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Enhancing Grain Yield Quality of Wheat with Exogenous Application of Salicylic Acid and Trehalose

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Abstract

Drought stress is a major constraint for crop production in arid and semiarid regions, such as Egypt. In this study, water stress induced noticeable increase in the root length and root/shoot ratio while decreased flag leaf area of both wheat cultivars during grain-filling. Moreover, water stress decreased wheat yield components (spike length, number of spikelets/main spike, 100 kernel weight, grain number/spike, grain yield/spike, grain yield/plant, straw yield/plant, crop yield/plant, harvest, mobilization and crop indices). The applied chemicals appeared to alleviate the negative effects of water stress on wheat productivity (particularly the sensitive one). The effect was more pronounced with salicylic acid and Trehalose treatment. This improvement would result from the repairing effect of the provided chemicals on growth and metabolism of wheat plants grown under water deficit condition. In response to the applied water stress and the used chemicals, the grain yield of the sensitive and tolerant wheat cultivar was strongly correlated with all the estimated yield components (shoot length, spike length, plant height, main spike weight, number of spikelets per main spike, 100 kernel weight, grain number per spike, grain weight per plant, straw weight per plant, crop yield per plant, harvest, mobilization and crop indices).

Keywords: Wheat; Drought; Yield

Abbreviations: HI: Harvest Index; SA: Salicylic Acid; Tre: Trehalose; WS: Water Stress

Introduction

Wheat is a staple crop that plays a major role in global food security [1]. In addition, wheat is one of the most important cereals in view of nutritional value [2]. It serves as a rich source of carbohydrates, essential amino acids, fiber components, minerals and vitamins in the human diet [3]. Furthermore, wheat grains contain unique phytochemicals that complement those in fruits and vegetables when consumed together [4]. In Egypt, wheat is considered the first strategic food crop [5] and it has maintained its position as the basic staple food in urban areas and mixed with maize in rural areas for bread making. In addition, wheat straw can be considered as an important fodder [6]. Although the land cultivated under wheat is continuously increasing to meet the food demand of over growing population, Egypt still import about 46.8% of its need from wheat yearly [4]. Therefore, improving wheat production has become apriority of national programs.

Drought is one of the greatest constraints on growth and development [7]. In Egypt, outside the Nile valley, severe drought, (resulting from a deficiency of soil water) is among the most common factors limiting wheat production. Furthermore, the beginning of the 21st century is marked by global scarcity of water resources and environment pollution. Abiotic stress is already a major limiting factor in plant growth and will soon become even more severe as desertification covers more and more of the world's terrestrial area [8].

Yield is the most important economic trait of wheat plants, and grain production is the main selection criteria for drought resistance [9]. Yield is the ultimate outcome of all the processes involved at all stages in growth and development of a crop, any one of which may limit the yield of a particular crop. Estimates of grain yield bring another complexity to stress response, not just because the crops must be grown in controlled environments for long periods of time, but because the conversion of shoot biomass to grain biomass is complex [10]. Water stress is well known to cause disturbance in metabolites transport to grains, reduce the number of reproductive tillers which limit their contribution to grain yield and cause pollen sterility. Furthermore, water stress during grain growth could have a sever effect on final yield compared with stress during the other stages [5].

An efficient use of limited water resources and better growth under limited water supply are desirable traits for crops in drought environments [11]. Crop production and sustainable development are severely constrained by water limitations during the growing season [12]. Drought stress has become the major limitation factor on plant yield at global scale [13,14]. Plant response to water deficit dependent on the length and severity of water lost and also on the species or genotype, as well as on the age and stage of its development [15]. The plant response to drought at the crop level is complex because it reflects the integration of stress effects at all underlying levels of organisations over space and time. Many studies about the effects of supplemental irrigation on yield performance have shown that proper supplement of irrigation can increase crop yield by improving soil water conditions [16].

Water stress is well known to cause disturbance in metabolites transport to grains, reduce the number of reproductive tillers which limit their contribution to grain yield and cause pollen sterility. Furthermore, water stress during grain growth could have a sever effect on final yield compared with stress during the other stages [5]. Stressing wheat plants resulted in significant and gradual decline in all yield components, such as number of tillers, number of spikes per plant, number of grains per plant, straw yield, grain yield and weight of 1000 grains [17]. Moreover, the yield components, like grain yield, grain number, grain size, and floret number, were decreased under pre-anthesis drought stress treatment in sunflower [13]. Also, in water-stressed soybean, the seed yield was far below when compared to well-watered control plants [18].

In addition, the yielded grains contained less nitrogen, phosphorus, potassium, calcium and magnesium contents, but higher sodium content when compared with control plants [19]. In connection, yield components of water-stressed mustard plants were generally reduced, while total seed protein content showed significant increase [20]. Numerous studies have demonstrated that the composition of fatty acids can be altered in response to drought, with the extent of those changes depending upon the plant species, organ or severity of the stress [21,22]. Dornbos & Mullen [23] reported that drought stress can increase stearic acid and decrease oleic acid. However, the increase or decrease depended on the severity of drought [24]. So far, few works have been done on the effect of environmental factors such as drought on fatty acid composition of wheat grains.

Materials and Methods

Plant material and growth conditions

Pure strains of *Triticum aestivum L.* Gemmieza-7 (drought sensitive cultivar) and Sahel-1 (salt tolerant cultivar) were kindly supplied by the Agricultural Research Center, Ministry of Agriculture, Giza, Egypt. For soaking experiment, a homogenous lot of *Triticum aestivum L.* (i.e. either sensitive or tolerant cultivar) grains were selected. The grains were separately surface sterilized by soaking in 0.01M $HgCl_2$ solution for three minutes, then washed thoroughly with distilled water. The sterilized grains from each cultivar were divided into two sets (≈500g per set for each cultivar). Grains of the 1st set were soaked in distilled water to serve as control, while those of the 2nd were soaked in salicylic acid (3mM) each for about 6 hours.

