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Calcium Silicate Application Enhances Oxidative Defense and Improves the Physiological and Growth Responses of Shallot (*Allium cepa* L. Aggregatum Group) Under Salinity Stress

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Abstract

Indonesia's shallot production still falls short of domestic demand, necessitating imports. Expanding cultivation into marginal coastal areas such as Yogyakarta is promising but constrained by soil salinity. Silicon can help by enhancing plant resistance to such abiotic stress. This study evaluated the physiological and biochemical responses of shallot plants (*Allium cepa* L. Aggregatum group) to the application of calcium silicate ($CaSiO_3$) under saline conditions. The experiment employed a completely randomized design with 2 factors: $CaSiO_3$ (0, 2, and 4 mM) and salinity (0, 2, 4, and 8 dS m^{-1}), each with 5 replications. Physiological parameters, antioxidant activity, and yield traits were analyzed using analysis of variance (ANOVA) followed by Duncan multiple range test (DMRT) at $p \leq 0.05$. $CaSiO_3$ significantly alleviated salt stress by enhancing superoxide dismutase (SOD) activity and membrane stability, improving photosynthetic efficiency, promoting growth, and yield components. Under high salinity, 4 mM $CaSiO_3$ reduced proline and H_2O_2 accumulation compared with untreated plants. These findings indicate that applying 4 mM $CaSiO_3$ can enhance shallot productivity and resilience in saline coastal soils, supporting sustainable shallot self-sufficiency in Indonesia.

Keywords: *Allium cepa*; antioxidant enzymes; photosynthesis; salt tolerance; silicon

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INTRODUCTION

The demand for shallots in Indonesia is increasing due to their widespread use in cooking (Parmawati et al., 2021). Shallot cultivation is a vital part of Indonesian agriculture (Puspitasari et al., 2022). Expanding cultivation to coastal areas is a promising strategy to boost production; however, it faces a major challenge in soil salinity (Tuhuteru et al., 2019). Indonesia possesses significant agricultural development opportunities in marginal lands, including coastal areas, arid zones, and wetlands. However, research by Arulmathi and Porkodi (2020) shows that the coastal soils often exhibit high salinity, low organic matter, and poor nutrient and water retention, leading to reduced plant productivity.

Syamsiah et al. (2020) indicate that elevated salinity reduces nutrient absorption, yield, and soil quality in shallots. Increased salinity causes imbalances, hyperosmotic stress, and various physiological disorders in plants, which can be alleviated by osmolytes (protective chemicals

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that assist in maintaining cellular water balance) and by preserving ion homeostasis (Kumar et al., 2021). Salinity stress significantly restricts plant growth and development, leading to decreased shoot and root growth, smaller leaf size, and stunted plant structures (Ahmed, 2022). Excess sodium and chlorine ions may inhibit vegetative development and diminish photosynthesis and stomatal conductance, which plants can counteract through several physiological mechanisms.

An electrically conductive value of the saturation point extract in the root zone that exceeds 4 dS m^{-1} (approximately 40 mM NaCl) at 25 °C, in conjunction with an exchangeable sodium percentage of 15%, is indicative of a saline soil. At this electrical conductivity level (ECe), the yield of most crops diminishes; however, specific crops demonstrate yield declines even at lower ECe values (Munns, 2002; Bakır et al., 2024). Rasool et al. (2013) classified salinity levels according to ECe (dS m^{-1}) as follows: > 32 (extreme salinity), 16 to 32 (severe salinity), 8 to 16 (high salinity), 4 to 8 (moderate salinity), 2 to 4 (low salinity), and 0 to 2 (non-saline).

Salinity stress markedly blocks shallot development and growth, affecting multiple parameters that include plant height, leaf count, root traits, and bulb yield (Trisnaningsih et al., 2023). Shallots demonstrate a degree of salinity resistance, with local varieties capable of enduring irrigation saline levels of up to 3 dS m^{-1} in specific soil types (Syamsiah et al., 2020). Ehtaiwwesh and Emsahel (2020) show that salinity generally inhibits seed germination in many plant species. This results in a decrease in both dry and fresh weight of the roots, and the inhibition of root growth (Zörb et al., 2019). At the physiological level, salinity stress results in reduced stomatal conductance, closure, the transpiration rate, and the rate of photosynthesis during the stage of vegetative growth, while elevating the respiration rate (Higbie et al., 2010). Stomatal conductance and chlorophyll production are also diminished (Lee et al., 2013). In response to salinity, plants may demonstrate increased leaf tissue thickness and elevated proline concentrations (Anwar et al., 2024) as key indicators of oxidative stress.

One promising approach to mitigate these adverse effects is the application of silicon. The silicon treatment can alleviate the detrimental impacts of salt stress in plants through various pathways (Coskun et al., 2016). For instance, nutrient and water absorption are restricted by osmotic stress and salinity (Wang et al., 2021). Silicon may enhance water absorption and maintain nutritional balance, thereby reducing the effect of osmotic stress (Wang et al., 2021). Furthermore, while plants exposed to salinity often experience reduced photosynthesis (Trivellini et al., 2023), the application of silicon promotes photosynthesis and increases chlorophyll content (Rahmawati and Yasvi, 2024). Moreover, silicon enhances the activity of antioxidant enzymes that produce antioxidants in response to salt stress, including superoxide dismutase (SOD) (Kalteh et al., 2014). In summary, silicon treatment enhances the photosynthetic process, leaf water content, membrane stability, and plant development in conditions of salinity stress (Akhoundnejad et al., 2018).

Adequate fertilization is a key factor in maximizing shallot yield (Gunadi et al., 2024). In particular, applying silicate fertilizers containing silicon can enhance shallot growth and yield, as silicon mitigates environmental stress in the production of crops (Wangiyana et al., 2021). Research indicates that silicon accumulation in shallot plants can increase resistance, growth, and productivity (Kovács et al., 2022). Under stress conditions such as salinity, plants may have difficulty absorbing nutrients from the soil. Therefore, nutrient fertilization can be a useful strategy, allowing plants to maintain growth and development by absorbing nutrients directly. Given this background, the present research aims to investigate the impact of calcium silicate ($CaSiO_3$) treatment on the oxidative defense, physiological responses, and growth of shallot plants (*Allium cepa* L.) under salinity stress. Researchers hypothesize that $CaSiO_3$ application will enhance salt tolerance in shallots by strengthening the antioxidant defense system and improving key physiological processes such as photosynthesis.

