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Comparative Effects of Nano-Silicon Application Methods on Salt Stress Tolerance in Lettuce (*Lactuca sativa* L.)*

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Abstract: The aim of this study was to evaluate the effectiveness of different doses and application methods of nano-silicon (Nano-Si) in enhancing salt tolerance in lettuce (*Lactuca sativa* L.) and to determine the most effective treatment under saline conditions. The curly lettuce cultivar Caipira (Şakira) (Batavia type) was used as the plant material. Salt stress was imposed by applying 100 mM NaCl 20 d after transplantation. Nano-Si was applied weekly at doses of 0, 0.5, 1.0, 2.0, and 3.0 mM using two different methods, foliar spraying and root drenching. At harvest, plants were evaluated for shoot fresh and dry weights, plant height, plant circumference, crown width, root collar diameter, leaf area, total yield, relative water content (RWC), shoot Na^+, Cl^-, K^+, and Ca^{2+} concentrations, malondialdehyde (MDA), total chlorophyll, and carotenoid contents. Salt stress significantly reduced growth parameters, total yield, RWC, K^+ and Ca^{2+} concentrations, and photosynthetic pigment content, whereas it increased Na^+ and Cl^- accumulation and MDA levels. In contrast, Nano-Si applications markedly improved plant performance under salt stress. Compared with salt stress alone, Nano-Si treatments increased fresh and dry weights by 37-286%, plant height and root collar diameter by 5-46%, plant circumference and crown width by 6-91%, leaf area by 100-271%, total yield by 74-219%, RWC by 16-56%, K^+ content by 24-57%, Ca^{2+} content by 56-164%, and photosynthetic pigments by 15-126%. Furthermore, Nano-Si applications reduced Na^+ uptake by 13-47% and Cl^- uptake by 1-40%, while decreasing MDA accumulation by 5-55%. Overall, Nano-Si effectively mitigated the detrimental effects of salt stress and enhanced the salt tolerance of lettuce. Among the treatments, foliar applications of 0.5 and 1.0 mM Nano-Si were the most effective and can be recommended as a promising strategy for improving lettuce performance under saline conditions.

Keywords: Foliar application, ion homeostasis, lipid peroxidation, relative water content, root drench, sodium chloride

1. Introduction

The rapid increase in the global population is intensifying the demand for food, with projections indicating a population of nearly 10 billion by 2050 and a corresponding 50% rise in food requirements (Tilman and Clark, 2014). However, climate change and environmental degradation increasingly constrain agricultural productivity, making it essential to improve efficiency within existing production systems. Among the major limiting factors, abiotic stresses such as salinity, drought, and high temperatures significantly threaten crop yield, quality, and global food security (Zhao et al., 2021).

Salinity is one of the most critical abiotic stressors, affecting approximately 20% of cultivated land worldwide. It disrupts plant growth and development by inducing osmotic and ionic stress, leading to nutrient imbalances and reduced photosynthetic activity. Excessive accumulation of sodium (Na^+) and chloride (Cl^-) ions interferes with the uptake of essential nutrients such as potassium (K^+) and calcium (Ca^{2+}), resulting in metabolic dysfunction and impaired cellular processes (Zhao et al., 2021). Furthermore, salinity induces oxidative stress through the overproduction of reactive oxygen species (ROS), which damage cellular components including

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lipids, proteins, and nucleic acids. Although plants possess enzymatic and non-enzymatic antioxidant defense systems, severe stress can overwhelm these protective mechanisms (Parihar et al., 2015).

Therefore, enhancing plant tolerance to salinity has become a key objective in sustainable agriculture (Van Zelm et al., 2020; Gong, 2021). In recent years, nanotechnology-based approaches have emerged as promising tools for improving plant performance under stress conditions. Nanoparticles, owing to their small size (1-100 nm) and high reactivity, can enhance nutrient use efficiency, regulate physiological and biochemical processes, and mitigate stress-induced damage. In particular, nanofertilizers have gained attention for their ability to improve crop productivity while reducing the environmental impacts associated with excessive fertilizer use (Derosa et al., 2010; Siddiqui et al., 2015; Saxena et al., 2016).

Silicon (Si), although not classified as an essential nutrient, is widely recognized as a beneficial element that enhances plant tolerance to various abiotic stresses, including salinity stress. It plays a crucial role in maintaining cell structure, improving ion balance, and enhancing physiological and biochemical stability under stress. Numerous studies have demonstrated that Si application can improve plant growth, yield, and stress resilience across different crop species (Özyazıcı, 2021, 2022; El-Ramady et al., 2022). Lettuce (*Lactuca sativa* L.) is a widely consumed leafy vegetable that is rich in vitamins, antioxidants, and bioactive compounds beneficial to human health. However, it is relatively sensitive to salinity, with adverse effects on germination, biomass accumulation, and overall plant development. Therefore, improving salt tolerance in lettuce is essential for sustaining its productivity under saline conditions (Hung et al., 2004; Barassi et al., 2006; Xu and Mou, 2015).

This study aimed to evaluate the effectiveness of nano-silicon (Nano-Si) application in improving the salt tolerance of lettuce (*L. sativa*). Specifically, the effects of different Nano-Si doses and application methods (foliar spray and root application) were investigated in terms of plant growth, yield, ion regulation, and physiological responses to salinity stress.

2. Materials and Methods

2.1. Plant material and stress treatment

Caipira (Şakira) lettuce (Batavia type) was used as the plant material. Seedlings were transplanted into 11-L plastic pots (57×16×12 cm) filled with a peat:perlite (2:1, v/v) substrate, with three plants per pot. The experiment was arranged in a completely randomized design with three replicates, and plants were irrigated with a standard nutrient solution from transplanting until the initiation of the treatments. Salt stress was imposed 20 days after transplanting by applying either 0 (control) or 100 mM NaCl. Nano-silicon was applied at concentrations of 0, 0.5, 1.0, 2.0, and 3.0 mM using either foliar spray or root drench methods. Nano-Si (particle size: 10-20 nm; purity: 99.5%; trace metals basis) was purchased from Sigma-Aldrich (St. Louis, MO, USA). Nano-Si treatments were initiated simultaneously with the onset of salt stress and repeated at 7-day intervals throughout the experimental period. Control plants received only the standard nutrient solution (Kusvuran and Dikmen Yılmaz, 2023). Plants were harvested seventeen days after the initiation of salt stress for morphological, physiological, and biochemical analyses. Plants were harvested 37 days after transplanting, which corresponds to the commercial harvest stage of the Caipira (Şakira) lettuce cultivar under greenhouse conditions. Accordingly, the fresh weight of the marketable aboveground biomass was used to calculate total yield (kg m^{-2}).

