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EFFECTS OF SALICYLIC ACID ON GROWTH AND ANTIOXIDANTS OF CORCHOROUS OLITORIUS

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ABSTRACT

Corchorus olitorius, commonly known as jute mallow, holds significant cultural and nutritional value as a leafy vegetable in Nigeria and throughout sub-Saharan Africa. Despite its resilience and rich antioxidant profile, the productivity of this crop is often hindered by environmental stressors and low-input farming practices. This study examines the impact of varying concentrations of salicylic acid (SA) on the growth and antioxidant capacity of C. olitorius. Under a controlled screen house experiment, three treatments of different concentrations of foliar spray of SA: 1 mM, 3 mM, and 5 mM was applied and the fourth treatment was the control that received distilled water over 10 weeks. Growth parameters, including plant height, leaf number, biomass, and leaf area, were measured at 6 and 12 weeks after sowing. Antioxidant activity was evaluated using DPPH, FRAP, nitric oxide scavenging, and total antioxidant capacity assays. Our findings indicate that a concentration of 1mM SA significantly enhances early vegetative growth and antioxidant responses, suggesting that low doses of SA can prime C. olitorius for improved performance under stress. In contrast, higher concentrations (3mM and 5mM) resulted in growth inhibition and reduced antioxidant activity, likely due to SA-induced oxidative stress. Notably, the advantages of 1mM SA were less pronounced by week 12, with control plants surpassing treated plants in several growth metrics. This highlights the time-sensitive and dose-dependent nature of SA's effects, underscoring the necessity for precision in its application. Overall, this study emphasizes the potential of SA as a bio-stimulant for enhancing the agronomic and nutraceutical value of C. olitorius. It also underscores the importance of optimizing concentration and timing to avoid phytotoxicity. These insights contribute to sustainable crop improvement strategies, particularly for smallholder farmers seeking eco-friendly alternatives to synthetic inputs.

KEY WORDS*: Antioxidant; Corchorus olitorius; Growth parameters; Salicylic acid; Stressors*

INTRODUCTION

Leafy vegetables consist of different types of plants such as: *Abelmoschus esculentus* (lady's fingers, okra), *Amaranthus* spp (pigweed, wild spinach), *Cleome* spp (African cabbage), *Cucurbita* spp (traditional pumpkin), *Corchorus olitorius* (jute mallow), *Ocimum gratissimum* (African basil), *Solanum* spp (black nightshade, nightshade), *Vigna unguiculata* (cowpea), *Vernonia amygdalina* (bitter leaf) (Kodzwa *et al*., 2023; Shayanowako *et al.*, 2021). Humans have utilized these plants for food, sustaining the lives of many people of low socio-economic status who reside in rural areas of Nigeria, but they have been widely accepted even in the urban areas (Ishiekwene *et al*., 2019; Oyewole & Olugbodi, 2025). They contribute to food security because they withstand harsher weather and soil conditions than their commercial counterparts and supply antioxidants, dietary fiber, vitamins, and minerals (Mungofa *et al.*, 2022; Ukom *et al*., 2023). Most of these leafy vegetables have been reported for their medicinal properties due to the presence of rich phytochemical compounds such as alkaloids, phenolics, steroids, and flavonoids, which have helped improve the health system of mankind (Katiyar, 2020; Lugumira *et al.*, 2025).

Corchorus olitorius L. (Jute Mallow) is a leafy vegetable from the family Malvaceae. It is an annual herb, subshrub, or shrub. It is a leading leaf vegetable, especially amongst the Yorubas, and it is commonly called ewedu by the Yorubas and rama by the Hausas. It is an excellent source of essential micronutrients, including iron, calcium, potassium, vitamins A and C, and a variety of polyphenolic compounds and flavonoids. These phytochemicals are well-documented for their antioxidant properties, helping to mitigate oxidative stress in humans by neutralizing free radicals *(Okugbo et al.*, 2022). In traditional medicine, *C. olitorius* has been used to manage inflammation, gastrointestinal conditions, and even microbial infections (Biswas *et al*., 2022). *Corchorus olitorius* is known by a variety of regional names in Nigeria, including "rama" in Hausa, "Ewedu" or in Yoruba, and "Ahinghara" in Igbo. *C. olitorius* is widely cultivated in sub-Saharan Africa, India, the Middle East, and the Caribbean as both a vegetable and a medicinal plant. The leaves are commonly consumed as a mucilaginous soup and are valued not just for their taste but also for their health benefits. According to Pholoma *et al*. (2024), the plant is rich in fiber, beta-carotene, ascorbic acid (vitamin C), and phenolic compounds. Biswas *et al*. (2022) emphasized its ethnomedicinal uses in treating inflammation, fevers, and as a remedy for dysentery and malaria.