MATERIALS AND METHOD

Experimental site, plant material, and growth conditions

This study was conducted at Universitas Gadjah Mada, Yogyakarta, utilizing greenhouse facilities at Sawitsari Research Center from 2022 to 2023. The ambient conditions at the research site consist of light intensity between 6,000 and 12,500 lx during daylight hours and temperatures that range from 23.5 to 34.6 °C.

The study utilized shallot (*A. cepa* L. Aggregatum group) bulbs of the Crok Kuning variety, sourced from the Agricultural Seed Center of Bantul, Yogyakarta. Silicon treatment involved the application of pure calcium silicate ($CaSiO_3$, Sigma-Aldrich), while salinity stress was induced using sodium chloride (NaCl, Sigma-Aldrich). A locally sourced organic compost ('S'per Kompos', Putra Sihobon Jaya, Ltd.) was also used. The soil medium was paddy field soil from the coastal area of Bantul, Yogyakarta, classified as loam with 54% sand, 20% clay, and 26% silt, as determined by the hydrometric method at the Soil, Fertilizer, Water, and Plant Laboratory, Agricultural Instrument Standards Implementation Center, Yogyakarta, Indonesia (June 5, 2023). The soil's physical and chemical properties were analyzed at the same laboratory, and the results are presented in Table 1.

Experimental design and treatments

A completely randomized design with a combination of 2 variables was used. The starting factor was silicon administered as $CaSiO_3$ at 3 dose levels (0, 2, and 4 mM). The second factor was salinity stress caused by NaCl at 4 levels (0, 2, 4, and 8 dS m^{-1}). Each experiment was replicated 5 times, resulting in a total of 60 polybags. Sixty polybags were prepared, each containing 5 kg of culture medium consisting of compost and soil (ratio 1:3). Soil was obtained from Kretek area, Bantul Regency, Yogyakarta. Shallot bulbs (diameter ±3.00 to 3.35 cm, weight ±2.0 to 2.5 g) were selected from the available mass, and 1 bulb was planted in each polybag. Fertilization was done twice: at 12 and 35 days after planting (DAP) using compost (4 g per polybag), urea (1.5 g per polybag), and NPK (1 g per polybag). Silicon and salinity treatments were applied simultaneously at 14 DAP by adding them to the soil medium.

Table 1. Physical and chemical properties of the soil

Parameter	Unit	Value
Sand	%	74
Clay	%	20
Silt	%	26
pH		6-8
Organic C	%	1.08
Exchangeable K	g^{-1}	0.79
Available P	mg kg^{-1}	1.41
Total N	%	0.04
Exchangeable Ca	g^{-1}	0.41
Exchangeable Na	g^{-1}	0.17
Si	ppm	0.1

Plant oxidative defense analysis

The proline content was assessed using the methodology described by Forlani and Funck (2020). Briefly, 0.5 g of leaf tissue was pulverized and homogenized in 10 ml of 3% sulfosalicylic acid solution, followed by filtration. Two milliliters of the filtrate were combined with 2 ml of ninhydrin reagent, which was prepared by heating to dissolve 1.25 g of ninhydrin in 35 ml of glacial acetic acid and 25 ml of phosphoric acid. The solution was incubated in a 95 °C water bath for 60 minutes and subsequently cooled in an ice bath to terminate the reaction. Toluene was used to extract the chilled mixture, and the toluene layer containing proline was collected. Absorbance was measured at 520 nm using a spectrophotometer. Each analysis was conducted with 5 replications per treatment. The concentration of proline was determined by comparing it to a standard curve prepared with known proline standards (Ábrahám et al., 2010).

SOD activity was assessed according to Gao et al. (1998). The assay mixture combination comprised 0.1 ml of pyrogallol solution (2 mM in 10 mM HCl) and 0.9 ml of 55.6 mM Tris-cacodylic acid buffer (TCB, pH 8.2) with 1.1 mM DTPA. The reaction commenced with the introduction of pyrogallol, and an increase in absorbance at 325 nm was observed at 25 °C utilizing a spectrophotometer (GENESYS 20 UV Scanning, Thermo Fisher Scientific). The autoxidation rate of pyrogallol served as the blank. In the absence of SOD, the change in absorbance was 0.02 per minute. Each treatment was replicated 5 times.

The concentration of hydrogen peroxide (H_2O_2) was assessed using the methodology established by Wang et al. (2022). Briefly, 0.25 g of fresh leaves were frozen in liquid nitrogen, subsequently pulverized and homogenized in 5 ml of 0.1% TCA. At 4 °C, the homogenate was centrifuged for 15 minutes at 12,000 rpm. The resultant mixture was incubated in an ice box for 1 hour to facilitate color development. Approximately 0.5 ml of the supernatant was combined with 0.5 ml of 10 mM phosphate buffer (pH 7) and 1 ml of 1 M KI solution. Absorbance was quantified at 390 nm. The blank solution contained 0.1% TCA. H_2O_2 concentrations were determined using a standard curve generated with known H_2O_2 standards. Every treatment was carried out 5 times in this parameter.

The membrane stability index (MSI) was assessed by quantifying electrolyte leakage from leaf tissue. Two hundred milligrams of leaf tissue were washed, segmented into 5 mm pieces, and subsequently submerged in 20 ml of distilled deionized water. Samples were cultured for 12 hours under controlled laboratory conditions, and the initial electrical conductivity (EC1) was assessed. The samples were subsequently heated to 100 °C for 15 minutes, cooled to 25 °C, and a second conductivity (EC2) measurement was recorded. In this parameter, each treatment was repeated 5 times. MSI was calculated using Equation 1 (Singh et al., 2022).

$$\text{MSI (\%)} = 1 - \left[\frac{EC1}{EC2}\right] \times 100\% \quad (1)$$

Plant physiological analysis

Photosynthetic parameters, including intracellular CO_2 concentration, transpiration rate, stomatal conductance, and assimilation rate, were measured using LI-6800 (portable photosynthetic system analyzer). The chlorophyll content was assessed in the third leaf using the method described by Harborne (1998). The absorbance of the extracts was measured at wavelengths of 470, 645, and 664 nm using a spectrophotometer (GENESYS 20 UV Scanning, Thermo Fisher Scientific). Every treatment in this parameter was carried out 5 times.