2.2. Assessment of growth, yield, and physiological parameters

In the lettuce experiment, plants were harvested 17 days after stress imposition. The fresh weights of the aboveground parts (shoots) were measured using a precision balance. Subsequently, the samples were oven-dried at 65 °C for 48 h to determine dry weights. Total yield (kg m^{-2}) was calculated from the marketable fresh weight of lettuce plants harvested from each experimental unit and expressed on an area basis according to the planting density. Plant height, plant circumference, and crown width were measured in centimeters (±0.5 cm) using a tape measure. Root collar diameter, defined as the stem thickness just above the root zone, was measured using a digital caliper and expressed in mm. Leaf area was determined using a CI-202 portable leaf area meter (CI BioScience, USA) and expressed as cm^2 $plant^{-1}$. The relative water content (RWC, %) was determined according to the methods described by Sanchez et al. (2004) and Türkan et al. (2005). For this purpose, the fresh weights of the leaf samples were recorded immediately after harvest. The samples were then dried at 65 °C for 48 h, and the dry weights were measured to calculate RWC.

2.3. Determination of lipid peroxidation level

Lipid peroxidation was determined by the measuring malondialdehyde (MDA) content, as described by Lutts et al. (1996). Briefly, 200 mg of fresh leaf tissue was homogenized in 5 mL of 0.1% (w/v) trichloroacetic acid (TCA) and centrifuged at 12.500 rpm for 20 min. From the obtained extract, 3 mL of the supernatant was mixed with 3 mL of 0.1% (w/v) thiobarbituric acid (TBA) prepared in 20% TCA. The reaction mixture was incubated in a water bath at 95 °C for 30 min and then cooled to room temperature. The absorbance was measured at 532 and 600 nm using a spectrophotometer.

2.4. Determination of chlorophyll and carotenoid contents

Photosynthetic pigment content was determined according to Kusvuran et al. (2021). Leaf samples (100-200 mg) were homogenized in 15 mL of 80% (v/v) acetone and filtered through Whatman No. 1 filter paper. The absorbance of the extracts was measured using a UV-Vis spectrophotometer at 652 nm for total chlorophyll, and 470 nm for carotenoids.

2.5. Determination of ion contents

The ion contents (Na^+, K^+, Ca^{2+}, and Cl^-) were determined following the method described by Dasgan and Koc (2009). The chloride (Cl^-) concentration in aboveground plant tissues (shoots) was determined according to Nielsen (2017) using the Mohr titration method.

2.6. Statistical analysis

The experiment was conducted using a completely randomized design with three replicates, each consisting of three plants. Statistical analyses were performed using the JMP software (version 8.0). Differences among treatment means were evaluated using the least significant difference (LSD) test at $p \leq 0.05$.

3. Results

3.1. Fresh and dry weights of plants

Nano-silicon application improved fresh and dry weights in control plants, with increases ranging from 4-34% for fresh weight and 3-49% for dry weight. Notably, the foliar application of 0.5 mM Nano-Si resulted in the highest increases (34% and 49%, respectively). In contrast, 100 mM NaCl treatment significantly reduced both fresh and dry weights in lettuce, with decreases of 74% and 72%, respectively, compared to control plants. The interaction between salinity and Nano-Si application significantly affected shoot fresh and dry weights ($p \leq 0.05$). Salt stress markedly reduced biomass accumulation, whereas Nano-Si applications significantly alleviated these adverse effects, particularly under foliar application. Under salt stress conditions, Nano-Si application markedly alleviated these reductions, resulting in overall improvements of up to 198% in fresh weight and 156% in dry weight compared with salt-stressed plants. Among the treatments, 0.5 and 1 mM Nano-Si were the most effective, showing the least reduction relative to control plants (1-23%) and substantial recovery (167-286%). Regarding application methods, foliar application was more effective than root application. Under salt stress, foliar treatments resulted in only a 12% reduction in fresh weight (corresponding to a 234% improvement), whereas root application led to a 31% reduction (corresponding to a 161% improvement). Similarly, dry weight decreased by 14% (200% improvement) under foliar application and by 38% (117% improvement) under root application compared to control plants (Figure 1).

3.2. Plant height

Nano-silicon application enhanced plant height in control plants, with increases ranging from 1-25%. The most pronounced effect was observed with foliar application of 0.5 mM Nano-Si, which resulted in a 25% increase. Plant height was significantly influenced by the interaction between salinity and Nano-Si treatments ($p \leq 0.05$). Salt stress significantly reduced the plant height in lettuce, with a decrease of approximately 40% compared to that in control plants. However, Nano-Si application mitigated the adverse effects of salinity. Under salt stress conditions, plant height decreased by an average of 27% relative to that of control plants, whereas Nano-Si treatments provided an overall improvement of approximately 20%. Among the treatments, 0.5 and 1 mM Nano-Si were the most effective, showing the least reduction (19-28%) and the highest recovery (19-34%) under salt stress. When the application methods were compared, foliar application was found to be more effective than the root application. Foliar treatments resulted in an average reduction of 19% (corresponding to a 33% improvement), whereas root application led to a 30% reduction in plant height compared to that in control plants (Figure 2).

3.3. Plant circumference and crown width

Nano-silicon applications increased plant circumference and crown width in control plants, with improvements of 5-22% and 3-15%, respectively. The most pronounced effect was observed with the foliar application of 0.5 mM

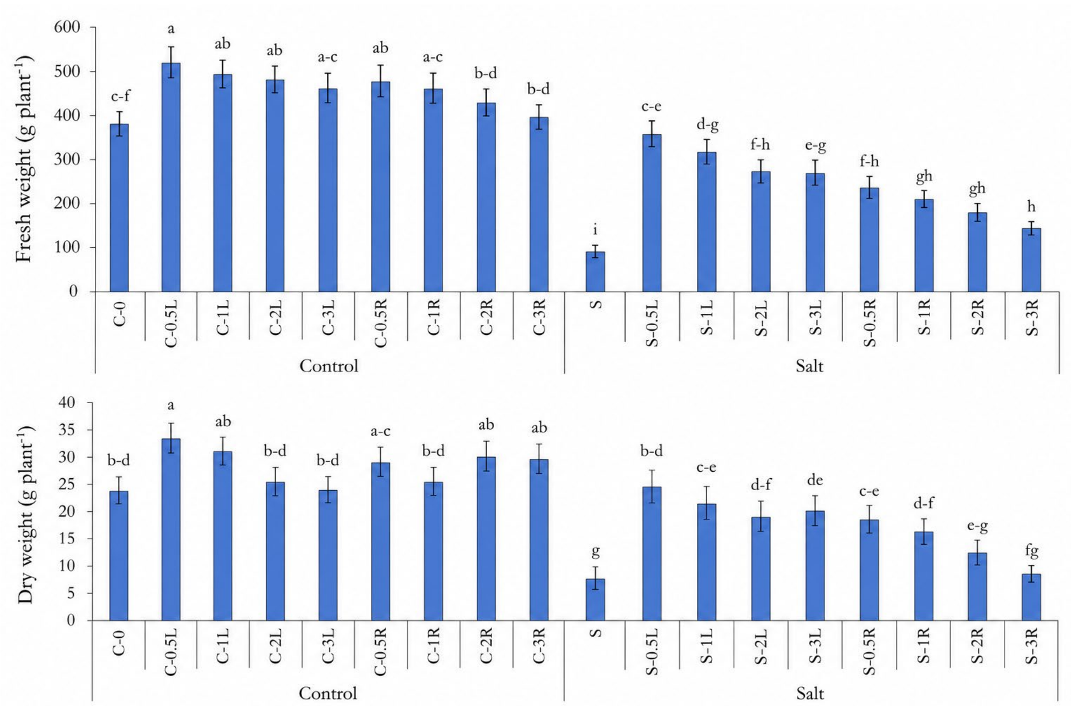

Figure 1. Effect of Nano-Si applications on fresh and dry weight of lettuce under salt stress conditions*
C: Control, S: Salt, R: Root, L: Leaf application, *: There is no significant difference at the 0.05 level between the means denoted by the same letter within the same group

