al.*, 2010; Javed *et al.*, 2014). Various physiological, biochemical and molecular adaptations are exhibited by drought stressed plants to grow well under drought stress (Arora *et al.*, 2002). Impaired crop growth and reduced productivity are mainly attributed in plants suffered drought (Ashraf *et al.*, 1998). Further, drought affects water and nutrients supply to plants thus affecting adversely plant development and yield (Erdem *et al.*, 2002). Reduction in rate of photosynthesis and stomatal conductance was observed in plants grown under drought stress (Kawamitsu *et al.*, 2000).

Fertilizer is one of the basic inputs of agriculture and its timely availability is very crucial for agricultural production (Bukhari *et al.*, 2015). Nitrogen being important constituents of plant cell components, like amino acid, protein and nucleic acid, required in great quantity by all crops plants. The deficiency of N is one of the greatest constraints in crop production. A close relationship was established between N and water availability (Saneoka *et al.*, 2004, Shabbir *et al.*, 2015),

Drenovsky *et al.* (2012) stated that crop yield cannot be increased without sufficient water availability. The affirmative supply of N via roots has been clearly illuminated in relation to plant growth and its corresponding physiological process (Saneoka *et al.*, 2004; Zhang *et al.*, 2009; Ahmed *et al.*, 2014). Palta *et al.* (2005) reported that foliar application of N before terminal drought increased the photosynthetic activity and seed yield in chickpea.

Potassium (K) is a third most important macronutrient needed for plant growth and development after N and (P) phosphorous (Waraich *et al.*, 2011). Its foliar application improved the tolerance of crop plants against various types of abiotic stresses subsequently growth and yield (Ashraf *et al.*, 2013). Mengel & Kirkby (2001) reported that K improves physiological processes by the regulation of turgor pressure and photosynthesis; translocation of cations and enzymes activation. Cakmak (2005) also observed that plant suffering from drought stress require more internal K. In legumes, devastating effects of drought can be alleviated by rich K supply (Sangakkara *et al.*, 2000). Foliar application of K not only increases the tolerance of plants to drought stress but also helpful in maintaining the osmotic potential and water uptake and has a positive effect on stomatal closure (Waraich *et al.*, 2011). Leaf water potential, osmotic potential and moisture status are maintained at low and higher level in plants treated with appropriate K under drought. Adequate K fertilization of crop plants may facilitate osmotic adjustment which maintain turgor potential at lower leaf water potential and ultimately improve the ability of plants to tolerate drought conditions (Ashraf *et al.*, 2013).

It has been well documented that N and K uptake efficiency remained constrained when applied through roots as compared to foliar application under drought (Waraich *et al.*, 2011, Ahmad *et al.*, 2014). Thus under

drought stress conditions foliar application of these elements may be more proficient to minimize drought stressed injuries (Hu *et al.*, 2008). The present study was conducted to investigate the effect of foliar N, K and their combination applications on photosynthetic activities, water relation and growth of sunflower facing soil moisture deficit conditions.

Material and Methods

Experiment layout and plant material: This experiment was conducted in wire-house of Crop Stress Management Group at Nuclear Institute for Agricultural and Biological (NIAB), Faisalabad, Pakistan. Plastic pots containing 1.5 kg soil + sand were used as growth medium under two (100% and 60% field capacities) water levels with two sunflower hybrids; Hysun-33 (drought tolerant) and LG-5551 (drought sensitive), selected through lab screening (Rai, 2015). Plants were grown for five weeks and data on various physiological indices like plant height stress tolerance index (PHSI), root length stress tolerance index (RLSI) and dry matter stress tolerance index (DMSI) were estimated as described by Ashraf *et al.* (1996). Before harvesting water relations and gas exchange characteristics were recorded. Six treatments of nutrient application were maintained during the course of study (Table 1.)

The seeds of the above mentioned sunflower hybrids were allowed to germinate under normal condition (at 100 % field capacity) for one week. Then treatment i.e., 100 and 60 % field capacity were maintained upto five weeks. The recommended dose (120-60-60 NPK kg ha⁻¹) was applied as Urea (N= 46 %), diammonium phosphate (N= 18 %; and P₂O₅ 46 %) and sulphate of potash (K₂O = 50%) at start of experiment. Foliar applications of N, K and N+K treatments (Table 1) were imposed after two weeks of sowing (after 14 days). Field capacity i.e. 100 and 60 % of the pots were maintained by adding (distilled water) lost water through evapotranspiration. The experiment was laid out using a completely randomized design (CRD) with three replicates. Plants were harvested after four to five week and on the basis of various physiological indices like PHSI, RLSI, DMSI, water relations and gas exchange characteristics the best nutrient combination was selected.

