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Antioxidant compounds enhanced the anatomical characteristics of Date Palm (Phoenix dactylifera L.) offshoot leaves under salinity stress.

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Abstract

This study was conducted to investigate the protective effects of foliar spraying with antioxidant compounds, including calcium chelate, potassium chelate, salicylic acid, and citric acid (1000 ppm each), on the anatomical characteristics of date palm offshoot leaves under salinity stress (soil electrical conductivity: 14 dS m^{-1} irrigation water: 4 dS m^{-1}), for both Barhee and Sayer cultivars. The use of antioxidants significantly increased leaf anatomical parameters. Salicylic acid treatment was the most effective, resulting in increased leaf blade thickness, vascular bundle diameter, xylem thickness, phloem thickness, epidermal thickness, and tannin cell density in both cultivars. In contrast, antioxidant treatments generally reduced stomatal density. Furthermore, the Barhee cultivar exhibited consistently superior anatomical responses compared with the Sayer cultivar. These findings demonstrate that exogenous application antioxidant-related compounds can alleviate salinity-induced structural alterations and contribute to improved anatomical adaptation in date palm offshoots..

Keywords: Antioxidant compounds, vascular bundles, tannin cells, salinity stress, and Stomatal density

Introduction

Date palm (*Phoenix dactylifera* L.) is one of the most important fruit crops worldwide, especially in hot, dry areas such as the Middle East and North Africa, as well as in some regions of South Asia (Al-habsi, 2025). The date palm has a high tolerance to harsh environmental conditions and has become a common food source and an important part of the economy of many regions (Koippully et al., 2025). Nevertheless, the widespread distribution of salinity in agricultural soils is one of the most serious challenges to date palm growth and yield (Gabash et al., 2024). Salinity stress not only regulates several plant physiological processes but also induces physiological and anatomical impairments, ultimately restricting crop production. Salinity affects water relations, disrupts ion balance, and has detrimental effects on the metabolic functions of the date palm (Shareef, 2024). Osmotic stress resulting from high salinity restricts plant water and nutrient uptake and can ultimately lead to physiological drought. This type of stress can lead to reduced leaf area, inhibited photosynthetic photochemistry, and changes in leaf anatomy (Hu et al., 2023). Hence, understanding the impact of salinity on the morphological characteristics of date palm leaves is essential for its management. In recent years, increasing attention has been directed toward the use of exogenous protective compounds to mitigate salinity stress damage, particularly those associated with antioxidant activity and signaling. Such compounds have been reported to enhance salt tolerance by improving ion homeostasis, water retention, and physiological responses (Zhou et al., 2024). Calcium chelate plays a pivotal role in plant stress responses, functioning not only as a structural component of cell walls and membranes but also as a second messenger in abiotic stress signaling pathways (Wdowiak et al., 2024). Calcium has been shown to play a crucial role in stabilizing membranes (plasma and organelle membranes) to minimize leakage and maintain cell integrity (Abdullah et al., 2023a). Moreover, calcium contributes to the modulation of antioxidant enzyme activities, indirectly by preventing the accumulation of ROS and, therefore, oxidative damage. Leaves from palms treated with calcium also had more lignified vascular bundles (associated with structural reinforcement) (Wdowiak et al., 2024). Potassium chelate is essential for osmoregulation, fluid retention, and charge balance of the plant. It is the primary inorganic solute that contributes to osmotic adjustment, particularly under salt stress (Abdullah et al., 2023b). Potassium status regulates stomatal behavior, adjusting stomatal density and function to optimize transpiration while minimizing water loss, thereby supporting steady CO_2 uptake (Shareef, 2019). As such, improved potassium status has also been

associated with wider blades and robust mesophyll tissue, which sustain leaf hydration and continued photosynthetic function (Hu et al., 2022). Salicylic acid (SA) is a well-recognized phytohormones and a potent signaling molecule that regulates numerous defense responses, including the up regulation of antioxidant defenses (Mishra et al., 2024). Histological studies reveal that exogenously applied SA stimulates the activity of enzymatic antioxidants (e.g., superoxide dismutase, catalase, ascorbate peroxidase) and induces the accumulation of non-enzymatic antioxidants (e.g., phenolic compounds, tannins) in leaf tissues (Gonz et al., 2022). Citric acid, as a natural organic acid and chelating agent, improves nutrient availability and alleviates ion toxicity (especially Na and Cl) in salt-affected soils (Hemdan et al., 2025). Citric acid can also increase endogenous levels of compatible solutes and antioxidant metabolites in leaf tissues (Mohamed et al., 2025). The synergistic performance of foliar-applied antioxidant compounds is anticipated to yield significant enhancements in date palm leaf anatomical traits under stress. Offering a mitigating anatomical support by reducing oxidative damage, reinforcing cell structure, regulating water balance, and sustaining cellular processes, these compounds support the long-term performance and functioning of palm leaves in adverse environmental conditions. Here, we present the comprehensive anatomical response of date palm leaves to exogenously applied antioxidant treatments, thus filling the information gap regarding the ultimate role of these inexpensive materials in promoting sustainable agriculture in the context of resilient farming in open orchards.

Materials and Methods

A field experiment was conducted at a private orchard in the Alhartha region – Basrah, Iraq (30°37'52.68"N and 47°45'8.15"E) during the 2024 growing season. Thirty-six uniform, average girth ± 10 cm, vigorous 3-4 years-old Barhee and Sayer date palm offshoots were used in the experiment. The selected offshoots were planted at 5 x 5m in silty clay loam soil, with an average soil EC of 12 dS m^{-1}, and the irrigation had an EC of approximately 4 dS m^{-1}. A drip irrigation system was then used for all treatments. On 1 March 2024, each treatment was replicated four times, with one offshoot per replicate. Each plant was treated with the following: Control (water spray), calcium chelate at 1000 ppm, potassium chelate at 1000 ppm, Salicylic acid at 1000 ppm, and citric acid at 1000 ppm. The average temperature in the field was 37ºC in May, 45ºC in July, and 35ºC in September. These averages were measured by the Hygro-thermometer at 11:00 A.M. on the day, with the air temperature corrected. Precipitation/Rainfall was 0 mm in all months of

the experiment sampling. After 6 months of treatment, the following data were recorded: leaf samples were collected for anatomical analysis. Including: Anatomical characteristics were Blade thickness, upper epidermis thickness (adaxial), lower epidermis thickness(abaxial), mesophyll thickness, cuticle thickness, vascular bundles diameter, tannin cells, and Stomatal density.