After soaking, thoroughly washed grains were drilled in 20 November 2011 and 2012 in plastic pots (20 cm in diameter) filled with 5.5kg soil (clay/sand 2/1, v/v), where fifteen grains were sown in each pot. The pots were then kept in a greenhouse at Botany Department, Faculty of Science, Mansoura University, Egypt. The plants were subjected to natural day/night conditions (minimum/maximum air temperature and relative humidity were 15/25 °C and 35/45%; respectively) at mid-day during the experimental period. The plants in all sets were irrigated to field capacity by tap water. After two weeks from sowing, thinning was started so that five uniform seedlings were left in each pot for the subsequent studies.

On the day 65 after planting (at the beginning of heading) the pots of the 1st set was allocated to four groups (20 pots per each group) as follows: control (cont.), water stress (WS), trehalose control, trehalose+water stress (trehalose+WS). The 2nd set group was allocated to four groups as follows: salicylic acidcontrol (SA), salicylic acid + water stress (SA+WS), control trehalose+salicylic acid (SA+ trehalose) and salicylic acid+trehalose+water stress (SA+trehalose+WS). For+trehalose (1.5mM) treatment, the plants were sprayed by trehalose 48hrs before starting the stress period and weekly during the stress period.

Water deficit was imposed by withholding water at the reproductive stage for 30 days within two periods: on the day 65 from planting (heading stage) and the day 80 from planting (anthesis stage). Each droughted pot received 500ml water at the end of 1st stress period. At the end of stress periods, rewatering to the field capacity was carried out. The undroughted (control) plants were irrigated to the field capacity during the stress period, and all plants were left to grow until grain maturation under normal irrigation with tap water. After thinning and at heading, the plants received 36kg Nha^{-1} as urea and 25kg Pha^{-1} as super-phosphate.

Yield analyses

Harvest index= Economic yield/Straw yield [25]

Crop index= Grain yield/Biological yield [25]

Mobilization index= Crop yield/Straw yield [26]

Relative grain yield= (Yield in treatment/Yield in control) X 100 [25]

Evapotranspiration efficiency= Water use efficiency for grain/Harvest index [27]

Statistical analysis

It should be mentioned that the sample numbers which were taken for investigation were as follows: ten for growth parameters, ten for agronomic traits and three for all chemical analyses and only the mean values were represented in the respective figures. The data were subjected to one-way analysis of variance (ANOVA),

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and different letters indicate significant differences between treatments at p≤0.05, according to CoHort/CoStat software, Version 6.311.

Results

Perusal of the data shown in Figure 1 cleared that, in comparison with control values, water stress induced a noticeable (p≤0.05) increase in the root length (non significant increase in case of sensitive cultivar), root/shoot ratio and decreased flag leaf areaof both wheat cultivars during grain-filling. In the majority of cases, application of SA and/or Tre induced a marked increase (p≤0.05) in the values of root length (non significant increase in case of sensitive cultivar), root/shoot ratio and flag leaf area. However, interaction of SA and Tre was the most effective treatment in increasing root length, root/shoot ratio and flag leaf area.

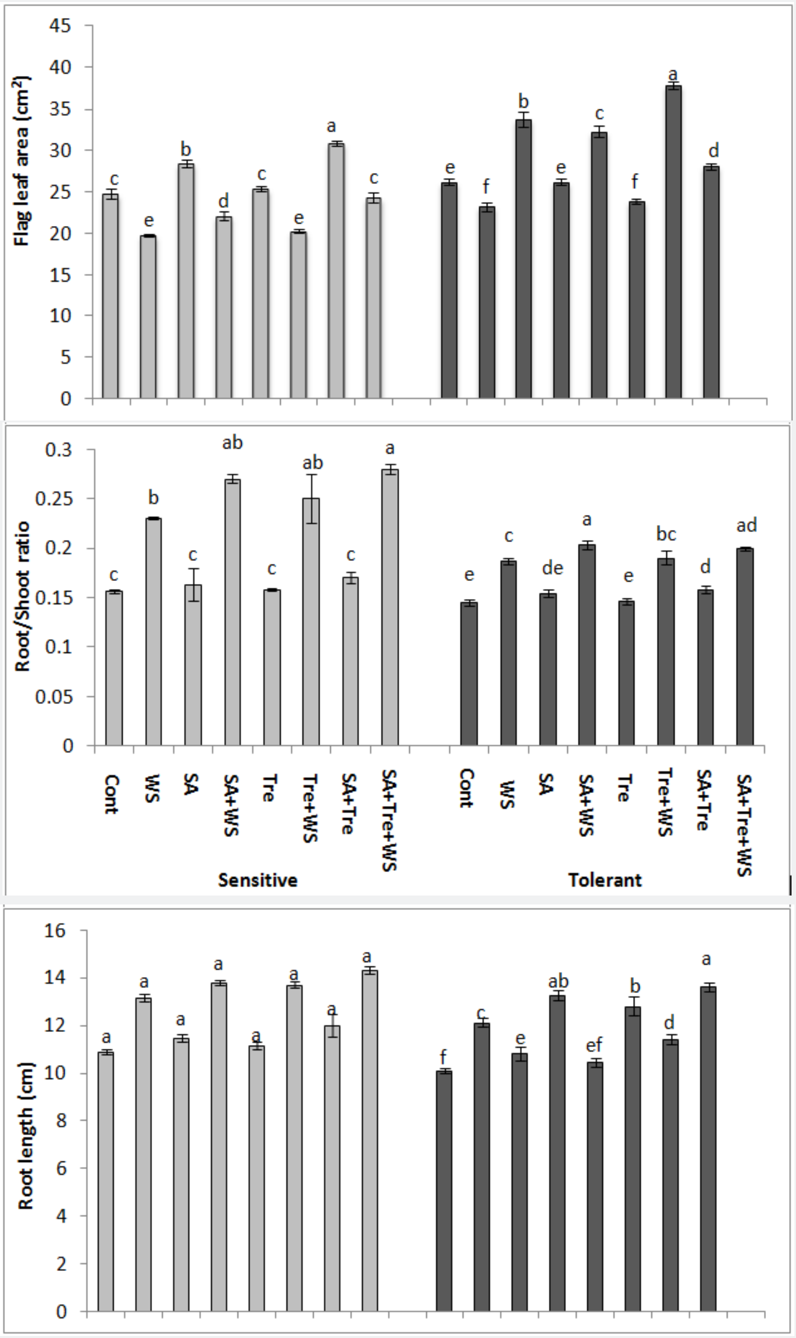

Figure 1: Effect of salicylic acid, trehalose and their interaction on root length (cm), root/shoot ratio and flag leaf area (cm^2) during grain-filling. Vertical bars represent standard error of the mean (n=10). Different letters indicate significant differences between treatments at p≤0.05, according to CoHort/ CoStat software, Version 6.311.

