

ENHANCEMENT IN COTTON GROWTH AND YIELD USING NOVEL GROWTH PROMOTING SUBSTANCES UNDER WATER LIMITED CONDITIONS

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Abstract

To investigate the role of different growth promoting substances including glycine betaine, salicylic acid and ascorbic acid individually and in different combinations on the growth and yield of promising cotton cultivars viz. Lalazar and FH-142 under limited water supply, a pot study was conducted at research farm of University College of Agriculture, University of Sargodha, Sargodha. Two watering regimes viz. well watered (normal irrigation condition) and irrigation at 60 % field capacity level (drought condition) was maintained at appearance of first true leaf of the cotton plant. Exogenous application of different plant growth regulators (PGRs) on two cotton cultivars (Lalazar and FH-142) included: distilled water spray (control), distilled water spray + tween-20 (0.1%), glycine betaine @ 100 mg L⁻¹ + tween-20 (0.1%), salicylic acid @ 100 mg L⁻¹ + tween-20 (0.1%), ascorbic acid @ 100 mg L⁻¹ + tween-20 (0.1%), glycine betaine and salicylic acid @ 50 mg L⁻¹ each + tween-20 (0.1%), glycine betaine and ascorbic acid @ 50 mg L⁻¹ each + tween-20 (0.1%), salicylic acid and ascorbic acid @ 50 mg L⁻¹ each + tween-20 (0.1%), glycine betaine + salicylic acid + ascorbic acid @ 33 mg L⁻¹ each + tween-20 (0.1%). Results indicated that different plant growth regulators significantly affected number of days to squaring, number of days to first boll splitting, number of monopodial and sympodial branches per plant, number of main stem nodes per plant, plant height, height to node ratio, membrane stability index, relative water contents, first fruiting branch node number, first fruiting branch node height, number of open bolls per plant, average boll weight, lint yield, cotton seed yield per plant and GOT (%). Yield attributes of cotton were enhanced with combined application of salicylic acid and ascorbic acid each at 50 mg L⁻¹ each under drought conditions. However, above stated yield and yield attributes were enhanced with exogenous application of glycine betaine, salicylic acid and ascorbic acid each at 33 mg L⁻¹ under normal irrigation conditions. FH-142 is suitable for good production under drought conditions with the tested PGRs.

Key words: Cotton, Drought stress, Growth, Plant growth regulator, Yield.

Introduction

To grow crops under water limited regimes, a multitude of strategies are currently underway throughout the world including generation of drought tolerant transgenics, exogenous application of bioregulators, water management practices, etc. Through all such means, water potential inside the plant body is maintained below that of soil so as to maintain turgidity of cells and hence overall maintenance of all metabolic functions therein (Brillante *et al.*, 2016). A variety of inorganic and organic compounds including organic compatible solutes have been recommended for protecting crops against abiotic stresses (Ashraf *et al.*, 2008). Accumulation of these organic compatible solutes not only depends upon environmental factors but also on type of a plant species or plant variety (Moghaieb *et al.*, 2004; Hassine *et al.*, 2008; Cui *et al.*, 2015; Yu *et al.*, 2016).

Cotton (*Gossypium hirsutum* L.), being a potential fiber crop of many countries, is believed to tolerate drought, but it responds well to sufficient quantity of water in terms of attaining an optimum quantity of lint yield (Lv *et al.*, 2007; Ahmed *et al.*, 2018). Cotton plant has been reported to respond to water scarcity by reducing stomatal area, increasing leaf temperature, decreasing maximum and actual quantum yield of PS-II, CO₂ assimilation rate and chlorophyll contents to some extent (Ullah *et al.*, 2008; Filippou *et al.*, 2014; Hejnák *et al.*, 2015). Adaptive mechanism of cotton to water limited condition is reported to be maintenance of higher transpiration in contrast with most of other crops like soybean, potato and wheat

(Subrahmanyam *et al.*, 2006; Liu *et al.*, 2014;). Larger stomatal area, greater stomatal conductance, higher flow rate of stem sap, higher transpiration rate to lower leaf temperature to protect photosynthetic apparatus from damaging effects of solar radiations and diheliotropism are the features related to adaptive strategies of cotton under water limited conditions (Inamullah & Isoda, 2005a, 2005b; Deeba *et al.*, 2012).

However, it is well established that different cultivars of cotton respond differentially to water limited conditions in terms of differences in maintenance of relative water contents in leaves and degree of reduction in chlorophyll, and protein and carotenoid contents (Ullah *et al.*, 2008). A lot of interest has emerged on the possibility of improving tolerance of crops against stressful environments by exogenous application of growth promoting substances (Athar *et al.*, 2008; Divi & Krishna, 2009; Ullah *et al.*, 2017). However, the development of strategies to improve drought tolerance without losing productivity is a major milestone yet to achieve. Among different meaningful strategies, improved crop varieties could be the main focus of plant breeders, but the application of drought mitigating substances has a substance in view of their potential for crop improvement under environmental cues as well as being a shot-gun cost-effective approach (Singh *et al.*, 2015). The quaternary ammonium compound glycine betaine has been widely used in the past for attaining sustainable crop production under harsh environmental cues (Ashraf & Foolad, 2007). However, variable results have been recorded by different researchers regarding application of

glycine betaine in different crops depending upon time, amount and environmental conditions (Ma *et al.*, 2007; Iqbal *et al.*, 2008; Wani *et al.*, 2013). Salicylic acid is another organic compound which occurs naturally in plants and it is known for protecting plants against oxidative stress (Dianat *et al.*, 2016; Jabeen *et al.*, 2018). Ascorbic acid (vitamin C) has also been one of the favorite organic compounds as plant growth regulator under both normal and stress conditions (Malik & Ashraf, 2012; Terzi *et al.*, 2015). In view of the literature, it is amply clear that exogenous application of all such plant regulators has been carried out individually and reports on the use of combinations of these compounds are rare. Thus, the present study was performed to examine the role of glycine betaine, salicylic acid and ascorbic acid individually and in different combinations on the yield and growth of some selected cotton cultivars under limited water supply.

Material and Methods

A pot experiment was conducted to assess the role of selected novel growth promoting substances in enhancing cotton productivity under limited water conditions at the Research Farm of University College of Agriculture, University of Sargodha during 2015-16. Two cotton cultivars viz. FH-142 and FH-Lalazar were sown during the cotton growing season. The arrangement of the experiment followed a completely randomized design with 2×2×10 factorial arrangement and each treatment replicated four times. Equal weight earthen pots (5.5 kg) were taken and each contained 8.5 kg dry sandy loam soil. Normal irrigation water was applied to each pot to get it completely saturated. Thereafter, all pots were kept in an open air to bring the soil moisture to field capacity. At this stage, 10 healthy seeds of each cultivar were sown in each pot. Thinning was done to maintain one healthy plant per pot after one week after germination. Two watering regimes viz. well watered (normal irrigation condition) and irrigation at 60 % field capacity level (limited water condition) was maintained at appearance of first true leaf of the cotton plant. Moisture contents of the pot soil were daily appraised and maintained at full field capacity or 60 % field capacity by weighing the pots. Exogenous application of different plant growth regulators (PGRs) included: distilled water spray, distilled water spray + tween-20 (0.1 %), glycine betaine @ 100 mg L⁻¹ + tween-20 (0.1 %), salicylic acid @ 100 mg L⁻¹ +tween-20 (0.1 %), ascorbic acid at 100 mg L⁻¹ +tween-20 (0.1 %), glycine betaine and salicylic acid @ 50 mg L⁻¹ each +tween-20 (0.1 %), glycine betaine and ascorbic acid @ 50 mg L⁻¹ each +tween-20 (0.1 %), salicylic acid and ascorbic acid @ 50 mg L⁻¹ each +tween-20 (0.1 %), glycine betaine + salicylic acid +ascorbic acid @ 33 mg L⁻¹ each+tween-20 (0.1 %). A control treatment without any chemical or water spray was also included. The chemical treatments were applied at 3, 6 and 9 weeks after emergence of crop seedlings. Recommended doses of nitrogen (160 kg N ha⁻¹), phosphorus (70 kg ha⁻¹) and potash (50 kg ha⁻¹) were applied to each pot to produce healthy plants. The NPK fertilizers were added to each pot based on the calculation of NPK fertilizers required

for acre furrow slice weight. Data of different phenological parameters including days taken to emergence, number of days taken to squaring, number of days taken to appearance of first flower, and number of days taken to first boll split, were recorded for each pot at particular phenological stage. Measurement of the height of first fruiting branch (cm) was done from each plant pseudo-node along with first fruiting branch node number. Other agronomic traits like number of sympodial branches per plant, number of monopodial branches per plant, plant height (cm) at maturity, number of main stem nodes per plant, number of opened and unopened bolls per plant, average boll weight (g), seed cotton yield per plant (g), cotton seed yield (g) per plant, lint yield per plant and ginning out turn (GOT) % were recorded using the standard procedures.