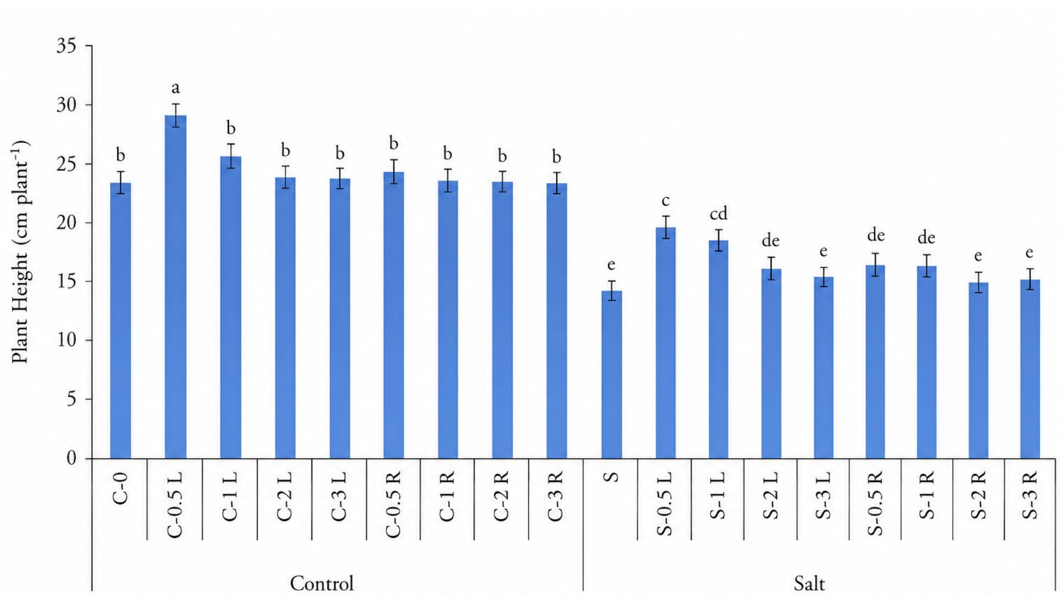

Figure 2. Effect of Nano-Si applications on plant height of lettuce under salt stress conditions*
C: Control, S: Salt, R: Root, L: Leaf application, *: There is no significant difference at the 0.05 level between the means denoted by the same letter within the same group

Nano-Si, resulting in increases of 22% (circumference) and 15% (crown width). The interaction between salinity and Nano-Si treatments significantly affected both plant circumference and crown width ($p \leq 0.05$). Salt stress (100 mM NaCl) significantly reduced both parameters, with decreases of 46% (20.20 cm) in plant circumference and 57% (15.33 cm) in crown width compared with control plants. However, Nano-Si treatment alleviated these adverse effects. Under salt stress, reductions averaged 31% for circumference and 35% for crown width relative to control plants, whereas Nano-Si applications provided overall improvements of 30% and 51%, respectively. Among the treatments, 0.5 and 1 mM Nano-Si were the most effective, showing the least reductions (9-32%) and the highest recovery (27-91%) under salinity. In terms of application methods, foliar application was more effective than root applications. For plant circumference, foliar treatment resulted in an average reduction of 15% (59% improvement), whereas root application caused a 28% reduction (34% improvement). Similarly, crown width decreased by 20% (86% improvement) under foliar application and by 38% (45% improvement) under root application compared to control plants (Figure 3).

3.4. Root collar diameter

Nano-silicon application increased root collar diameter in control plants by an average of 9%, with the highest increase (24%) observed with 0.5 mM foliar application. Root collar diameter was significantly affected by the interaction between salinity and Nano-Si application ($p \leq 0.05$). Salt stress (100 mM NaCl) significantly reduced the root collar diameter by 36% compared to that in control plants. However, Nano-Si treatment mitigated these negative effects. Under salt stress, root collar diameter decreased by an average of 18% relative to control plants, whereas Nano-Si application provided an overall improvement of 28%. Among the treatments, 0.5 and 1 mM Nano-Si were the most effective, showing the least reduction (6-15%) and the highest recovery (31-46%). Foliar application was more effective than root application, resulting in an average reduction of 8% (43% improvement), whereas root application led to a 15% reduction (33% improvement) compared with the control plants (Figure 4).

3.5. Leaf area

Nano-silicon application increased the leaf area of control plants by an average of 13%, with the highest increase (30%) observed with a 0.5 mM foliar application. Salt stress significantly reduced leaf area by 77% compared to that in control plants. Leaf area was significantly influenced by the interaction between salinity and Nano-Si treatments ($p \leq 0.05$). However, Nano-Si treatment alleviated these adverse effects, limiting the reduction to 38% and providing an overall improvement of 163% under salt stress conditions. Among the treatments, 0.5 and 1 mM Nano-Si were the most effective, showing the least reduction (13-43%) and the greatest recovery (140-271%). Foliar application was more effective than root application, resulting in an average reduction of 23% (230% improvement), whereas root application led to a 42% reduction (146% improvement) compared with control plants (Figure 5).

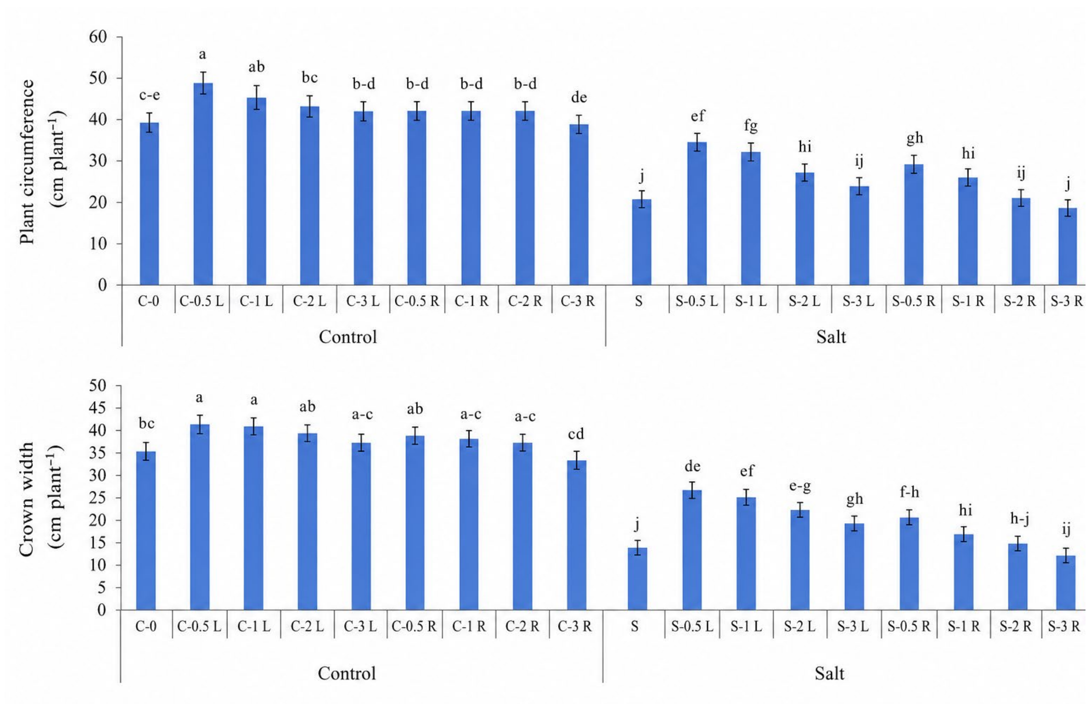

Figure 3. Effect of Nano-Si applications on plant circumference and crown width of lettuce under salt stress conditions*