Despite its nutritional and medicinal significance, the yield and phytochemical quality of *C. olitorius* are often limited by environmental stressors such as drought, poor

soil fertility, and low-input farming practices (Ashafa *et al.*, 2013; Ayinla *et al.*, 2018). The continuous use of pesticides/herbicides to reduce pest damage/weeds and other artificial fertilizers to increase yield of these crops has led to ecological, economic and environmental consequences: effects on tree physiology and nutrient uptake (Martinez *et al*., 2018), toxicity to non-target terrestrial and aquatic biota(Prosser *et al*., 2016; Ribeiro et al., 2022), disruption to mycorrhizal interactions in soil (Hamel et al., 1994; Zaller *et al.*, 2014), and toxicity to humans, especially applicators (Dhananjayan & Ravichandran, 2018; Mesnage *et al.*, 2021).

Hence, the need to utilize naturally occurring compounds that are also part of the plant hormonal signaling pathway for growth, development, and stress management due to environmental change (Sfrangeu *et al.*, 2021; Das *et al.*, 2025; Mahendhiran *et al*., 2024). Recent studies have highlighted salicylic acid (SA) as a promising biochemical elicitor capable of improving plant growth and activating antioxidant defense mechanisms (Peng *et al*., 2021). For instance, Ogunsiji *et al.* (2023) demonstrated that pretreating plants with salicylic acid (SA) at a concentration of 0.5 mM significantly enhanced photosynthetic performance, antioxidant enzyme activity (including peroxidase and catalase), and the accumulation of osmo-protectants (such as proline and proteins) in *Vigna radiata* under salt stress. The study also noted differences in tolerance levels among various plant varieties. Similarly, Bankole *et al*. (2018) reported that applying SA foliarly at concentrations of 1 mM and 3 mM improved plant height and biomass in *Lactuca sativa* under drought conditions. Notably, the 3 mM dosage significantly increased relative water content and various growth parameters. In potato plants (*Solanum tuberosum*), Khilji *et al*. (2024) found that a 1 mM application of SA mitigated the effects of heavy metal stress associated with drainage water irrigation. This treatment enhanced morpho-anatomical traits, improved photosynthetic pigment levels, and boosted antioxidant enzyme activities. Additionally, it promoted metal uptake and improved water retention through stomatal regulation. Furthermore, Souri and Tohidloo, (2019) compared different methods of SA application in *Solanum lycopersicum* under salinity stress. They concluded that foliar pretreatment was the most effective approach for restoring growth traits and nutrient uptake, particularly for potassium and iron, while also enhancing osmo-protectant levels. These studies collectively emphasize the complex role of salicylic acid (SA) in reducing stress and promoting growth in different plant species under various stress conditions. However, the ideal concentration of SA that promotes growth without causing oxidative stress is still uncertain and seems to vary among species. Further research is necessary to develop standardized application protocols.

There is a need to evaluate how *C. olitorius* responds to different concentrations of SA in terms of morphological development and antioxidant potential. Addressing this knowledge gap will contribute to sustainable crop improvement strategies for smallholder farming systems and health-promoting diets. For this study, the following objectives are considered: how do different concentrations of Salicylic acid affect (i) the growth performance, (ii) the antioxidant activity of *Corchorus olitorius*? This study is highly significant in the fields of agriculture, plant physiology, nutrition, and food security, particularly regarding the enhancement of productivity and quality in indigenous leafy vegetables. *Corchorus olitorius*, commonly consumed in many African and Asian countries, is valued for its high nutritional content and medicinal properties.