Cell membrane stability was determined by taking two sets of leaf samples each 200 mg were weighed. An aliquot of 10 ml double distilled water was added to each of the two test tubes. From the two samples of each treatment, one test tube was kept at 40°C in a water bath for 30 min and then measured the electrical conductivity (C1). The other test tube was incubated at 100°C for 15 min and measured the electrical conductivity (C2). MSI was calculated following Premachandra *et al.*, (1990) using the following formula:

$$\text{Membrane Stability Index (MSI)} = \frac{[1 - C1]}{C2} \times 100$$

The relative water content was estimated following Schonfeld *et al.*, (1988).

$$\text{RWC (\%)} = \frac{(\text{FW} - \text{DW})}{(\text{TW} - \text{DW})} \times 100$$

Leaf relative water content was calculated after 7 days of exogenous application of all PGRs.

Statistical Analysis

The data recorded for different attributes were subjected to analysis of variance test. The Tukey HSD test was employed to appraise significant differences among the treatment means (Steel *et al.*, 1997).

Results

Days to emergence of cotton seed was similar under drought and normal irrigation conditions and emergence was completed within 5 or 6 days of sowing (Table 1). Similarly, there was no difference between the cultivars for days to emergence. Both varieties completed emergence in 5 days. Foliar-applied PGRs caused earlier start of squaring when compared with the control treatments. Ascorbic acid at 100 mg L⁻¹ recorded minimum number of days to start squaring in cv. Lalazar under normal irrigation (37 days) and drought conditions (34 days). Similarly, FH-142 showed a minimum number of days to start squaring under normal irrigation (36 days) and drought (33 days) where ascorbic acid at 100 mg L⁻¹ was sprayed (Table 1). Data showed that drought conditions caused early start of squaring in both cultivars (36

days) when compared with normal irrigation condition (39 days). Minimum days (43) to start flowering were recorded in pots where salicylic acid at 100 mg L^{-1} was applied on cv. Lalazar under drought conditions that was similar to that caused by a mixture of glycine betaine and ascorbic acid each at 50 mg L^{-1} when applied under normal irrigation conditions. In contrast, combination of glycine betaine, salicylic acid and ascorbic acid caused delay in flowering in cv. FH-142 under normal irrigation conditions which was similar to that of control or distilled water spray. Cultivar Lalazar had no difference in days to start flowering under normal irrigation and drought conditions and it started flowering after 47 days. The cotton cv. FH-142 started flowering earlier (after 46 days) under drought conditions than that under normal irrigation condition which occurred after 50 days of sowing. Means of cultivars showed that cv. Lalazar started flowering 1 day earlier than cv. FH-142, whereas drought conditions caused 2 days earlier flowering than that by normal irrigation conditions (Table 2). Foliar application of ascorbic acid at 100 mg L^{-1} and a mixture of glycine betaine, salicylic acid and ascorbic acid each at 33 mg L^{-1} recorded minimum number of days to split boll in cv. Lalazar under normal irrigation (107 days) and under drought (109 days) in comparison with all other treatments. The cotton cultivar FH-142 under irrigation conditions recorded minimum number of days (107 days) to split boll where the mixture of three selected PGRs was sprayed (Table 2). Mean values showed that there was a difference of 1-2 days in boll splitting between irrigation conditions and cotton cultivars (Table 2).

It is evident from the data in Table 3 that cv. Lalazar recorded maximum plant height (106.75 cm) under normal irrigation conditions with the combination of glycine betaine, salicylic acid and ascorbic acid at 33 mg L^{-1} each. The height of Lalazar plants under drought conditions was improved with the application of PGRs compared with control. Maximum plant height was recorded where the mixture of salicylic acid and ascorbic acid each at 50 mg L^{-1} was sprayed (Table 3). The performance of FH-142 varied under normal irrigation and drought conditions with the application of PGRs. The mixture of glycine betaine, salicylic acid and ascorbic acid each at 33 mg L^{-1} recorded maximum plant height (103 cm) under normal irrigation condition, whereas the mixture of glycine betaine and ascorbic acid each at 50 mg L^{-1} gave maximum plant height (94 cm) of cv. FH-142 under drought conditions followed by the combination of glycine betaine, salicylic acid and ascorbic acid each at 33 mg L^{-1} (93.25 cm plant height). Mean values showed that both cultivars had lower height under drought conditions when compared with normal irrigation conditions. However, mean of cultivars showed that FH-142 got more height than that of Lalazar. The PGRs application did not increase the monopodial branches in both cultivars. In cv. FH-142, application of glycine betaine at 100 mg L^{-1} recorded a maximum number of monopodial branches under normal irrigation, and the monopodial branches were more under normal irrigation conditions than those under drought (Table 3). Under normal irrigation conditions, foliar-applied mixture of glycine betaine, salicylic acid and ascorbic acid each at 33 mg L^{-1} counted 18 sympodial branches in cv. Lalazar followed by the combined application of salicylic acid and ascorbic acid

each at 50 mg L^{-1} which recorded 17 sympodial branches and 22 in cv. FH-142 with the mixture of three selected PGRs at 33 mg L^{-1} each. Under drought conditions, the combination of salicylic acid and ascorbic acid each at 50 mg L^{-1} recorded a maximum number of sympodial branches (16) in cv. Lalazar. The mixture of glycine betaine and salicylic acid at 50 mg L^{-1} recorded maximum 17 sympodial branches under drought conditions (Table 4). Lalazar and FH-142 produced more sympodial branches under normal irrigation conditions than those under drought stress.

Cultivar Lalazar has maximum relative water contents (75%) under normal irrigation conditions with foliar-applied mixture of three selected PGRs each at 33 mg L^{-1} followed by glycine betaine at 100 mg L^{-1} that resulted in 74% relative water contents (Table 4). Under drought conditions, PGRs slightly increased the relative water contents of cv. Lalazar compared with the control. In cotton cultivar FH-142, the mixture of three selected PGRs each at 33 mg L^{-1} and glycine betaine at 100 mg L^{-1} recorded maximum relative water content (74%) under normal irrigation conditions. Under drought conditions, PGRs had no effect on relative contents of cv. FH-142. Mean values in Table 5 show that normal irrigation caused more relative contents than that of drought, and there was a difference of 1.8% in relative water contents between the two cultivars (Table 4). Application of the mixture of salicylic acid and ascorbic acid each at 50 mg L^{-1} improved membrane stability index (68.2) markedly in cv. Lalazar under normal irrigation condition, whereas, the mixture of glycine betaine, salicylic acid and ascorbic acid each at 33 mg L^{-1} resulted in a maximum stability index of cell membrane (61.7) in cv. Lalazar under drought conditions (Table 5). In cv. FH-142, application of the mixture of glycine betaine, salicylic acid and ascorbic acid each at 33 mg L^{-1} resulted in stability index of 62.2 under normal irrigation condition, whereas the mixture of salicylic acid and ascorbic acid each at 50 mg L^{-1} showed the membrane stability index (64) under drought conditions. Mean values showed that cv. Lalazar was more tolerant to stress than cv. FH-142 in terms of membrane stability (Table 5).