C: Control, S: Salt, R: Root, L: Leaf application, *: There is no significant difference at the 0.05 level between the means denoted by the same letter within the same group

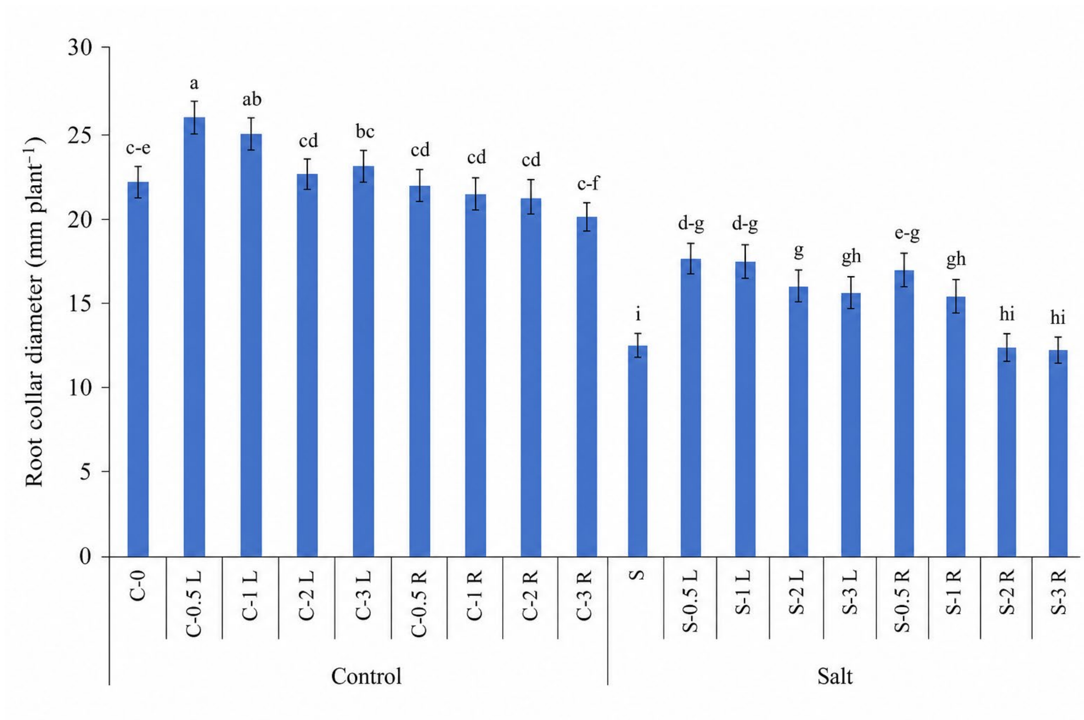

Figure 4. Effect of Nano-Si applications on root collar diameter under salt stress conditions*

C: Control, S: Salt, R: Root, L: Leaf application, *: There is no significant difference at the 0.05 level between the means denoted by the same letter within the same group

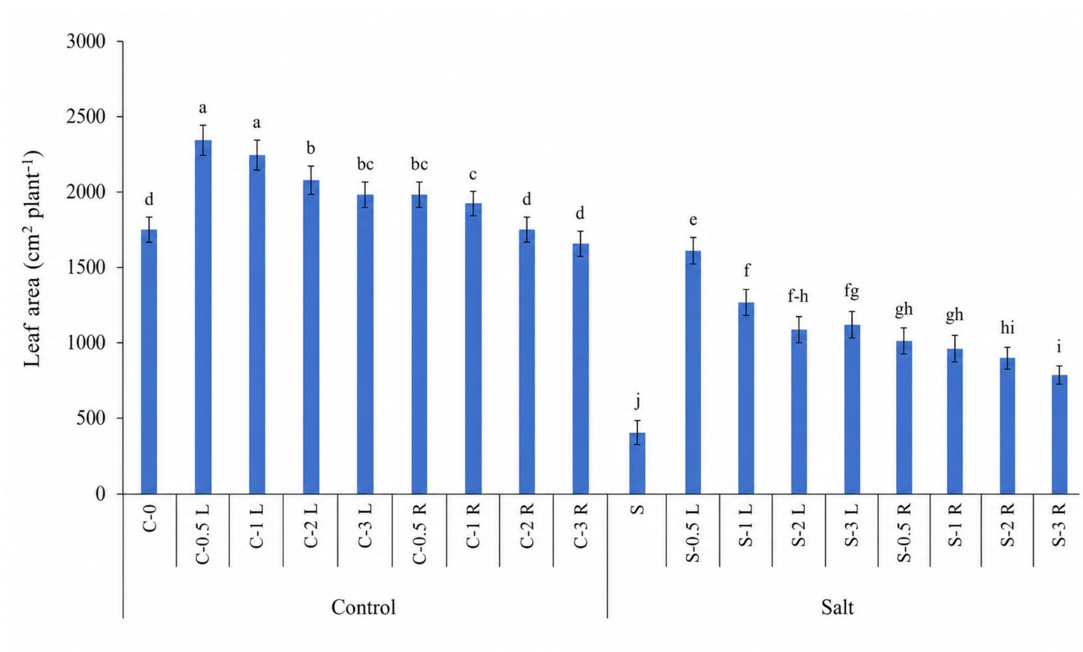

Figure 5. Effect of Nano-Si applications on leaf area under salt stress conditions*

C: Control, S: Salt, R: Root, L: Leaf application, *: There is no significant difference at the 0.05 level between the means denoted by the same letter within the same group

3.6. Relative water content

Nano-silicon application slightly increased leaf RWC in control plants by an average of 2%, with the highest increase (4%) observed with the 0.5 mM foliar application. Salt stress reduced the RWC by 40% compared to that in control plants. Relative water content was significantly affected by the interaction between salinity and Nano-Si application ($p \leq 0.05$). However, Nano-Si treatments mitigated this effect, limiting the reduction to 16% and providing an overall improvement of 40% under salt stress conditions. The most effective treatments were 0.5 and 1 mM Nano-Si, which showed the lowest reduction (6–18%) and the highest recovery (36-56%). Foliar application was more effective than root application, resulting in a 12% reduction (46% improvement), whereas root application caused a 20% reduction (34% improvement) compared to control plants (Figure 6).

