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EFFECT OF PLANT GROWTH REGULATORS AND DROUGHT STRESS ON GROUNDNUT (*ARACHIS HYPOGAEA* L.) GENOTYPES

AYUB KHAN[1*], JEHAN BAKHT[2], ASGAHRI BANO[3] AND NASRULLAH JAN MALIK[4]

[1]*Agricultural Research Institute (N), Mingora, Swat, KPK, Pakistan.*
[2]*Institute of Biotechnology and Genetic Engineering, KPK Agricultural University, Peshawar, Pakistan.*
[3]*Department of Plant Sciences, Quaid-E-Azam University, Islamabad, Pakistan.*
[4]*Agriculture Research System, Agricultural University Peshawar, KPK, Pakistan.*

Abstract

Experiments were carried out to investigate the physiological (dry weight of root, stem, peg, flowers plant^{-1}, fruits sets percent, pod yield (kg plot^{-1}), 100-kernel weight, days to flowering and maturity) and biochemical (endogenous proline level) traits of groundnut cultivar Swat Phalli-96 under drought stress. The result showed that drought stress significantly ($p \leq 0.05$) reduced dry weight of root, peg/flowers plant^{-1}, fruits sets percent, pod yield (kg plot^{-1}), 100-kernel weight, days to flowering and maturity. GA and IAA applied as seed treatment or foliar spray had no significant ($p>0.05$) effect on various parameters under drought stress conditions. Foliar application of ABA (10^{-4} M) partially reduced the adverse effect of drought stress on growth and yield components. Foliar application of ABA to plants when subsequently exposed to drought stress resulted in elevated levels of endogenous shoot and root proline levels.

Introduction

Drought is one of the most important abiotic stress which affect crop growth and yield (Lutts *et al.,* 2004, Wahid, 2004; Luo *et al.,* 2005; Parida & Das, 2005; Mahajan & Tuteja, 2005; Castillo *et al.*, 2007; Khan *et al.*, 2010; Bakht *et al.*, 2010). The stage at which the drought stress occurs plays a major role in the final yield of the crop. Numerous physiological and biochemical changes occur in response to drought stress in various plant species. Many defense mechanisms i.e., osmoregulation, ion homeostasis, antioxidant and hormonal systems are invoked in tolerant plants which allow the plants to stay alive and properly develop prior to reproductive stages (Reddy *et al.,* 2004; Sairam & Tyagi, 2004; Mahajan & Tuteja, 2005; Ashraf, 2010). The alteration of protein synthesis or degradation is one of the fundamental metabolic processes that may influence drought tolerance. Both quantitative and qualitative changes of proteins were detected during water stress (Riccardi *et al*., 1998). Evidence is increasing in favor of a relationship between the accumulation of drought-induced proteins and physiological adaptations to water limitation (Riccardi *et al*., 1998; Han & Kermode, 1999). Research into the plant response to water stress is becoming increasingly important as most climatic change scenario suggest an increase in aridity in many areas of the globe. Agriculture is the major user of water resources in many regions of the world. With increasing aridity and growing population, water will become an increasingly scarce commodity in the near future. Even though in viable agriculture, severe water deficits should be a rare (but catastrophic) event, a better understanding of the effects of drought on plants is vital for improved management practices in breeding efforts in agriculture and for predicting the fate of natural vegetation under climatic change (Passioura, 2002).

Groundnut is cultivated mostly in the arid and semiarid regions of the world facing soil moisture deficit. Groundnut yield is a function of many plant and environmental factors, which are often inter-related. Several workers reported yield reduction with water stress induced at different stages of growth in groundnut (Balasubramanyam & Yayock, 1981). Under stress many pods are partly empty and assimilates are mostly transferred to the shell, hence maintaining nearly constant pod weight while reducing shelling percentage i.e., compensation of pod weight with shelling percentage. Kernel size itself seems to be slightly reduced under some conditions of stress.