The highest number of main stem nodes was 29 and 26 under normal irrigation and drought conditions, respectively in cv. Lalazar with the combined application of salicylic acid and ascorbic acid each at 50 mg L^{-1} . In contrast, in case of cv. FH-142, maximum number of main stem nodes (32) was recorded where mixture of three selected PGRs was applied at 33 mg L^{-1} each under normal irrigation conditions. Under drought conditions maximum nodes were (28) obtained with the combined applications of glycine betaine and ascorbic acid each at 50 mg L^{-1} . Mean comparison shows that both cultivars had less number of main stem nodes under drought conditions as compared to that under normal irrigation conditions, and cv. FH-142 produced more nodes than did Lalazar. Foliar-applied glycine betaine and salicylic acid each at 50 mg L^{-1} has a minimum value of first fruiting branch node number in cv. FH-142 under drought conditions when compared to rest of the treatments (Table 6). However, foliar-applied PGRs had no significant effect on first fruiting branch node height and height to node ratio under normal and drought conditions in the tested cultivars (Table 6 and 7).

Table 1. Effect of PGRs on days to emergence and days to squaring in two cotton cultivars under normal irrigation and drought conditions.

Treatment	Days to emergence						Days to squaring					
	Lalazar			FH-142			Lalazar			FH-142		
	Normal irrigation	Drought	Mean	Normal irrigation	Drought	Mean	Normal irrigation	Drought	Mean	Normal irrigation	Drought	Mean
T ₁	5.8 a	5.3 a	5.5 a	5.5 a	6.0 a	5.7 a	43.75 a	39.0 c-h	43.0 ab	38.75 c-i	41.1 a	
T ₂	5.3 a	5.3 a	5.5 a	5.5 a	5.3 a	5.4 a	41.0 a-e	35.2 i-n	36.7 g-n	34.7 klmn	36.9 de	
T ₃	5.0 a	6.3 a	5.5 a	5.5 a	5.5 a	5.6 a	38.5 c-j	36.5 g-n	38.0 d-l	36.25 h-n	37.3 de	
T ₄	5.0 a	5.0 a	5.3 a	5.3 a	6.0 a	5.3 a	37.75 d-m	34.7 klmn	36.7 g-n	34.25 mn	35.9 ef	
T ₅	6.3 a	5.8 a	5.3 a	5.3 a	6.3 a	5.9 a	37.0 f-n	37.0 j-n	41.7 abc	38.0 d-l	37.9 cd	
T ₆	6.5 a	5.5 a	5.0 a	5.0 a	5.8 a	5.7 a	37.25 f-m	34.5 lmn	36.0 h-n	33.5 n	35.3 f	
T ₇	6.0 a	5.3 a	5.3 a	5.3 a	4.5 a	5.3 a	38.0 d-l	36.0 h-n	37.7 d-m	35.25 i-n	36.8 def	
T ₈	6.0 a	5.8 a	6.0 a	6.0 a	5.5 a	5.8 a	37.5 e-m	35.7 h-n	37.0 f-n	34.7 klmn	36.3 ef	
T ₉	5.0 a	5.0 a	5.5 a	5.5 a	5.0 a	5.1 a	41.2 abcd	38.2 c-k	40.5 a-f	38.5 c-j	39.6 b	
T ₁₀	5.8 a	5.5 a	6.3 a	6.3 a	5.3 a	5.7 a	40.0 b-g	37.5 e-m	41.0 a-e	39.0 c-h	39.4 bc	
Mean of cultivars	5.55 a	5.50 a	5.50 a	5.50 a	5.48 a	5.50 a	37.5 a	37.5 a	37.5 a	37.5 a	37.5 a	
Means of irrigation conditions	Normal irrigation = 5.58 a	Drought = 5.48 a	Normal irrigation = 5.58 a	Drought = 5.48 a	Normal irrigation = 5.58 a	Drought = 5.48 a	Normal irrigation = 39 a	Drought = 36.2 b	Normal irrigation = 39 a	Drought = 36.2 b	Normal irrigation = 39 a	

T₁ = Control (Untreated), T₂ = Distilled water spray, T₃ = Distilled Water spray + Tween-20 (0.1 %), T₄ = Glycine betaine @ 100 mg L⁻¹ + Tween-20 (0.1 %), T₅ = Salicylic acid @ 100 mg L⁻¹ + Tween-20 (0.1 %), T₆ = Ascorbic acid @ 100 mg L⁻¹ + Tween-20 (0.1 %), T₇ = Glycine betaine and Salicylic acid @ 50 mg L⁻¹ each + Tween-20 (0.1 %), T₈ = Glycine betaine and Ascorbic acid @ 50 mg L⁻¹ each + Tween-20 (0.1 %), T₉ = Salicylic acid and Ascorbic acid @ 50 mg L⁻¹ each + Tween-20 (0.1 %), T₁₀ = Glycine betaine + Salicylic acid + Ascorbic acid @ 33 mg L⁻¹ each + Tween-20 (0.1 %)

Table 2. Effect of PGRs on days to flowering and days to first boll splitting in two cotton cultivars under normal irrigation and drought conditions.

Treatment	Days to flowering						Days to first boll splitting					
	Lalazar			FH-142			Lalazar			FH-142		
	Normal irrigation	Drought	Mean	Normal irrigation	Drought	Mean	Normal irrigation	Drought	Mean	Normal irrigation	Drought	Mean
T ₁	50.5 abc	42.75 c	52.5 a	52.5 a	46.25 abc	48.0 a	118.8 a	119.5 a	117.3 a	118.5 a	118.5 a	
T ₂	49.5 abc	49.25 abc	51.25 ab	51.25 ab	49 abc	49.8 a	111.5 a	113.3 a	118.5 a	119.5 a	115.1 ab	
T ₃	45.25 abc	47.0 abc	51.75 a	51.75 a	46.75 abc	47.7 a	116 a	116.3 a	111.5 a	113.0 a	114.2 abc	
T ₄	49.25 abc	45.5 abc	49.5 abc	49.5 abc	46.25 abc	47.6 a	109.0 a	110.3 a	112.3 a	113.3 a	111.2 bcd	
T ₅	52.25 a	43.0 bc	49.25 abc	49.25 abc	46.0 abc	47.6 a	109.0 a	112.8 a	115.3 a	115.8 a	113.2 abcd	
T ₆	49.5 abc	45.0 abc	45.52 abc	45.52 abc	47.25 abc	46.8 a	107.5 a	108.8 a	111.3 a	113.3 a	110.2 bcd	
T ₇	44.25 abc	47.5 abc	50.0 abc	50.0 abc	46.5 abc	47.1 a	111.75 a	112.5 a	113.0 a	114.5 a	112.9 abcd	
T ₈	43.25 bc	45.0 abc	50.75 abc	50.75 abc	46.5 abc	46.4 a	108.5 a	108.5 a	108.8 a	110.5 a	109.1 cd	
T ₉	47.5 abc	48.0 abc	47.0 abc	47.0 abc	48.5 abc	47.8 a	110.2 a	111.5 a	111.3 a	112.3 a	111.3 bcd	
T ₁₀	46.0 abc	50.0 abc	51.0 abc	51.0 abc	45.25 abc	48.1 a	107.0 a	108.5 a	107.3 a	107.5 a	107.8 d	
Mean of cultivars	47.01 b	48.33 a	48.33 a	48.33 a	46.45 b	48.1 a	111.6 a	111.6 a	111.6 a	111.6 a	111.6 a	
Means of irrigation conditions	Normal irrigation = 48.78 a	Drought = 46.45 b	Normal irrigation = 48.78 a	Drought = 46.45 b	Normal irrigation = 48.78 a	Drought = 46.45 b	Normal irrigation = 111.7 a	Drought = 112.9 a	Normal irrigation = 111.7 a	Drought = 112.9 a	Normal irrigation = 111.7 a	

T₁ = Control (Untreated), T₂ = Distilled water spray, T₃ = Distilled Water spray + Tween-20 (0.1 %), T₄ = Glycine betaine @ 100 mg L⁻¹ + Tween-20 (0.1 %), T₅ = Salicylic acid @ 100 mg L⁻¹ + Tween-20 (0.1 %), T₆ = Ascorbic acid @ 100 mg L⁻¹ + Tween-20 (0.1 %), T₇ = Glycine betaine and Salicylic acid @ 50 mg L⁻¹ each + Tween-20 (0.1 %), T₈ = Glycine betaine and Ascorbic acid @ 50 mg L⁻¹ each + Tween-20 (0.1 %), T₉ = Salicylic acid and Ascorbic acid @ 50 mg L⁻¹ each + Tween-20 (0.1 %), T₁₀ = Glycine betaine + Salicylic acid + Ascorbic acid @ 33 mg L⁻¹ each + Tween-20 (0.1 %)

Table 3. Effect of PGRs on plant height (cm) and number of monopodial branches in two cotton cultivars under normal irrigation and drought conditions.