3.7. Total yield

Nano-silicon application increased the total yield in control plants by an average of 13%, with the highest increase (32%; 17.46 kg m^{-2}) observed with the 0.5 mM foliar application. Salt stress significantly reduced the total yield by 68% (4.27 kg m^{-2}) compared to that of the control plants. The interaction between salinity and Nano-Si treatments had a significant effect on total yield ($p \leq 0.05$). However, Nano-Si treatment mitigated this effect, limiting the reduction to 19% and providing an overall improvement of 150% under salt stress conditions. The most effective treatments were 0.5 and 1 mM Nano-Si, showing only an 11% reduction and achieving a 173% improvement. Foliar application was more effective than root application, resulting in a 10% reduction (179% improvement), whereas root application led to a 28% reduction (123% improvement) compared with the control (Figure 7).

3.8. Lipid peroxidation

Salt stress significantly increased the MDA content to 11.51 µmol g^{-1} fresh weight (336% increase). Nano-Si application significantly alleviated salt-induced lipid peroxidation, resulting in an overall 30% reduction in MDA accumulation compared with salt-stressed plants (Figure 8). MDA content was significantly influenced by the interaction between salinity and Nano-Si treatments ($p \leq 0.05$). The lowest MDA content under salt stress was observed with 0.5 mM foliar Nano-Si application (5.14 µmol g^{-1} fresh weight), corresponding to a 94% reduction and 55% improvement compared to that of the control plants. Among application methods, foliar treatment was more effective, resulting in a 159% increase (40% decrease), whereas root application led to a 257% increase (18% decrease) (Figure 8).

3.9. Total chlorophyll and carotenoid content

Nano-Si application increased total chlorophyll and carotenoid contents in control plants by 4% and 18%, respectively, with the highest increases (7% and 37%) observed with 0.5 mM foliar application. Total chlorophyll and carotenoid contents were significantly affected by the interaction between salinity and Nano-Si application ($p \leq 0.05$). Salt stress significantly reduced the total chlorophyll (35%) and carotenoid content (56%) compared to that in the control plants. However, Nano-Si treatments mitigated

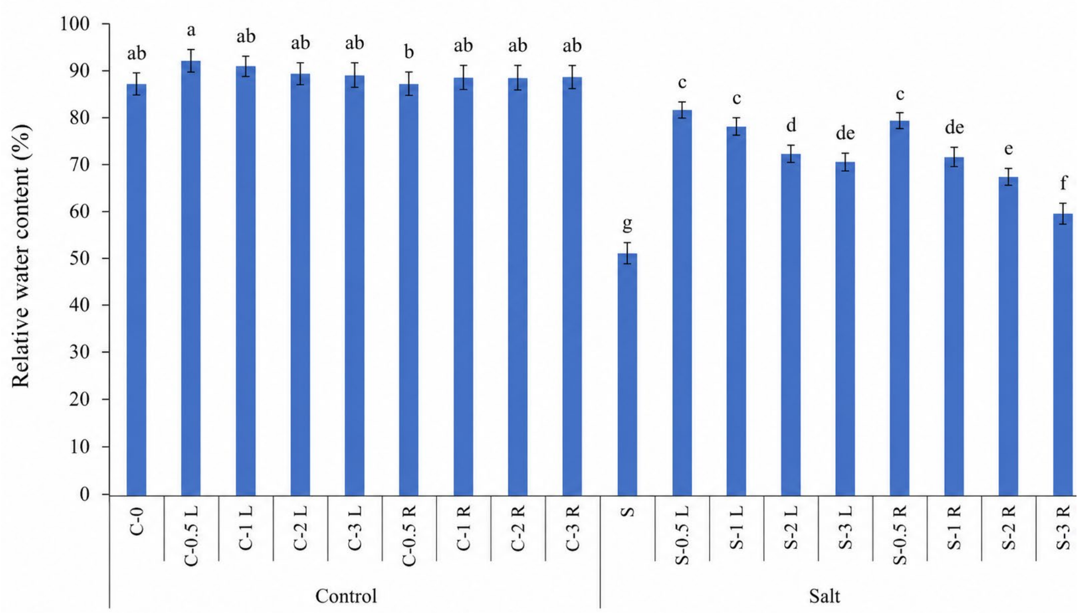

Figure 6. Effect of Nano-Si applications on RWC under salt stress conditions*

C: Control, S: Salt, R: Root, L: Leaf application, *: There is no significant difference at the 0.05 level between the means denoted by the same letter within the same group

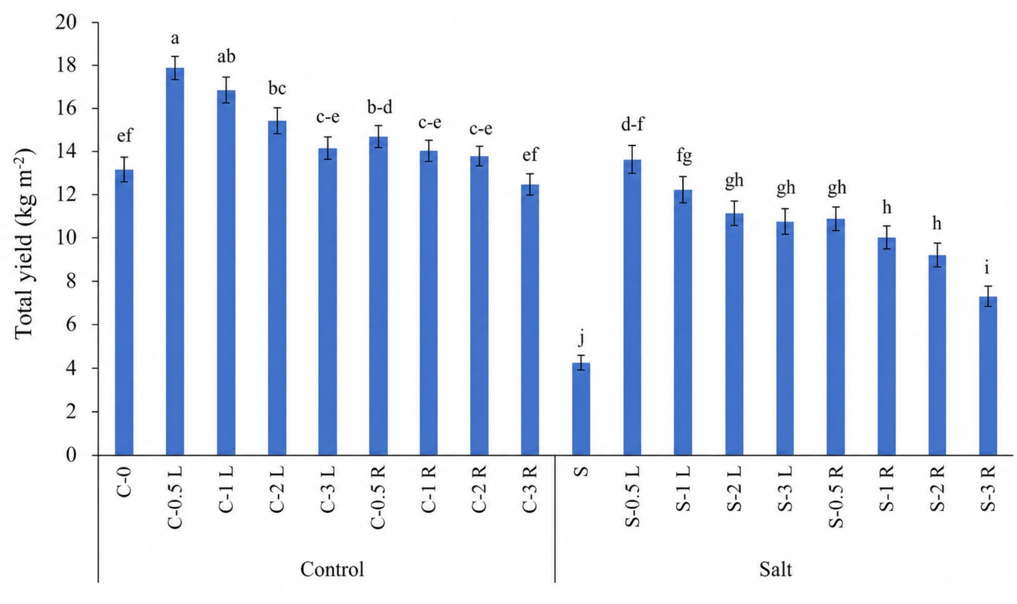

Figure 7. Effect of Nano-Si applications on total yield under salt stress conditions*

C: Control, S: Salt, R: Root, L: Leaf application, *: There is no significant difference at the 0.05 level between the means denoted by the same letter within the same group

