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Original Research Article

INTERACTIVE EFFECTS OF KINETIN AND SPERMINE ON ANATOMICAL ADAPTATIONS AND PRODUCTIVITY TO SEAWATER SALINITY IN WHEAT

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Abstract: A pot experiment was conducted to evaluate the beneficial effect of grain presoaking in kinetin (0.1mM), spermine (0.3mM) or their interaction on wheat plants (*Triticum aestivum* L.) variety Sakha 93 on some anatomical features in flag leaf as and pedicle of main shoot as well as grain yield at ear emergence (after 65 days from sowing) by measuring leaf thickness, ground tissue thickness, number of hairs, meta-xylem vessel area, xylem vessel area, phloem tissue area, vascular bundle tissues area, number of motor cells as well as number of opened and closed stomata on both upper and lower epidermis and some anatomical features of peduncle (peduncle diameter, tracheids area, meta-xylem vessel area, xylem area, phloem area, vascular area, number of vascular bundle as well as opened and closed stomata) of wheat plants. Wheat plants respond to seawater salinity with characteristic modifications in their anatomy to counter the ill effect of seawater stress. Therefore, Irrigation of wheat plants with seawater caused significant increase in leaf and ground tissue thickness in flag leaves as well as meta-xylem vessel area, xylem vessel area, vascular bundle area in flag leaf and peduncle of main shoot of wheat plants. However, irrigation of wheat plants with seawater decreased phloem area in flag leaves and peduncle of the main shoot of wheat plants. The application of kinetin, spermine or their interaction induced some modifications in the anatomical features of the flag leaf and peduncle of the main shoot which appeared to be an adaptive response to salinity stress caused by seawater. Furthermore, grain priming with kinetin, spermine or their interaction increased phloem thickness in both leaf and peduncle of main shoot and consequently induced rapid rate of translocation of photo-assimilates from flag leaf to developing grains in spikes and consequently increase productivity of wheat plants irrigated by seawater.

Key Words: Anatomy, Kinetin, Wheat, Seawater, Spermine.

INTRODUCTION

Anatomical changes induced by water deficits in higher plants were better-observed indicators, which can be directly applied to agriculture and handled. To aim at exploring efficient anatomical indices, much information has been documented, but more attention should be paid to link them with physiological and molecular one [1]. It is evident that there are extensive changes in leaf anatomy of the plants growing under salt stress [2]. Plants encountering stressful environmental conditions, such as high salinity, respond to this external stressor with characteristic modifications in their anatomy [3]. Irrigation of wheat plants with NaCl, particularly at 99 mM, induced noticeable increases in flag leaf blade, mesophyll tissue thickness, as well as the number of motor cells and hairs on lower epidermis. Furthermore, treatment with NaCl generally reduced the peduncle diameter, xylem tissue thickness, number of hairs as well as number of stomata on the peduncle of the main shoot [4]. Well-developed motor cells for extensive leaf rolling was apparent in *Festuca novae* [5] and *Deschampsia antarctica* [6] plants grown under water stress.

In principal, the production of cereal grains is directly correlated to the growth of shoot as the main factory in which assimilates are produced. However, the photosynthetic ability of these shoots depends mainly on their pigments content [7,8]. Considering the conclusion of that the accumulation of dry matter in the grains requires the production of assimilates in the leaves, their translocation to the rachilla of the spikelets, movement into the endosperm and embryo of the grain and synthesis of materials to be stored. Leaf structure seems to be of great importance in the regard. Thus conductive canals between source and sink in wheat cultivars have been reported to the developing grains [9].

Çavuşoğlu *et al.,* [2] reported that salinization of the nutrient medium caused changes in the anatomic properties of the leaves of barely plants, where salinity increased stomatal number, stomatal index and epidermis cell width on the upper surface in comparison with the leaves of control. In addition, the salt treatment increased the distance between vascular bundles and decreased the stomatal width. Also, salinity caused insignificant increase in stomatal length and leaf thickness. Hameed *et al.,* [10] showed an increment in midrib and lamina thickness of *Imperata cylindrica* plants with increasing salt level. Furthermore, salt treatment resulted in extensive leaf rolling and reduced epidermal thickness.