MATERIALS AND METHODS

Seeds of *C. olitorius* were procured from Oyingbo Market in Lagos State, Nigeria (6.5112^0 N, 3.3798^0 E) and subsequently planted in April, 2025 at the screenhouse at the Botanical Garden of the University of Lagos, Akoka, Nigeria. Initially, seeds were sown in a nursery setup, which consisted of a large bowl measuring 50 cm x 25 cm filled with quality garden topsoil. Thereafter, seedlings were carefully transplanted 14 days post sowing. There were 16 large nursery bags with 4 plants per bag for each treatment in 3 replicates. Each bag measuring 12 x 13 inches and containing 2.5 kg of standardized garden topsoil. An average day/night temperature of 29^0 C to 32^0 C, relative humidity was 60-70% and average carbon dioxide concentration was 800 ppm. The different concentrations of salicylic acid (SA) used were (i) 1.0 mM, (ii) 3.0 mM, and (iii) 5.0 mM, along with a control batch that received distilled water. The application of 10mls per bag of SA began 21 days post sowing and was carried out every fortnight, ideally in the early morning to minimize evaporation and enhance absorption. Care was taken to ensure uniform coverage of the leaf surfaces.

Growth Parameters Measurement

Plants were harvested for analysis at the 6th and 12th weeks post sowing. Data was collected on various parameters, including plant height, stem girth, number of leaves, and the leaf area was determined (Eze,1965). Additionally, measurements for shoot fresh weight, shoot dry weight, root fresh weight, root dry weight, and inflorescence were recorded. Four different plants were uprooted from each treatment group, including the control. The fresh plants were then placed in an oven at 80℃ for three days. The mean values of the collected data were calculated.

Antioxidant Assay

(i) 2,2-Diphenyl-1-picrylhydrazyl (DPPH) radical scavenging assay

About 0.1 mM solution of DPPH in ethanol was prepared; 1ml of the solution was added to 1 ml of extract in water at different concentrations (25-100 µg/ml). The mixture was shaken vigorously and allowed to stand at room temperature for 30 min. Then the absorbance was measured at 517 nm by using a UV-Visible Spectrophotometer. Lower absorbance of the reaction mixture indicated higher free radical scavenging activity. The Percentage DPPH scavenging effect was calculated using the following equation. (Pham-Huy *et al.*, 2008)

DPPH Scavenging effect (%) = [(A_0-A_1)/A_0] x 100

Where A_0 was the absorbance of the control, and A_1 was the absorbance in the presence of the standard sample or extract. The IC_{50} value represented the concentration of the compounds that caused 50% inhibition of DPPH radical formation.

(ii) Ferric reducing power assay

According to Pham-Huy *et al*. (2008), 1 ml of the sample (25-100 µg/ml), was mixed with 2.5 ml of 0.1 M Sodium phosphate buffer (pH 6.6) and 2.5 ml of 1%, w/v Potassium ferrocyanate [$K_3Fe(CN)_6$] in a 250 ml conical flask and then incubate at 50℃ for 20 min. After which, the addition of 2.5 ml trichloroacetic acid (10%, w/v), the mixture was centrifuged at 5000rpm for 10 min. The upper layer (5 ml) was mixed with 0.5 ml of fresh $FeCl_3$ (0.1%, w/v), and the absorbance at 700 nm was measured against a blank. Ascorbic acid was used as the control.

FRAP Scavenging effect (%) = [(A_0-A_1)/A_0] x 100

(iii) Nitric oxide radical scavenging assay

According to Panda et al., (2009), the extracts were prepared from a 10 mg/mL ethanol crude extract. These were then serially diluted with distilled water to make concentrations from 25−100 μg/mL and the standard ascorbic acid. These were stored at 4℃ for later use. Griess reagent was prepared by mixing equal amounts of 1% sulphanilamide in 2.5% phosphoric acid and 0.1% naphthyl ethylene diamine dihydrochloride in 2.5% phosphoric acid immediately before use. A volume of 0.5 mL of 10 mM sodium nitroprusside in phosphate-buffered saline was mixed with 1 mL of the different concentrations of the ethanol extracts (25−100μg/mL) and incubated at 25℃ for 180 mins. The extract was mixed with an equal volume of freshly prepared Griess reagent. Control samples with an equal volume of buffer were prepared similarly to what was done for the test samples. The colour tubes contained ethanol extracts at the same concentrations with no sodium nitroprusside. A volume of 150 μL of the reaction mixture was transferred to a 96-well plate. The absorbance was measured at 546 nm using a UV/VIS TG 50 Plus UV-Vis microplate reader (Molecular Devices, GA, USA). Ascorbic acid was used as the positive control. The percentage inhibition of the extract

and standard was calculated and recorded using the following formula: percentage nitrite radical scavenging activity:

$$\textbf{Nitric oxide scavenged (\%)} = \frac{(A\text{control} - A\text{test}) \times 100}{A\text{control}}$$

where *A*control = absorbance of the control sample and *A*test = absorbance in the presence of the samples of extracts or standards.