Water relations parameters: The third leaf from top (fully expanded and photosynthetically active) of plants from each treatment was used to determine the leaf water potential (Ψ_w). The measurements were made from 8.00 to 10.00 a.m. with Scholander type pressure chamber, the same leaf, was frozen at -20°C for 72 h used for osmotic potential determinations (Ψ_s). The frozen leaf material was thawed and cell sap was extracted while crushing the leaves with a glass rod and then sap was sucked with a disposable syringe. The sap so extracted was directly used for the determination of Ψ_s using an osmometer (Wescor 5500). Turgor potential (Ψ_p) was calculated as the difference between Ψ_s and Ψ_w values, i.e. $\Psi_p = \Psi_w - \Psi_s$

Photosynthetic activity/Gas exchange characteristics: A fully expanded photosynthetically active leaf of each plant (third leaf from top) was used to measure the instantaneous photosynthetic rate (P_N), transpiration (E) and stomatal conductance (g_s) by using an open system LCA-4 ADC portable infrared gas analyzer (Analytical Development Company, Hoddesdon, England). These measurements were taken from 9.00 to 11.00 a.m. with the following adjustments: molar flow of air per unit leaf area 403.3 mmol m⁻²s⁻¹, atmospheric pressure 99.9 kPa, water vapor pressure into chamber ranged from 6.0 to 8.9 m bar, PAR at leaf surface was maximum up to 1711 mol m⁻² s⁻¹, temperature of leaf ranged from 28.4 to 32.4°C, ambient temperature ranged from 22.4 to 27.9°C and ambient CO₂ concentration was 352 mol mol⁻¹.

Results

Physiological indices: Growth parameters such as plant height, root length and dry matter were recorded and different physiological indices i.e. PHSI, RLSI and DMSI were calculated. The results showed that all these parameters were significantly affected by different N and K foliar application treatments in both the hybrids (Fig. 1). The highest values for PHSI (81%), RLSI (96%) and DMSI (86%) were observed with supplementary foliar application of 0.5% N + 0.5% K. The minimum values for these indices (62%), (78%) and (62%), respectively were recorded with no spray. Sunflower hybrid Hysun-33 exhibited the higher value (76%) for PHSI, (89%) for RLSI and (75%) for DMSI as compared to LG-5551 (70%), (77%) and (69%) respectively (Fig. 1).

Table 1. Treatments: Nutrient application detail.

Sr. #	Description	Rates	Designation
1.	Recommended dose of NPK as soil application	120-60-60 NPK kg ha ⁻¹	No spray
2.	Recommended dose of NPK as soil application + water spray	120-60-60 NPK kg ha ⁻¹ + Distilled water Spray	Water spray
3.	Recommended dose of NPK as soil application + N as foliar application	120-60-60 NPK kg ha ⁻¹ + 1% N as foliar application	1 % N spray
4.	Recommended dose of NPK as soil application + K as foliar application	120-60-60 NPK kg ha ⁻¹ + 1 % K as foliar application	1 % K spray
5.	Recommended dose of NPK as soil application + NK ₁ as foliar application	120-60-60 NPK kg ha ⁻¹ +NK (0.5 % each) as foliar application	0.5 % N + 0.5% K as foliar spray
6.	Recommended dose of NPK as soil application + NK ₂ as foliar application	120-60-60 NPK kg ha ⁻¹ +NK (1.0 % each) as foliar application	1.0 % N + 1.0 % K as foliar spray