Salinity was found to increase epidermal thickness, mesophyll thickness, palisade cell length, palisade diameter and spongy cell diameter in leaves of bean and cotton plants [11]. In leaves of potato, salt stress caused smaller intercellular spaces and a

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reduction in chloroplast number [12]. Moreover, salinity reduced stomatal density in the leaves of tomato plants [13]. Recently, it was shown that seed priming with plant growth regulators (PGRs) could counteract the deleterious effects of salinity on germination and plant growth, since the major effect of salinity is the inhibition of crop growth by the reduced hormone delivery from roots to leaves [14]. These PGRs have been found to play a key role in the integration of the responses expressed by plants under stress conditions [15]. Moreover, PGRs may also enhance germination and adaptation of plants to stress conditions. Polyamines have been proposed as a new category of PGRs that are purported to be involved in a large spectrum of physiological processes in plant growth and development, such as cell division, embryogenesis, morphogenesis, reproductive organ development, root growth, tuberization, floral initiation and development as well as fruit development and ripening [16].

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 salinity 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 [17]. Stressing wheat plants with different levels of saline solution 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. In addition, the yielded grains contained less nitrogen, phosphorus, potassium, calcium and magnesium contents, but higher sodium content when compared with control plants [18].

The primary objective of the present study is to assess up to what extent seed priming with kinetin, spermine or their interaction could ameliorate the deleterious effects of seawater irrigation on some anatomical features in flag leaf as well as pedicle of main shoot at ear emergence wheat plants.

MATERIALS AND METHODS

Plant material and growth conditions

For soaking experiment, a homogenous lot of *Triticum aestivum* L. var. Sakha 93 grains were selected. The grains were surface sterilized by soaking in 0.01 M HgCl2 solution for three minutes, then washed thoroughly with distilled water. The sterilized grains were divided into four sets. Grains of the 1st set were soaked in distilled water to serve as control, while those of the 2nd, 3rd or 4th set were soaked in 0.1 mM kinetin, 0.3 mM spermine or 0.1 mM kinetin + 0.3 mM spermine; respectively, each for about 12 hours. After soaking, thoroughly washed grains were drilled in 15th November 2011 in plastic pots (20 cm in diameter) filled with 5.5 kg soil (clay/sand 2/1, v/v), where fifteen grains was 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. The plants of each set were sub-divided into two groups. The 1st group in each set was still irrigated with normal tap water serving as control, whereas the 2nd one was irrigated with 25% seawater. The resulting eight treatments were marked as following: 1. Cont., 2. SW, 3.Cont. K, 4. SW+K, 5. Cont. Spm, 6. SW+ Spm, 7. Cont. K+ Spm, 8. SW+ K+ Spm. Irrigation with seawater was applied after 30 days from sowing with a periodical soil washing (each two weeks) with tap water. After thinning and at heading, the plants received 36 kg N ha^{-1} as urea and 25 kg P ha^{-1} as super-phosphate. Measurements were carried out at ear emergence (i.e. 65 d from planting). Samples from the flag leaf as well as pedicel of the main shoot were taken at ear emergence (65d after sowing) as triplicates for anatomical studies. For anatomical study, samples from the flag leaf and peduncle of the main shoot at heading stage were used. Measurements of all anatomical parameters studied were carried out in the keel region of the flag leaf and in the peduncle of main shoot of wheat plants. Also, all measurements were well photographed using light microscope.

Anatomical Studies

For anatomical studies, samples from fresh plant materials were used. Samples were killed and fixed in Formalin- Acetic acid –Alcohol (FAA) for at least 48 hours. Dehydration, sectioning staining and mounting procedures was followed according to the method described by Sass [19]. Sections were cut at thickness 15 µm, and then stained with safranin and light green combination. Canda Balsam was used as mounting medium. Sections were estimated by the aid of light microscope. Measurements of all anatomical parameters were calculated in keel region to µm.

Measurements of conductive canals area in flag leaf and peduncle

A new technique developed using the image analysis for measuring the anatomical features of leaf and peduncle of the wheat plants was performed using the following steps:

1. Image acquisition-obtaining precise microscopic images (transmitted) of the leaf and peduncle to

determine (the areas of metaxylem, tracheids, xylem, phloem, and vascular bundle of the leaf and peduncle).

2. Color planes HSL extraction- this steps aims at extraction of saturation plane from HSL images. (Note: Because each color plane is made up of 8 bits, the color plane extracted will appear as an 8-bit grayscale image).

2. Bright points filtering out- In this step bright points in the image that are associated with the periodic structure of the web are filtered out.

These bright and relatively small points could be confused with pores if they were not removed. The Gaussian model for the background is applied locally to the image to establish the threshold for each area of the image. The result is a binary image where objects pores are segmented from the background are converted into black segments.