Treatment	Plant height (cm)						Number of monopodial branches			
	Lalazar		FH-142		Mean	Lalazar		FH-142		Mean
	Normal irrigation	Drought	Normal irrigation	Drought		Normal irrigation	Drought	Normal irrigation	Drought	
T ₁	55.75 h-i	53 jkl	74.75 c-l	54.5 ijkl	59.5 f	3.5 ab	2.75 ab	3 ab	2.75 ab	3 b
T ₂	62 f-l	47.75 kl	72.75	52.37jkl	58.71 f	3.75 ab	3 ab	4 ab	4.25 ab	3.75 ab
T ₃	78.5 b-j	47.25 l	65.75 d-l	60.65 g-l	63.03 ef	4.75 ab	3.75 ab	4 ab	3 ab	3.87 ab
T ₄	75.5 b-k	67.25 c-l	76.75 b-j	64.55 e-l	71.01 cde	3 ab	2.5 ab	5.75 a	4 ab	3.81 ab
T ₅	77.5 b-j	67.87 c-l	62.75 f-l	55.5 h-l	65.90 def	4.75 ab	3 ab	4.75 ab	3 ab	3.87 ab
T ₆	81.25 a-i	55 h-l	76 b-j	63.62 f-l	68.96 cdef	4.5 ab	4.5 ab	5 ab	3.75 ab	4.43 a
T ₇	72.75 c-l	63.5 f-l	91.75 a-e	71.32 c-l	74.83 cd	5 ab	3.5 ab	3.5 ab	3.75 ab	3.93 ab
T ₈	67.25 c-l	65.25 e-l	94.25 abc	94 abc	80.18 bc	4.75 ab	4.25 ab	4 ab	2.75 ab	3.93 ab
T ₉	87.75 a-g	86.3 a-g	88.75 a-f	82.5 a-h	86.32 ab	5 ab	4 ab	4.25 ab	2.75 ab	4 ab
T ₁₀	106.75 a	72 c-l	103 ab	93.25 abcd	93.75 a	2.25 b	4.25 ab	4.75 ab	3.75 ab	3.75 ab
Mean of cultivars	59.50 b	59.50 b	74.93 a	74.93 a	74.93 a	3.83 a	3.83 a	3.83 a	3.83 a	3.83 a
Means of irrigation conditions	Normal irrigation = 78.57 a	Normal irrigation = 78.57 a	Drought = 65.87 b	Drought = 65.87 b	Normal irrigation = 4.21 a	Normal irrigation = 4.21 a	Drought = 3.46 b	Drought = 3.46 b	Normal irrigation = 4.21 a	Drought = 3.46 b

T₁ = Control (Untreated), T₂ = Distilled water spray + Tween-20 (0.1 %), T₃ = Glycine betaine @ 100 mg L⁻¹ + Tween-20 (0.1 %), T₄ = Salicylic acid @ 100 mg L⁻¹ + Tween-20 (0.1 %), T₅ = Ascorbic acid @ 100 mg L⁻¹ + Tween-20 (0.1 %), T₆ = Glycine betaine and Salicylic acid @ 50 mg L⁻¹ each + Tween-20 (0.1 %), T₇ = Glycine betaine and Ascorbic acid @ 50 mg L⁻¹ each + Tween-20 (0.1 %), T₈ = Glycine betaine and Salicylic acid @ 50 mg L⁻¹ each + Tween-20 (0.1 %), T₉ = Salicylic acid and Ascorbic acid @ 50 mg L⁻¹ each + Tween-20 (0.1 %), T₁₀ = Glycine betaine + Salicylic acid + Ascorbic acid @ 33 mg L⁻¹ each + Tween-20 (0.1 %)

Table 4. Effect of PGRs on number of sympodial branches and relative water contents (%) in two cotton cultivars under normal irrigation and drought conditions.

Treatment	Number of sympodial branches						Relative water contents (%)			
	Lalazar		FH-142		Mean	Lalazar		FH-142		Mean
	Normal irrigation	Drought	Normal irrigation	Drought		Normal irrigation	Drought	Normal irrigation	Drought	
T ₁	15.75 b-i	12.5 fghi	17 b-f	13.25 e-i	14.62 bcd	69.1 d-h	61.0 lmn	69.2 c-g	60.1 mn	64.9 d
T ₂	14 d-i	11.5 ghi	16.5 b-g	11 i	13.25 d	71.8 abcd	67.5 f-j	71.5 abcd	66.7 e-i	69.4 ab
T ₃	16 b-i	11.25 hi	14.75 b-i	13.5 e-i	13.87 cd	70.5 b-f	59.7 n	70.8 a-f	59.7 n	65.2 d
T ₄	15.75 b-i	14.25 c-i	18 a-e	13.75 d-i	15.43 bc	74.1 ab	65.7 g-k	74.1 ab	66.0 g-k	70.0 a
T ₅	15.25 b-i	16 b-i	14 d-i	14.25 c-i	14.87 bcd	71.0 a-f	65.4 ijkl	71.1 a-f	64.5 ijkl	68.0 bc
T ₆	16.5 b-g	12.25 fghi	18.75 abcd	13.75 d-i	15.31 bc	73.4 abc	64.3 i-n	73.3 abc	64.5 h-l	68.9 abc
T ₇	16.25 b-h	14.25 c-i	19.25 abc	14.25 c-i	16 ab	72.4 abcd	64.5 i-m	72.4 abcd	64.1 i-m	68.4 bc
T ₈	15 b-i	14 d-i	19.5 ab	17.25 a-f	16.43 ab	71.4 a-e	65.7 g-k	71.5 a-e	65.5 g-k	68.5 abc
T ₉	17.75 a-e	16 b-i	16.75 b-f	14.25 c-i	16.18 ab	72.8 abcd	63.0 klmn	73.3 abc	63.1 i-n	68.1 c
T ₁₀	18.75 abcd	14.75 b-i	22.25 a	14.5 b-i	17.56 a	75.0 a	63.3 j-n	74.6 a	62.7 klmn	68.9 abc
Mean of cultivars	14.88 b	14.88 b	15.82 a	15.82 a	15.82 a	68.1 a	68.1 a	68.1 a	68.1 a	68.1 a
Means of irrigation conditions	Normal irrigation = 16.88 a	Normal irrigation = 16.88 a	Drought = 13.82 b	Drought = 13.82 b	Normal irrigation = 71.8 a	Normal irrigation = 71.8 a	Drought = 63.4 b	Drought = 63.4 b	Normal irrigation = 71.8 a	Drought = 63.4 b

T₁ = Control (Untreated), T₂ = Distilled water spray + Tween-20 (0.1 %), T₃ = Glycine betaine @ 100 mg L⁻¹ + Tween-20 (0.1 %), T₄ = Salicylic acid @ 100 mg L⁻¹ + Tween-20 (0.1 %), T₅ = Ascorbic acid @ 100 mg L⁻¹ + Tween-20 (0.1 %), T₆ = Glycine betaine and Salicylic acid @ 50 mg L⁻¹ each + Tween-20 (0.1 %), T₇ = Glycine betaine and Ascorbic acid @ 50 mg L⁻¹ each + Tween-20 (0.1 %), T₈ = Glycine betaine and Salicylic acid @ 50 mg L⁻¹ each + Tween-20 (0.1 %), T₉ = Salicylic acid and Ascorbic acid @ 50 mg L⁻¹ each + Tween-20 (0.1 %), T₁₀ = Glycine betaine + Salicylic acid + Ascorbic acid @ 33 mg L⁻¹ each + Tween-20 (0.1 %)

Table 5. Effect of PGRs on and membrane stability index and number of main stem nodes in two cotton cultivars under normal irrigation and drought conditions.