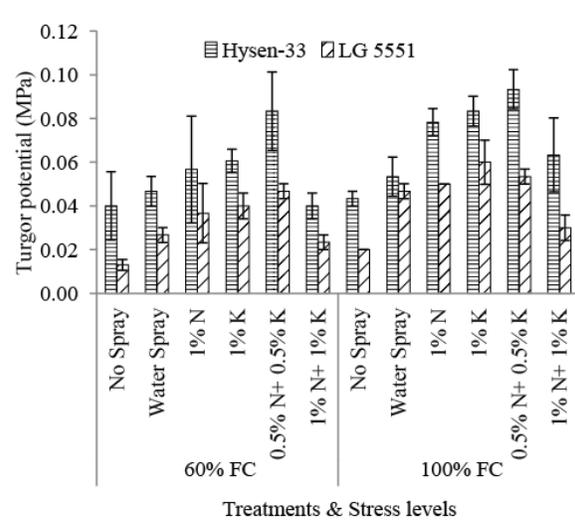
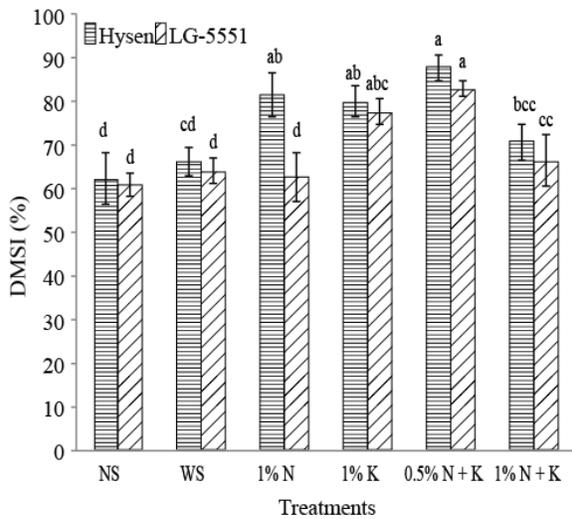
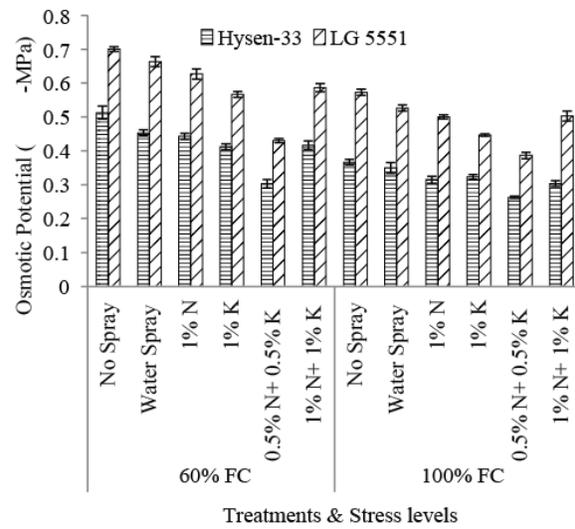
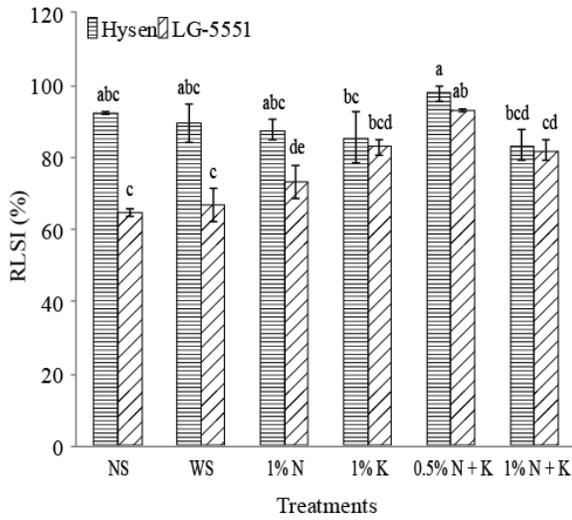
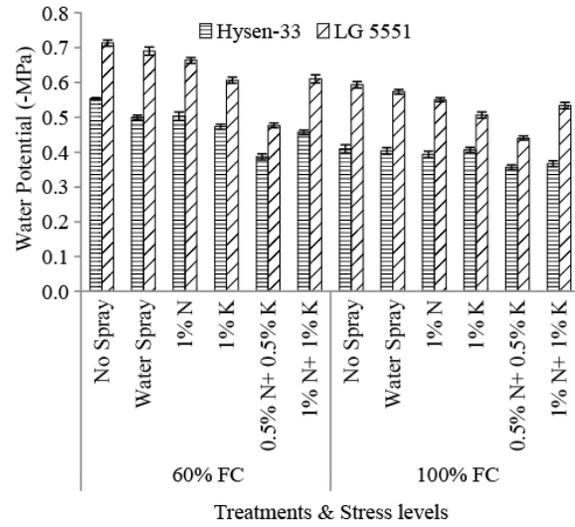
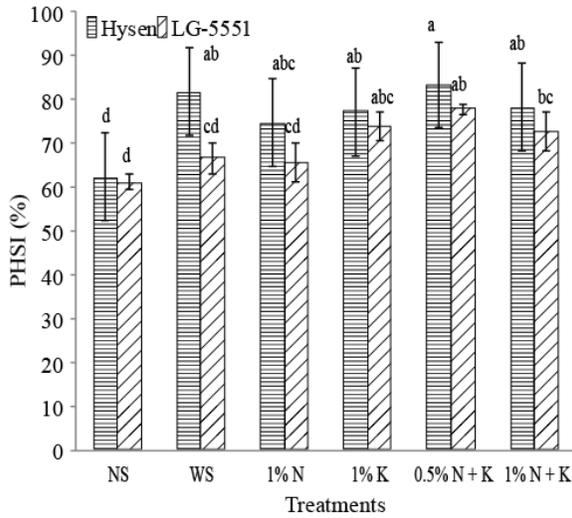