Anatomical studies

According to (Cárcamo et al., 2012), segments of the third leaflet and root of the offshoot were collected and immediately cut and fixed in formaldehyde, glacial acetic acid, and 70% ethanol (FAA) (5:5:90) for 48 h. Subsequently, they were dehydrated in an ascending series of alcohol (70-80-90-100%). After that, the case was infiltrated with 1:1 (xylene: alcohol 100%) overnight. Tissues embedded in paraffin. Finally, paraffin blocks were cut with a rotary microtome (MSE, mod., Germany). For light microscopy, semi-thin sections (10-12 µm) were fixed in a Myer's albumin solution. The samples were stained with safranin and fast green and examined with a photomicroscope (Olympus, mod.). Photographs, representative images, and average measurements were obtained using the quantitative anatomical data image analysis software Motic Images Edn-2 and J image.

Statistical analysis

Data were analyzed using SPSS software version 21.0 (SPSS Inc., Chicago, IL, USA) with an analysis of variance (ANOVA). Mean comparisons were performed using Tukey's pst hoc test at a significance level of $P \leq 0.05$.

Results

Leaf Blade and Mesophyll Thickness

Anatomical characteristics of date palm offshoots under severe salinity stress (soil EC: 14 dS m^{-1}, water EC: 4 dS m^{-1}) showed highly significant effects of treatment and cultivar on blade and mesophyll thickness (Table 1, Figs 2-5). For the Barhee cultivar, salicylic acid-treated plants formed a significantly thicker blade (395.83 µm) than all the other treatments (calcium chelate (354.25µm) > citric acid (331.50 µm) > potassium chelate (304.00µm) > control (278.83 µm), again with each treatment significantly different ($P < 0.05$). Similarly, the blade thickness of salicylic acid was again higher for the Sayer cultivar (375.00 µm, 32.7% higher than the control (282.50 µm). Responses to different treatments were calcium chelate 348.00 µm; citric acid,

potassium chelate. Significantly, Sayer consistently showed lower absolute thickness values than Barhee across all treatments.

Adaxial and Abaxial Surface Cuticle Thickness

While cuticle thickness had opposite tendencies in the two cultivars (Table 1), for Barhee, the upper (adaxial) cuticle was thickest in the control treatment (4.25µm) and significantly decreased with the foliar application of salicylic acid (1.50 µm, $P < 0.05$). In Sayer, conversely, the thickest upper cuticle was found in the control (5.00 µm), while salicylic acid produced the thinnest (2.00 µm), followed by intermediate values with calcium chelate and citric acid (3.75 µm) and potassium chelate (2.50 µm); lower (abaxial) cuticle exhibited a similar trend, the control (3.05 µm) and treatments (1.10µm). The lower cuticle in Sayer decreased from the control value of 3.80 µm to 1.20 µm with salicylic acid (all significantly different).

Table 1. The effect of calcium chelate, potassium chelate, salicylic acid, and citric acid at 1000 ppm on the thickness of the blade, mesophyll, and upper and lower cuticle thickness

Cultivars	Treatments (1000 ppm)	Blade (µm)	Mesophyll (µm)	Upper cuticle thickness (adaxial) (µm)	Lower cuticle thickness (abaxial) (µm)
Barhee	Control	298.83±4.49d	278.83±4.49e	4.25±0.95a	3.05±0.95a
	Calcium ch.	374.25±6.03b	354.25±6.03b	3.75±0.95ab	2.55±0.95ab
	Potassium ch.	324.00±5.43d	304.00±5.43d	2.50±0.57bcd	1.40±0.46b
	Salicylic acid	395.83±4.95a	375.83±4.95a	1.50±0.57d	1.10±0.11b
	Citric acid	351.50±10.90c	331.50±10.90c	3.75±0.95ab	2.55±0.95ab
Sayer	Control	282.50±8.41e	262.50±8.416e	5.00±0.81a	3.80±0.81a
	Calcium ch.	348.00±15.00c	328.00±15.00c	2.50±0.57bcd	1.40±0.46b
	Potassium ch.	301.25±16.39e	281.25±16.39e	3.50±0.57abc	2.30±0.57ab
	Salicylic acid	375.00±2.88b	355.00±2.88b	2.00±0.30cd	1.20±0.23b
	Citric acid	325.62±9.65e	305.62±9.65d	3.50±0.57abc	2.30±0.57ab

Mean ± standard deviation. Different letters indicate significant differences according to Tukey's test at the 0.05 level.