Growth analysis

Growth parameters were measured at 49 DAP. This included plant height, number of leaves, total fresh weight, bulb weight, root length, shoot dry weight, root dry weight, and root-to-shoot ratio (Olas et al., 2020). In this parameter, each treatment was repeated 5 times.

Statistical analysis

The influences of silicon and salinity on the parameters were examined using analysis of variance (ANOVA), followed by Duncan multiple range test (DMRT) for post-hoc comparisons. A significance level of $p < 0.05$ was used for all statistical tests, which were performed using IBM-SPSS Statistics version 27.0 (Compton, 2012).

RESULTS AND DISCUSSION

Plant oxidative defense

A significant difference in $CaSiO_3$ level was observed due to salinity-induced stress in nearly all oxidative defense parameters. $CaSiO_3$ showed a significant interaction with salinity level for MSI, indicating that its effect on membrane stability depended on the salinity condition (Table 2).

Salinity treatment increased proline and H_2O_2 levels but decreased SOD activity and the MSI. Conversely, $CaSiO_3$ treatment decreased proline and H_2O_2 levels while increasing SOD activity and the MSI. Proline is considered a crucial osmoprotectant in plants experiencing abiotic stress, especially salinity.

The data in Table 3 detail the proline content in *A. cepa* subjected to varied salinity levels and $CaSiO_3$ addition. A clear link between salinity and $CaSiO_3$ levels was discovered, suggesting a complex regulation of proline production in response to the combined impacts of stress and mitigation interventions. Without $CaSiO_3$ (0 mM), proline accumulation progressively increased with rising salinity, reaching a maximum of 0.71±0.05 μmol g^{-1} FW at 8 dS m^{-1} salinity. This pattern aligns with the established understanding that salt stress leads to oxidative

Table 2. The effect of $CaSiO_3$ on the oxidative defense of *A. cepa* under varying salinity

Treatment	Proline content (μmol g^{-1} FW)	SOD (U mg^{-1})	H_2O_2 (μmol g^{-1} FW)	MSI (%)
Salinity/S (dS m^{-1})	*	*	*	*
0	0.39^{d}	17.91^{a}	2.36^{d}	73.45^{a}
2	0.42^{c}	16.46^{a}	2.72^{c}	69.77^{b}
4	0.54^{b}	15.41^{b}	2.86^{bc}	65.01^{c}
8	0.63^{a}	14.06^{c}	2.98^{a}	63.98^{d}
$CaSiO_3$/C (mM)	*	*	*	ns
0	0.57^{a}	12.63^{c}	3.34^{a}	65.65^{b}
2	0.41^{b}	15.60^{b}	2.78^{b}	67.74^{b}
4	0.34^{c}	18.90^{a}	2.08^{c}	70.78^{a}
S × C	*	*	*	*

Note: Different lowercase letters indicate significant differences ($p \leq 0.05$) based on the DMRT test. * = Significant, ns = Non-significant

Table 3. The oxidative defense of *A. cepa* treated with $CaSiO_3$ and salinity

Parameter	$CaSiO_3$ (mM)	Salinity (dS m^{-1})			
		0	2	4	8
Proline content (μmol g^{-1} FW)	0	0.43±0.09^{d}	0.48±0.06^{de}	0.69±0.07^{ef}	0.71±0.05^{f}
	2	0.35±0.05^{b}	0.37±0.08^{c}	0.39±0.01^{cd}	0.56±0.08^{e}
	4	0.29±0.05^{a}	0.35±0.04^{b}	0.36±0.06^{b}	0.37±0.01^{c}
SOD (U mg^{-1})	0	14.36±0.78^{abc}	13.47±1.74^{ab}	12.57±1.85^{ab}	11.22±0.78^{a}
	2	17.06±1.82^{efg}	16.61±1.35^{defg}	15.71±1.74^{bcde}	13.02±1.22^{abcd}
	4	19.75±1.39^{g}	18.86±0.78^{fg}	17.96±1.95^{efg}	17.76±1.35^{efg}
H_2O_2 (μmol g^{-1} FW)	0	2.93±0.55^{de}	3.33±0.54^{defg}	3.51±0.71^{defgh}	3.60±0.76^{h}
	2	2.38±0.95^{ab}	2.80±0.56^{c}	2.92±0.31^{cd}	3.02±0.15^{de}
	4	1.78±0.97^{a}	2.09±0.49^{b}	2.15±0.12^{b}	2.33±0.59^{bc}
MSI (%)	0	70.86±0.49^{e}	68.41±0.87^{d}	63.71±0.92^{b}	58.29±0.47^{a}
	2	74.67±0.26^{f}	68.61±1.26^{d}	63.84±1.11^{b}	60.18±0.49^{a}
	4	77.52±1.15^{g}	70.83±1.14^{e}	66.34±0.37^{c}	63.38±0.39^{b}

Note: The mean (n = 5) value followed by the same letter of each parameter indicates no significant difference at a 95% confidence level based on ANOVA and DMRT

and osmotic imbalance, which, in turn, stimulates proline synthesis as a protective mechanism.

The use of $CaSiO_3$ resulted in a significant decrease in proline content at all salinity levels. At a dosage of 2 mM $CaSiO_3$, proline levels ranged from 0.35±0.05 to 0.56±0.08 μmol g^{-1} FW. A notable decrease was recorded at 4 mM $CaSiO_3$, with values dropping to a minimum of 0.29±0.05 μmol g^{-1} FW under non-saline conditions. This evidence suggests that $CaSiO_3$ plays a crucial role in mitigating the stress caused by salinity, possibly stabilizing membranes, eliminating reactive oxygen species (ROS), or altering ion homeostasis, which may, in turn, reduce the reliance on proline-mediated stress protection (Singh et al., 2022).

Salinity stimulates the formation of ROS, causing oxidative damage, which plants resist through enzymatic and non-enzymatic antioxidant processes. The elevation in proline is significant both as an osmoprotectant and as a non-enzymatic ROS scavenger (Anwar et al., 2024). One important antioxidant enzyme that combats superoxide radicals generated under abiotic stressors, such as salinity, is SOD. Table 2 presents the SOD activity in *A. cepa* subjected to escalating salinity levels alongside different concentrations of $CaSiO_3$. A clear interactive effect between salinity stress and silicon application was observed, indicating the oxidative stress status and the protective response in shallot's physiology.