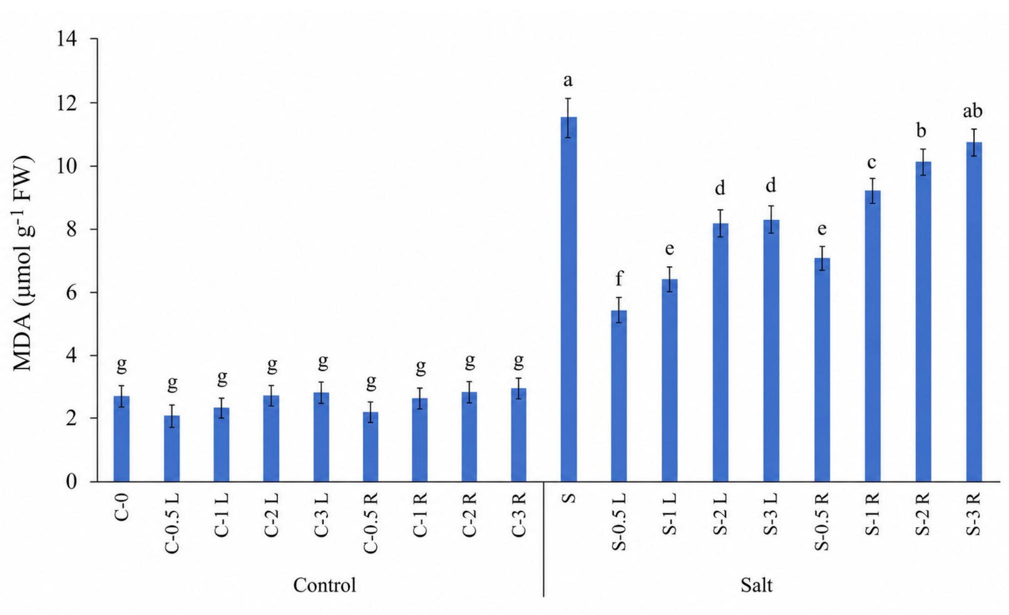

Figure 8. Effect of Nano-Si applications on MDA content under salt stress conditions*

C: Control, S: Salt, R: Root, L: Leaf application, *: There is no significant difference at the 0.05 level between the means denoted by the same letter within the same group

these effects, limiting the reductions to 17% and 35%, and providing improvements of 26 and 60%, respectively. The most effective treatments were 0.5 and 1 mM Nano-Si, which showed minimal reductions (~15%) and improvements ranging from 20% to 126%. Foliar application was more effective than root application, resulting in lower reductions in both chlorophyll (11% vs. 24%) and carotenoid contents (21% vs. 39%), and greater recovery under salt stress conditions (Figure 9).

3.10. Ion contents

Nano-Si application decreased Na^+ (9%) and Cl^- (17%) contents while increasing K^+ and Ca^{2+} levels (both ~12%) in the control plants. Under salt stress (100 mM NaCl), the Na^+ and Cl^- contents increased dramatically by 1512% and 1114%, respectively, whereas the K^+ and Ca^{2+} contents decreased by 36% and 64%, respectively. The interaction between salinity and Nano-Si treatments significantly affected Na^+, Cl^-, K^+, and

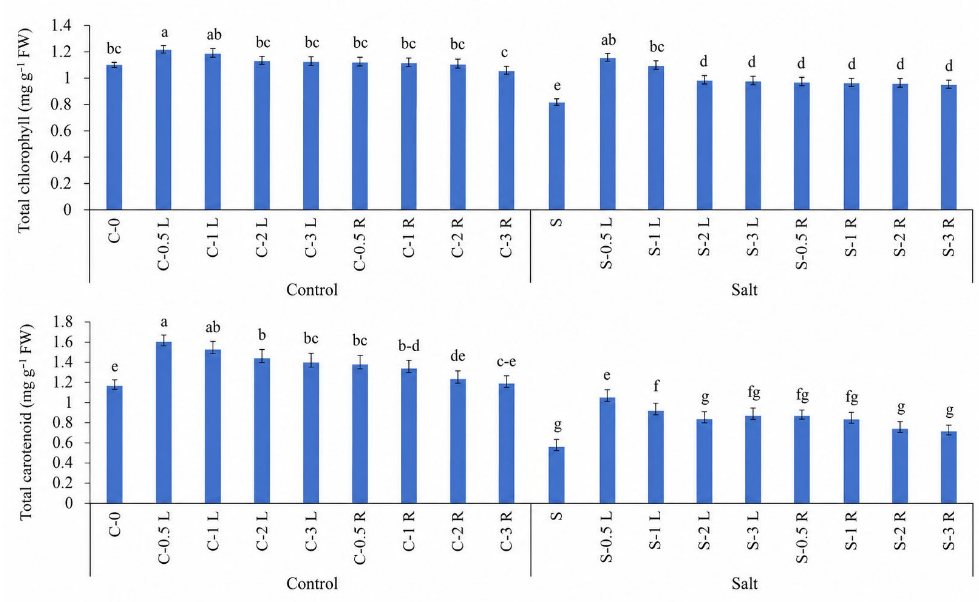

Figure 9. Effect of Nano-Si applications on total chlorophyll and carotenoid content under salt stress conditions*

C: Control, S: Salt, R: Root, L: Leaf application, *: There is no significant difference at the 0.05 level between the means denoted by the same letter within the same group

Ca^{2+} concentrations ($p \leq 0.05$). Nano-Si treatments mitigated these effects, limiting Na^+ and Cl^- accumulation (3-48% reduction) and preserving K^+ and Ca^{2+} levels (24-164% increase). Among treatments, 0.5 mM Nano-Si was most effective in restricting toxic ion uptake, reducing Na^+ and Cl^- levels by up to 37% under salt stress. Foliar application showed greater efficiency, decreasing Na^+ and Cl^- by 33% and 26%, respectively, compared to 26% and 7% in root application. Additionally, foliar application enhanced K^+ and Ca^{2+} contents by 51% and 137%, respectively, whereas root application resulted in increases of 32% and 90%, respectively (Figure 10).