Diameter of the vascular bundle and vascular elements development

Furthermore, the treatment effects on vascular architecture were highly significant (Table 2). In Barhee, the effect of salicylic acid was significantly highest in terms of the greatest vascular bundle diameter (355.83 µm) compared with calcium chelate (334.25 µm), citric acid (311.50 µm), potassium chelate (284.00 µm), and control (258.83µm), showing an increase of 37.5% compared with control ($P < 0.05$). In Barhee, the xylem thickness ranged between 131.41µm (control) and 179.91µm (salicylic acid), and the thickness of the phloem increased from 80.70µm (control) to 104.95µm (salicylic acid). For Sayer, salicylic acid also had the largest vascular bundle diameter (335.00µm), followed by calcium chelate (308.00 µm), citric acid (285.62 µm), potassium chelate (261.25 µm), and control (242.50 µm). In Sayer, the increases in xylem and phloem tissues were proportional (salicylic acid: xylem 37.5% higher, phloem 30.2% higher compared to control, $P < 0.05$). **Among cultivars, Barhee exhibited larger vascular structures than Sayer under the same treatment.**

Upper and Lower Epidermis Thickness

Foliar application of antioxidant compounds significantly increased both upper and lower epidermis thickness in Barhee and Sayer cultivars compared to untreated controls under salinity stress (Fig. 1). Salicylic acid produced the highest increase, significantly surpassing all other treatments, followed by calcium chelate, citric acid, and potassium chelate in descending order of effectiveness. Statistical analysis revealed significant differences among treatments ($P \leq 0.05$), with salicylic acid demonstrating superior performance in enhancing epidermal tissue development. The Barhee cultivar exhibited significantly greater epidermal thickness than Sayer across all treatments.

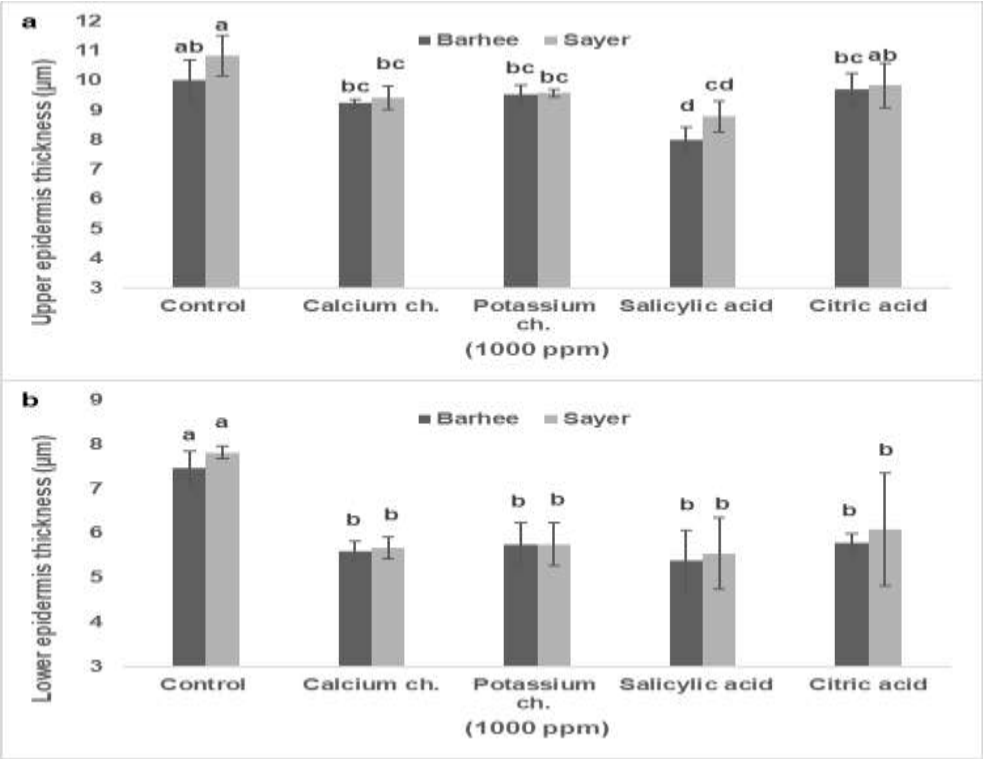

Fig 1. The influence of different treatments (calcium chelate, potassium chelate, salicylic acid, and citric acid at (1000 ppm) on the anatomical features of the Barhee and Sayer. offshoots leaf (Upper epidermis thickness and Lower epidermis thickness). Mean ± standard deviation. Letters indicate a significant difference based on Tukey's test at the 0.05 level in plants.

Table 2 Effect of calcium chelate, potassium chelate, salicylic acid, and citric acid at 1000 ppm on vascular bundle diameters, and Xylem and Phloem thickness in the Barhee and Sayer offshoots leaf

Cultivars	**Treatments (1000 ppm)**	**Vascular bundles diameter (µm)**	**Xylem thickness (µm)**	**Phloem thickness (µm)**
Barhee	Control	258.83±4.49e	131.41±2.24e	80.70±1.12e
	Calcium chelated	334.25±6.03b	169.12±3.01b	99.56±1.50b
	Potassium chelated	284.00±5.43d	144.00±2.71d	87.00±1.35d
	Salicylic acid	355.83±4.95a	179.91±2.47a	104.95±1.23a
	Citric acid	311.50±10.90c	157.75±5.45c	93.87±2.72c
Sayer	Control	242.50±8.41e	123.25±4.20e	76.62±2.10e
	Calcium chelated	308.00±15.00c	156.00±7.50c	93.00±3.75c
	Potassium chelated	261.25±16.39e	132.62±8.19e	81.31±4.09e
	Salicylic acid	335.00±2.88b	169.50±1.44b	99.75±0.72b
	Citric acid	285.62±9.65d	144.81±4.82d	87.40±2.41d

Mean ± standard deviation. Different letters indicate significant differences according to Tukey's test at the 0.05 level.