In the absence of $CaSiO_3$ (0 mM), SOD activity decreased as salinity increased, ranging from 14.36±0.78 U mg^{-1} at 0 dS m^{-1} to 11.22±0.78 U mg^{-1} at 8 dS m^{-1} (Table 3). This reduction suggests that increased salinity levels may impair the antioxidative defense system, possibly due to oxidative damage or inhibited SOD biosynthesis. This vulnerability under salinity stress highlights the susceptibility of shallot plants to ROS accumulation when protective agents are absent. The addition of $CaSiO_3$ resulted in a considerable increase in SOD activity at all salinity levels. At a concentration of 2 mM $CaSiO_3$, SOD activity varied between 17.06±1.82 and 13.02±1.22 U mg^{-1}. In contrast, at 4 mM, SOD activity increased, reaching a maximum of 19.75±1.39 U mg^{-1} in non-saline conditions. Interestingly, even at the highest salinity concentration of 8 dS m^{-1}, SOD activity was still recorded at 17.76±1.35 U mg^{-1} in the 4 mM treatment group (Table 3).

The increase in SOD activity with higher $CaSiO_3$ concentrations indicates a potential function of silicon in enhancing the system of antioxidants under saltwater conditions. Silicon has a critical function in controlling the production and activity of antioxidant enzymes, enhancing nutrient balance, and reducing Na^+ toxicity, which collectively contribute to mitigating the effect of oxidative stress (Souza Junior et al., 2023). It also regulates and enhances the activity of antioxidant enzymes (Avestan et al., 2019; Shinga et al., 2025). SOD, a key enzyme, initiates ROS detoxification by converting superoxide anions into H_2O_2. Superoxide radicals are converted into H_2O_2 and O_2 by SOD, which prevents cellular damage (Akhoundnejad et al., 2018).

The substance H_2O_2 is used as a signaling molecule and a marker of oxidative stress in plants, particularly in response to abiotic conditions such as salinity. Table 2 breaks down

the H_2O_2 concentration in *A. cepa* subjected to varying levels of salinity and $CaSiO_3$ applications. The identified trends offer a detailed understanding of the plant's oxidative status and silicon's potential role in alleviating salinity-induced stress. In the absence of $CaSiO_3$ (0 mM), H_2O_2 levels increased in response to salinity, peaking at 3.60±0.76 μmol g^{-1} FW at a salt concentration of 8 dS m^{-1} (Table 3). This trend suggests a salt-induced oxidative burst, likely resulting from the excessive generation of ROS coupled with a constrained antioxidant capacity. The higher level of H_2O_2 in response to salinity indicates that shallot plants undergo considerable oxidative stress when protective treatments are not applied.

$CaSiO_3$ significantly decreased H_2O_2 accumulation at all salinity levels (Table 2). At a concentration of 2 mM $CaSiO_3$, the H_2O_2 levels varied between 2.38±0.95 and 3.02±0.15 μmol g^{-1} FW, indicating a moderate reduction relative to the control. The reduction was significantly more pronounced at 4 mM $CaSiO_3$, with H_2O_2 content declining significantly to 1.78±0.97 μmol g^{-1} FW in non-saline conditions, while remaining comparatively low (2.33±0.59 μmol g^{-1} FW) even under high salinity (8 dS m^{-1}) (Table 3). $CaSiO_3$ can reduce the adverse effects of salinity on plants, partially through the reduction of H_2O_2 levels (Abdelaal et al., 2020). The application of silicon under stress from salinity can significantly enhance the functioning of the protection system, as this environment is characterized by the accumulation of ROS, including H_2O_2 (Akhter et al., 2022).

Applying $CaSiO_3$ can potentially enhance the value of the MSI in *A. cepa* subjected to salinity stress (Table 2). Salinity and $CaSiO_3$ treatments resulted in the highest MSI value (77.52%) at 0 dS m^{-1} and 4 mM, respectively, and the lowest (58.29%) at 8 dS m^{-1} and 0 mM (Table 3). The MSI has emerged as a reliable physiological parameter for assessing salt tolerance in *A. cepa*. When a plant is exposed to salt stress conditions, its MSI indicates the ability to maintain the integrity of its cellular membranes (Sanwal et al., 2022).

One of the most significant physiological indicators of cell membrane integrity is the MSI, which is particularly useful in the context of abiotic stress factors such as salinity. This research illustrates that the relationship between different concentrations of $CaSiO_3$ and salinity had a significant impact on the MSI of *A. cepa* (Table 2). The detrimental effects of salt stress on membrane quality were underscored by the significant decrease in MSI that was observed as salinity levels increased across all $CaSiO_3$ regimens. The control group (0 mM $CaSiO_3$) exhibited a decrease in MSI from 70.86% without $CaSiO_3$ to 58.29% at 8 dS m^{-1}, suggesting a notable destabilization of the membrane. This trend was reduced in plants treated with $CaSiO_3$.

The MSI values showed a modest rise under similar salinity conditions at a concentration of 2 mM $CaSiO_3$, varying from 74.67% without $CaSiO_3$ to 60.18% at 8 dS m^{-1}. The 4 mM $CaSiO_3$ treatment exhibited the highest MSI values, maintaining a level of 77.52% under non-saline conditions, which decreased to 63.38% at the maximum salinity level (Table 3). ROS causes peroxidation of membrane lipids, which decreases MSI. The increase in MSI by silicon directly reflects the reduction in oxidative stress (lower H_2O_2) and the cellular protection provided by silicon (Akhoundnejad et al., 2018).

Plant physiological responses

Salinity treatment reduced the assimilation rate, transpiration rate, intercellular CO_2 concentration, stomatal conductance, and chlorophyll content. While $CaSiO_3$ increased assimilation rate, intercellular CO_2, stomatal conductance, and chlorophyll content, but decreased transpiration rate (Table 4).

Chlorophyll content exhibited a marked reduction with increased salinity across all treatment groups, corroborating the deterioration of photosynthetic pigments under salinity stress. $CaSiO_3$ supplementation enhanced chlorophyll content. At 0 mM $CaSiO_3$, chlorophyll content diminished from 3.62 mg g^{-1} FW (0 dS m^{-1}) to 2.16 mg g^{-1} FW (8 dS m^{-1}). At 4 mM $CaSiO_3$, values varied from 4.55 to 2.34 mg g^{-1} FW across different salinity levels, indicating enhanced pigment stability (Table 5).