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Changes in yield components

To study the impact of grain priming with SA and spraying the wheat plants with Tre on yield components of both wheat plants grown under water stress, the shoot length, main spike length, plant height, main spike weight, number of spikes per plant, number of spikelets per main spike, number of spikelets per plant, number of grains per main spike, number of grains per plant, grain yield per main spike, grain yield per plant, individual grain biomass, 100-kernel weight, straw yield per main spike, straw yield per plant, crop yield per main spike, crop yield per plant, mobilization index, crop index, harvest index, as well as relative grain yield and evapotranspiration efficiency were estimated. Ten plants and ten grains from each treatment were randomly taken to estimate yield and yield components of wheat plants. The obtained results were tabulated and represented in suitable (Figure 1-7).

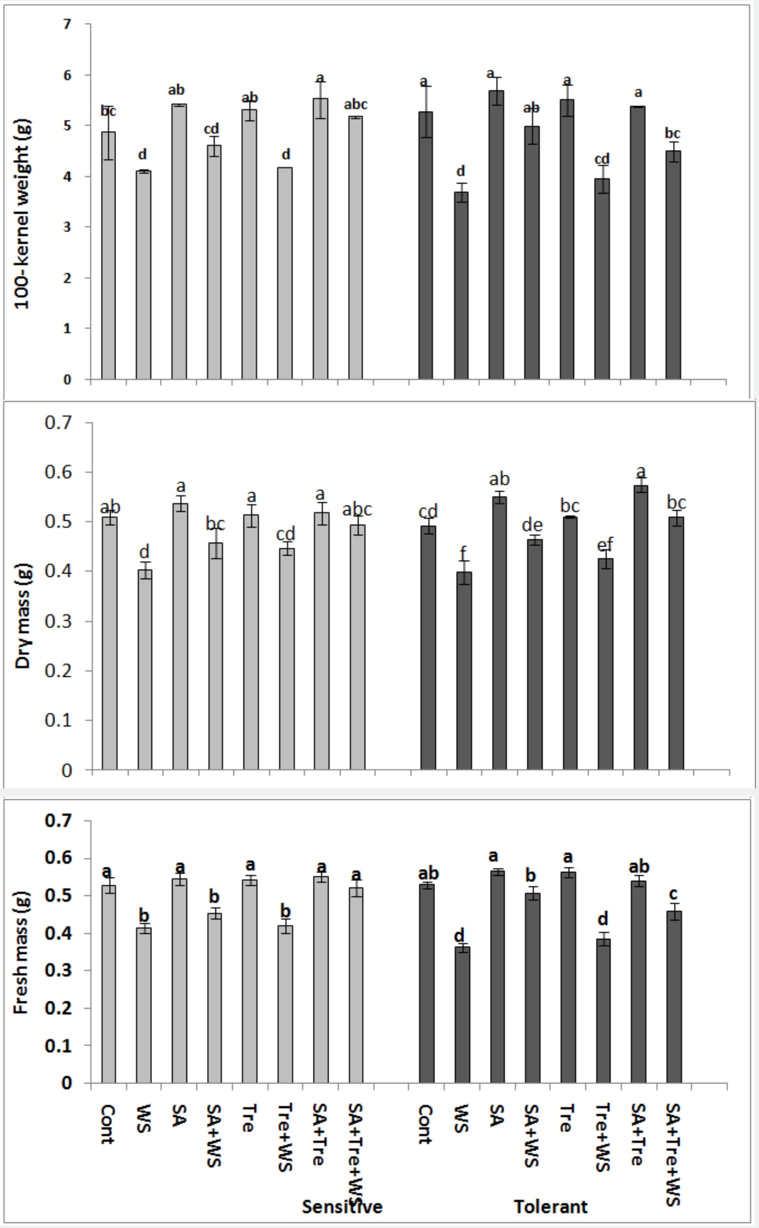

Figure 2: Effect of salicylic acid, trehalose and their interaction on grain biomass (g) and100-kernel weight (g) of yielded grains of droughted wheat cultivars. Vertical bars represent standard error of the mean (n=10). Different letters indicate significant differences between treatments at p≤0.05, according to CoHort/ CoStat software, Version 6.311.

The data in (Figure 2-9) revealed that water stress caused significant reduction ($p \leq 0.05$) in all yield components of wheat plants. With regard to the wheat cultivar, the sensitive one was more affected by water stress than the tolerant. In consequence to the previous determinations, SA and/or Tre induced additional increase ($p \leq 0.05$) in yield components of stressed wheat plants. Moreover, the treatment of SA and Tre improved all yield components more than that of SA or Tre only.