Density and distribution of tannin cells

Tannin cell density, a crucial indicator of antioxidant protection, fluctuated across treatments and cultivars (Table 3). In Barhee, the highest density (68.00 cells mm^{-2}) was achieved with salicylic acid treatment, followed by calcium chelate (62.50 cells mm^{-2}), citric acid (43.00 cells mm^{-2}), potassium chelate (44.75 cells mm^{-2}) and control (41.50 cells mm^{-2}), indicated for comparative control (Difference between salicylic acid and control; 63.9% increase, $P < 0.05$). Salicylic acid (58.50 cells mm^{-2}), calcium chelate (55.00 cells mm^{-2}), and other treatments (37.00 cells mm^{-2}) ($P < 0.05$). In Sayer, it was further noted that across all treatments, Barhee had a much higher density of tannin cells than Sayer, suggesting that Barhee had a better capacity to take up

phenolic compounds at high salinity. So was the spatial pattern of tannic cell distribution. The control treatments showed a localized distribution around vascular bundles in both cultivars, with a high level of scattered debris cells (45-47% of total and 25-30%, respectively). Salicylic acid, on the other hand, generated a comparative “denser, more uniform” 95% in Barhee and 92% in Sayer, with a broadly distributed mesophyll cell distribution (47%) suggesting a strong tissue-wide activation of antioxidant defenses. Calcium chelate also resulted in the "regular-dense" (80% homogeneity) distribution, as did potassium chelate and citric acid, which also exhibited a moderate tendency to enhancement (magnitude >50%). Statistical analysis confirmed that salicylic acid provided an "optimal distribution", calcium chelate an "excellent distribution", whereas other treatments provided "good" to "very good" distributions–all were significantly different from a "poor distribution" of control ($P < 0.05$).

Stomatal Density

For both cultivars, stomatal density were also significantly affected by treatment (Figures 6–7). In Barhee, control plants exhibited the highest stomatal density (150 stomata mm^{-2}), likely a stress-induced response. Salicylic acid treatment significantly reduced stomatal density to 120 stomata mm^{-2} ($P < 0.05$), followed by calcium chelate (130 stomata mm^{-2}), citric acid (135 stomata mm^{-2}), and potassium chelate (140 stomata mm^{-2}).

Similar trends were observed in Sayer, with the control showing elevated stomatal density, and treatments reducing this parameter in proportion to their antioxidant efficacy.

Table 3. Density, spatial distribution pattern, and degree of homogeneity of tannin cells in leaf tissues of Barhee and Sayer offshoots under severe salt stress by foliar spraying of antioxidant compounds.

Cultivars	Treatments (1000 ppm)	Density (cells mm^{-2})	Most Concentrated Area	Distribution Mode	Degree of homogeneity	Classification
Barhee	Control	41.50±1.29fg	About VB (45%)	dispersed	25%	Poor distribution
	Calcium chelated	62.50±1.29b	Mesophilic/VB (42% each)	Regular dense	80%	Excellent distribution
	Potassium chelated	44.75±2.50e	About VB (46%)	Moderate Regular	65%	Good distribution
	Salicylic acid	68.00±1.41a	Mesophilic (47%)	Comprehensive Dense	95%	Perfect distribution
	Citric acid	43.00±1.41ef	About VB (46%)	Regular-Dense	75%	Very good distribution
Sayer	Control	37.00±0.81h	About VB (47%)	dispersed	30%	Poor distribution
	Calcium chelated	55.00±1.82d	Mesophilic/VB (43%)	Regular dense	80%	Excellent distribution
	Potassium chelated	39.75±0.95gh	About VB (45%)	Mild	65%	Good distribution
	Salicylic acid	58.50±1.29c	Mesophilic (47%)	Very dense	92%	Perfect distribution
	Citric acid	39.00±0.81gh	About VB/Mesophile	Regular-Dense	75%	Very good distribution

Mean ± standard deviation. Different letters indicate significant differences according to Tukey's test at the 0.05 level.

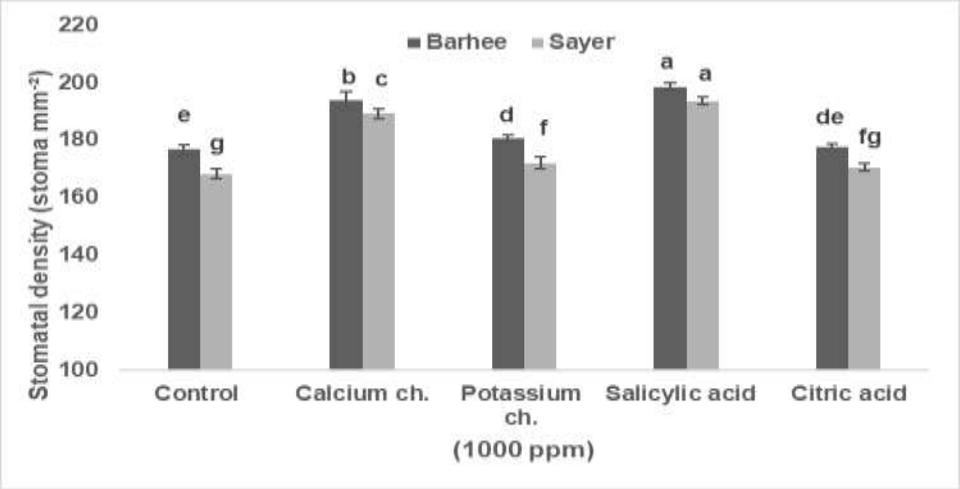

Fig 2. Stomatal density in Barhee and Sayer Date palm offshoots leaves under different treatments (control, calcium, potassium, salicylic acid, and citric acid). Mean ± standard deviation. Different letters indicate significant differences according to Tukey's test at the 0.05 level.