Fig. 1. Effect of N, K and their combinations foliar supplementation on PHSI, RLSI and DMSI of two sunflower hybrids grown under normal (100% field capacity) and water stress (60% field capacity) conditions.

Fig. 2. Effect of N, K and their combinations foliar supplementation on water potential (ψ_w), osmotic potential (ψ_s) and Turgor potential (ψ_p) of two sunflower hybrids grown under normal (100% field capacity) and water stress (60% field capacity) conditions.

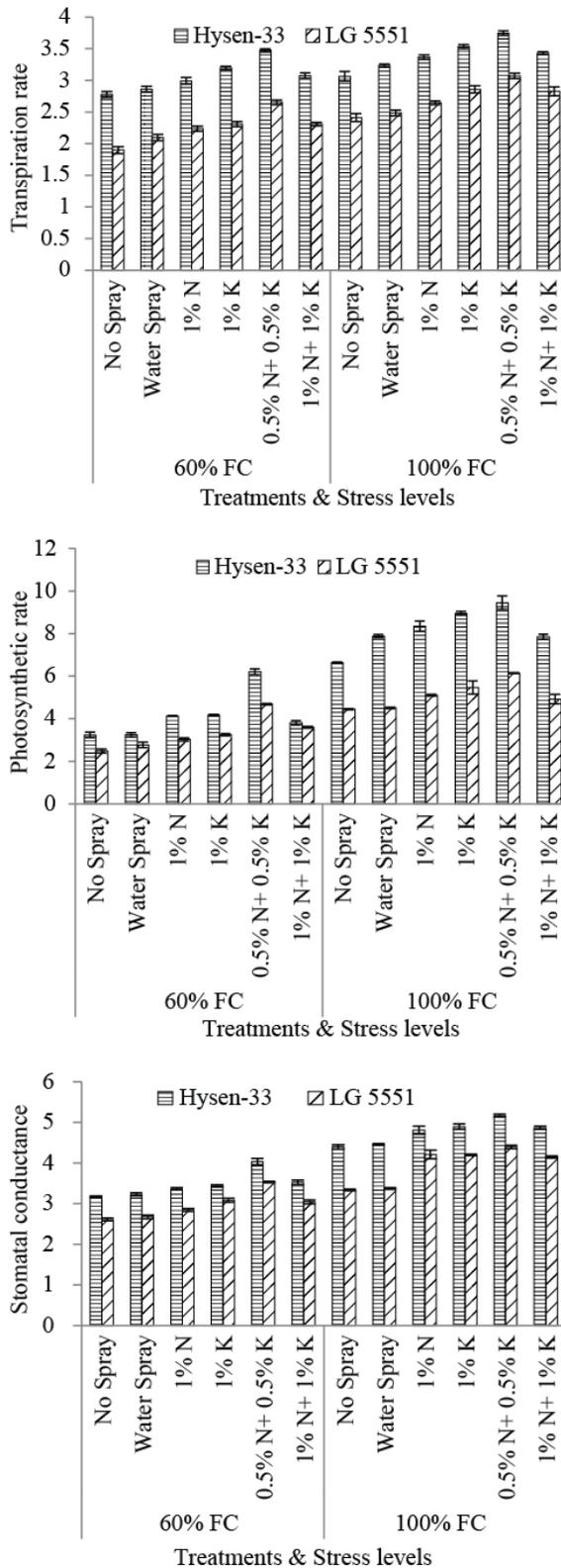


Fig. 3. Effect of N, K and their combinations foliar supplementation on Photosynthetic rate (P_N), Transpiration rate (E) and Stomatal conductance (g_s) of two sunflower hybrids grown under normal (100% field capacity) and water stress (60% field capacity) conditions.

Water relations parameters: Water stress significantly lowered the water relations parameters in sunflower as compared to normally irrigated plants however, different combinations of supplementary foliar application of N and K were found effective in maintenance of turgor. However, the highest value of ψ_w (-0.73 MPa) was recorded in plants sprayed with 0.5 % N+ 0.5 % K as compared to other treatments and the minimum (-0.84 MPa) was recorded in plants with no spray under control and stressed conditions (Fig. 2).

It is evident from the Fig. 2 that supplemental foliar application of N+K (0.5% each) did not affect ψ_s significantly in both sunflower hybrids. However, the highest value of ψ_s (-1.13 MPa) was recorded in plants sprayed with water. It was observed that supplementary foliar application of different treatments of N and K such as 1% N, 1% K, N+K (0.5% each) and N+K (1% each) has no considerable effects on ψ_s (Fig. 2).

Highly significant effect of drought stress was found on leaf turgor potential ψ_p in both the sunflower hybrids. The highest value of ψ_p (0.43 MPa) was recorded in plants supplied with 0.5% N+ 0.5% K (Fig. 2). A significant increase (0.36 MPa) in ψ_p was noted by foliar application of 1% N that was statistically at par with 1% K (0.36 MPa). The lowest value of ψ_p (0.32 MPa) was recorded in plants with no spray that did not differ significantly with water spray (0.32 MPa). The sunflower hybrid Hysun-33 showed maximum values for water relations as compared to LG-5551 under both normal and water stressed conditions (Fig. 2).

Photosynthetic activity/Gas exchange characteristics:

Data represented in Fig. 3 indicated that water stress significantly reduced the gas exchange characteristics i.e. P_N , E and g_{sin} both sunflower hybrids. Different combination of supplementary foliar application of N and K significantly increased all gas exchange characteristics. The highest values for P_N ($6.62 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$), E ($3.23 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$) and g_s ($4.28 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$) were observed with foliar application of 0.5% N+ 0.5% K however, the lowest values for these variables ($4.20 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$), ($2.53 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$) and ($3.38 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$) respectively was recorded with no spray (Fig. 3). Sunflower hybrids Hysun-33 maintained the highest values for these variables as compared to LG-5551 under both normal and water stressed conditions (Fig. 3).

Discussion

In our study significant effects of drought stress and supplementary foliar application of N and K was noted on physiological indices of both sunflower hybrids. The highest values for physiological indices were observed in plants sprayed with N+K (0.5% each) as compared to other treatments. These results are in accordance with Abou El Defan *et al.* (1999) and El-Sabbagh *et al.* (2002) who also described improvements in physiological indices with foliar spray of nutrients that are attributed due to their effective role in enhancing physiological processes involved in plant growth and delayed plant leaves senescence as well increasing photosynthetic activity. Dry and fresh weights of plants decreased under water stress.

However, a significant increase in dry matter stress tolerance index (DMSI) in both sunflower hybrids was observed with foliar application of 0.5% N+ 0.5% K in our study. Supplementary potassium fertilization at a lower dose had a stimulatory effect on DMSI but at higher dose it did not have a significant impact, which confirms the results obtained by Kaya *et al.* (2003). A pronounced effect was exerted by N fertilization on photosynthesis and dry matter partitioning between shoots and roots (Costa *et al.*, 2002). Many studies confirmed significant increase in root length under drought stress conditions (Türkan *et al.*, 2004; Ahmed *et al.*, 2009; Tuna *et al.*, 2010). Supplementary foliar application of nutrient plays a critical role in plants by increasing plant resistance to drought stress (Marschner, 1995).