Intercellular CO_2 concentration and stomatal conductance decreased during salinity stress; however, increased $CaSiO_3$ concentrations mitigated this decline (Table 4). The maximum stomatal conductance (0.39 mol m^{-2} s^{-1}) and intercellular CO_2 (378.74 μmol mol^{-1}) were recorded in 4 mM $CaSiO_3$ under non-saline conditions. At 8 dS m^{-1}, stomatal conductance and intercellular CO_2 were sustained at 0.11 mol m^{-2} s^{-1} and 291.86 μmol mol^{-1}, respectively, with 4 mM $CaSiO_3$, in contrast to markedly reduced values in the control group (Table 5). Salinity significantly decreased transpiration and photosynthetic assimilation rates in *A. cepa*.

Table 4. The effect of $CaSiO_3$ on the physiological responses of *A. cepa* under varying salinity

Treatment	Chlorophyll content (mg g^{-1} FW)	Assimilation rate (µmol m^{-2} s^{-1})	Intercellular CO_2 (µmol mol^{-1})	Transpiration rate (mol m^{-2} s^{-1})	Stomatal conductance (mol m^{-2} s^{-1})
Salinity/S (dS m^{-1})	*	*	*	*	*
0	4.45^{d}	5.07^{c}	373.75^{d}	0.0025^{d}	0.28^{c}
2	3.21^{c}	4.40^{b}	349.91^{c}	0.0022^{c}	0.16^{bc}
4	3.11^{b}	4.24^{b}	334.39^{b}	0.0021^{b}	0.12^{b}
8	2.32^{a}	2.81^{a}	284.46^{a}	0.0020^{a}	0.08^{a}
$CaSiO_3$/C (mM)	*	*	*	*	*
0	2.93^{a}	3.72^{a}	329.58^{a}	0.0032^{a}	0.12^{a}
2	3.11^{b}	3.92^{b}	336.51^{b}	0.0021^{b}	0.15^{b}
4	3.42^{c}	4.78^{c}	341.79^{c}	0.0014^{c}	0.21^{c}
S × C	*	*	*	*	*

Note: Values in each column superscripted with different lowercase letters indicate significant differences ($p \leq 0.05$) based on the DMRT test. * = Significant

Table 5. The physiological responses of *A. cepa* treated with $CaSiO_3$ and salinity

Parameter	$CaSiO_3$ (mM)	Salinity (dS m^{-1})			
		0	2	4	8
Chlorophyll content (mg g^{-1} FW)	0	3.62 ± 0.08^{g}	2.96 ± 0.11^{de}	2.60 ± 0.08^{c}	2.16 ± 0.32^{a}
	2	3.84 ± 0.18^{g}	3.13 ± 0.09^{ef}	2.80 ± 0.11^{d}	2.24 ± 0.10^{ab}
	4	4.55 ± 0.04^{h}	3.26 ± 0.08^{f}	2.91 ± 0.09^{d}	2.34 ± 0.06^{b}
Stomatal conductance (mol m^{-2} s^{-1})	0	0.21 ± 0.001^{de}	0.14 ± 0.001^{abc}	0.09 ± 0.005^{ab}	0.07 ± 0.001^{a}
	2	0.26 ± 0.008^{e}	0.16 ± 0.007^{bc}	0.13 ± 0.002^{abc}	0.08 ± 0.001^{ab}
	4	0.39 ± 0.004^{f}	0.18 ± 0.002^{d}	0.14 ± 0.001^{abc}	0.11 ± 0.004^{ab}
Intercellular CO_2 (µmol mol^{-1})	0	365.18 ± 1.51^{f}	342.51 ± 1.41^{de}	333.78 ± 1.67^{c}	276.92 ± 2.11^{a}
	2	377.48 ± 1.32^{g}	348.79 ± 1.05^{e}	334.11 ± 1.78^{c}	284.50 ± 1.38^{ab}
	4	378.74 ± 2.57^{g}	358.48 ± 2.05^{f}	335.29 ± 1.34^{cd}	291.86 ± 1.42^{b}
Transpiration rate (mol m^{-2} s^{-1})	0	0.0036 ± 0.00008^{h}	0.0026 ± 0.0006^{fg}	0.0021 ± 0.0001^{de}	0.0015 ± 0.0001^{bc}
	2	0.0035 ± 0.0005^{h}	0.0024 ± 0.0003^{ef}	0.0020 ± 0.0004^{d}	0.0012 ± 0.0003^{ab}
	4	0.0028 ± 0.0002^{g}	0.0022 ± 0.0001^{de}	0.0019 ± 0.0002^{cd}	0.0011 ± 0.0004^{a}
Assimilation rate (µmol m^{-2} s^{-1})	0	4.39 ± 0.11^{d}	3.91 ± 0.22^{c}	3.88 ± 0.35^{c}	2.67 ± 0.29^{a}
	2	4.96 ± 0.22^{ef}	4.01 ± 0.33^{c}	3.97 ± 0.14^{c}	2.78 ± 0.16^{a}
	4	5.87 ± 0.25^{g}	5.30 ± 0.11^{f}	4.88 ± 0.18^{e}	3.07 ± 0.21^{b}

Note: The mean (n = 5) value followed by the same letter of each parameter indicates no significant difference at a 95% confidence level based on ANOVA and DMRT

Nonetheless, the use of $CaSiO_3$ mitigated these reductions. The assimilation rate decreased from 4.39 to 2.67 µmol m^{-2} s^{-1} with rising salinity in the absence of $CaSiO_3$ but remained higher (5.87 to 3.07 µmol m^{-2} s^{-1}) under the 4 mM treatment (Table 5). Transpiration also decreased across varying salinity levels; however, with $CaSiO_3$, plants sustained increased rates. The improved performance following $CaSiO_3$ treatment may be ascribed to greater water usage efficiency, fewer stomatal constraints, and superior cellular osmotic adjustment.