Treatment	Membrane stability index						Number of main stem nodes					
	Lalazar		FH-142		Mean	Drought	Lalazar		FH-142		Mean	Drought
	Normal irrigation	Drought	Normal irrigation	Drought			Normal irrigation	Drought	Normal irrigation	Drought		
T ₁	61.30 a-i	48.21 ijkl	63.09 a-h	39.22 l	52.95 de	25 b-i	21.75 ghi	26.5 b-h	23.75 e-i	24.25 cd	23.81 d	24.31 cd
T ₂	60.85 a-i	50.32 g-l	60.56 a-j	46.06 jkl	54.45 cde	25.5 b-i	20 i	28.25 a-f	21.5 ghi	23.81 d	24.31 cd	24.31 cd
T ₃	62.99 a-h	38.70 l	59.61 a-j	42.25 kl	50.77 e	26.25 b-h	21.25 hi	25.75	24 d-i	26.25 abc	24.87 bcd	26.56 abc
T ₄	68.82 a-e	60.73 a-j	68.10 a-e	52.55 f-l	62.5 ab	25.75 b-i	24 d-i	30.25 ab	23.75 e-i	24.87 bcd	26.56 abc	26.56 abc
T ₅	66.41 a-f	48.24 ijkl	61.46 a-i	54.60 e-k	57.68 bcd	27.25 a-g	24.25 c-i	29.75 abcd	24.25 c-i	26.56 abc	24.87 bcd	26.56 abc
T ₆	72.034 ab	54.3 b-j	71.2 abc	38.82 l	60.02 abc	29 a-f	23.25 fghi	29.75 abcd	24.25 c-i	26.56 abc	24.87 bcd	26.56 abc
T ₇	71.85 ab	54.24 e-k	63.95 a-g	50.28 g-l	60.08 abc	28 a-f	24.5 b-i	29.25 a-e	25.75 b-i	26.87 ab	27.06 ab	27.06 ab
T ₈	67.57 a-e	48.80 h-l	66.38 a-f	51.03 g-l	58.44 bcd	25 b-i	24.5 b-i	30 abc	28.75 a-f	27.06 ab	27.06 ab	27.06 ab
T ₉	72.71 a	55.82 d-k	66.36 a-f	55.85 d-k	62.6 ab	29.75 b-i	26.5 b-h	25.25 b-i	24 d-i	26.37 abc	26.37 abc	26.37 abc
T ₁₀	70.07 abcd	60.79 a-j	72.64 ab	56.94 c-k	65.1 a	27.75 a-f	25.5 b-i	32.5 a	26 b-h	27.93 a	27.93 a	27.93 a
Mean of cultivars	59.9 a	57.03 b	57.03 b	57.03 b	57.03 b	25.23 b	26.42 a	25.23 b	26.42 a	25.23 b	26.42 a	26.42 a
Means of irrigation conditions	Normal irrigation = 66.3 a	Drought = 50.5 b	Normal irrigation = 66.3 a	Drought = 50.5 b	Normal irrigation = 66.3 a	Normal irrigation = 27.52 a	Drought = 24.13 b	Normal irrigation = 27.52 a	Drought = 24.13 b	Normal irrigation = 27.52 a	Drought = 24.13 b	Normal irrigation = 27.52 a

T₁ = Control (Untreated), T₂ = Distilled water spray + Tween-20 (0.1 %), T₃ = Distilled water spray + Tween-20 (0.1 %) + Salicylic acid @ 100 mg L⁻¹ + Tween-20 (0.1 %), T₄ = Glycine betaine @ 100 mg L⁻¹ + Tween-20 (0.1 %), T₅ = Salicylic acid @ 100 mg L⁻¹ + Tween-20 (0.1 %), T₆ = Ascorbic acid @ 100 mg L⁻¹ + Tween-20 (0.1 %), T₇ = Glycine betaine and Salicylic acid @ 50 mg L⁻¹ each + Tween-20 (0.1 %), T₈ = Glycine betaine and Ascorbic acid @ 50 mg L⁻¹ each + Tween-20 (0.1 %), T₉ = Salicylic acid and Ascorbic acid @ 50 mg L⁻¹ each + Tween-20 (0.1 %), T₁₀ = Glycine betaine + Salicylic acid + Ascorbic acid @ 33 mg L⁻¹ each + Tween-20 (0.1 %)

Table 6. Effect of PGRs on height to node ratio and first fruiting branch node number in two cotton cultivars under normal irrigation and drought conditions.

Treatment	Height to node ratio						First fruiting branch node number					
	Lalazar		FH-142		Mean	Drought	Lalazar		FH-142		Mean	Drought
	Normal irrigation	Drought	Normal irrigation	Drought			Normal irrigation	Drought	Normal irrigation	Drought		
T ₁	2.23 c	2.48 bc	2.84 abc	2.30 bc	2.46 c	6.75 ab	7 ab	7.25 ab	6 ab	6.75 ab	6.75 ab	7.31 a
T ₂	2.45 bc	2.40 bc	2.60 abc	2.52 bc	2.49 c	6.5 ab	7.5 ab	8 a	7.25 ab	7.31 a	7.31 a	7 ab
T ₃	3.02 abc	2.23 c	2.57 abc	2.54 abc	2.59 c	6.75 ab	7.5 ab	6.5 ab	7.25 ab	7 ab	7 ab	7 ab
T ₄	2.98 abc	2.82 abc	2.56 abc	2.58 abc	2.73 c	7.5 ab	6 ab	7.5 ab	6.25 ab	6.81 ab	6.81 ab	6.81 ab
T ₅	2.84 abc	2.82 abc	2.66 abc	2.30 bc	2.65 c	5.5 ab	6.75 ab	7 ab	5.75 ab	6.25 ab	6.25 ab	6.25 ab
T ₆	2.80 abc	2.36 bc	2.56 abc	2.61 abc	2.58 c	7.75 a	6 ab	7.25 ab	6.75 ab	6.93 ab	6.93 ab	6.93 ab
T ₇	2.60 abc	2.61 abc	3.19 abc	2.76 abc	2.79 bc	6 ab	6.25 ab	4.25 b	6.5 ab	5.75 b	5.75 b	5.75 b
T ₈	2.71 abc	2.66 abc	3.17 abc	3.28 abc	2.95 abc	6.5 ab	5.75 ab	6 ab	6.5 ab	6.18 ab	6.18 ab	6.18 ab
T ₉	2.95 abc	3.27 abc	3.54 abc	3.45 abc	3.30 ab	5.5 ab	6.5 ab	7 ab	6.75 ab	6.43 ab	6.43 ab	6.43 ab
T ₁₀	3.85 a	2.83 abc	3.18 abc	3.59 ab	3.36 a	6 ab	6.25 ab	4.75 ab	6 ab	5.75 b	5.75 b	5.75 b
Mean of cultivars	2.74 a	2.84 a	2.84 a	2.84 a	2.84 a	6.51 a	6.51 a	6.51 a	6.51 a	6.51 a	6.51 a	6.52 a
Means of irrigation conditions	Normal irrigation = 2.86 a	Drought = 2.72 a	Normal irrigation = 2.86 a	Drought = 2.72 a	Normal irrigation = 2.86 a	Normal irrigation = 6.51 a	Drought = 6.52 a	Normal irrigation = 6.51 a	Drought = 6.52 a	Normal irrigation = 6.51 a	Drought = 6.52 a	Normal irrigation = 6.51 a