Images are two-dimensional computer arrays of numbers. Each point in the image has x and y coordinates so that pixels are often specified by (x, y). Images can be of several types but in this analysis only 2 types are considered: integer or grey level images, and binary images. Integer or grey level images are typically the most common type. Each of the pixels has an integer value which might be between 0 and 255 or possibly something larger. Usually each possible value is associated with a shade of grey between black (0) and white (the maximum value). Binary images contain only 0's and 1's and are the same as 1 bit integer images. Binary images are usually created by the image analysis technique. Very often we want to identify some parts of the image and, for example, measure their geometrical features. The way this is done is to create a binary image with 1's in the feature area and 0's everywhere else. This removes all the information from the image except the part we want, which is where the features are and then makes the measurements on the binary image [20].

It was also important to implement techniques of image segmentation to measure and count special features contained in the image. Image segmentation implies separating the parts of the image which are of interest from the rest.

In order to segment the image, a threshold of darkness was established using image processing of the grey levels. In other words, all grey levels darker than some value G were considered as ink and everything else as a background. The image was then converted to binary, which gives us a binary image containing 1's in the places where the original image was < G and 0's where the original image was > G. In other words, measures and counts of clusters of 1's in this binary image (representing the needed areas) were made.

The raw data from each measurement was in pixels. These were converted into real area units by calibration. This was done by measuring the size in pixels and calculating a scale factor in mm (or μm) per pixel. In this case, the lengths are multiplied by this value and the areas are multiplied by its square. Sometimes the scaling is different in the x and y directions and two scale factors have to be used.

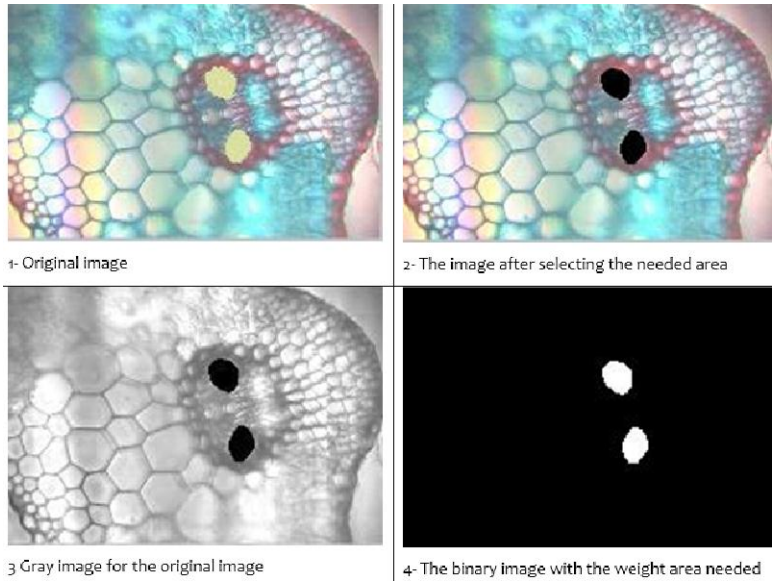

Figure 1: The image analysis steps for estimating the area of meta-xylem (in microns) for flag leaf of wheat cultivars.

Statistical analysis

A test for significant differences between means at P ≤ 0.05 was performed using least significant difference (LSD) test. The correlation coefficients were estimated according to SPSS programme.

RESULTS

Changes in anatomical characteristics

It was found that seawater stress markedly affected the anatomical features of wheat flag leaves (Table 1 and Plate 1). Irrigation of wheat plants with 25% seawater caused noticeable increase (P < 0.05) in leaf thickness, ground tissue thickness, meta-xylem vessel area, xylem area, vascular bundle area as well as the number of motor cells, hairs and closed stomata on both upper and lower epidermis of wheat flag leaves. On the other hand, salinity stress decreased (P< 0.05) phloem area and the number of opened stomata on both upper and lower epidermis.

Grain presoaking in kinetin, spermine or their interaction seemed to alleviate the adverse effects of salinity stress on the anatomical features of wheat flag leaves. Treatment with kinetin + spermine appeared to be the most effective in the recovery of seawater-induced alterations in anatomical features of wheat flag leaves (Table 1 and Figs 2& 3).

Table 1: Effect of grain presoaking in kinetin, spermine or their interaction on flag leaf anatomy of wheat plants (at heading stage) irrigated with seawater.