The overall physiological enhancements resulting from $CaSiO_3$ application, especially at 4 mM, underscore its effectiveness in mitigating the adverse effects of salt-induced stress. These enhancements indicate silicon's role in many protective mechanisms, including improved antioxidant defense, osmotic equilibrium, and stabilization of chloroplast membranes and enzymes associated with photosynthesis. The process by which plants transform sunlight into chemical energy is called photosynthesis. Salinity stress significantly impacts plant photosynthesis, affecting various components of the photosynthetic process (Wungrampha et al., 2018). It can cause irreversible harm at any formative stage, leading to diminished development and productivity (Wungrampha et al., 2018; Zahra et al., 2022).

The ultrastructure of chloroplasts can be altered by salt stress, which can result in the dilatation of thylakoid membranes and a reduction in the number of grana (Alkhatib et al., 2021). Photosynthesis is conducted within chloroplasts. The growth of a variety of higher plants can be facilitated by silicon in the presence of abiotic stresses, such as salinity (Mir et al., 2022). Silicon has the potential to improve plant growth in conditions of salinity stress (Dhiman et al., 2021). Plants experiencing salt stress exhibited a significant rise in their assimilation rate, stomatal conductance, and internal CO_2 content when exogenously supplied silicon was administered (Xue et al., 2021). The size of chloroplasts and the quantity of grana in leaves can be increased to boost the photosynthetic capability of plants subjected to silicon treatment (Rastogi et al., 2021). Silicon accumulation in leaves enhances photosynthetic potential and efficiency by widening leaf angles, reducing shading, and maintaining leaf uprightness (El-Sayed et al., 2019).

According to Rastogi et al. (2021), silicate can decrease Na^+ uptake by slowing down transpiration, as evidenced by the study's findings that stomatal conductance values started to rise dramatically when the dose of silicon was administered to salt-stressed plants. The addition of silicon to saline growth media enhances photosynthetic activity (Nabati et al., 2013). Silicate amendments enhance stomatal conductance in salt-stressed plants, indicating that silicate may mitigate Na^+ uptake by lowering transpiration rates, which consequently results in diminished growth and net photosynthesis (Yeo et al., 1999).

Jang et al. (2020) found that the amount of silicon associated with cellulose in the epidermal cell wall affects the transpiration rate, which allows plants to resist salinity stress. They also found that applying silicon can reduce water loss in plants by altering the morphological structure of leaf epidermal cells (Shen et al., 2022). Silicon is stored in the leaves, resulting in decreased transpiration and consequently the accumulation of dilute salts in saline environments (Rastogi et al., 2021). The epidermis serves to restrict excessive water loss, while silica interacts with cellulose within the epidermal cells of the leaf blade (Kumar et al., 2021). The influence of silicon on stomatal opening can also be a contributing factor to the decrease in transpiration (Gao et al., 2006). *Allium cepa* enhances water efficiency under salinity stress by minimizing excessive transpiration and optimizing stomatal conductance for photosynthesis.

Plant growth characters

Plant growth is significantly impeded by salinity, which interferes with physiological and biochemical processes, thereby affecting biomass accumulation and yield. This research assessed the impact of $CaSiO_3$ on key growth parameters of *A. cepa* across varying salinity levels. The findings demonstrate that elevated salt concentrations negatively influenced all growth parameters; however, the addition of $CaSiO_3$ partially mitigated these effects.

Table 6 shows a significant difference in $CaSiO_3$ due to salinity stress in almost all plant growth characters, except for the number of leaves and root length. Figure 1 shows that the salinity treatment reduced root length, bulb weight, total fresh weight, number of leaves, and plant height. $CaSiO_3$ treatment increases plant height, number of leaves, fresh weight,

Table 6. The effect of $CaSiO_3$ on growth characteristics of *A. cepa* under varying salinity

Treatment	Plant height (cm)	Number of leaves	Total fresh weight (g)	Bulb weight (g)	Root length (cm)
Salinity/S (dS m^{-1})	*	*	*	*	*
0	40.74^{d}	49.75^{d}	110.67^{d}	41.52^{d}	27.88^{d}
2	37.04^{c}	37.50^{c}	66.70^{c}	35.73^{c}	25.01^{c}
4	35.85^{b}	27.16^{b}	55.77^{b}	27.89^{b}	23.41^{b}
8	34.50^{a}	22.41^{a}	42.36^{a}	17.42^{a}	19.77^{a}
$CaSiO_3$/C (mM)	*	ns	*	*	ns
0	35.37^{a}	31.50^{a}	59.25^{a}	24.97^{a}	22.50^{a}
2	37.85^{b}	33.58^{a}	70.60^{b}	30.47^{b}	24.98^{a}
4	39.87^{b}	37.58^{b}	76.77^{c}	36.98^{c}	26.83^{b}
S × C	*	*	*	*	*

Note: Values in each column superscripted with different lowercase letters indicate significant differences ($p \leq 0.05$) based on the DMRT test. * = Significant, ns = Non-significant

Figure 1. The morphology of *A. cepa* treated with $CaSiO_3$ and salinity

bulb weight, and root length. The morphology of *A. cepa* plants that are subjected to salinity stress is significantly influenced by $CaSiO_3$, which substantially enhances the plant's structure in terms of height (Figure 2).

The maximum plant height (41.76 cm) was observed in the control treatment (0 dS m^{-1} salinity), while the lowest (35.72 cm) was recorded at the highest salinity level (8 dS m^{-1}) (Figure 2). Leaf production was diminished with increased salt levels. In the absence of $CaSiO_3$, the number of leaves decreased from 46.33 at 0 dS m^{-1} to 21.40 at 8 dS m^{-1}. Nonetheless, the use of $CaSiO_3$ enhanced leaf retention during salt stress. The maximum leaf number (51.23) was recorded in plants subjected to 4 mM $CaSiO_3$ at 0 dS m^{-1}, and even at 8 dS m^{-1}, the leaf number (25.10) exceeded that of untreated controls (Table 7).

Biomass production, encompassing both fresh and bulb weight, exhibited a comparable trend (Table 6). Fresh weight diminished markedly with increasing salinity; however, plants subjected to 4 mM $CaSiO_3$ exhibited considerably increased fresh weights (120.62 g at 0 dS m^{-1} and 50.27 g at 8 dS m^{-1}) in comparison to untreated controls (98.58 and 32.35 g, respectively). The weight of the bulb, an essential yield component, exhibited a downward trend, decreasing from 34.24 to 14.99 g (0 mM $CaSiO_3$), but increased with $CaSiO_3$ treatment (47.44 to 19.59 g at 4 mM) (Table 7).