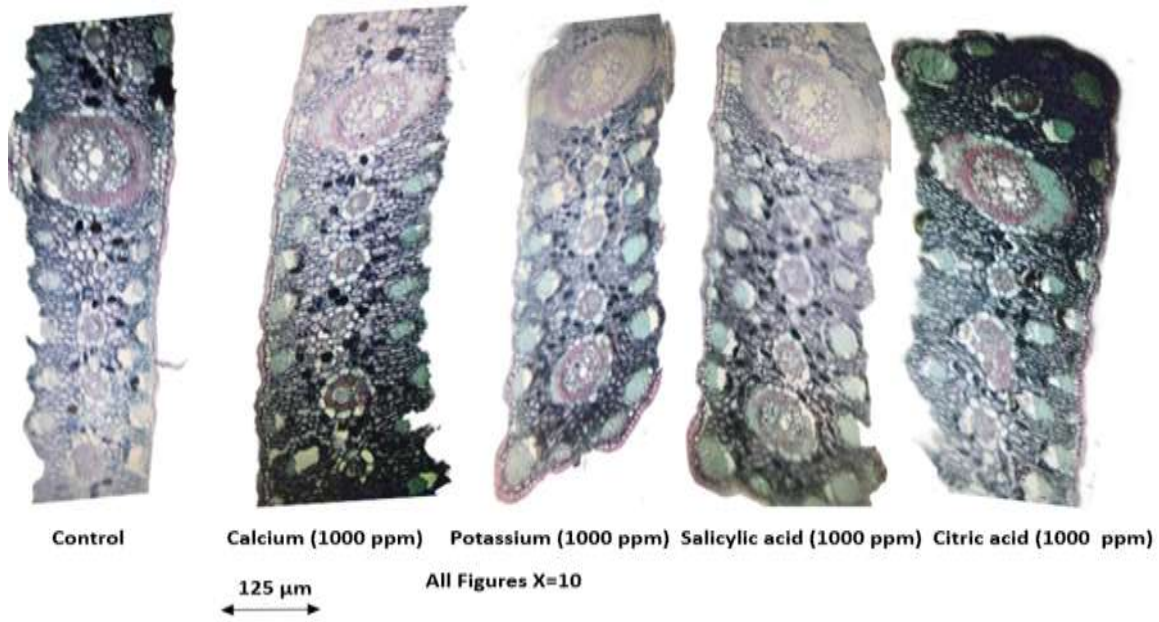

Fig 3. Cross-sections of Berhee offshoot pinnae under the effect of anti-salinity compounds and salt stress, Control, Calcium chelated, potassium chelated, Citric acid, and Salicylic acid.

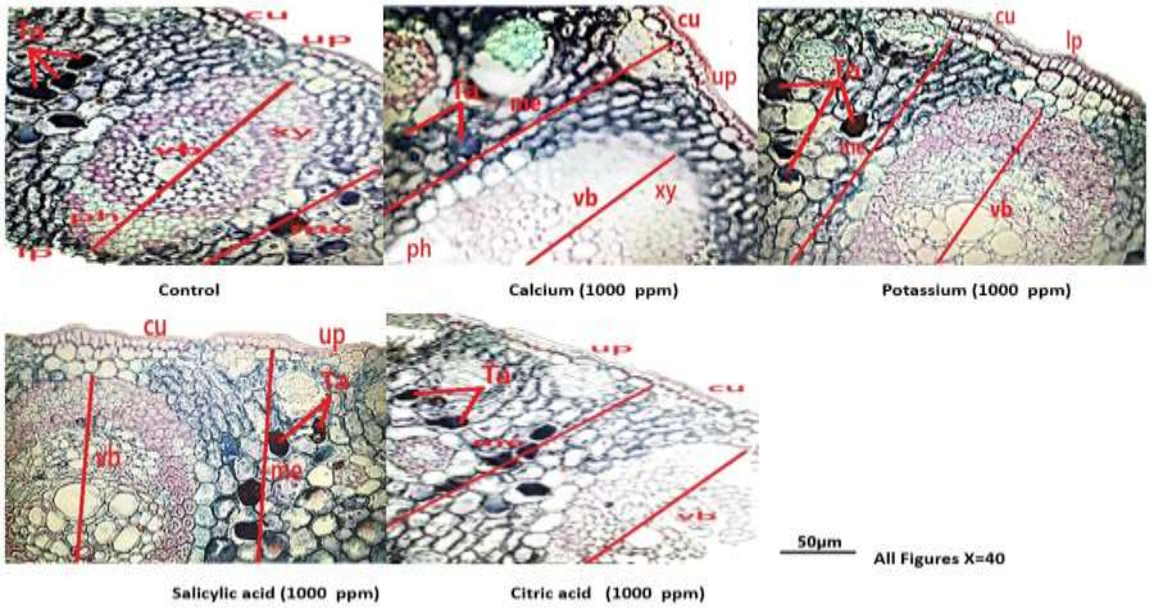

Fig 4. Cross-sections of Berhee offshoot pinnae under the effect of anti-salinity compounds and salt stress, Control, Calcium chelated, potassium chelated, Citric acid, and Salicylic acid. cu: cuticle, lp: lower epiderm, up: upper epiderm, me: mesophyll tissue, vb: vascular bundles, Ta: Tannin, Ph: Phloem, and xy: Xylem.

Fig 5. Cross-sections of Sayer offshoot pinnae under the effect of anti-salinity compounds and salt stress, Control, Calcium chelated, potassium chelated, Citric acid, and Salicylic acid.