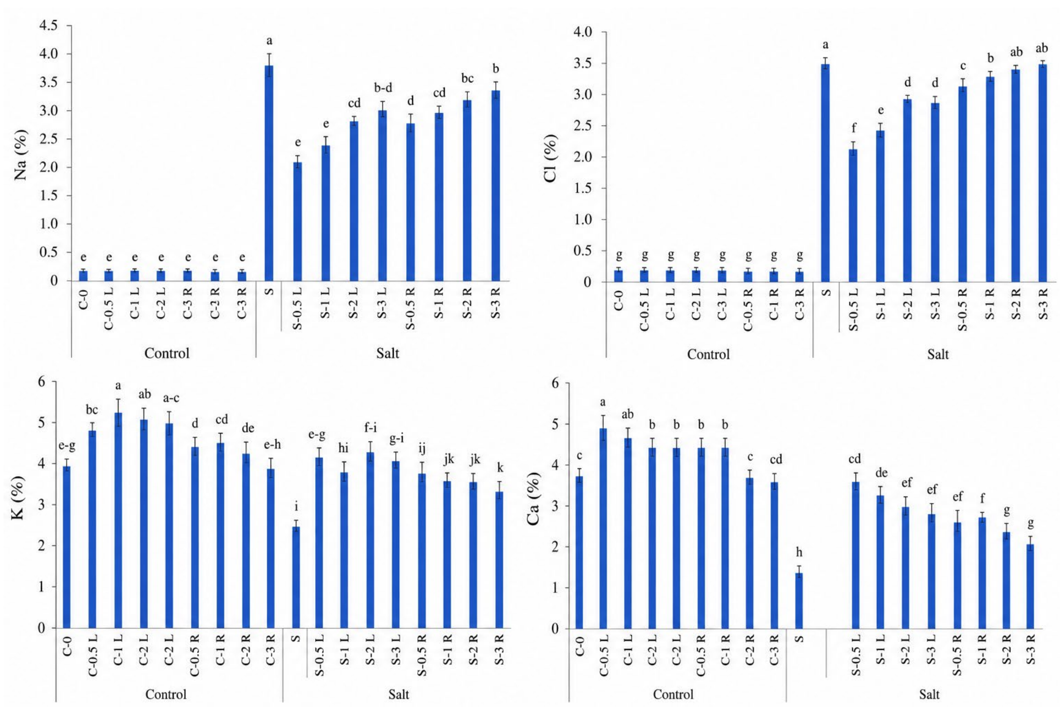

Figure 10. Effect of Nano-Si applications on ion contents under salt stress conditions*

C: Control, S: Salt, R: Root, L: Leaf application, *: There is no significant difference at the 0.05 level between the means denoted by the same letter within the same group

4. Discussion and Conclusion

Salinity is one of the most important environmental constraints limiting lettuce production because it simultaneously affects plant growth, water relations, nutrient acquisition, and cellular metabolism. Rather than acting through a single mechanism, salt stress triggers a complex cascade of osmotic, ionic, and oxidative disturbances that collectively suppress plant productivity. The present study demonstrated that Nano-Si effectively mitigated these detrimental effects through coordinated improvements in morphological, physiological, and biochemical traits. The superior performance of foliar Nano-Si, particularly at 0.5 and 1.0 mM, further indicates that both application method and concentration play decisive roles in maximizing silicon-mediated salt tolerance. These findings support the concept that Si nanoparticles function as multifunctional stress modulators capable of regulating several physiological processes simultaneously rather than acting solely as a nutrient supplement (Rastogi et al., 2019; El-Ramady et al., 2022).

Vegetative growth is generally the first process to be inhibited under saline conditions because reductions in external water potential rapidly limit water uptake and decrease cell turgor pressure, thereby restricting both cell division and cell elongation. Simultaneously, excessive Na^{+} and Cl^{-} accumulation interferes with nutrient acquisition and metabolic activity, resulting in lower photosynthetic carbon assimilation and reduced biomass production (Parihar et al., 2015; Zhao et al., 2021). Lettuce is particularly sensitive to these alterations because commercial production depends almost entirely on rapid vegetative growth and leaf biomass accumulation. Consequently, even moderate reductions in cell expansion and assimilate production may substantially reduce marketable productivity. Similar growth inhibition under saline conditions has previously been reported in lettuce, confirming that osmotic and ionic stress collectively limit vegetative development throughout the cultivation period (Garrido et al., 2014; Sardar et al., 2023).

The improvement in biomass accumulation following Nano-Si application suggests that Si nanoparticles effectively alleviated the physiological limitations imposed by salinity. Silicon has been reported to improve structural stability of epidermal tissues through silica deposition beneath the cuticle, thereby reducing excessive transpiration while maintaining tissue hydration. At the same time, Si contributes to membrane stabilization and preserves enzyme activities associated with photosynthetic carbon fixation and primary metabolism. These responses allow plants to maintain active cell expansion and biomass accumulation despite osmotic stress. Compared with conventional Si fertilizers, nanoparticles possess considerably greater surface area and reactivity, facilitating improved absorption and mobility within plant tissues. Consequently, Nano-Si frequently exhibits higher physiological efficiency at relatively low concentrations than bulk Si formulations (Merwad et al., 2018; El-Ramady et al., 2022; Siddiqi et al., 2025).

The positive responses observed in plant height, plant circumference, crown width and root collar diameter indicate that Nano-Si not only preserved biomass accumulation but also maintained normal vegetative architecture under saline conditions. Plant height is closely associated with sustained cell elongation, whereas plant circumference and crown width reflect successful leaf initiation and canopy expansion. Root collar diameter is likewise considered an important indicator of vascular development and assimilate transport capacity. Therefore, simultaneous improvements in these morphological characteristics suggest that Nano-Si alleviated multiple physiological constraints rather than affecting only a single growth process. Similar observations have been reported in tomato, cucumber and strawberry, where Nano-Si promoted shoot development by maintaining tissue hydration, improving nutrient utilization and reducing metabolic disruption under abiotic stress conditions (Haghighi and Pessarakli, 2013; Avestan et al., 2019; Alam et al., 2022).

Among the evaluated growth parameters, maintenance of leaf area is particularly important because it directly determines the photosynthetically active surface responsible for light interception and carbon assimilation. Under saline conditions, reduced turgor pressure, impaired cell enlargement and accelerated leaf senescence collectively restrict canopy development, thereby decreasing radiation interception and biomass production (Xu and Mou, 2015). The substantial preservation of leaf area following Nano-Si application therefore represents more than a simple morphological response; rather, it reflects the maintenance of functional photosynthetic capacity throughout plant development. By sustaining leaf expansion, Nano-Si likely enabled greater assimilate production, which subsequently supported biomass accumulation and commercial yield. Recent studies have similarly demonstrated that Si nanoparticles preserve canopy architecture under salinity by improving water relations and delaying stress-induced leaf senescence (Avestan et al., 2019; Alam et al., 2022; Simko et al., 2025).

The improvement in total yield observed in Nano-Si-treated plants is best interpreted as the cumulative consequence of multiple coordinated physiological responses. In lettuce, commercial yield depends largely on the accumulation of succulent shoot biomass rather than reproductive development. Therefore, maintenance of vegetative growth, canopy expansion, water status and photosynthetic activity directly contributes to marketable productivity. Rather than acting as an independent response, the increased yield obtained in Nano-Si-treated plants most likely resulted from the combined preservation of cellular metabolism, carbon assimilation and biomass partitioning under saline conditions. This integrated response further explains why moderate Nano-Si concentrations consistently produced greater benefits than higher concentrations. Silicon nanoparticles generally exhibit dose-dependent physiological activity, and excessive concentrations may reduce uptake efficiency, increase particle aggregation or alter nutrient balance, thereby diminishing their beneficial effects. Similar concentration-dependent responses have been described in soybean, cucumber, pea and several horticultural crops exposed to salinity (Farhangi-Abriz and Torabian, 2018; Alsaeedi et al., 2019; Ismail et al., 2022; Cakmakci and Sensoy, 2025).

Maintenance of plant water status is one of the primary adaptive responses determining plant survival under saline environments. Salinity decreases soil osmotic potential, thereby limiting root water uptake and reducing cellular turgor, which ultimately restricts physiological processes including cell expansion, stomatal conductance and photosynthesis. Therefore, RWC is widely accepted as a reliable indicator of plant water status and salt tolerance. The higher RWC observed following Nano-Si application suggests that Si nanoparticles effectively improved the ability of lettuce plants to maintain tissue hydration under osmotic stress. This protective effect may result from reduced cuticular transpiration, enhanced root hydraulic conductivity and improved regulation of water transport through aquaporin channels. Silicon has also been reported to preserve membrane permeability and facilitate water movement by maintaining membrane stability under saline conditions (Haghighi and Pessarakli, 2013; Kalteh et al., 2014; Mir et al., 2022; Atta et al., 2023). Consequently, maintenance of cellular hydration likely contributed to sustained metabolic activity and continued vegetative growth in Nano-Si-treated plants.