Salinity significantly inhibited root dry weight, shoot dry weight, and root length, but these effects were mitigated by $CaSiO_3$ (Table 8). Root length decreased from 24.50 to 18.00 cm in untreated plants, while it grew to 30.83 cm at 0 dS m^{-1} and stayed increased at 21.16 cm at 8 dS m^{-1} after 4 mM $CaSiO_3$ treatment (Table 7). Likewise, the dry weights of the shoot and root were maximal in the 4 mM $CaSiO_3$ group (2.73 and 0.52 g at 0 dS m^{-1}, respectively), signifying improved biomass distribution and stress tolerance (Table 9).

While salinity treatment decreased plant growth, including shoot dry weight, root dry weight, and root-to-shoot ratio, the addition of

Table 7. The growth characteristics of *A. cepa* treated with $CaSiO_3$ and salinity

Parameter	$CaSiO_3$ (mM)	Salinity (dS m^{-1})			
		0	2	4	8
Number of leaves	0	46.33±1.45^{fg}	32.00±1.32^{d}	24.25±1.15^{bc}	21.40±1.82^{a}
	2	50.50±2.64^{h}	37.06±0.57^{e}	25.50±2.64^{bc}	22.08±1.52^{ab}
	4	51.23±1.37^{h}	43.16±2.08^{f}	32.20±2.00^{d}	25.10±1.48^{bc}
Fresh weight (g)	0	98.58±8.16^{f}	57.01±2.29^{cd}	50.65±3.14^{c}	32.35±3.78^{a}
	2	112.88±4.35^{fg}	67.57±3.78^{de}	56.74±2.16^{cd}	44.54±2.42^{b}
	4	120.62±2.08^{g}	75.61±9.45^{e}	60.02±7.56^{de}	50.27±2.08^{c}
Bulb weight (g)	0	34.24±5.71^{e}	28.08±5.32^{cd}	20.59±6.51^{ab}	14.99±4.82^{a}
	2	42.72±2.77^{f}	32.88±3.92^{bc}	29.99±2.64^{e}	15.61±4.22^{a}
	4	47.44±0.61^{fg}	45.40±4.16^{f}	32.10±5.13^{e}	19.59±4.74^{ab}
Root length (cm)	0	24.50±1.08^{def}	23.33±1.50^{cde}	22.00±1.15^{bcd}	18.00±0.64^{a}
	2	27.20±1.32^{fg}	26.50±1.52^{f}	23.06±1.25^{cde}	20.16±2.06^{ab}
	4	30.83±1.80^{g}	27.33±1.79^{fg}	25.16±1.25^{ef}	21.16±3.22^{bc}

Note: The mean (n = 5) value followed by the same letter of each parameter indicates no significant difference at a 95% confidence level based on ANOVA and DMRT

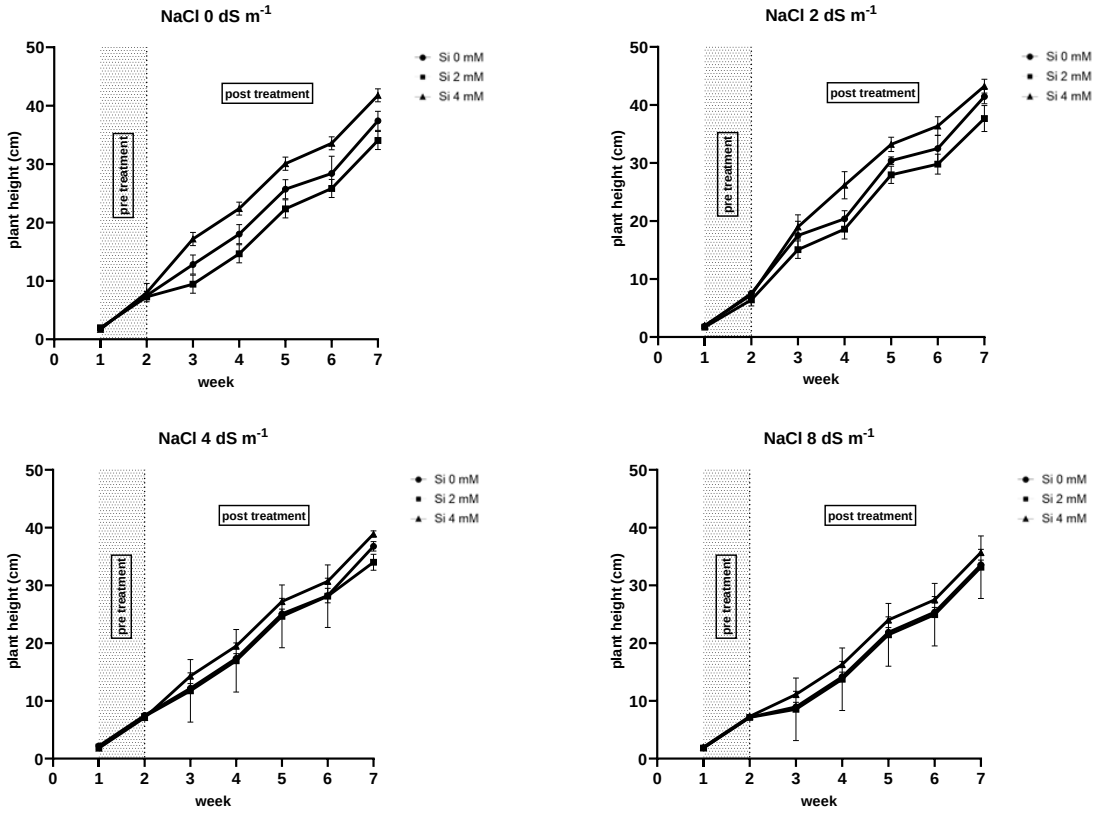

Figure 2. The plant height of *A. cepa* treated with $CaSiO_3$ and salinity
Note: The mean (n = 5) according to a 95% confidence level based on ANOVA and DMRT

$CaSiO_3$ helped to improve these parameters. The interaction of salinity and $CaSiO_3$ significantly decreased root-to-shoot ratio, shoot dry weight, and root dry weight (Table 9).