T₁ = Control (Untreated), T₂ = Distilled water spray + Tween-20 (0.1 %), T₃ = Distilled water spray + Tween-20 (0.1 %) + Salicylic acid @ 100 mg L⁻¹ + Tween-20 (0.1 %), T₄ = Glycine betaine @ 100 mg L⁻¹ + Tween-20 (0.1 %), T₅ = Salicylic acid @ 100 mg L⁻¹ + Tween-20 (0.1 %), T₆ = Ascorbic acid @ 100 mg L⁻¹ + Tween-20 (0.1 %), T₇ = Glycine betaine and Salicylic acid @ 50 mg L⁻¹ each + Tween-20 (0.1 %), T₈ = Glycine betaine and Ascorbic acid @ 50 mg L⁻¹ each + Tween-20 (0.1 %), T₉ = Salicylic acid and Ascorbic acid @ 50 mg L⁻¹ each + Tween-20 (0.1 %), T₁₀ = Glycine betaine + Salicylic acid + Ascorbic acid @ 33 mg L⁻¹ each + Tween-20 (0.1 %)

Under normal irrigation conditions, maximum number of opened bolls per plant was 31 and 21 in cv. FH-142 and cv. Lalazar, respectively, where the mixture of three selected PGRs at 33 mg L⁻¹ each was applied. In contrast, under drought conditions maximum number of opened bolls per plant (17) was obtained with the foliar-applied salicylic acid and ascorbic acid each at 50 mg L⁻¹ in cv. Lalazar (Table 7). Cotton cv. FH-142 produced more number of opened bolls per plant as compared to Lalazar, and drought conditions significantly reduced the number of opened boll compared to that by normal irrigation conditions (Table 7). Under normal irrigation and drought conditions, the number of unopened bolls per plant was almost similar, and 1 or 2 bolls remained unopened in each treatment (Table 8).

Foliar-applied salicylic acid and ascorbic acid each at 50 mg L⁻¹ weighed highest average boll weight in cv. Lalazar under normal irrigation (3.67 g) and under drought (3.43 g) conditions (Table 8). In cv. FH-142, the mixture of three PGRs at 33 mg L⁻¹ each gave highest average boll weight under both irrigation regimes. Data given in Table 9 showed that lint yield per plant was significantly affected by exogenous application of different PGRs. Maximum lint yield was 29.36 and 47.52 g per plant in Lalazar and FH-142, respectively under normal irrigation conditions where the mixture of three PGRs at 33 mg L⁻¹ each was applied. In cotton cultivar FH-142, the combined application of glycine betaine and ascorbic acid each at 50 mg L⁻¹ produced maximum lint yield per plant (28.55 g) under drought conditions (Table 9). Performance of FH-142 was better than that of Lalazar in terms of lint yield per plant. Foliar application of PGRs produced more cotton seed yield compared to the control treatments. The mixture of glycine betaine, salicylic acid and ascorbic acid at 33 mg L⁻¹ each recorded maximum cotton seed yield in both cultivars under normal irrigation conditions (Table 10). Under drought conditions, maximum cotton seed yield per plant (34.53 g) was recorded in cv. Lalazar with the combined application of salicylic acid and ascorbic acid each at 50 mg L⁻¹, and in cv. FH-142 maximum cotton seed yield (44.45 g per plant) was obtained with the foliar-applied glycine betaine and salicylic acid each at 50 mg L⁻¹. Mean values showed that cotton seed yield per plant was higher under normal irrigation conditions than that of drought conditions in both cultivars. However, the performance of FH-142 was better in terms of cotton seed yield and ginning out turn (GOT) percentage. The exogenous application of PGRs did not enhance the GOT (Table 10).

Discussion

The present investigation regarding the use of novel growth promoting substances for enhancing growth and yield of cotton grown under water scarcity suggests that exogenously applied growth promoting substances such as glycine betaine, salicylic acid and ascorbic acid may counter the harmful effects of water scarcity. In view of our results, days to plant emergence are similar for both cultivars due to the presence of appropriate moisture contents at the time of germination and seedling emergence till first true leaves or occurrence of drought. Our findings agree with the findings reported earlier by Hubbard *et al.*, (2012) that emergence is delayed or

inhibited under deficit water. As far as the reproductive stages of cotton are concerned, it is very sensitive to water scarcity as reported by Snowden *et al.*, (2014). These findings are in agreement with our findings regarding number of days to squaring and initiation of flowering. Early squaring in cotton may be due to early diversion of plants towards reproductive growth due to less availability of water (Rao *et al.*, 2016). In our study, early flowering was achieved where salicylic acid was exogenously applied at 100 mg L⁻¹. It shows linkage of early flowering of cotton with the possible role of applied PGRs. In earlier findings, it is evident that salicylic acid regulates time of flowering in Arabidopsis. It was attributed to involvement of photoperiod and autonomous pathway without involvement of genes for flowering time (Martínez *et al.*, 2004). Under normal irrigation and water limited conditions, days to first boll split was similar in both cultivars. It can be attributed to less sensitivity of cotton fruit to water stress as reported by Van Iersel & Oosterhuis (1996). However, it is obvious from the data that PGRs alone and in combinations resulted into different days to first boll split. Maximum number of days to first boll split was found in case of distilled water spray. Early maturity of cotton bolls can be due to use of different combination of PGRs. Combination of salicylic acid, ascorbic acid and glycine betaine yielded 10.7 days earlier than did by control. Our results are in conformity with those reported by Noreen *et al.*, (2013) who found significant effect of exogenous application of PGRs on days to first cotton boll bursting under drought.

Both cultivars differed in plant height. Decreased plant height under water deficit as compared to that under normal irrigation can be attributed to undesirable effects of water stress on cellular and metabolic structure. Similar findings were reported by Pettigrew (2004). Minimum plant height was achieved in the treatment where distilled water was sprayed, while maximum plant height where combined glycine betaine, salicylic acid and ascorbic was exogenously applied. This indicates that there is a role of PGRs to maintain plant height under water deficit conditions. Similar findings were reported by Noreen *et al.*, (2015) who concluded that foliar-applied PGRs mitigate the adverse effects of drought and enhance the growth parameters in cotton. Numbers of monopodial and sympodial branches are very important as they closely relate to yield characteristics. In our study, there were no differences in the cultivars in monopodial branches, while the opposite was true for sympodial branches. However, in normal and drought conditions, monopodial and sympodial branches are different from each other. It might be linked to less height of plants under drought with less vegetative growth as compared to that under normal irrigation. However, PGRs improved number of sympodial branches and monopodial branches as compared to the control. Exogenous application of glycine betaine, salicylic acid and ascorbic acid in combination produced more total number of branches including monopodial and sympodial branches. Thus, the results suggest the role of PGRs in improving cotton growth under water deficit conditions. Our findings are in accordance with those reported by Noreen *et al.*, (2015) who sprayed salicylic acid and glycine betaine separately on cotton crop at 45 days after sowing and recorded significant cotton growth under drought stress when compared with untreated plants under water stress.

Table 7. Effect of PGRs on first fruiting branch node height (cm) and number of open bolls per plant in two cotton cultivars under normal irrigation and drought conditions.