Parameters / Treatments	Leaf thickness (µm)	Ground tissue thickness (µm)	Metaxylem vessel area (µm²)	Xylem area (µm²)	Phloem area (µm²)	Vascular bundle area (µm²)	Number of motor cells	Number of hairs	Number of stomata on lower epidermis		Number of stomata on upper epidermis	
									Opened	Closed	Opened	Closed
Cont	315	298	852	3054	8703	13793	44.8	13.3	13.8	5.4	19.2	5.7
SW	331	310	973	3895	7478	15009	48.1	15.1	11.2	7.9	16.4	8.5
K	339	317	981	4253	9417	15817	52.4	17.9	16.1	3.3	22.1	2.9
SW + K	376	341	10211	4971	9152	17043	53.2	20.4	13.6	5.9	17.8	7.2
Spm	301	290	863	3093	6931	12011	49.7	13.6	15.2	3.9	19.5	4.7
SW + Spm	309	294	904	3004	7542	12039	52.9	16.8	14.4	4.8	19.3	5.6
K + Spm	389	360	10524	5261	10234	18221	53.6	19.2	17.2	2.0	22.5	2.7
SW +K +Spm	404	375	11098	6109	9871	19028	57.2	22.4	12.1	7.2	17.4	7.8
LSD at P ≤ 0.05	12.8	11.2	97.9	324.2	509.8	602.4	2.9	1.7	1.4	0.4	1.5	0.6

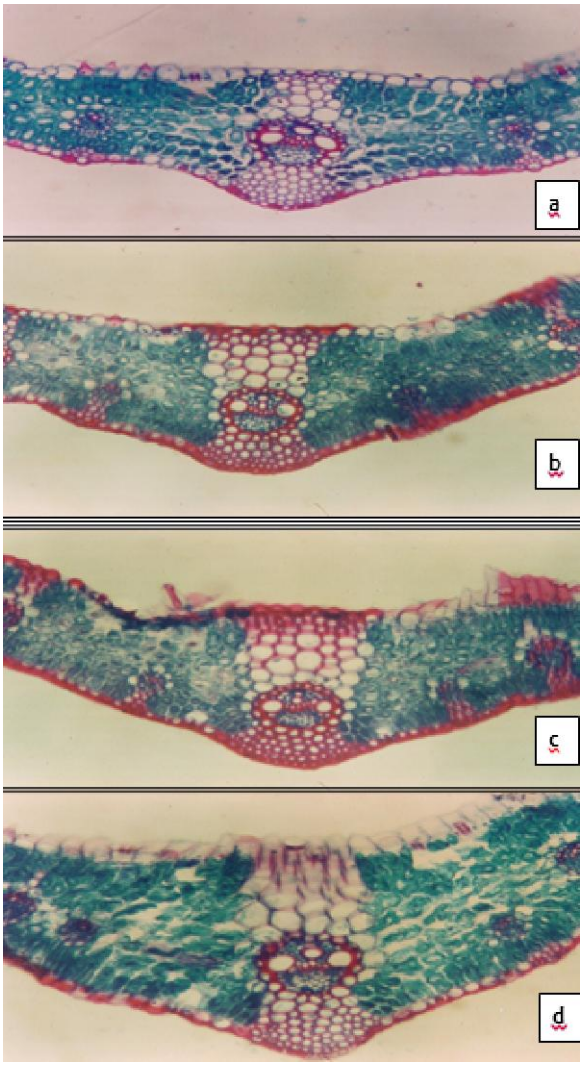

Figure 2: Effect of grain presoaking in kinetin on flag leaf anatomy of wheat plants (at heading stage) irrigated with seawater. (Cross section X = 40).
a) Control b) 25% Seawater
c) Kinetin d) 25% Seawater + kinetin