Abscisic acid (ABA) plays an important role in abiotic stress (Bakht *et al.,* 2006; Bakht *et al.,* 2011). Root-originated xylem sap ABA can move to crop reproductive structures and accumulate there to a high level under drought conditions in wheat crop (Lie *et al.*, 2003). This elevated ABA content in the crop reproductive structures had been thought to be involved in controlling kernel pod^{-1} abortion, presumably *via* inhibition of cell division in the young ovaries (Setter & Ammgam, 2001; Lie *et al.*, 2003). In addition, exogenous application of ABA to developed maize ovaries inhibits cell division in the embryo and endosperm, and this effect is probably due to a depression cell-cycle gene expression by high levels of ABA (Setter & Ammgam, 2001). These studies suggested that drought induced increase in xylem sap (ABA) might affect expansion growth of crop reproductive structure resulting in a weak sink intensity, which fails to attract assimilate from source organs and eventually leads to abortion.

A comprehensive knowledge of the biochemical basis of drought tolerance of different genotypes will help to identify suitable drought tolerant species for drought affected areas. Biochemical (proline) changes in plants growing under water stress conditions have been broadly investigated in many crop species (Castillo *et al.,* 2007; Cha-um *et al.,* 2007a and b; Hu *et al.,* 2007; Teixeira & Pereira, 2007; Wang *et al.,* 2008; Cha-um *et al.,* 2009a and b; Cha-um *et al.,* 2010). Such parameter in crop species under drought condition has been developed as indicator for tolerance selection in breeding programs (Ashraf & Haris, 2004; Parida & Das, 2005; Ashraf & Foolad, 2007). The present study investigates the response of groundnut genotypes to seed and foliar application of different phytohormones under drought conditions.

[*]Corresponding author e-mail: peerayub@gmail.com

Materials and Methods

Pot experiments were conducted to study the response of groundnut genotype Swat Phalli-96, to various growth regulators (Gibrrellic acid (GA), Indole-3-acetic acid (IAA) and Abscisic acid (ABA) applications and induced drought stress. These experiments were carried out at Agriculture Research Institute Mingora, Swat KPK (1150 asl, 34° 10' to 35° 56' North latitude and 72° 7' to 73°0' East longitude) Pakistan using randomized complete block design (RCBD) with three replications. The seeds of groundnut cultivar Swat Phalli-96 were sown in earthen pots measuring 30 cm x 40 cm, containing soil and farmyard manure in the ratio of 3:1. Recommended agronomic practices were carried out uniformly for all the treatment. Seed were soaked in 10^{-4} M solution of different growth regulators viz., GA, IAA and ABA for 6 h prior to sowing. For control seeds were soaked in distilled water. For foliar application, groundnut plants were sprayed with GA, IAA and ABA having the same concentration as used for seed soaking at 40 days after sowing. Plants were sprayed between 10.00 -12.00 h. During spray soil in the pots was covered with aluminum foil to avoid contamination of soil with the applied growth-regulators/hormones. Drought stress was imposed at three critical growth stages i.e. -40 days after sowing (DAS;flowering initiation), 41-60 DAS (flowering and peg formation stage) and 61-80 DAS (pod development stage). Soil moisture was measured at the start, mid and end of each induced drought period (Fig. 1). Data on days to first flowering was recorded by counting the number of days from sowing, till the appearance of first flower in 50% plants in each plot. Days to maturity data were noted from the date of sowing till the crop reached physiological maturity. Height of three plants in each treatment was taken from the ground to the tip to record data on plant height. The length of 20 pods randomly selected in each treatment was measured. At maturity plant were harvested and yield was calculated, whereas the harvested plants were dried till constant weight**.** Endogenous proline concentration in leaves was determined according to the method of Bates *et al.*, (1973).

Statistical analysis: All data are presented as mean values of three replicates. Data were analyzed statistically for analysis of variance (ANOVA) following the method described by Gomez & Gomaz (1984). MSTATC computer software was used to carry out statistical analysis (Russel & Eisensmith, 1983). The significance of differences among means was compared by using Least Significant Difference (LSD) test (Steel & Torrie, 1997).