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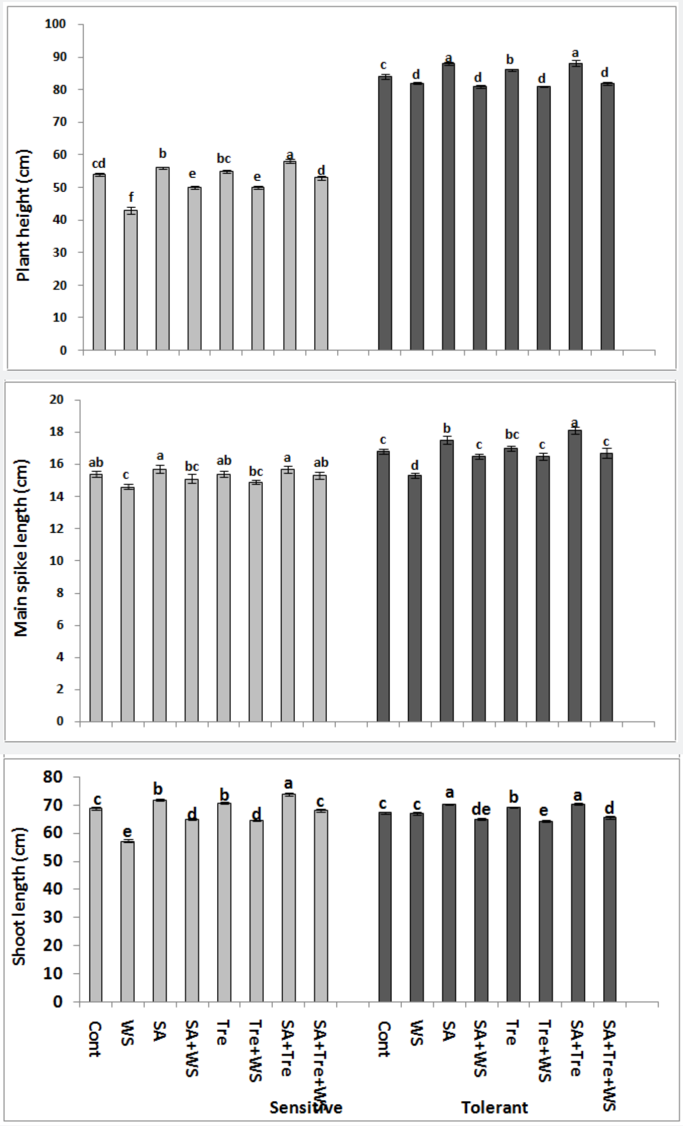

Figure 3: Effect of salicylic acid, trehalose and their interaction on shoot length (cm), main spike length (cm) and plant height (cm) of yielded grains of droughted wheat cultivars. Vertical bars represent standard error of the mean (n=10). Different letters indicate significant differences between treatments at p≤0.05, according to CoHort/ CoStat software, Version 6.311

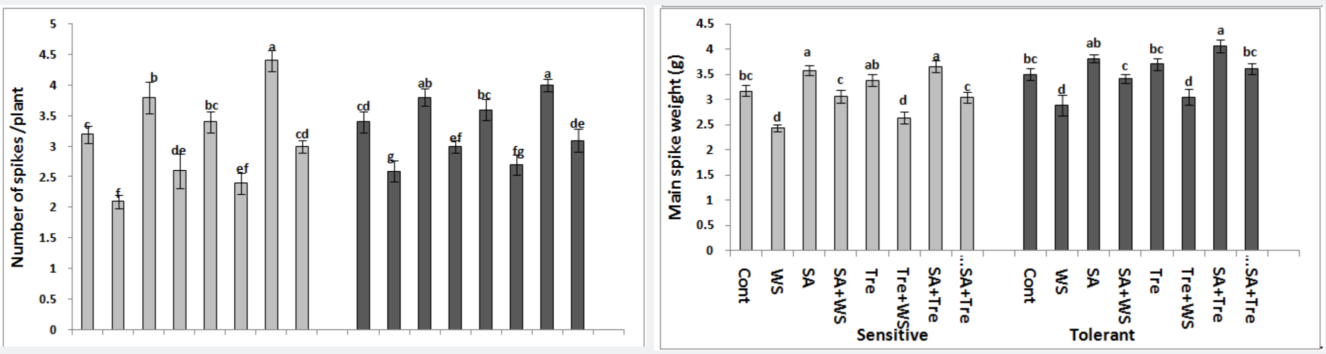

Figure 4: Effect of salicylic acid, trehalose and their interaction on main spike weight (g), number of spikes/plant and number of spikelets/ main spike of yielded grains of droughted wheat cultivars. Vertical bars represent standard error of the mean (n=10). Different letters indicate significant differences between treatments at p≤0.05, according to CoHort/ CoStat software, Version 6.311.

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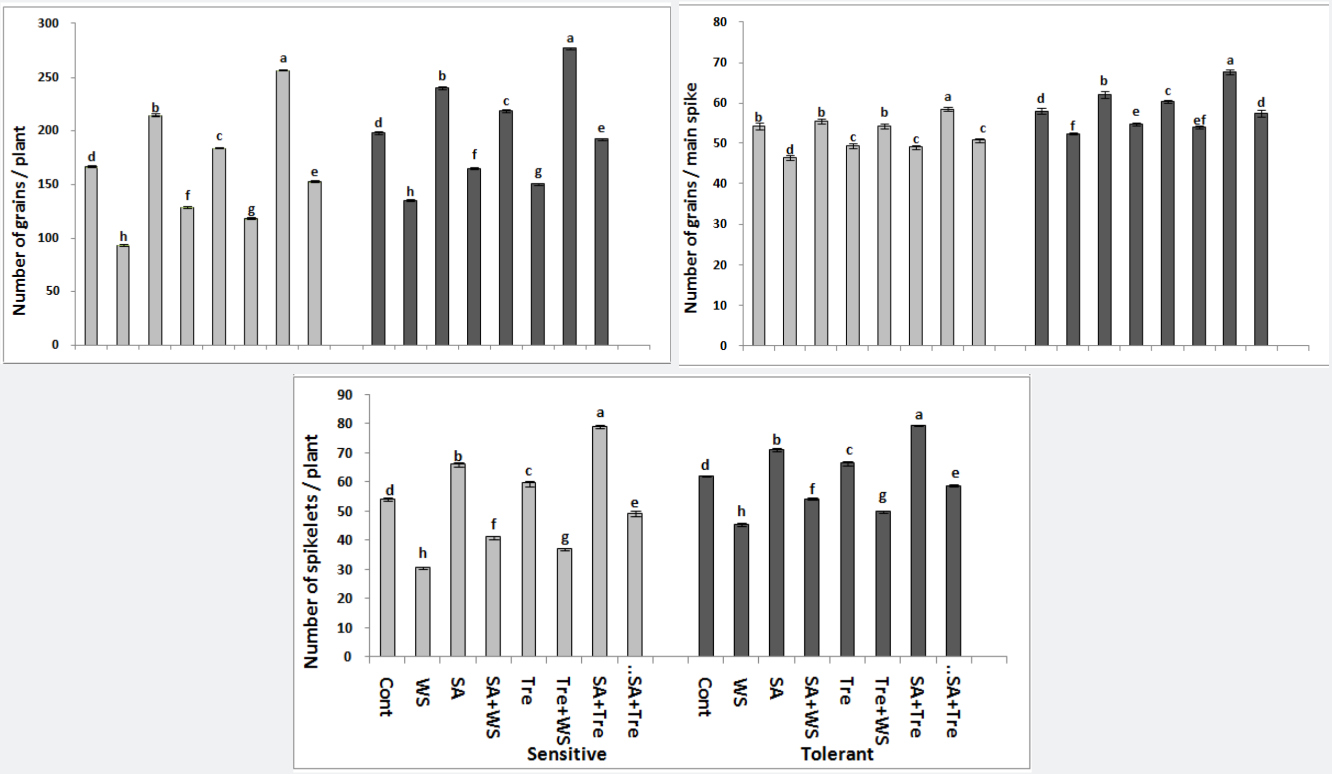