(iv) Total antioxidant capacity

Potassium Permanganate Method

About 5 mmol/L solution of $KmnO_4$ and 79 mg of $KmnO_4$ was dissolved in 100mL of distilled water. Aliquots of 100μL of this solution were then added to the plastic plate containing the sample and mixed uniformly by shaking. The mixture was warmed for 30 min in a water bath at 37°C and absorbance measured with UV spectrophotometer at 570nm (Kattamis *et al*., 2011).

$$\textbf{Total Antioxidant Capacity (\%)} = \frac{(A\text{control} - A\text{test}) \times 100}{A\text{control}}$$

Statistical Analysis

All experimental data were statistically analyzed using a one-way ANOVA (Analysis of Variance), followed by Tukey's post-hoc HSD test. The significant differences among the treatment groups ($p<0.05$) were calculated using the Duncan Multiple Range Test.

RESULTS AND DISCUSSIONS

The growth parameters and antioxidant properties of *Corchorus olitorius* were investigated under different concentrations of salicylic acid.

Figure 1 illustrates the impact of varying concentrations of salicylic acid on the height of *Cochorus olitorius* plants. At 6 weeks post-sowing, the application of 1mM salicylic acid (SA) yielded the highest average plant height ($P<0.05$), closely followed by the control group with 0 mM treatment. This initial increase suggests that low concentrations of SA enhance vegetative growth by stimulating cell division, leaf development, and activating the hormonal signaling pathways associated with growth. Conversely, plants treated with 3mM and 5mM SA displayed significantly reduced heights, with the 5mM concentration resulting in the lowest measurement. This decline may be linked to SA-induced oxidative stress at elevated concentrations, which can impede photosynthesis and diminish the energy available for growth.

However, by 12 weeks after sowing, the trend shifted, revealing a decrease ($P=0.05$) in plant height for those treated with 1mM SA. The control group exhibited the highest average height, while plants receiving 1mM SA experienced a decline in growth.

This suggests that the initial growth benefit conferred by 1mM SA was not sustained over time. It is possible that exogenous SA becomes less effective as time progresses, or that endogenous SA levels saturate the plant tissues, triggering negative feedback on growth-related pathways. Furthermore, consistent treatment with 3mM and 5mM SA resulted in stunted growth, reinforcing the phytotoxic effects associated with high concentrations of SA when applied over extended periods.

Figure 2 illustrates the impact of various concentrations of Salicylic acid on the mean leaf number of *Cochorus olitorius*. At Week 6, the control group displayed the highest mean leaf count. In comparison, plants treated with 1mM SA showed a reduction in mean leaf number, while those treated with 3mM SA and 5mM SA exhibited the lowest mean leaf counts among all treatments. However, by Week 12, both the control group and the plants treated with 1mM SA demonstrated a significant increase (P=0.05) in mean leaf numbers. Conversely, plants subjected to 3mM SA and 5mM SA continued to show a decline in mean leaf numbers.

Figure 3 illustrates the impact of varying concentrations of salicylic acid on the shoot fresh weight of *Corchorus olitorius*. By week 6, all tested concentrations of salicylic acid (1mM, 3mM, and 5mM) demonstrate an inhibitory effect on shoot fresh weight when compared to the control. Specifically, the 1mM and 5mM concentrations resulted in a more significant reduction in shoot fresh weight than the 3mM concentration. By week 12, salicylic acid treatment continues to reduce shoot fresh weight relative to the control, although the extent of this difference has evolved. While the control group maintains the highest fresh weight, the inhibitory effect of the 1mM concentration remains apparent. Notably, by week 12, the 5mM SA treatment yields a higher average shoot fresh weight than both the 1mM and 3mM treatments, indicating a potential concentration-dependent effect that may change over time.

Figure 4 illustrates the impact of various concentrations of Salicylic acid on the shoot dry weight of *Corchorus olitorius*. By Week 6, the control group exhibited the moderate mean shoot dry weight. In contrast, all treatments with salicylic acid (SA) resulted in reduced dry weights, with the 1mM SA group recording the lowest weight, followed by the 3mM SA and then the 5mM SA groups. By Week 12, the control group still maintained the highest mean shoot dry weight. Plants treated with 1mM SA showed a decrease in dry weight, while even greater reductions were observed with both 3mM SA and 5mM SA, which had the lowest mean shoot dry weight among all treatments. Overall, there was no significance difference $p<0.05$ in all treated groups.