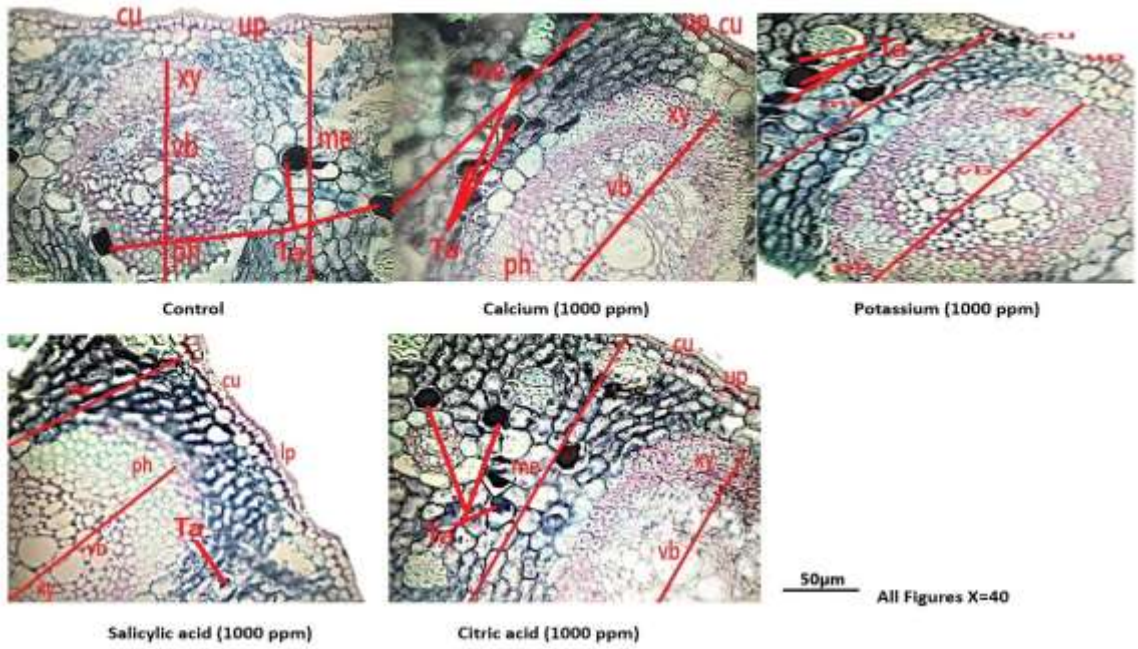

Fig 6. Cross-sections of Sayer offshoot pinnae under the effect of anti-salinity compounds and salt stress, Control, Calcium chelated, potassium chelated, Citric acid, and Salicylic acid. cu: cuticle, lp: lower epiderm, up: upper epiderm, me: mesophyll tissue, vb: vascular bundles, Ta: Tannin, Ph: Phloem, and xy: Xylem

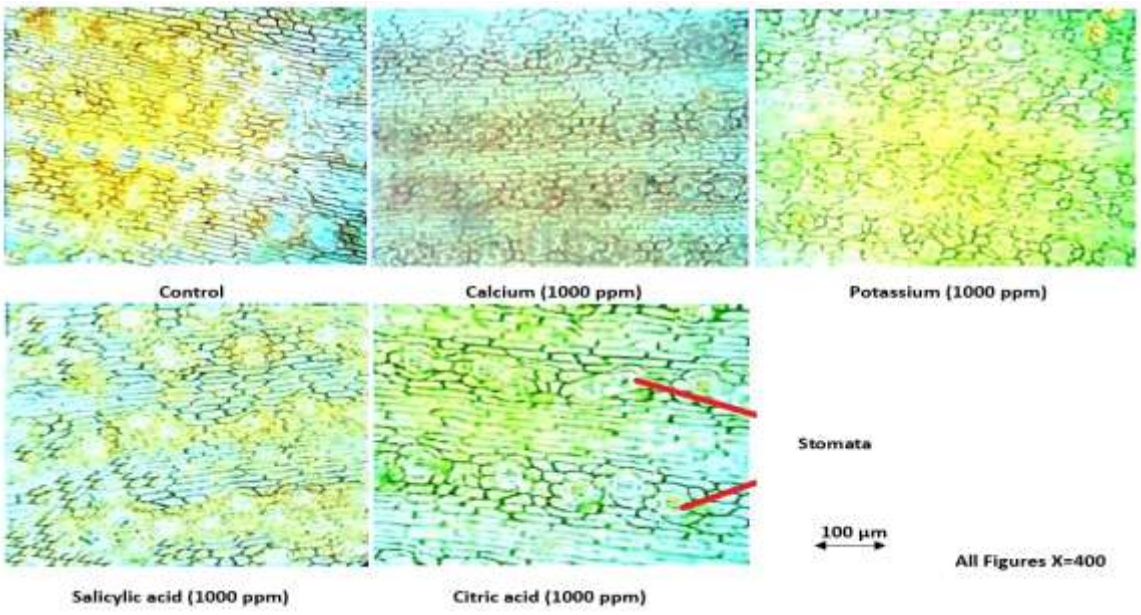

Fig 7. Stomata of Berhee offshoot leaflet (A lower surface of leaflet) under the effect of anti-salinity compounds and salt stress, Control, Calcium chelated, potassium chelated, Citric acid, and Salicylic acid.