Figure 5: Effect of salicylic acid, trehalose and their interaction on number of spikelets / plant, number of grains / main spike and number of grains / plant of yielded grains of droughted wheat cultivars. Vertical bars represent standard error of the mean (n=10). Different letters indicate significant differences between treatments at p≤0.05, according to CoHort/ CoStat software, Version 6.311.

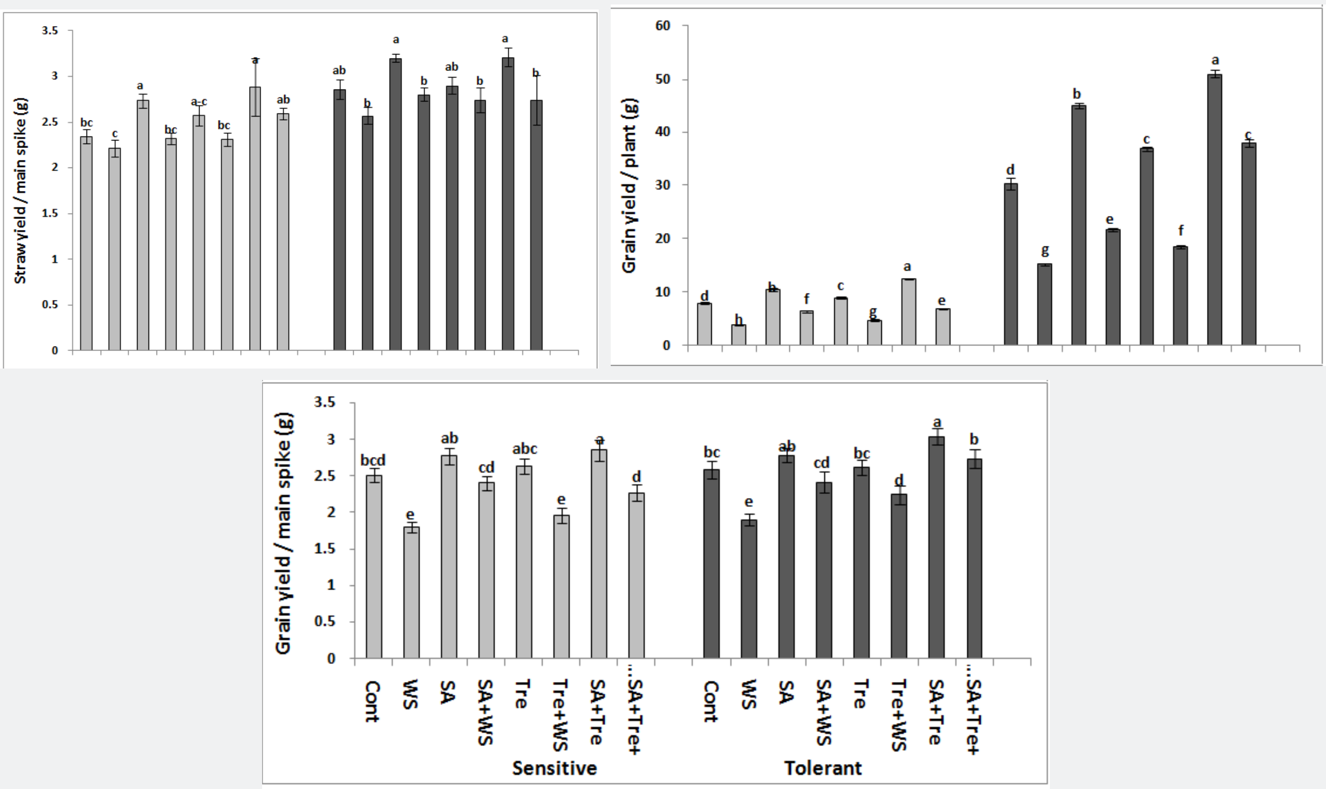

Figure 6: Effect of salicylic acid, trehalose and their interaction on grain yield/main spike (g), grain yield/plant (g) and straw yield/main spike (g) of yielded grains of droughted wheat cultivars. Vertical bars represent standard error of the mean (n=10). Different letters indicate significant differences between treatments at p≤0.05, according to CoHort/ CoStat software, Version 6.311.