Disruption of ion homeostasis represents another major limitation imposed by salinity. Excessive accumulation of Na^+ and Cl^- not only causes ionic toxicity but also competitively inhibits the uptake of essential nutrients such as K^+ and Ca^{2+}, resulting in impaired enzyme activity, membrane instability and reduced photosynthetic efficiency. Potassium plays indispensable roles in stomatal regulation, osmotic adjustment, protein synthesis and enzyme activation, whereas calcium contributes to membrane integrity, cell wall stability and intracellular signaling. Therefore, maintenance of an appropriate K^+/Na^+ balance is considered one of the principal determinants of salt tolerance (Arif et al., 2020; Zhao et al., 2021). The improved ionic balance observed following Nano-Si application indicates that Si nanoparticles facilitated selective nutrient acquisition while simultaneously restricting the transport of toxic ions into aerial tissues.

The beneficial influence of Nano-Si on ion homeostasis is likely associated with several complementary physiological and molecular mechanisms. Silicon has been reported to promote the formation of apoplastic barriers through silica deposition and enhanced lignification, thereby limiting passive Na^+ movement towards the xylem. In addition, increasing evidence indicates that Si regulates the activity or expression of ion transporters involved in sodium exclusion and compartmentalization, including members of the Salt Overly Sensitive, High-Affinity Potassium Transporter and Na^+/H^+ Exchanger transporter families. Such regulation reduces cytosolic Na^+ accumulation while improving K^+ retention and vacuolar sequestration of toxic ions. Enhanced calcium accumulation may further stabilize plasma membranes and improve the selectivity of membrane transport proteins, thereby indirectly contributing to nutrient homeostasis (Farhangi-Abriz and Torabian, 2018; Alsaeedi et al., 2019; Ismail et al., 2022; Siddiqi et al., 2025). Similar improvements in mineral nutrition and ion balance following Nano-Si application have recently been demonstrated in lettuce, further supporting the hypothesis that Si nanoparticles enhance salt tolerance primarily by preserving ionic equilibrium rather than merely reducing sodium uptake (Cakmakci and Sensoy, 2025).

Oxidative stress is another inevitable consequence of prolonged salt exposure because excessive production of ROS exceeds the detoxification capacity of plant antioxidant systems. Reactive oxygen species attack membrane lipids, proteins and nucleic acids, leading to membrane deterioration and metabolic dysfunction. Malondialdehyde, a major product of lipid peroxidation, is therefore widely used as a biochemical indicator of oxidative membrane damage. The reduction in MDA accumulation following Nano-Si application indicates that Si nanoparticles effectively alleviated oxidative injury and maintained membrane integrity under saline conditions. Rather than acting as a direct antioxidant, Si appears to enhance the efficiency of endogenous defense systems by stimulating antioxidant enzymes such as superoxide dismutase, catalase, peroxidase and ascorbate peroxidase, thereby improving ROS detoxification before irreversible cellular damage occurs (Farhangi-Abriz and Torabian, 2018; Gözen and Kuşvuran, 2021; Ismail et al., 2022; Mir et al., 2022). Preservation of membrane integrity further supports nutrient transport, photosynthetic activity and cellular metabolism, collectively contributing to improved plant performance under salt stress.

Photosynthetic pigments are particularly susceptible to salinity because ionic toxicity and oxidative stress disrupt chloroplast ultrastructure and accelerate chlorophyll degradation. Consequently, decreases in chlorophyll and carotenoid contents reduce photosynthetic efficiency and carbon assimilation, ultimately limiting biomass production. The enhanced pigment concentrations observed following Nano-Si application suggest that Si preserved chloroplast function and delayed stress-induced pigment degradation. Carotenoids additionally serve as non-enzymatic antioxidants by dissipating excess excitation energy and scavenging singlet oxygen, thereby protecting photosystem II against photooxidative damage. Maintenance of chlorophyll and carotenoid contents therefore reflects not only improved pigment stability but also sustained photosynthetic functionality under saline conditions (Ndiso et al., 2016; Avestan et al., 2019; Bayram et al., 2021; Simko et al., 2025). Improved photosynthetic efficiency is likely one of the key factors explaining the superior biomass accumulation and marketable yield observed following Nano-Si treatments.

An important outcome of the present study was the consistently greater effectiveness of foliar Nano-Si application compared with root application across most evaluated parameters. Although both methods alleviated salinity-induced damage, foliar treatments generally produced superior improvements in growth, water relations, ion homeostasis and oxidative stress tolerance. This response may be attributed to the rapid absorption of nanoparticles through leaf tissues, allowing direct interaction with photosynthetically active organs while avoiding potential immobilization within the root zone or growing substrate. Foliar application may therefore increase Si use efficiency by facilitating faster translocation to metabolically active tissues where protection against oxidative damage and maintenance of photosynthetic activity are particularly critical. Recent studies conducted on lettuce and other horticultural crops similarly reported greater physiological efficiency of foliar Si application under abiotic stress conditions, emphasizing its potential as a practical management strategy for improving crop performance in salt-affected production systems (Cakmakci and Sensoy, 2025; Simko et al., 2025).

The present study demonstrated that Nano-Si is an effective strategy for enhancing salt tolerance in lettuce by simultaneously improving morphological growth, water status, ion homeostasis, membrane stability and photosynthetic performance. The beneficial effects of Nano-Si were not restricted to a single physiological process but resulted from coordinated regulation of water relations, mineral nutrition, oxidative stress protection and photosynthetic capacity. Among the evaluated treatments, foliar application of 0.5 and 1.0 mM Nano-Si consistently provided the greatest improvements, indicating that moderate Nano-Si concentrations are sufficient to maximize stress mitigation under saline conditions. These findings highlight the considerable potential of foliar Nano-Si application as an environmentally friendly and sustainable agronomic approach for lettuce cultivation in salt-affected regions. Future studies should focus on elucidating the molecular mechanisms regulating silicon-mediated ion transport, antioxidant defense and water transport pathways, while validating the effectiveness of Nano-Si under field conditions and different environmental scenarios.

Ethical Statement

The authors declare that ethical approval is not required for this research.

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Author Contributions Statement

Methodology, Investigation, Data Curation, Formal Analysis, Writing-Original Draft, *G. TORUN*; Conceptualization, Methodology, Investigation, Data Curation, Formal Analysis, Visualization, Supervision, Writing-Original Draft, Writing-Review & Editing, *Ş. KUŞVURAN*; Writing-Original Draft, Writing-Review & Editing, *E. DERELLİ TÜFEKÇİ*. All authors have seen, read and agreed to the published version of the manuscript.

Statement of Conflicts of Interest

The authors declare no conflicts of interest.

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