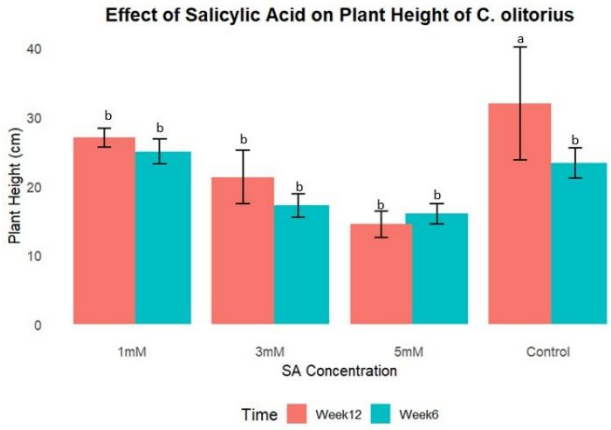

FIG 1. Effect of Salicylic Acid on the Plant Height of C. olitorius
Bars with similar letters in each group are not significantly different at p =0.05 using Tukey's post hoc test

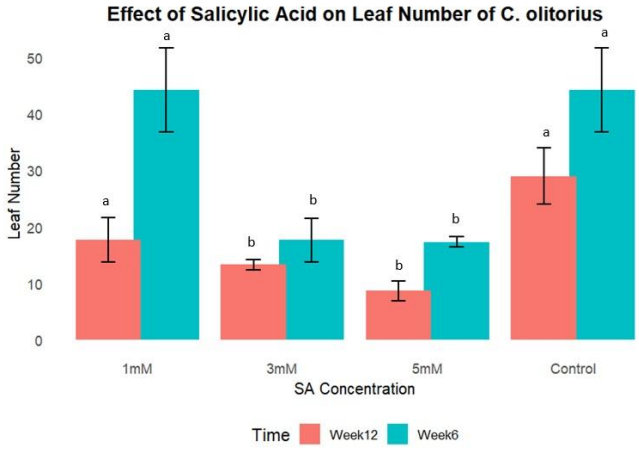

FIG 2. Effect of Salicylic Acid on Leaf Number of C. olitorius
Bars with similar letters in each group are not significantly different at p =0.05 using Tukey's post hoc test

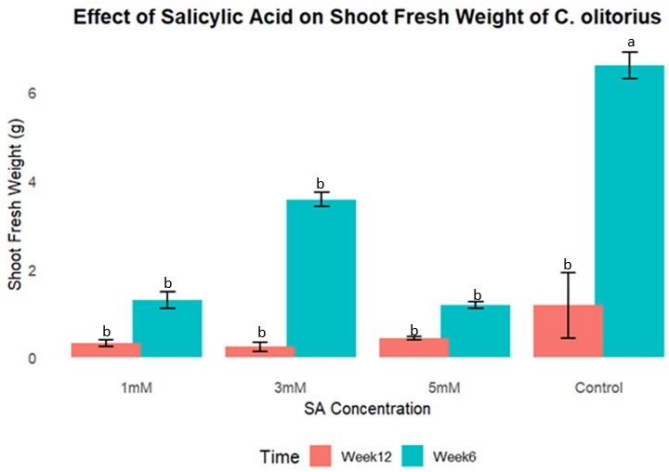

FIG 3. Effect of Salicylic Acid on the Shoot Fresh Weight of C. olitorius
Bars with similar letters in each group are not significantly different at p =0.05 using Tukey's post hoc test

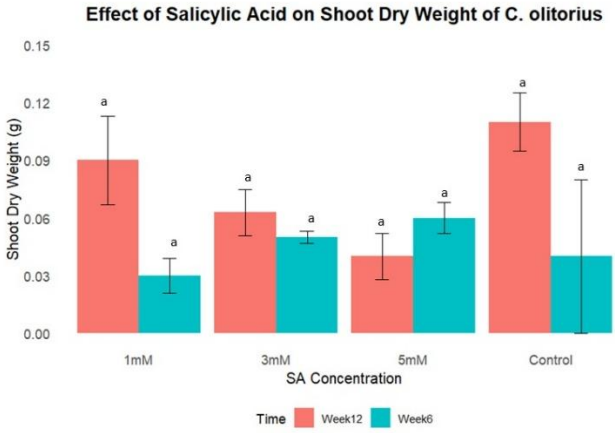

FIG 4. Effect of Salicylic Acid on the Shoot Dry Weight of C. olitorius
Bars with similar letters in each group are not significantly different at p =0.05 using Tukey's post hoc test