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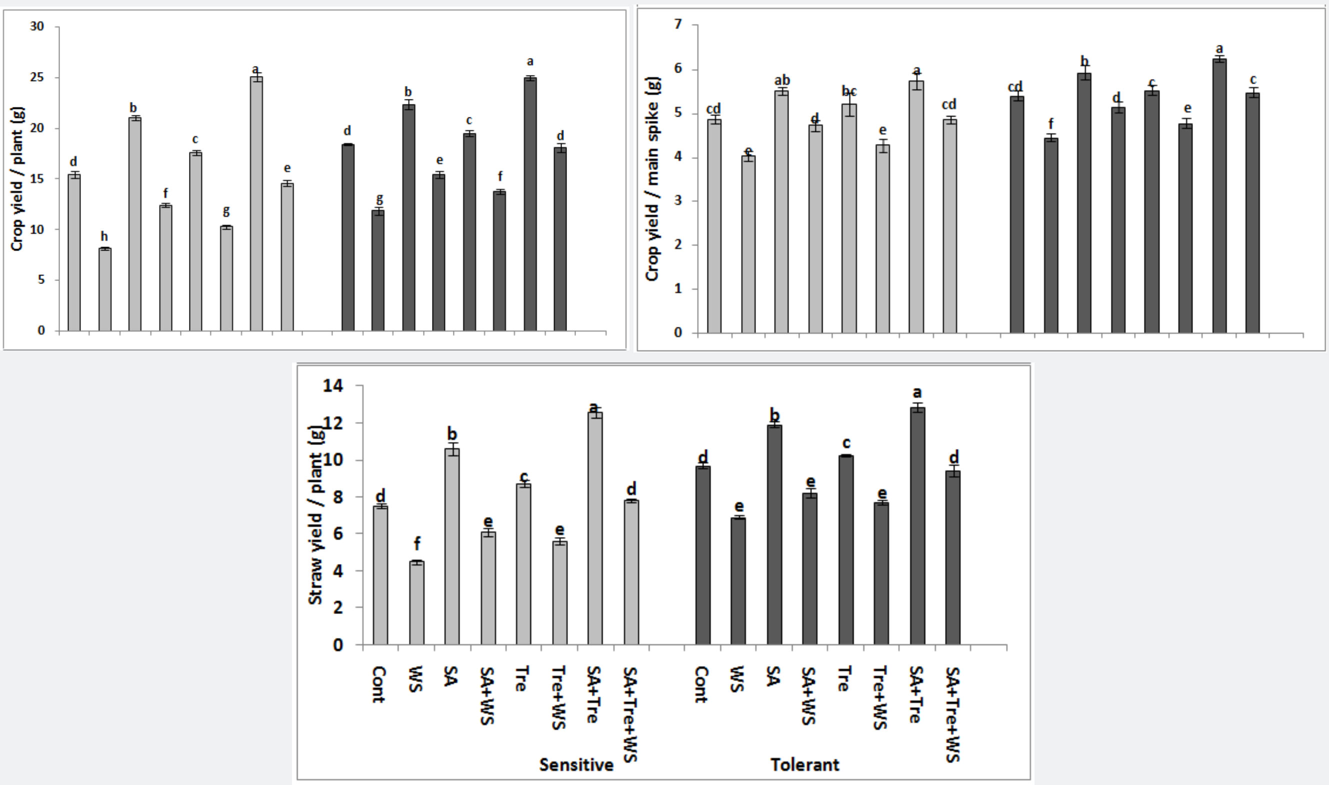

Figure 7: Effect of salicylic acid, trehalose and their interaction on straw yield/plant (g), crop yield/main spike (g)) and crop yield/plant (g) of yielded grains of droughted wheat cultivars. Vertical bars represent standard error of the mean (n=10). Different letters indicate significant differences between treatments at p≤0.05, according to CoHort/ CoStat software, Version 6.311.

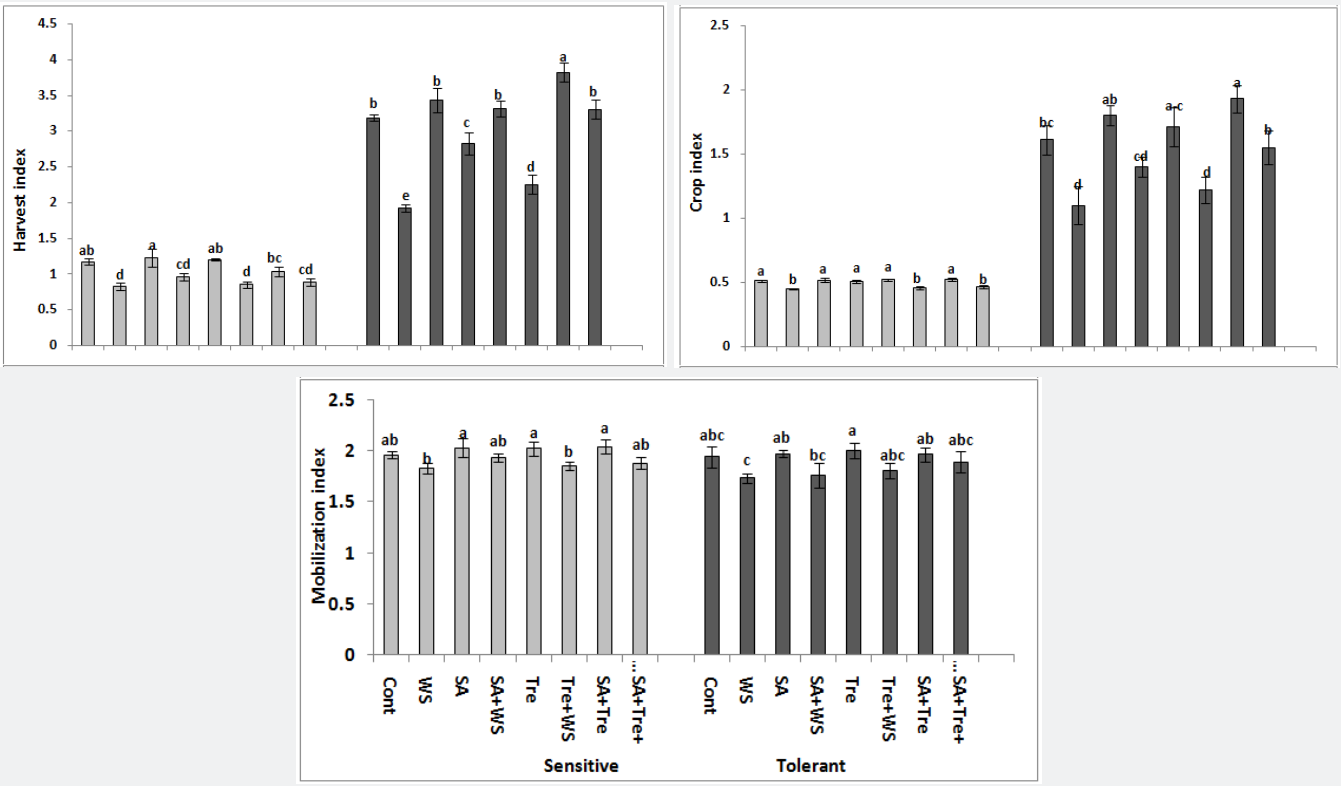

Figure 8: Effect of salicylic acid, trehalose and their interaction on mobilization index, crop index and harvest index of yielded grains of droughted wheat cultivars. Vertical bars represent standard error of the mean (n=10). Different letters indicate significant differences between treatments at p≤0.05, according to CoHort/ CoStat software, Version 6.311.