Results and Discussion

Plant growth and yield: Seed soaking or foliar application of GA and IAA had no significant ($p>0.05$) effect on any parameter under study, therefore, the present paper only reports the effect of foliar application of ABA. Drought and ABA application had a significant ($p<0.05$) effect on pod yield (Fig. 2). Drought stress reduced 46% pod yield when compared with control. There was an increase of 11% in pod yield when drought stress and ABA was applied at 41-60 DAS compared with drought stressed plants alone (Fig. 2). Wheat and other grain crops under water deficit during grain filling substantially affects grain weight (Rahman & Yoshida, 1985) due to early plant senescence, cessation of grain filling (Hossain *et al.*, 1990) and shortening of the grain filling period (Royo *et al*., 2000).

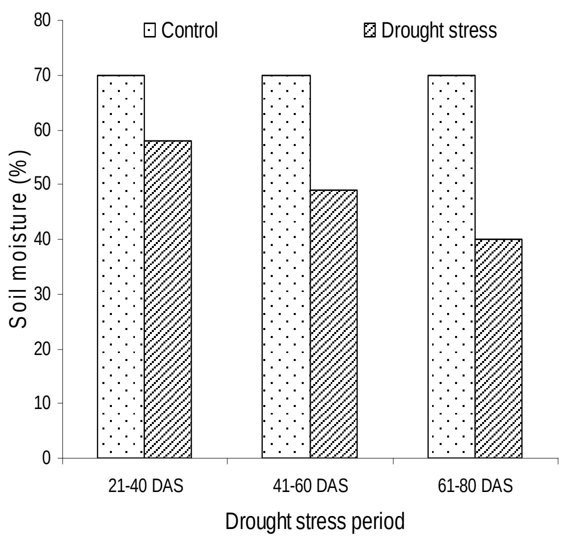

Fig. 1. Soil moisture (%) in pot as affected by different drought stress period.

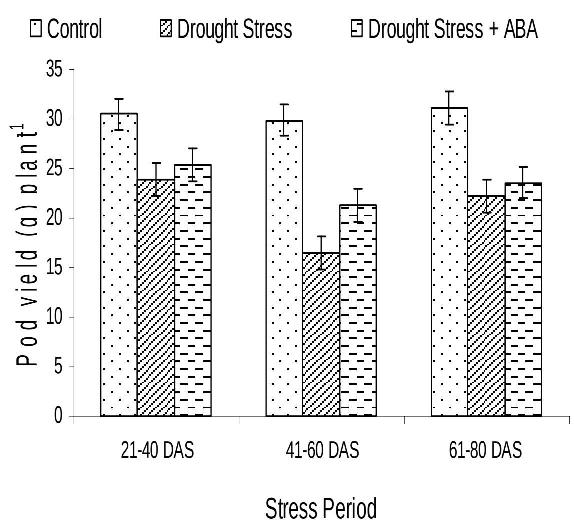

Fig. 2. Effect of drought stress and foliar application of ABA on pod yield (g) plant^{-1} of groundnut variety Swat Phalli-96. The bars show ± 1 LSD at $p<0.05$.