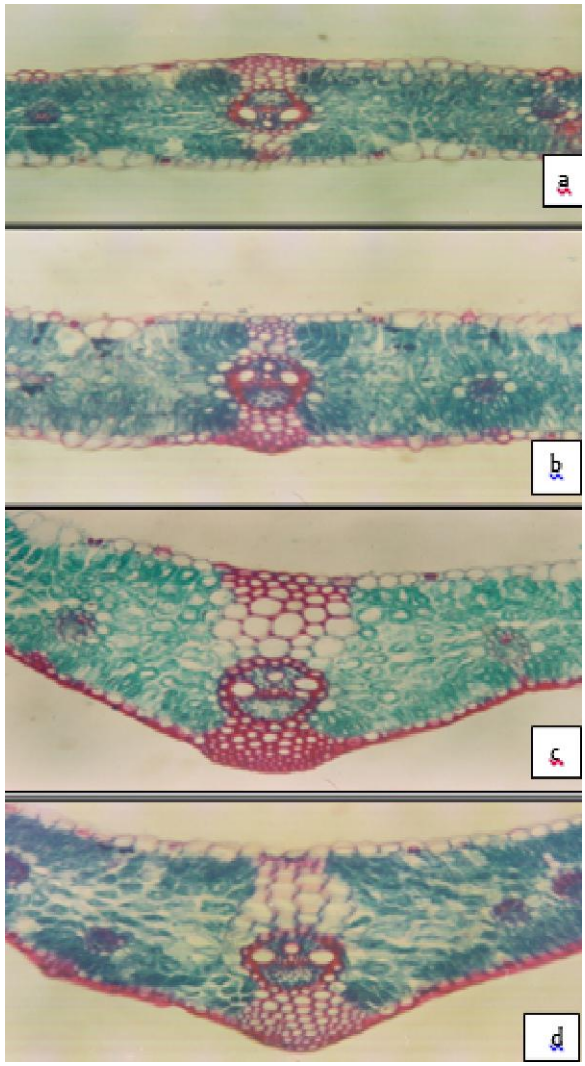

Figure 3: Effect of grain presoaking in spermine and kinetin + spermine on flag leaf anatomy of wheat plants (at heading stage) irrigated with seawater. (Cross section X = 40).
a) Spermine b) 25% Seawater + spermine
c) Kinetin+ spermine d) 25% Seawater + kinetin+ spermine

As compared to control values, salinity stress caused noticeable changes in the anatomical features of the peduncle of wheat plants (Table 2 and Fig. 4). Salinity stress generally led to a massive decrease (P< 0.05) in peduncle diameter, phloem area, vascular bundle area as well as number of vascular bundles and number of opened stomata. At the same time, seawater stress significantly increased (P< 0.05) meta-xylem vessel area, xylem area, as well as the number of hairs and closed stomata on the peduncle surface of wheat plants.

Table 2: Effect of grain presoaking in kinetin, spermine or their interaction on peduncle anatomy of wheat plants (at heading stage) irrigated with seawater.

Parameters / Treatments	Peduncle diameter (mm)	Metaxylem vessel area (µm²)	Xylem area (µm²)	Phloem area (µm²)	Vascular bundle area (µm²)	Number of vascular bundles	Number of hairs	Number of stomata	
								Opened	Closed
Cont	1.21	340	983	844	3056	48.9	12.7	13.6	15.3
SW	1.17	378	906	734	2752	44.5	13.2	10.6	18.5
K	1.27	389	1028	987	3847	53.2	13.5	13.1	16.1
SW + K	1.22	409	1099	901	3289	49.3	14.1	9.2	19.8
Spm	1.23	356	1003	892	3504	50.6	13.6	12.3	15.8
SW + Spm	1.20	393	1070	851	3159	48.3	14.1	10.0	19.1
K + Spm	1.30	402	1105	1032	4023	55.4	14.4	12.1	16.9
SW + K + Spm	1.24	411	1081	923	3373	52.9	15.6	9.0	20.1
LSD at P ≤ 0.05	0.28	32.4	66.2	73.7	356.1	3.72	1.36	1.98	2.14

In general, the anatomical features of the peduncle of salinized wheat plants were mitigated by kinetin, spermine or their interaction (Table 2 and Figs 4 & 5). The magnitude of response was more pronounced with kinetin + spermine treatment.

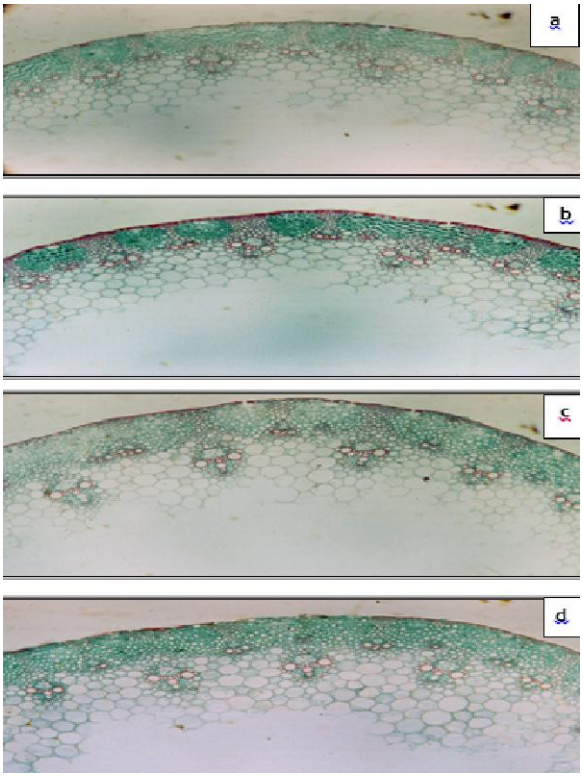

Figure 4. Effect of grain presoaking in kinetin on peduncle anatomy of wheat plants (at heading stage) irrigated with seawater. (Cross section X = 40).
a) Control b) 25% Seawater
c) Kinetin d) 25% Seawater + kinetin