Plants water relations were significantly influenced by water stress and are usually considered as an indicator of turgor maintenance in plants under water deficit conditions. In our study leaf osmotic potential significantly decreased in both sunflower hybrids under water stress. The highest ψ_s was recorded in Hysen-33 as compared to LG-5551 under both control and water stress conditions. Decreased ψ_s adversely affects the capability of plants to take up water from the surroundings (Munns, 2002). In the present study foliar application of N and K proved beneficial in improving water relation parameters under water stress. Maximum improvement in water relations parameters was observed in plants sprayed with N+K (0.5% each) as compared to other treatments. Previous studies described synergistic effects of combined N and K application on plant growth (Fridgen & Varco, 2004).

The decrease in Ψ_w caused a parallel decrease in osmotic potential (Ψ_s) of sunflower hybrids. However, supplemental foliar application of nutrients (N+K) had no significant effect on Ψ_s in this study. In contrast, Sultana *et al.* (2001) and Akram *et al.* (2009), who suggested that stress mediated decrease in Ψ_s can be minimized by the supplemental supply of nutrients. An adequate K status may facilitate osmotic adjustment, which maintains higher turgor pressure, relative water contents (RWC) and lower Ψ_s , thus improving the ability of plants to tolerate drought stress (Egilla *et al.*, 2005).

Decrease in P_N reduced the plant growth as also observed by (Ali *et al.*, 2007; Ahmed *et al.*, 2010; Demirevska *et al.*, 2010). The results of present study are in accordance with Cai *et al.* (2007), and Rapacz *et al.* (2010) who also demonstrated that reduction in P_N under water stress conditions may take place mainly due to closing of stomata that results in decreased E . Decrease in the absorption of essential elements and photosynthetic capacity under water stress was also reported by Kandil *et al.* (2001). Our results are in agreement with those of Wang *et al.*, 2010 who also observed reduction in P_N , E and g_s under water stress. In the present study, water stress significantly reduced the P_N , E and g_s in both sunflower hybrids. Reduction in g_s under low availability of water was also reported by Scheiber *et al.* (2008). Maintenance of plant water status is a vital phenomenon for normal growth of plants under stressful environment.

Disturbances in water balance in plants lead to impaired functioning of different gas exchange attributes, ultimately resulting in reduced plant growth (Demirevska *et al.*, 2010; Rapacz *et al.*, 2010). Under such conditions supplementary foliar application of N and K at the rate of 0.5% N + 0.5% K was found highly effective in ameliorating the adverse effects of water stress on gas exchange characteristics. Foliar spray of fertilizers partially minimized the nutrients deficiency in plants, increased photosynthesis and photosynthesis related parameters (Sultana *et al.*, 2001). Lejay *et al.* (2003) demonstrated a positive correlation between N status of leaf and photosynthetic activity of plants and expression of some transporter depends on the amount of available carbon. Walia *et al.* (2006) found that foliar application of urea also appeared to counteract the stress induced damage by maintaining growth and photosynthesis at moderate stress. Sugiharto *et al.* (1990) found a significant positive correlation between the photosynthetic capacity of leaves and their N nutrition suggesting that most of the N is used for synthesis of components of the photosynthetic apparatus (Pandey and Tyagi, 1999). An increase in P_N with foliar application of nutrients is interrelated with enhanced E and g_s under water stress conditions (Khan *et al.*, 2002). Unfavourable environmental conditions prompted injurious effects on photosynthetic activities of plants (Akhtar *et al.*, 2001; Kaymakanova & Stoeva, 2008). However supplemental foliar application of K fertilizer proved beneficial in minimizing these harmful effects. From the results of present study it can be concluded that N and K foliar supplementation @ 0.5% each is effective in improving photosynthetic activity/gas exchange characteristics under both normal and water stressed environments in sunflower plants.

Conclusion

The results of present study conclude that water stress significantly reduced the physiological indices, water relations and gas exchange characteristics of both sunflower hybrids. Foliar supplementation of 0.5% N + 0.5% K significantly enhanced the water relations and photosynthetic activity which improve the plant growth under water deficit conditions in sunflower hybrids.

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