Pod length was significantly ($p<0.05$) reduced by drought stress. Drought stress applied at 41-60 DAS resulted a 9% decrease in pod length when compared with drought stress applied at 21-40 DAS. Drought stressed plants sprayed with ABA, resulted in 7% increase in pod length compared with plants experiencing drought stress alone. Statistical analysis of the data indicated that there was an increase of 6% in number of flowers plant^{-1} when drought stress was applied at 41-60 DAS compared with 21-40 DAS. Drought stress applied at 61-80 DAS significantly ($p<0.05$) increased the number of flowers plant^{-1} by 8% over that of previous value. ABA applied to drought stressed plants increased number of flowers plant^{-1} by 16% compared with drought stressed plants alone (Fig. 3). There was a decrease of 6% in fruit set when drought stress was applied at 41-60 DAS compared with 21-40 DAS (Fig. 4). Similarly, fruit set was increased by 7% when drought stress was applied at 61-80 DAS compared with 41-60 DAS. When the effect of drought stress and ABA application was taken into consideration, drought stress decreased fruit set by 74% when compared with the control. Application of ABA to drought stressed resulted in 13% increase in fruit set when compared with drought stressed plants alone. Drought stress had decreased kernels weight by 8% when compared with control. ABA application showed 5% increase in kernel weight when compared with drought stressed plants alone (Fig. 5). Drought stress applied at 41-60 DAS was less detrimental to the production of haulm yield. A decrease of 5% in haulm yield was recorded when compared with the drought stress imposed at 61-80 DAS. It is interesting to note that stress applied at 41-60 DAS performed better with respect to haulm yield compared with stress applied at other growth stages of the groundnut plant. When drought stressed plants were sprayed with ABA, a decrease of 15% in haulm yield was recorded when compared with drought stress alone treatment (Fig. 6). Similar results are also reported by Rahman & Yoshida, (1985), Hossain *et al.*, (1990) and Royo *et al.*, (2000).

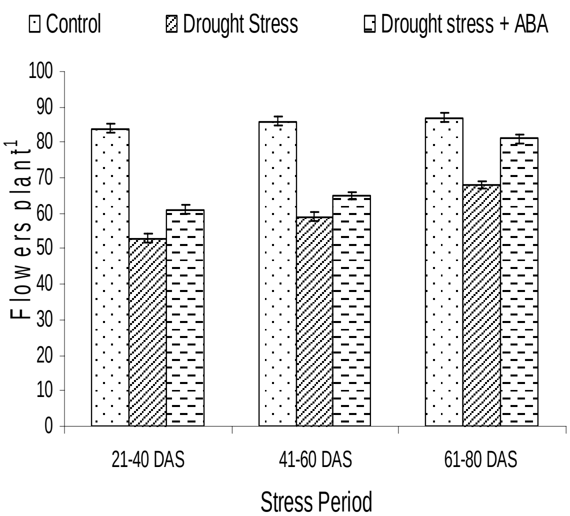

Fig. 3. Effect of drought stress and foliar application of ABA on flowers plant^{-1} of groundnut variety Swat Phalli-96. The bars show ± 1 LSD at $p<0.05$.

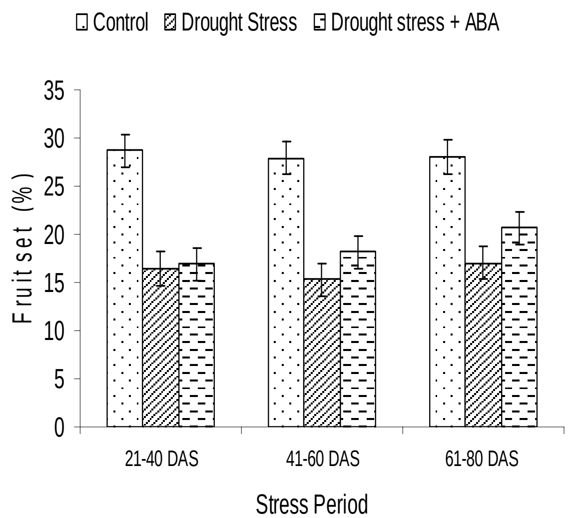

Fig. 4. Effect of drought stress and foliar application of ABA on fruit set (%) of groundnut variety Swat Phalli-96. The bars show ± LSD at $p<0.05$.

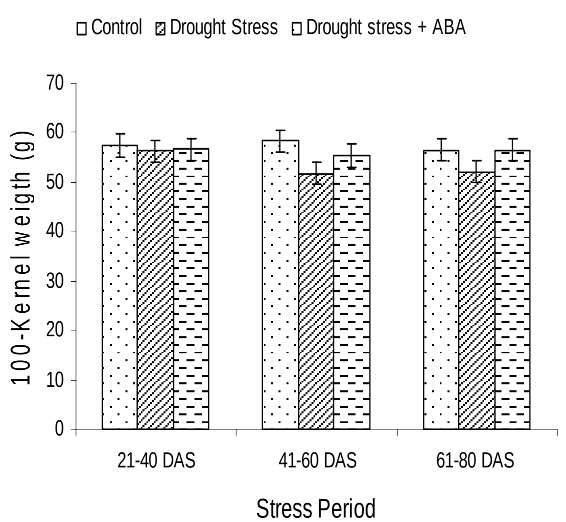

Fig. 5. Effect of drought stress and foliar application of ABA on 100-kemets weight of groundnut variety Swat Phalli-96. The bars show ± 1 LSD at $p<0.05$.