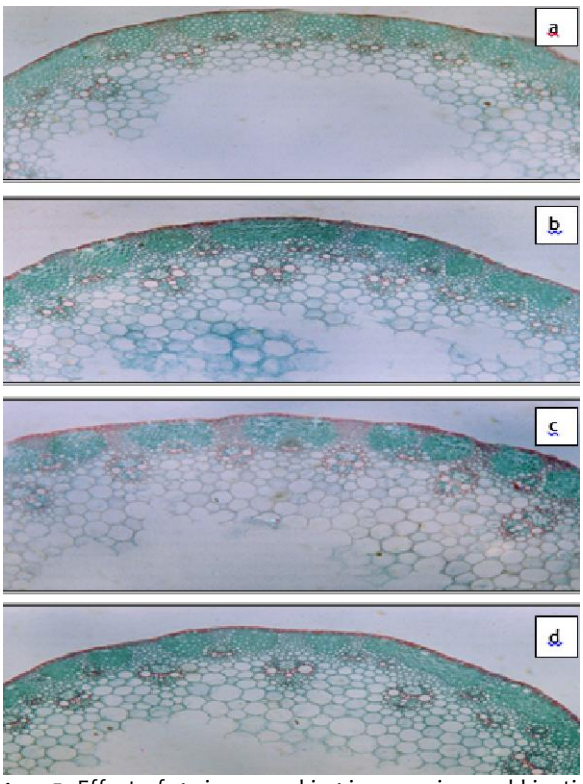

Figure 5: Effect of grain presoaking in spermine and kinetin + spermine on peduncle anatomy of wheat plants (at heading stage) irrigated with seawater. (Cross section X = 40).
a) Spermine b) 25% Seawater + spermine
c) Kinetin+ spermine d) 25% Seawater + kinetin+ spermine

Changes in grain yield

Seawater stress at 25% caused significant reduction (P ≤ 0.05) in grain yield of wheat plants (Fig. 6). On the other hand, grain presoaking in kinetin, spermine or their interaction caused marked increase

(P ≤ 0.05) in grain yield of wheat plants irrigated with seawater (Table 3).

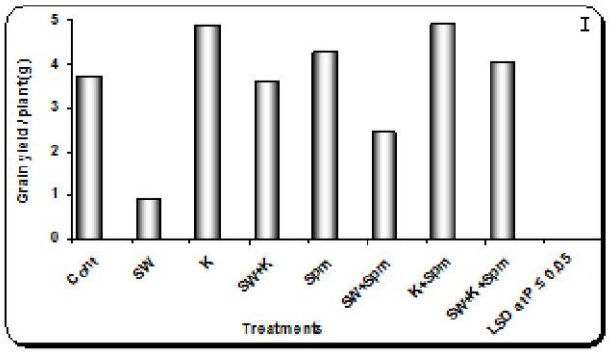

Figure 6: Effect of grain presoaking in kinetin, spermine or their interaction on grain yield per main spike of wheat plants irrigated with seawater. Vertical bars represent LSD at P ≤ 0.05.

The correlation coefficient between grain yield and the anatomical features of wheat flag leaf

For identifying the factors which were related to the economic yield, the correlation coefficients between the grain yield and anatomical features in response to the applied salinity stress and the used chemicals (kinetin, spermine or their interaction) were estimated from the measured mean values. For anatomical features of flag leaf, the grain yield was strongly correlated with phloem area (r = 0.60) as well as the number of opened stomata on both the lower (r = 0.75) and upper (r =0.74) epidermis. Conversely, negative correlation was recorded for the number of closed stomata on both lower (r = -0.72) and upper (r = -0.73) epidermis of wheat flag leaf. Grain yield of wheat plants was also observed to be positively correlated with most anatomical criteria of peduncle, such as peduncle diameter (r = 0.90), meta-xylem vessel area (r = 0.88), xylem area (r = 0.88), phloem area (r = 0.92), vascular bundle area (r = 0.88) and the number of vascular bundles (r = 0.92). Moreover, grain yield was positively correlated with both the number (r = 0.78) and frequency (r = 0.77) of chloroplasts in mesophyll cells of wheat flag leaf at heading stage.