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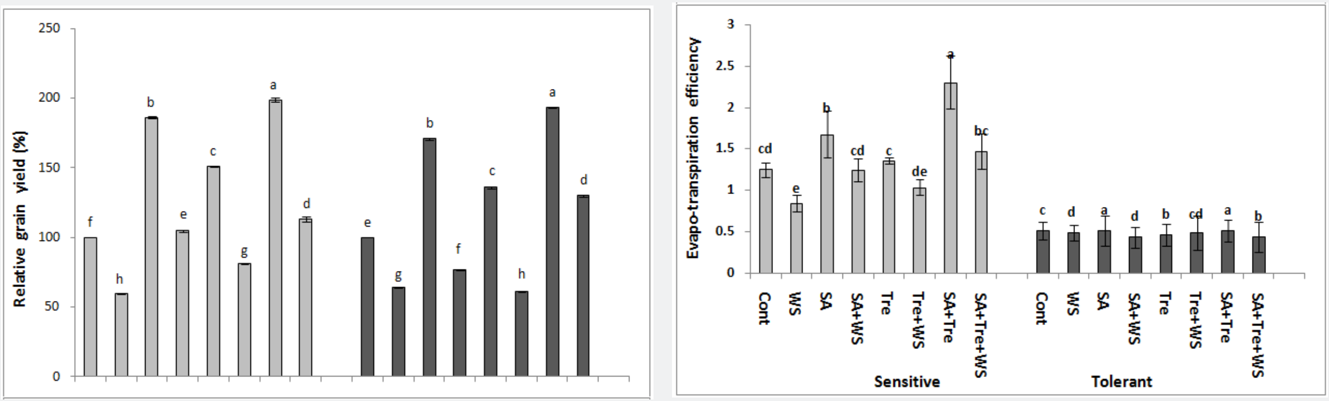

Figure 9: Effect of salicylic acid, trehalose and their interaction on evapo-transpiration efficiency and relative grain yield (%) of yielded grains of droughted wheat cultivars. Vertical bars represent standard error of the mean (n=10). Different letters indicate significant differences between treatments at p≤0.05, according to CoHort/ CoStat software, Version 6.311.

Discussion

The response and adaptation of plants to stress conditions are very complex and highly variable [28]. In a word, the study of the physiological mechanisms of wheat anti-drought has much work to do [13]. Research on the response of plants to drought with understanding the mechanisms of drought resistance is necessary in order to find ways for improving plant growth for almost million square kilometers of arid and dry land in Egypt. Drought is one of the major abiotic stresses which adversely affects crop growth and yield and thus a constraint for plant productivity worldwide [29,30]. In the present study, water stress greatly increased root length, root to shoot ratio was found to (Figure 1). In agreement with these results, stressing mulberry plants generally decreased root growth parameters as revealed by Das et al. [31].

Continuation of root growth under drought stress through stimulation in root length is an adaptive mechanism that facilitates water uptake from deeper soil layers. These results are in accordance with those obtained by Sundaravalli et al. [32] and Yin et al. [33]. Plants must develop vigorous root system that allows them to grow and overcome any stress conditions. Moreover, a large root system may improve a plant's competitive ability for water during drought stress, which is a plant's individual survival strategy in natural selection. According to life-history strategy theory, plants with a large root system are partitioning more photosynthetic products to roots, which imply a reduced partition to reproductive growth.

Stressing wheat plants significantly increased root/shoot ratio when compared with the control plants Figure 2. This finding coincided with that of many other researchers [17,34]. In this respect, Jabeen et al. [35] suggested that the increase in root/shoot ratio under water stress conditions may be due to;

(i) The increased accumulation of assimilates diverted to root growth,

(ii) Differential sensitivities of root and shoot to endogenous ABA and/or

(iii) Greater osmotic adjustment in roots compared with shoots. Matching these assumptions, El-Hendawy [36] assumed that although stress can induce rapid reduction in root growth; shoot growth decreases proportionally more than root growth, causing an increase in the root/shoot ratio.

Flag leaf plays the key and the most important role in plant life as it transport assimilates to spike and developing grains. The performance of flag leaf under certain growth condition reflects the overall viability and development of the whole plant. The general pattern of plant response to stress is a reduction in the rate of leaf surface expansion, followed by a cessation of expansion as the stress intensifies [37]. The retardation of leaf growth in stressed plants could be attributed to decreased turgor that may diminish cell production within the leaves. The adverse impact of drought on leaf growth may be also due to photosynthetic decline, osmotic stress, imposed constraints on plant processes and interference with nutrient availability [38].

The observed increase in growth of droughted wheat plants resulting from SA (antitranspirant) application may be due to its effect on improvement of turgidly at a time when the growth of that particular plant part was more dependent on water status than in photosynthesis [39]. The repairing effect of SA on the growth of water-stressed wheat plants may be due to the increased water uptake by root system. In addition, Aldesuquy & Ibrahim [40] proposed that exogenous application of hormones during water stress may reduce water loss rates and cause a concomitant increase in leaf water potential and carbon gain rates. Moreover, salicylic acid with decreasing evapotranspiration and increasing root development can help root to absorb more nutrients [41]. Also, Shakirova et al. [42] revealed that salicylic acid increased cell division in meristem of wheat seedling and improved plant growth.

The inhibition of leaf area in response to drought stress was alleviated when the grains were soaked in SA (Figure 1). This recovery may result from the role of SA in stimulating the rate of movement of nutrients and hormones from the plant root

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towards the developing leaves, and thus accelerating the rate of leaf expansion. Also, Movaghatian & Khorsandi [43] stated that seed priming with SA improved and enhanced germination traits of *Triticum aestivum L*. under various drought stress levels, thus, reducing some of the inhibitory effects of drought stress. Moreover, Arzandi [44] stated that salicylic acid spray was able to reduce the effect of deficit watering stress and increase plant dry weight and root length of *coriandrum sativum*.