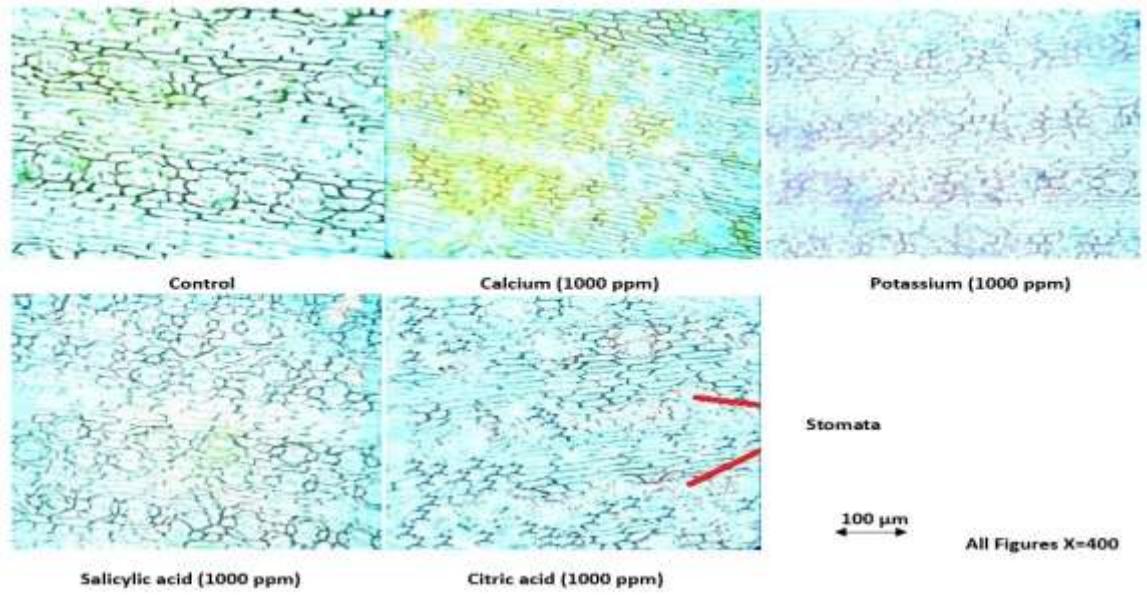

Fig 8. Stomata of Sayer offshoot leaflet (A lower surface of leaflet) under the effect of anti-salinity compounds and salt stress, Control, Calcium chelated, potassium chelated Citric acid, and Salicylic acid.

Discussion

The current results indicated that stages of salinity reactions changed by application antioxidant compounds (calcium chelated, potassium chelated, salicylic acid and citric acid) on anatomical properties of leaves of date palm offshoots under severe salt stress (EC soil: 14 dS m^{-1}, EC irrigation water: 4 dS m^{-1}). These findings are in line with current conceptual physiology of salt stress and the anatomical structure of plants, which allow structural defence against damage caused by toxic ions, oxidative stress, and salinity-mediated metabolic disturbances, acting through multi-mechanism protective compounds (Shareef et al., 2021; Liu et al., 2024). SA clearly outperformed the control treatment in both cultivars, increasing blade thickness and intermediate tissue by 32.5% and 32.7%, respectively, in Barhee and Sayer. These enhancements can be ascribed to SA's central position as a plant hormone and signalling molecule involved in the control of a wide variety of stress defence responses. Experiments show that, at the cellular level, SA activates the antioxidant enzyme system (SOD, CAT, APX), thereby reducing ROS levels and preventing oxidative damage to cell membranes, proteins, or nucleic acids (Rao et al., 2025). Increased blade thickness and intermediate tissue are indicative of cellular structural and cellular elongation of visceral parenchyma cells, likely due to lower oxidative damage and

improved ionic balance within the cells. Since median tissue is a photosynthetic source, its increase in clonal plants can enhance representation capacity and sustain photosynthetic efficiency under salt stress. The research indicates that mesenchymal tissues of salt-tolerant plants maintain potassium (K^+) levels without activating H^+-ATPase pumps, an energy-saving mechanism that helps sustain growth under stress (Atta et al., 2023). The next is calcium, which increases blade and mesophyll thickness. Calcium plays an important structural role, stabilizing plasma membranes and cellular organelles, reducing leakage of cellular contents, and maintaining cell integrity under stress conditions (Perez-rivera et al., 2025). Apart from its structural role, calcium also plays a signalling role in response to abiotic stress, including salinity tolerance pathways such as the SOS (Salt Overly Sensitive) pathway in plants (Bachani et al., 2022). Antioxidant compound treatments alleviated oxidative and ionic stress in plants and enabled them to form a thin but optimally functional cuticle. Furthermore, cuticle thickness is not always a good predictor of endurance; rather, the compromise in the cuticle's physicochemical properties (e.g., permeability, elasticity, chemical composition) is more crucial than absolute thickness (Skrzydeł et al., 2021). In this study, vascular bundle diameter and thickness of xylem and phloem tissues were significantly improved upon immediate application of SA and chelated calcium. This enhances the plant's anatomical features, which are essential for maintaining water and inorganic salts during salt stress. The vascular bundles are the equivalent of the plant's vital arteries, through which xylem conducts water and minerals from the roots to the aerial parts of the plant, and phloem transports photosynthetic products from the source leaves to the sinks during growth, development, and storage. Salt stress influences the capacity of vascular transport through decreases in water potential, increased viscosity of xylem sap owing to ion accumulation, and the formation of aerobic emboli in xylem vessels. The substantial increase in the thicknesses of the upper and lower epidermis in date palm apices after foliar application of antioxidative compounds, especially salicylic acid, is indicative of basic anatomical modifications that allow plants to preserve their structural and functional integrity under extreme salinity stress. The apparent decrease in epidermal thickness observed in untreated control plants under salinity is consistent with previous reports indicating that salt stress perturbs cell division and expansion in tissues such as the epidermis (Tariq et al., 2025). The limited epidermal formation under seawater salinity is part of a complex chain reaction that is initiated when the plant is subjected to abiotic stress, whereby water potential is compromised, leading to osmotic stress, ionic stress caused by excess Na^+ accumulation (toxicity), ultimately leading to reactive oxygen species accumulation