DISCUSSION

Anatomical changes induced by water deficit in higher plants were better-observed indicators which can be directly applied to agriculture and handled. To aim at exploring efficient anatomical indices, much information has been documented, but more attention should be paid to link them with physiological and molecular parameters [1]. Concerning the anatomical characteristics of wheat flag leaves, seawater stress caused noticeable increases in leaf thickness, ground tissue thickness, meta-xylem vessel area, xylem area, vascular bundle area as well as the number of hairs, motor cells and closed stomata on both upper and lower epidermis. On the other hand, salinity stress decreased phloem area and the number of opened stomata on both upper and lower epidermis of wheat flag leaves. Furthermore, seawater stress led to massive decrease in peduncle diameter, phloem area, vascular bundle area as well as the number of vascular bundles and the number of opened stomata. At the same time, seawater stress significantly increased meta-xylem vessel area, xylem area, as well as the number of hairs and closed stomata on the peduncle surface of wheat plants. In general, the anatomical features of the flag leaf and peduncle of salinized wheat plants were repaired by kinetin, Spm or their interaction. These results were generally in agreement with those obtained by Aldesuquy [21].

The increase recorded in leaf and ground tissue thickness in response to seawater stress could be considered as a strategy exerted by wheat plants to cope with high salt levels. This increase tended to compensate for the negative effect of salinity on leaf cell metabolism [22]. In this regard, salinity was found to enhance the thickness of jojoba (*Simmondsia chinensis*) leaves [23]. Parés *et al.,* [24] noticed that plants exposed to high salinity levels in the irrigation water showed an increase in leaf thickness which could be interpreted as a morphogenetic response counteracting the negative effect of salinity. From another point of view, Hameed *et al.,* [10] reported that increased thickness may help in storing ions inside the plant body due to increased vacuolar volume, especially when the plant species is characterized by having no glands to remove salt from the leaves. For that, Wignarajah *et al.,* [25] found that the leaves of the salt-treated French bean plants became thicker than those of the unstressed plants.

Grain presoaking in kinetin, Spm or their interaction caused additional increase in both leaf thickness and ground tissue thickness of stressed wheat plants, indicating better adaptability potential to prevent undue water loss under saline environments. The increase in the leaf thickness might be due to the increase in the number and/or size of cells forming the mesophyll tissue [26].

The increase in flag leaf thickness due to the exogenous application of kinetin and/or Spm was obvious to be positively correlated with the ability of these growth regulators to increase both flag leaf area and specific leaf area. Moreover, this stimulative effect of the applied chemicals on flag leaf and ground tissue thickness was in close parallelism with their ability to increase chloroplast frequency in mesophyll tissue, photosynthetic pigments and consequently carbohydrates content, which are all reflected in improved yield and yield components of stressed or

unstressed plants treated with these chemicals. The conductive canals between source and sink in wheat cultivars have been reported to play a prominent role in the translocation of photosynthates to the developing grains [9]. The reduction of peduncle diameter in response to seawater irrigation could be ascribed to the deleterious impact of salt stress on both cell division and cell enlargement or to the salt-induced inhibition of meristematic division [27]. According to Cutler *et al.,* [28], reduction in cell size appears to be a major response of cells to water deficiency.

Exogenous application of kinetin, either alone or in combination with Spm, increased peduncle diameter of stressed and unstressed wheat plants. This stimulating effect of both growth regulators on peduncle diameter may be ascribed to their ability to enhance cell division, extension and enlargement.

The obtained results revealed that salt stress caused reduction in vascular bundle area and the number of vascular bundles in the peduncle of wheat plants. This may probably be due to the decrease in the number of additive divisions in the cambium [29]. Furthermore, Hameed *et al.,* [10] reported that high salt concentrations reduced the cambium activity in *Populus euphratica* plants.

According to Evans *et al.,* [9], phloem area was found to be directly proportional to the rate of assimilate import to the ears of wheat plants. In this respect, Aldesuquy and Baka [7] reported that the flag leaf of salt-stressed wheat plants showed some negative changes in conductive tissues, where the decrease in the phloem area would lead to a slow rate of translocation of photo-assimilates towards the developing grains.

Data from anatomical analysis of different plant species showed that xylem vessel area is decreased when grown under stress conditions [30]. However, our results revealed that seawater stress increased meta-xylem vessel and whole xylem area in both the flag leaves and peduncle of wheat plants. In this connection, Tyree *et al.,* [31] found that vessels with large diameter are evolutionarily favored for efficient water conduction.

The applied chemicals induced additional increase in the area of conductive canals (xylem and phloem) as well as the number of vascular bundles in flag leaf and peduncle of wheat plants. In addition, the application of these chemicals caused positive correlations between grain yield and vascular bundle area, xylem area and phloem area in both leaf and peduncle of stressed and unstressed wheat plants. This furnishes better translocation of assimilates from flag leaf (as source) towards the developing grains (as sink) through the conductive canals, where there is a good source-sink relationship [4].