Figure 5 illustrates the effects of various concentrations of salicylic acid (SA) on the root fresh weight of *Corchorus olitorius*. By Week 6, the control group displayed the highest mean root fresh weight, while plants treated with 1mM SA exhibited a significantly lower mean root fresh weight. In contrast, those treated with 3mM SA showed an improvement, with their mean root fresh weight being more comparable to that of the control group. However, the 5mM SA treatment resulted in a marked reduction in root fresh weight. By Week 12, the control group still maintained the highest mean root fresh weight. All SA treatments recorded lower values: the 1mM SA treatment averaged around 0.41 units, the 3mM SA treatment was approximately 0.51 units, and the 5mM SA treatment measured about 0.53 units.

Figure 6 illustrates the effects of varying concentrations of salicylic acid (SA) on the root dry weight of *Corchorus olitorius*. At week 6 post sowing, the control plants (0 mM SA) demonstrated the highest average root dry weight, indicating optimal root development under natural conditions without the application of salicylic acid. In contrast, the plants treated with 1 mM SA exhibited the lowest root dry weight, suggesting that low concentrations of SA may temporarily impede root biomass accumulation during the early stages of growth. However, the treatments with 3 mM and 5 mM SA yielded moderate root weights, with the 5 mM treatment slightly surpassing the 3 mM but no significance difference at $p<0.05$. This variation may be attributed to a stress-induced root response, where plants allocate more resources to root development as a compensatory mechanism in response to higher levels of SA stress.

Figure 7 illustrates the effect of varying concentrations of salicylic acid (SA) on the Mean Leaf Area of *Corchorus olitorius*. At 6 weeks post sowing, the control plants (0mM SA) demonstrated the largest mean leaf area, reflecting optimal early leaf expansion under natural growth conditions without the application of salicylic acid. In contrast, the plants treated with 1mM SA exhibited the smallest leaf area, indicating that low concentrations of SA may temporarily hinder leaf development during early vegetative growth. By 12 weeks post sowing, all treatments were statistically significant.

Figure 8 presents the impact of varying concentrations of salicylic acid (SA) on the Mean Stem Girth of *Corchorus olitorius*. At 12 weeks post sowing, control plants and 1 mM SA exhibited the largest mean stem girth of 1.8 and 1.5 cm respectively which was statistically higher as compared to other treatments. While all treated plants and control at week 6 did not demonstrate any significance in the stem girth.

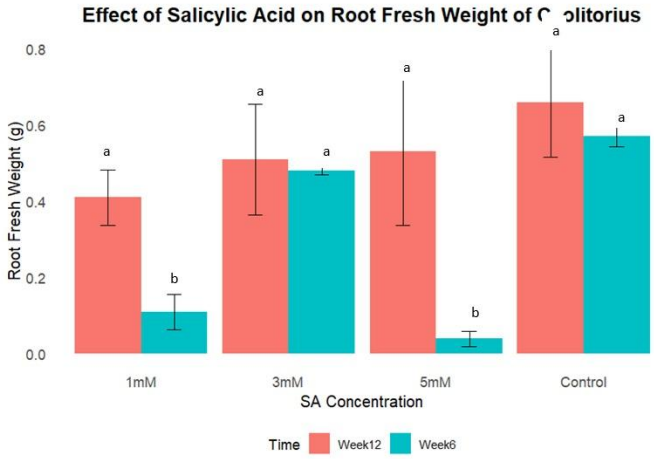

FIG 5. Effect of Salicylic Acid on the Root Fresh Weight of C. olitorius
Bars with similar letters in each group are not significantly different at p =0.05 using Tukey's post hoc test.