Treatment	First fruiting branch node height (cm)						Number of open bolls per plant					
	Lalazar			FH-142			Lalazar			FH-142		
	Normal irrigation	Drought	Mean	Normal irrigation	Drought	Mean	Normal irrigation	Drought	Mean	Normal irrigation	Drought	Mean
T ₁	11.8 ab	13.2 ab	12.5 a	12.5 ab	12.5 ab	12.5 a	13 g-n	9.25 l-p	16.75 c-h	7 op	11.5 f	
T ₂	11.5 ab	11.2 ab	11.6 a	13.0 ab	10.6 ab	11.6 a	11.75 h-o	9.5 l-p	15.25 e-k	6.25 p	10.68 f	
T ₃	12.0 ab	14.2 a	13.3 a	13.3 ab	13.8 a	13.3 a	18.5 b-f	7 op	13.75 f-l	10 l-p	12.31 ef	
T ₄	11.5 ab	11.7 ab	12.0 a	13.0 ab	11.8 ab	12.0 a	16 e-i	10.75 j-p	21.75 bc	12.25 h-n	15.18 cd	
T ₅	10.5 ab	11.2 ab	11.6 a	11.8 ab	13.0 ab	11.6 a	16 e-i	10 l-p	9.5 l-p	9.25 l-p	11.18 f	
T ₆	13.0 ab	9.7 ab	11.3 a	12.8 ab	9.5 ab	11.3 a	19.5 bcde	8 nop	17.75 c-g	10.5 j-p	13.93 de	
T ₇	12.5 ab	13.7 a	11.7 a	8.5 b	11.8 ab	11.7 a	16 e-i	11.25 i-p	18.75 b-f	11.75 h-o	14.43 d	
T ₈	14.2 a	11.7 ab	12.3 a	11.8 ab	11.3 ab	12.3 a	13.25 g-m	8.25 mnop	29.75 a	23.5 b	18.68 ab	
T ₉	12.0 ab	10.7 ab	12.1 a	12.0 ab	13.8 a	12.1 a	19.5 bcde	17.5 o-g	16.5 d-h	14.25 f-l	16.93 bc	
T ₁₀	11.7 ab	12.5 ab	12.1 a	12.5 ab	11.6 ab	12.1 a	21.5 bcd	10.25 k-p	31.75 a	15.5 e-j	19.75 a	
Mean of cultivars	12.06 a	12.06 a	12.04 a	12.04 a	12.04 a	12.04 a	13.33 b	13.33 b	15.58 a	15.58 a	15.58 a	
Means of irrigation conditions	Normal irrigation = 12.10 a	Drought = 12.0 a	Normal irrigation = 12.10 a	Drought = 12.0 a	Normal irrigation = 12.10 a	Drought = 12.0 a	Normal irrigation = 17.82 a	Drought = 11.10 b	Normal irrigation = 17.82 a	Drought = 11.10 b	Normal irrigation = 17.82 a	Drought = 11.10 b

T₁ = Control (Untreated), T₂ = Distilled water spray + Tween-20 (0.1%), T₃ = Distilled water spray + Tween-20 (0.1%), T₄ = Glycine betaine @ 100 mg L⁻¹ + Tween-20 (0.1%), T₅ = Salicylic acid @ 100 mg L⁻¹ + Tween-20 (0.1%), T₆ = Ascorbic acid @ 100 mg L⁻¹ + Tween-20 (0.1%), T₇ = Glycine betaine and Salicylic acid @ 50 mg L⁻¹ each + Tween-20 (0.1%), T₈ = Glycine betaine and Ascorbic acid @ 50 mg L⁻¹ each + Tween-20 (0.1%), T₉ = Salicylic acid and Ascorbic acid @ 50 mg L⁻¹ each + Tween-20 (0.1%), T₁₀ = Glycine betaine + Salicylic acid + Ascorbic acid @ 33 mg L⁻¹ each + Tween-20 (0.1%)

Table 8. Effect of PGRs on number of unopen bolls per plant and average boll weight (g) in two cotton cultivars under normal irrigation and drought conditions.

Treatment	Number of unopen bolls per plant						Average boll weight (g)					
	Lalazar			FH-142			Lalazar			FH-142		
	Normal irrigation	Drought	Mean	Normal irrigation	Drought	Mean	Normal irrigation	Drought	Mean	Normal irrigation	Drought	Mean
T ₁	2 a	2 a	2.06 a	2.25 a	2 a	2.06 a	3.28 abcd	2.39 cdef	3.06 a-f	2.89 a-f	2.90 bc	
T ₂	2 a	1.75 a	2 a	2.5 a	1.75 a	2 a	3.12 a-f	2.09 f	3.10 a-f	3.01 a-f	2.83 c	
T ₃	2 a	2 a	2 a	2.25 a	1.75 a	2 a	3.13 a-f	2.31 a-f	3.24 abcd	2.70 a-f	2.84 c	
T ₄	2 a	1.75 a	2.06 a	2.5 a	2 a	2.06 a	3.47 ab	2.44 b-f	3.06 a-f	2.80 a-f	2.94 bc	
T ₅	2.5 a	1.75 a	2.12 a	2.25 a	2 a	2.12 a	3.57 a	3.11 a-f	2.90 a-f	2.74 a-f	3.08 abc	
T ₆	2 a	1.5 a	1.81 a	2 a	1.75 a	1.81 a	3.33 abcd	2.76 a-f	2.87 a-f	2.76 a-f	2.93 bc	
T ₇	2.5 a	1.5 a	2.12 a	2.5 a	2 a	2.12 a	3.49 ab	2.14 ef	3.30 abcd	3.03 a-f	2.99 abc	
T ₈	1.5 a	2 a	1.93 a	2.5 a	1.75 a	1.93 a	3.53 a	3.10 a-f	3.24 abcd	3.12 a-f	3.24 abc	
T ₉	1.5 a	1.75 a	1.75 a	2 a	1.75 a	1.75 a	3.67 a	3.43 abc	3.17 a-e	3.09 a-f	3.34 ab	
T ₁₀	2.25 a	2.25 a	2.12 a	2 a	2 a	2.12 a	3.41 abc	3.27 abcd	3.42 abc	3.49 ab	3.39 a	
Mean of cultivars	1.92 a	1.92 a	2.07 b	2.07 b	2.07 b	2.07 b	3.05 a	3.05 a	3.26 a	3.26 a	3.26 a	
Means of irrigation conditions	Normal irrigation = 2.15 a	Drought = 1.85 b	Normal irrigation = 2.15 a	Drought = 1.85 b	Normal irrigation = 2.15 a	Drought = 1.85 b	Normal irrigation = 3.26 a	Drought = 2.83 b	Normal irrigation = 3.26 a	Drought = 2.83 b	Normal irrigation = 3.26 a	Drought = 2.83 b

T₁ = Control (Untreated), T₂ = Distilled water spray + Tween-20 (0.1%), T₃ = Distilled water spray + Tween-20 (0.1%), T₄ = Glycine betaine @ 100 mg L⁻¹ + Tween-20 (0.1%), T₅ = Salicylic acid @ 100 mg L⁻¹ + Tween-20 (0.1%), T₆ = Ascorbic acid @ 100 mg L⁻¹ + Tween-20 (0.1%), T₇ = Glycine betaine and Salicylic acid @ 50 mg L⁻¹ each + Tween-20 (0.1%), T₈ = Glycine betaine and Ascorbic acid @ 50 mg L⁻¹ each + Tween-20 (0.1%), T₉ = Salicylic acid and Ascorbic acid @ 50 mg L⁻¹ each + Tween-20 (0.1%), T₁₀ = Glycine betaine + Salicylic acid + Ascorbic acid @ 33 mg L⁻¹ each + Tween-20 (0.1%)

Table 9. Effect of PGRs on lint yield per plant (g) per pot in two cotton cultivars under normal irrigation and drought conditions.