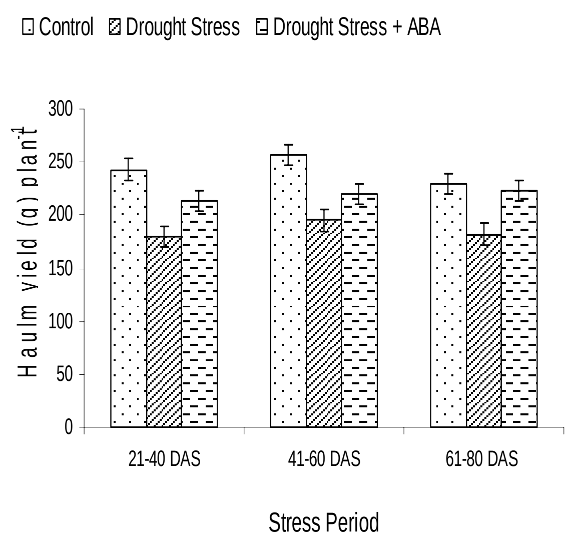

Fig. 6. Effect of drought stress and foliar application of ABA on haulm yield (g) plant^{-1} of groundnut variety Swat Phalli-96. The bars show ± 1 LSD at $p<0.05$.

Plants exposed to drought stress took 11% more days to flowering when compared with control. Similarly, when ABA was sprayed on drought stressed plants, days to flowering were reduced by 8% compared with the drought stressed plants alone. Days to flowering were more when drought stressed was imposed at 21-40 days after sowing (DAS) compared with drought stress applied at other growth stages and ABA application (Table 1). Plants exposed to drought stress took minimum days to maturity (Table 1). Similarly, when drought stressed plants were sprayed with ABA, an increase of 5% in days to maturity was observed compared with drought stress treatments (Table 2).

Table 1. Days to flowering of groundnut variety Swat Phalli-96 as affected by drought stress and ABA application.

Treatment	Growth stages (days after sowing)			
	21-40 DAS	**41-60 DAS**	**61-80 DAS**	**Mean**
Control	33.75 b	34.00 b	33.00 b	33.58 b
Drought Stress	41.00 a	35.00 ab	36.00 ab	37.67 a
Drought stress + ABA	36.00 ab	34.00 b	35.00 ab	35.00 b
Mean	36.92 a	34.33 b	34.67 b	

Means followed by different letters are significantly different at 5% level of probability using least significant difference (LSD) test
LSD value for growth stages at $p<0.05$ = 2.065
LSD value for drought stress at $p<0.05$ = 2.065
LSD value for interactions at $p<0.05$ = 3.577

Table 2. Days to maturity of groundnut variety Swat Phalli-96 as affected by drought stress and ABA application.

Treatment	Growth stages (days after sowing)			
	21-40 DAS	**41-60 DAS**	**61-80 DAS**	**Mean**
Control	160 ab	159 ab	161 a	160 a
Drought stress	146 d	152 c	155 bc	151 b
Drought stress + ABA	157 a-c	160 ab	159 ab	159 a
Mean	154	157	158	

Means followed by different letters are significantly different at 5% level of probability using least significant difference (LSD) test
LSD value for growth stages at $p<0.05$ = 3.416
LSD value for drought stress at $p<0.05$ = 3.416
LSD value for interactions at $p<0.05$ = 5.917