causing membrane damage and affecting organelles and affecting meristematic activity followed by tissue differentiation (Shahid et al., 2020). The better performance of salicylic acid in inducing epidermal tissue formation is, at least mechanistically, explained by the fact that salicylate functions as a signaling regulator of several physiological processes through the activation of antioxidant enzyme systems (i.e. superoxide dismutase, catalase, ascorbate peroxidase), the stimulation of osmolytes and compatible solutes as well as hetero-molecular roles of salicylate and signaling roles to modulate the expression of homeostasis regulating membrane transporters and stress-responsive genes as mechanisms to promote cellular protection and growth-restraining mechanisms at the same time (Shareef et al., 2022). Additionally, salicylic acid helps maintain cellular redox homeostasis, plasma membrane stability, and biosynthesis of plant cell wall components in the epidermis, thereby promoting cell division and expansion required for stress-induced tissue growth (Li et al., 2022). Density and distribution of tannin cells are among the most significant anatomical results reported in this study, and they exhibited remarkable qualitative and quantitative differences. Under SA treatment, the tannin cell density increased by 63.9% compared to the control, with the tannin pattern for the Barhee cultivar changing from "sparse" to "dense and uniform", with 95% homogeneity. These alterations indicate an evolutionarily evolved tissue-level response against salinity-induced oxidative stress. Tannin cells are specialized cells that store phenolic compounds that can bind and neutralize reactive oxygen species (ROS) and free radicals (Mora et al., 2022). Salt stress increases ROS (e.g., O_2^-, H_2O_2, OH^-) production by disrupting electron transport chains in chloroplasts and mitochondria (Shareef and Al-Khayri, 2021). If these reactive species are not neutralized, they will initiate lipid peroxidation, attack proteins, hydrolyze nucleic acids, and eventually induce apoptosis. The change in the spatial distribution of tannin cells — from preferentially around vascular bundles in the control to evenly distributed throughout the mesothelial tissue in SA treatment — also indicates a shift in defense strategy. In control plants subjected to stress, tannin is sequestered at vascular bundles to serve as a local line of defense against oxidative damage and toxic sap-borne ions in the biotransporter tissues. The widespread distribution of tannins in the intermediate tissue suggests a holistic histological defense in SA plants against oxidative damage to photosynthetic visceral cells. Intense tannin accumulations in mesenchymal cells were recently shown to play a role in the isolation of excess sodium ions (Na+) and to contribute to osmoregulation, thereby enhancing salinity tolerance in non-salt-secreting mangroves (Zhu et al., 2023). The results corroborated the notion that tannin cells constitute a double defence mechanism for date palm

against salt stress, acting as antioxidants and as isolated chambers for toxic ions. The density of stomata was highest for the control treatment, and this density was reduced by regulation. Our results are consistent with modern physiological knowledge of stomatal responses to salt stress, indicating that a decrease in stomatal density is a more permanent adaptation that minimizes water loss from transpiration while maintaining sufficient gas exchange for photosynthesis. The development and density of stomata are regulated by a complex genetic pathway that includes several key transcription factors, including SPEECHLESS (SPCH), the master regulator of stomatal development. Under water or salt stress, an increase in abscisic acid (ABA) levels activates SnRK2 kinases, which subsequently inhibit SPCH activity and reduce new stomatal formation (Song et al., 2022). Evidence for signaling crosstalk suggests that SA interacts with the ABA pathway in stomata regulation. Although this appears paradoxical in control plants, because high stomata density predicts a performance loss, this is an example of a disordered stress response in which the plant unsuccessfully attempts to compensate for the decreased efficiency of individual stomata (by increasing stomata density), resulting in excessive water loss. In contrast, SA plants showed lower stomatal density and sensitivity, which improved water-use efficiency and reduced photosynthesis inhibition. The responses of Barhee and Sayer cultivars to the applied antioxidants were significantly different, which was consistent with their evident anatomical differences. Barhee had the highest values for most anatomical traits (blade thickness, vascular bundle diameter, and tannin cell density) compared with Sayer, regardless of treatment. Such phylogenetic differences correspond to genetic differences in morphology and physiology as well as the genetic basis of salinity tolerance.

Conclusion

This study reinforces the hypothesis that bioactive compounds applied through foliage can stimulate positive anatomical modifications, ultimately enhancing the date palm's potential to adapt to extreme salinity or other stresses to which it is subjected, constituting a valuable step towards more sustainable date palm agriculture in these ever-salinizing agricultural landscapes.

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المركبات المضادة للأكسدة تعزز الخصائص التشريحية لأوراق فسائل نخيل التمر (*Phoenix dactylifera* L.) تحت إجهاد الملوحة

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الخلاصة

أُجريت هذه الدراسة لمعرفة التأثيرات الوقائية للرش الورقي بمركبات مضادة للأكسدة التي شملت مخلِب الكالسيوم ومخلِب البوتاسيوم وحامض الساليسيليك وحامض الستريك (1000 جزء في المليون لكل منها) على الخصائص التشريحية لأوراق فسائل نخيل التمر أثناء تعرضها لإجهاد ملوحة ضار (التوصيل الكهربائي للتربة: 14 ديسيمنز م$^{-1}$؛ ولمياه الري: 4 ديسيمنز م$^{-1}$)، لكلا صنفي البرحي والساير. أدت معالجات مضادات الأكسدة إلى زيادة ملحوظة في المعايير التشريحية المتتالية. وقد حقق حامض الساليسيليك أهم التحسينات في جميع المعايير المقاسة تقريبًا. وقيِم تأثير معالجة حامض الساليسيليك على المعايير الهيكلية للأوراق في صنفين بصريًا للاختلافات في سمك النصل، وقطر الحزمة الوعائية، وسمك الخشب، وسمك اللحاء، وسمك البشرة وكثافة خلايا التانين في كل صنف. أدى العلاج بحامض الساليسيليك إلى زيادة سمك النصل، وقطر الحزمة الوعائية، وسمك الخشب، وسمك اللحاء، وسمك البشرة وكثافة خلايا التانين مقارنة بالشاهد غير المعالج في كلا الصنفين. ويظهر انخفاض كثافة الثغور نتيجة للمعالجات بمضادات الأكسدة فائدةً في كفاءة استخدام المياه خلال ظروف الإجهاد. وقد دلت الاستجابات التشريحية المتفوقة باستمرار لصنف البرحي مقارنة بصنف الساير تحت كل معالجة على التباين الجيني في تحمل الإجهاد. وتزيد هذه التعديلات الهيكلية المتأثرة بفعالية جزيئات مضادات الأكسدة الخارجية في تخفيف التغيرات الهيكلية والاختلالات الفسيولوجية الناجمة عن الملوحة في فسائل نخيل التمر النامية.

الكلمات المفتاحية: المركبات المضادة للأكسدة، الحزم الوعائية، خلايا التانين، إجهاد الملوحة، وكثافة الثغور.