Treatment	Lint yield per plant (g)				Mean
	L.alazar		FH-142		
	Normal irrigation	Drought	Normal irrigation	Drought	
T ₁	17.41 e-m	8.58 no	21.32 c-g	9.31 no	14.15 e
T ₂	14.75 f-n	8.34 no	19.49 e-j	7.60 no	12.54 e
T ₃	23.34 bcde	6.57 o	17.81 e-l	11.14 k-o	14.71 de
T ₄	22.68 b-f	10.48 lmno	27.62 bcd	13.14 h-o	18.48 c
T ₅	23.29 bcde	11.32 k-o	11.75 j-o	9.09 no	13.86 e
T ₆	10.86 k-o	8.12 no	20.39 d-i	11.36 k-o	12.68 e
T ₇	22.53 b-f	9.55 mno	24.38 bcde	14.12 g-o	17.64 cd
T ₈	18.14 e-l	9.24 no	40.25 a	28.55 bc	24.04 ab
T ₉	28.33 bcd	23.47 bcde	20.99 c-h	18.47 e-k	22.81 b
T ₁₀	29.36 b	12.71 i-o	47.52 a	17.74 e-l	26.83 a
Mean of cultivars	15.95 b	19.60 a			
Means of irrigation conditions	Normal irrigation = 23.11 a	Drought = 12.44 b			

T₁ = Control (Untreated), T₂ = Distilled water spray, T₃ = Distilled water spray + Tween-20 (0.1 %), T₄ = Glycine betaine @ 100 mg L⁻¹ + Tween-20 (0.1 %), T₅ = Salicylic acid @ 100 mg L⁻¹ + Tween-20 (0.1 %), T₆ = Ascorbic acid @ 100 mg L⁻¹ + Tween-20 (0.1 %), T₇ = Glycine betaine and Salicylic acid @ 50 mg L⁻¹ each + Tween-20 (0.1 %), T₈ = Glycine betaine and Ascorbic acid @ 50 mg L⁻¹ each + Tween-20 (0.1 %), T₉ = Salicylic acid and Ascorbic acid @ 50 mg L⁻¹ each + Tween-20 (0.1 %), T₁₀ = Glycine betaine + Salicylic acid + Ascorbic acid @ 33 mg L⁻¹ each + Tween-20 (0.1 %)

Table 10. Effect of PGRs on cotton seed yield per plant (g) and GOT (%) in two cotton cultivars under normal irrigation and drought conditions.

Treatment	Cotton seed yield per plant (g)				GOT (%)				
	L.alazar		FH-142		L.alazar		FH-142		
	Normal irrigation	Drought	Normal irrigation	Drought	Normal irrigation	Drought	Normal Irrigation	Drought	
T ₁	24.60 j-n	13.42 rst	29.69 f-j	11.03 st	40.21 a	37.93 ab	43.90 a	39.21 ab	40.31 ab
T ₂	21.75 k-o	11.67 st	27.51 h-l	10.91 st	40.42 a	41.58 a	41.46 a	41.04 a	41.12 ab
T ₃	34.66 defg	9.43 t	26.69 i-m	14.87 o-t	40.24 a	41.06 a	40.03 a	42.84 a	41.04 ab
T ₄	32.32 d-i	15.52 o-t	38.38 bcd	20.87 k-q	41.25 a	40.30 a	41.86 a	38.64 ab	40.51 ab
T ₅	33.72 d-i	19.68 m-r	15.75 o-t	14.44 p-t	30.75 b	36.51 ab	42.73 a	42.26	38.06 b
T ₆	37.10 cde	13.88 qrst	30.61 e-j	17.64 n-s	42.70 a	36.91 ab	39.98 a	39.16 ab	39.68 ab
T ₇	32.97 d-i	13.95 qrst	36.62 cdef	21.39 k-p	40.60 a	40.71 a	39.97 a	39.76 ab	40.26 ab
T ₈	27.86 g-k	16.27 o-t	56.25 a	44.45 b	39.44 ab	36.21 ab	41.71 a	39.10 ab	39.11 ab
T ₉	42.67 bc	34.53 d-h	30.52 e-j	25.04 j-m	39.90 a	40.47 a	40.75 a	42.45 a	40.89 ab
T ₁₀	43.65 bc	20.80 l-q	60.73 a	32.83 d-i	41.45 a	38.99 ab	44.87 a	41.84 a	41.78 a
Mean of cultivars	25.02 b	28.30 a	39.38 b	41.17 a					
Means of irrigation conditions	Normal irrigation = 34.20 a	Drought = 19.12 b	Normal irrigation = 40.55 a	Drought = 40 a					

T₁ = Control (Untreated), T₂ = Distilled water spray, T₃ = Distilled water spray + Tween-20 (0.1 %), T₄ = Glycine betaine @ 100 mg L⁻¹ + Tween-20 (0.1 %), T₅ = Salicylic acid @ 100 mg L⁻¹ + Tween-20 (0.1 %), T₆ = Ascorbic acid @ 100 mg L⁻¹ + Tween-20 (0.1 %), T₇ = Glycine betaine and Salicylic acid @ 50 mg L⁻¹ each + Tween-20 (0.1 %), T₈ = Glycine betaine and Ascorbic acid @ 50 mg L⁻¹ each + Tween-20 (0.1 %), T₉ = Salicylic acid and Ascorbic acid @ 50 mg L⁻¹ each + Tween-20 (0.1 %), T₁₀ = Glycine betaine + Salicylic acid + Ascorbic acid @ 33 mg L⁻¹ each + Tween-20 (0.1 %)

Leaf relative contents indicate the status of water in plants and exogenous application of PGRs is believed to improve relative water contents in most plants especially under drought stress conditions (Zhang *et al.*, 2014; Cui *et al.*, 2015; Hafez & Gharib, 2016). Hafez & Gharib (2016) reported that exogenous application of ascorbic acid ameliorated the negative effect of water stress and improved the leaf water contents of wheat under drought stress. Our results also showed that foliar applied mixture of three PGRs even at minimum concentration (33 mg L⁻¹ each) improved the relative water contents under normal irrigation and drought conditions. Enhanced growth of cotton with foliar-applied PGRs under drought is associated with greater osmotic adjustment to promote water retention in leaves and cellular membrane stability. Similar findings were reported by Burgess & Huang (2014) who found that sequential application of PGRs and osmoregulants improved cellular membrane stability of creeping bentgrass (*Agrostis stolonifera*), indicating less membrane damage during drought stress. In another study, water scarcity caused a significant reduction in growth, yield and yield components of cotton (Çakir, 2004). The adverse effects of drought stress were reported to be alleviated to a greater extent by foliar-applied PGRs (Umar, 2006; Noreen *et al.*, 2013). In our study, yield and yield components of cotton like open boll per plant, average boll weight and cotton seed yield were improved with foliar-applied mixture of glycine betaine, salicylic acid and ascorbic acid each at 33 mg L⁻¹ under normal irrigation conditions. However, under drought conditions, maximum number of main stem node, opened boll per plant, average boll weight and cotton seed yield was recorded with the mixture of salicylic acid and ascorbic acid each at 50 mg L⁻¹. This might have been due to the water status in cotton plants under normal irrigation conditions where low concentration of PGRs (33 mg L⁻¹) enhanced growth and yield parameters. In contrast, under drought conditions, low water status might not have been mitigated by water stress at this concentration. So, high concentration of salicylic acid and ascorbic acid at 50 mg L⁻¹ each caused alleviation of the drought stress and improved the yield and yield components. The overall performance of FH-142 was better compared with cv. Lalazar under drought conditions with foliar-applied PGRs. This variation might have been due to better drought tolerance of FH-142 or capacity of this cultivar to build drought tolerance mechanism with foliar-applied PGRs. These results are in agreement with those of Ali & Ashraf (2011) who found that foliar-applied PGRs were very effective in improving water stress tolerance in maize plants which was ascribed to regulation in water relation and photosynthetic parameters as well as antioxidant defence system.

Many studies have been published on the effect of glycine betaine, salicylic acid and ascorbic acid, individually on the growth and yield of major field crops like cotton, wheat, rice and maize under stress conditions (Umar, 2006; Noreen *et al.*, 2013; Zhang *et al.*, 2014) and the results showed that various PGRs mitigate the adverse effects of stress environment on different crops. No data have been found on the effect of different PGRs in different combinations on the growth and yield of crops. So, the combination of effective PGRs may have synergistic effect on crop productivity when used in

different combinations and concentrations. Our results showed that combined application of selected PGRs especially salicylic acid and ascorbic acid at 50 mg L⁻¹ each even at low concentration (33 mg L⁻¹ each) improved the cotton productivity under water stress conditions. However, under normal irrigation conditions, the selected PGRs also improved the cotton productivity when used as single or in combination. It is concluded that salicylic acid and ascorbic acid each at 50 mg L⁻¹ could be effectively employed to reduce the adverse effects of drought stress on cotton crop and cv. FH-142 is suitable for good production under drought conditions.

Acknowledgment

The authors gratefully acknowledge the financial support of Pakistan Academy of Sciences.

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