Root-to-shoot ratio elucidates biomass allocation mechanisms during stress conditions. Salinity generally diminished the root-to-shoot ratio, especially in the absence of $CaSiO_3$. The treatments of $CaSiO_3$, particularly at 4 mM, maintained increased root-to-shoot ratio values across all salinity levels (Table 9). Salinity stress markedly reduced plant morphology, growth, and productivity (Sun et al., 2025). This study indicated a reduction in plant growth under

Table 8. The effect of $CaSiO_3$ on the shoot and root dry weight of *A. cepa* under varying salinity

Treatment	Shoot dry weight (g)	Root dry weight (g)	Root-to-shoot ratio
Salinity/S (dS m^{-1})	*	*	*
0	2.37^{d}	0.42^{d}	0.17^{b}
2	1.87^{c}	0.29^{c}	0.15^{a}
4	1.77^{b}	0.26^{b}	0.14^{a}
8	1.61^{a}	0.22^{a}	0.13^{a}
$CaSiO_3$/C (mM)	*	*	*
0	1.77^{a}	0.23^{a}	0.13^{a}
2	1.83^{b}	0.30^{b}	0.16^{b}
4	2.11^{c}	0.35^{c}	0.17^{b}
S × C	*	*	*

Note: Values in each column superscripted with different lowercase letters indicate significant differences ($p \leq 0.05$) based on the DMRT test. * = Significant

Table 9. Shoot and root dry weight of *A. cepa* treated with $CaSiO_3$ and salinity

Parameter	$CaSiO_3$ (mM)	Salinity (dS m^{-1})			
		0	2	4	8
Shoot dry weight (g)	0	1.92 ± 0.09^{cd}	1.88 ± 0.14^{bcd}	1.72 ± 0.12^{abc}	1.59 ± 0.21^{a}
	2	2.47 ± 0.13^{fg}	1.91 ± 0.28^{cd}	1.82 ± 0.67^{bcd}	1.63 ± 0.20^{ab}
	4	2.73 ± 0.10^{g}	2.03 ± 0.62^{ef}	1.97 ± 0.24^{cde}	1.73 ± 0.29^{abc}
Root dry weight (g)	0	0.29 ± 0.02^{ef}	0.25 ± 0.03^{bc}	0.22 ± 0.08^{ab}	0.18 ± 0.05^{a}
	2	0.45 ± 0.02^{h}	0.28 ± 0.04^{de}	0.25 ± 0.02^{bc}	0.23 ± 0.01^{abc}
	4	0.52 ± 0.01^{h}	0.34 ± 0.02^{g}	0.31 ± 0.02^{fg}	0.25 ± 0.01^{bc}
Root-to-shoot ratio	0	0.15 ± 0.07^{bc}	0.13 ± 0.05^{ab}	0.12 ± 0.08^{a}	0.11 ± 0.09^{a}
	2	0.18 ± 0.01^{d}	0.16 ± 0.03^{c}	0.15 ± 0.04^{bc}	0.15 ± 0.05^{ab}
	4	0.19 ± 0.02^{d}	0.16 ± 0.03^{c}	0.15 ± 0.03^{bc}	0.14 ± 0.05^{ab}

Note: The mean (n = 5) value followed by the same letter of each parameter indicates no significant difference at a 95% confidence level based on ANOVA and DMRT

salinity treatments. The results of this study indicate that silicon application can help maintain the growth of *A. cepa* under salinity stress.

Dry weight is a widely recognized indicator of growth, whether it pertains to the entire plant or individual components. Plant dry weight accounts for approximately 90% of the photosynthetic products. The analysis of growth through dry weight measurement aims to assess the capacity of plants to generate photosynthates (Table 9). Treatment involving elevated salt concentrations generally leads to heightened accumulation of Na^+ and Cl^-, alongside a reduction in specific cations, including K^+ and Ca^{2+} (Alharbi et al., 2022). Elevated Na^+ concentrations can adversely affect plant cells by impairing cellular metabolism, inhibiting growth, and promoting excessive ROS production (Munns, 2002). Under salinity stress, plants are required to allocate extra cellular resources to sustain elevated cytosolic K^+ levels and reduced Na^+ concentrations. Silicon application can decrease sodium ion accumulation in roots and/or shoots. The improvement of salinity tolerance in plants is contingent upon this mechanism, which is caused by silicon (Assaha et al., 2017).

The height of shallot plants is substantially reduced by salinity stress (Trisnaningsih et al., 2023). Silicon treatment in $CaSiO_3$ can sustain plant height and the number of shallot leaves in saline soil conditions (Indarwati et al., 2021). Syamsiah et al. (2020) have demonstrated that local shallot varieties can tolerate salinity levels of irrigation water as high as 3 dS m^{-1}. According to Venâncio et al. (2022), silicon fertilization has the potential to enhance yield and mitigate the detrimental impacts of salinity on shallot growth. The morphology of roots may be advantageously affected by the administration of silicon (Tripathi et al., 2021).

The number of leaves decreased because of the salinity stress treatment. This reduction leads to a redistribution of assimilates from new leaves to other parts of the plant (Ahmed, 2022). This assertion is corroborated by Trouvelot et al. (2014), who state that plants utilize carbohydrates

generated through photosynthesis during the division of meristem cells. The shoot meristem generates new leaves from primordial leaves that are formed. This process occurs when the meristem is engaged in cell division, resulting in the development of leaf seedlings from the shoots. In general, the growth of roots and branches is influenced by salinity and $CaSiO_3$ stress. This is the consequence of the potential for a decrease in root dry weight in conjunction with an equivalent shoot dry weight. This study compared the reduction in root dry weight with the reduction in shoot dry weight.

CONCLUSIONS

The application of $CaSiO_3$ effectively mitigates salt stress in *A. cepa* by enhancing antioxidant defense (SOD activity and membrane stability), improving photosynthetic performance, and promoting overall growth and yield components. Under salinity stress, $CaSiO_3$ application reduced proline and H_2O_2 accumulation while improving physiological efficiency. These findings suggest that applying 4 mM $CaSiO_3$ can be recommended to enhance shallot productivity in saline coastal soils of Indonesia. Future studies should focus on field-scale validation, long-term soil effects, and variety responses to $CaSiO_3$ under varying salinity levels.

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