Trehalose application alleviated adverse effects of drought stress in seedlings of different *Brassica* species by improving their growth and physiological attributes [45]. In connection, Alam et al. [45] stated that combination of Tre with drought showed improved seedlings fresh weight and dry weight in *Brassica* species. Since trehalose-producing resurrection plants show exceptionally high drought tolerance, we hypothesized that the introduction of trehalose-synthesizing capacity into crop plants may increase their growth performance under drought stress.

Yield is the most important economic trait of wheat plants and grain production is the main selection criteria for drought resistance [9]. Yield is a result of the integration of metabolic reactions in plants; consequently any factor that influences this metabolic activity at any period of plant growth can affect the yield [46]. Crop plants are especially sensitive to drought stress during the early reproductive stage, which causes significant yield loss in cereal production and detrimental effects on grain qua [47,48]. This is a well established fact that yield of crop plants in drying soil reduces even in tolerant lines of that crop species [14,49]. In this investigation, yield and yield attributes (shoot length, spike length, plant height, main spike weight, number of spikelets per main spike, 100 kernel weight, grain number per spike, grain weight per plant, straw weight per plant, crop yield per plant, harvest, mobilization and crop indices) are reduced due to water stress in both wheat cultivars (2 up to 8). The decrease in yield and yield components in different crops under similar conditions has also been reported by many workers [50-52]. These workers clearly indicated that drought tolerant genotypes showed less reduction in yield plants in respect of susceptible ones. Moreover, it is well known that drought can reduce the final grain yield by influencing wheat growth in different growth stages.

Drought stress during the early stage of reproductive growth tends to reduce yield by reducing seed number. During seed development stress reduces yield by reducing seed size. Prolonged moisture stress during reproductive growth can severely reduce yield because of reduced seed number and seed size [53]. A positive and significant relation was recorded between dry shoot weight and achene yield per plant. The yield components, like grain yield, grain number, grain size, and floret number, are decreased under drought stress treatment in sunflower [13]. Moreover, Amin et al. [54] attributed the increase in yield and yield characteristics of onion plants with foliar spray of salicylic acid to an increase in photosynthesis and assimilation and translocation of assimilates, and greater nutrient uptake and increased cytoplasmic streaming and cell integrity.

Water stress reduced harvest, mobilization and crop indices in the two wheat cultivars (Figure 7). This was in agreement with Jaleel et al. [12] who reported that, water stress decreased harvest index, and biomass yield in two varieties of *Catharanthus roseus*. However, in crops, the detrimental effects of water deficits on the harvest index (HI) also minimize the impact of the water limitation on crop productivity and increase the efficiency of water use [55]. Therefore, increasing transpiration, transpiration efficiency and harvest index are three important avenues for the important of agricultural productivity [49]. Additionally, the aerial environment plays a role in determining the ratio of carbon gain to water use, because the vapor pressure deficits between the leaf and the air determine the transpiration rate [16]. Moreover, results showed that the decrease in harvest index with extending interval of irrigation may be attributed to leaf abscission under water deficiency conditions which can be led to weight loss in plant shoot and decreasing of biological yield [56].

The application of SA and/or Tre enhanced the yield and yield components of the two unstressed and stressed wheat cultivars. Moreover, the treatment of SA and Tre improved all yield components more than that of SA or Tre only. This result conforms with the findings of Elwan & El-Hamahmy [57] who reported that SA application at low concentration positively improved the growth attributes and fruit yield of pepper (*Capsicum annuum* L.). Moreover, our results regarding the positive effects of SA application on grain weight were in agreement with those reported by Ali & Mahmoud [58]. In this respect, Arfan et al. [51] studied the effect of exogenous application of salicylic acid (SA) through the rooting medium of two wheat cultivars differing in salinity tolerance. They found that increase in grain yield along with increase in 100-grain weight, number of grains and number of spikelets per spike with 0.25mM SA application under saline conditions suggested that improvement in salt-induced reduction in grain yield with SA application was mainly due to increase in grain size and number. Moreover, Dawood et al. [59] in a study reported that all experimental treatments of SA significantly increased seed yield and yield components of sunflower.

The beneficial effect of SA and Tre on grain yield may be due to translocation of more photo assimilates to grains during grain filling, thereby increasing grain weight. These results are similar to those of Zhou et al. [60] who reported that maize stem injected with SA produced 9% more grain weight than those with sucrose and distilled water treatments. The second possible mechanism of SA-induced yield enhancement might be an increase in the number of spikelets and number of grains, because SA has the capacity to both directly or indirectly regulate yield. These results are in a good agreement with those obtained by Kumar et al. [61] and Khan et al. [62] with maize. Also, improvement of chlorophyll content, enzyme activity, maintenance of water balance and sufficient build up of food reserves for formation of more seeds might be reason for increase in seeds number with foliar application of salicylic acid [63]. The increase in translocation of photosynthates to the seed under foliar application of salicylic

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acid might be responsible for increasing the 1000-seed weight in our study.

The promoting effect of SA on the flag leaf blade area and blades area/plant of barley mentioned that enhancing effect of SA on the availability and movement of nutrients could result in stimulating different nutrients in the leaves and consequently promote yield and yield components [64]. The above-mentioned results are in accord with those obtained by many investigators [49]. Also, it was reported that drought during the grain-filling stage reduced grain weight to a greater extent and strongly reduced yielding capacity [65]. In addition, Savin & Nicolas [66] investigated the effects of drought stress on grain growth, starch and nitrogen accumulation in barley cultivars. Water deficits decreased both individual grain weight and grain yield. Nitrogen content per grain was quite high and similar for all treatments, and nitrogen percentage increased when stress was severe enough to reduce starch accumulation. This confirms that starch accumulation is more sensitive to post-anthesis stress than nitrogen accumulation.

Application of SA and/or Tre appeared to mitigate the deleterious effects of water stress on grain biomass of the two wheat cultivars. The repairing effect of SA may be attributed to the fact that SA reduces the rate of transpiration from leaves [67], which could possibly lead to the accumulation of excessive water, thus resulting consequently in an increase in grain fresh mass [68]. Furthermore, Tre application may act in the same manner as SA in inducing drastic reduction in the rate of transpiration [69,70].

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