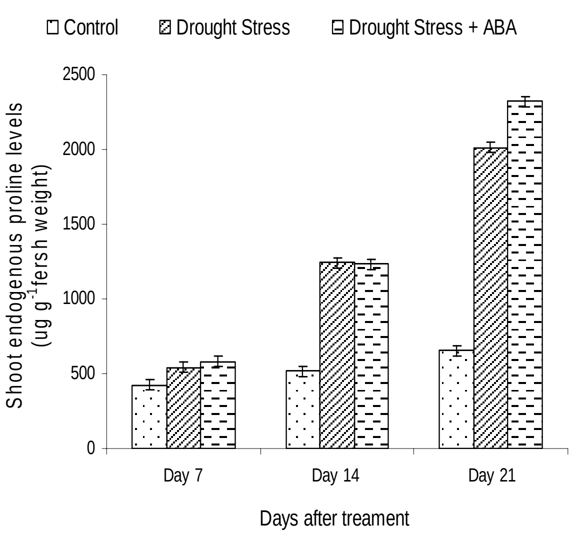

Fig. 7. Effect of drought stress and foliar application of ABA on shoot endogenous praline levels (ug g^{-1} fresh weight). The bars show ± 1 LSD at $p<0.05$.

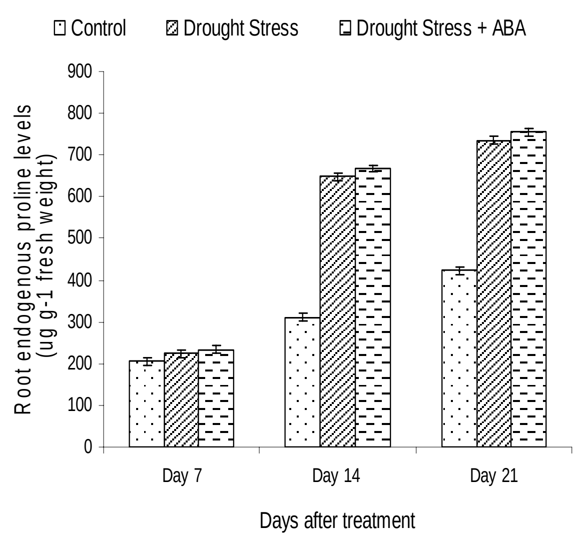

Fig. 8. Effect of drought stress and foliar application of root endogenous praline levels (ug g^{-1} fresh weight). The bars show ± 1 LSD at $p<0.05$.

Endogenous proline level: Endogenous proline levels were measured from the plants exposed to drought stress at 41-60 DAS and applied with GA, IAA and ABA either as seed soaking or foliar spray. Data was recorded on 7, 14 and 21 days post treatments of drought stress and phytohormone application. The data indicated that only foliar application of ABA showed significant ($p<0.05$) effect on the endogenous proline levels whereas, the other treatments were non-significant ($p>0.05$). Therefore, for simplicity only results of the foliar application of ABA (10^{-4} M) are presented here.

Drought stress also had a significant ($p<0.05$) effect on shoot proline levels (Fig. 7). A broad similar pattern for proline production was also observed when root proline levels were compared (Fig. 8). A significant ($p<0.05$) time-dependent increase was observed in control but a much larger change was observed in untreated drought stressed plants. By day 7 root proline levels in untreated drought stressed plants was 64.84% higher than control and increased to 73% by day 21. Endogenous proline levels were significantly ($p<0.05$) affected by the application of ABA and drought stress by day 21 when compared with drought stressed plants alone. By day 21, there was a significant ($p<0.05$) increase of 92.84% in endogenous proline levels of shoot in plants treated with drought stress + ABA over drought stressed plants alone (Fig. 7). It was revealed that non-significant ($p>0.05$) increase was observed in roots proline levels between drought stressed plants and plants treated with drought stress + ABA at any time period. Similar results are also reported by Castillo *et al.*, (2007) Cha-um *et al.*, (2007a and b), Hu *et al.*, (2007), Teixeira & Pereira, (2007), Wang *et al.*, (2008), Cha-um *et al.*, (2009 a and b) and Cha-um *et al.*, (2010).

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