Irrigation with seawater induced an increase in the number of closed stomata along with the decrease in the number of opened stomata in flag leaf and peduncle of wheat plants. This may be due to an increase in ABA content in leaves under salt stress [32].

The higher numbers of hairs on flag leaf and peduncle surfaces as a result of seawater stress could be attributed to the fact that hairs may increase the accumulation of ions inside it to avoid the harmful effects of Na^+ and Cl^- ions. This increase in the number of hairs on flag leaf and peduncle in seawater-irrigated plants would be expected to increase the leaf resistance to excessive ion toxicity [33]. In this connection, Udovenko *et al., [*34] found that salinity increased the number of hairs on peduncle surface.

Application of kinetin, Spm or their interaction resulted in additional increase in the number of hairs on both flag leaf and peduncle surface of wheat plants irrigated with seawater, and this may increase the ability of these stressed plants to cope with the elevated salt amounts in the growing medium. These results were in accordance with those obtained by Aldesuquy *et al.,* [35] working on wheat plants.

The enhanced effect of seawater irrigation on the number of motor cells on upper and lower surfaces of flag leaves could be considered as another adaptive response of wheat plants to seawater stress, where the role of motor cells is important in leaf rolling. Like muscle cells which have excitation-contraction coupling, motor cells in higher plants exhibit rapid movement resulting from excitation turgor loss [36]. Regarding the mechanism of movement in *Mimosa*, Sibaoka [36] reported that the motor cells contain a fibrillar structure, and the contraction of these fibrils may open pores in the membrane of the motor cells upon activation. Outward bulk flow of the vacuolar sap through these pores, due to the pressure inside the cell, results in turgor loss from the motor cells and then the bending of the organ.

The increased frequency of motor cells due to the application of kinetin and/or Spm can be regarded as an additive criterion to salt tolerance of wheat plants. Motor cells play an important role in leaf rolling to avoid water loss under stress conditions [8]. The presence of more motor cells in salt-stressed plants is a significant adaptation against water loss under physiological drought conditions due to salt stress.

The reduction in wheat yield and yield components in response to seawater stress might be attributed to the inhibitory effect of salinity on plant growth due to the suppression of many metabolic processes including protein, nucleic acids and PA synthesis lowered activity of mitochondria and chloroplasts [37], decreasing transpiration, stomatal conductance and photosynthesis [38] , restriction in the absorption of water by plant roots and water use efficiency [39] , the toxic effects of certain ions present in soil solution and/or imbalance in phytohormone levels through its effects on either biosynthesis or destruction of plant hormones [40]. Shani and Dudley [41] related the yield loss to reduced photosynthesis, high energy and carbohydrates expenses in osmoregulation and interference with cell functions under saline conditions.

An improvement in wheat productivity due to increased rates of photosynthates translocation from leaves to grains caused by hormone pretreatment had also been suggested by Ray and Choudhuri [42] and also by Aldesuquy and Ibrahim [43]. It was proposed that under salt stress conditions, the thickness of assimilate-conducting pathway (phloem area) is reduced [43], and leaves start behaving as sinks rather than sources [44]. This causes inhibition of assimilate movement towards the developing reproductive organs, which might be the reason for the observed decrease in yield. On the other hand, these adverse effects of high salinity were alleviated by the hormone treatment, primarily by rejuvenation of the sink potential and enhancement of the duration or rate of dry mass accumulation in developing reproductive organs [45].

CONCLUSION

In conclusion, wheat plants respond to seawater salinity with characteristic modifications in their anatomy to counter the ill effect of seawater stress. Therefore, Irrigation of wheat plants with seawater at 25% caused significant increase in leaf and ground tissue thickness in flag leaves as well as meta-xylem vessel area, xylem vessel area, vascular bundle area in flag leaf and peduncle of main shoot of wheat plants. However, irrigation of wheat plants with seawater decreased phloem area in flag leaves and peduncle of the main shoot of wheat plants. The application of kinetin, spermine or their interaction induced some modifications in the anatomical features of the flag leaf and peduncle of the main shoot which appeared to be an adaptive response to salinity stress caused by seawater. Furthermore, grain priming with kinetin, spermine or their interaction increased phloem thickness in both leaf and peduncle of main shoot and consequently induced rapid rate of translocation of photo-assimilates from flag leaf to developing grains in spikes and consequently increase productivity of wheat plants irrigated by seawater.

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