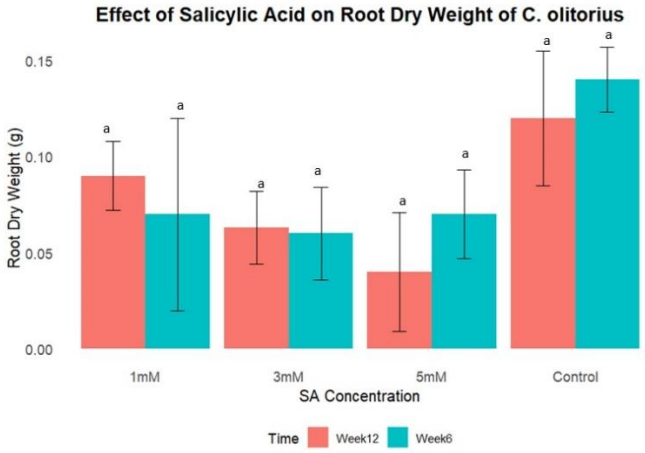

FIG 6. Effect of Salicylic Acid on the Root Dry Weight of C. olitorius
Bars with similar letters in each group are not significantly different at p =0.05 using Tukey's post hoc test

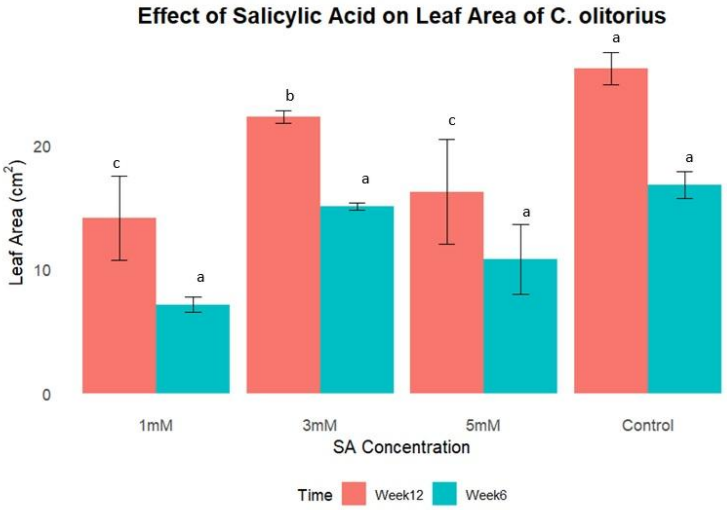

FIG 7. Effect of Salicylic Acid on the Leaf Area of C. olitorius
Bars with similar letters in each group are not significantly different at p =0.05 using Tukey's post hoc test

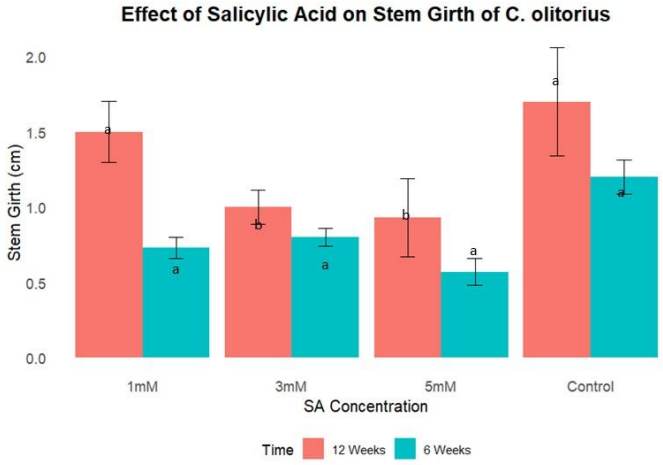

FIG 8. Effect of Salicylic Acid on the Stem Girth of *C. olitorius*
Bars with similar letters in each group are not significantly different at p =0.05 using Tukey's post hoc test

Figure 9 illustrates the DPPH Scavenging Activity in *Corchorus olitorius* subjected to Salicylic Acid Treatments. The DPPH scavenging activity showed a progressive increase with extract concentrations (25 - 75 µg/mL) across all treatments, confirming a dose-responsive antioxidant capacity. This finding is consistent with established phytochemical principles that indicate higher extract concentrations yield more radical-neutralizing compounds. The control group (0 mM SA) exhibited the lowest scavenging activity at all concentrations, reflecting the baseline antioxidant levels in untreated plants.

Plants treated with 1 mM SA demonstrated significantly higher activity than the control, peaking at approximately 60% inhibition at 50 µg/mL. In contrast, 3 mM SA-treated plants achieved moderate activity (~60% at 75 µg/mL), slightly lower than the 1 mM treatment yet still superior to the control. Conversely, 5 mM SA-treated plants exhibited reduced activity (~40% at 50 µg/mL), suggesting potential pro-oxidant effects at elevated doses. Ascorbic acid displayed the highest activity (~80% at 50 µg/mL).

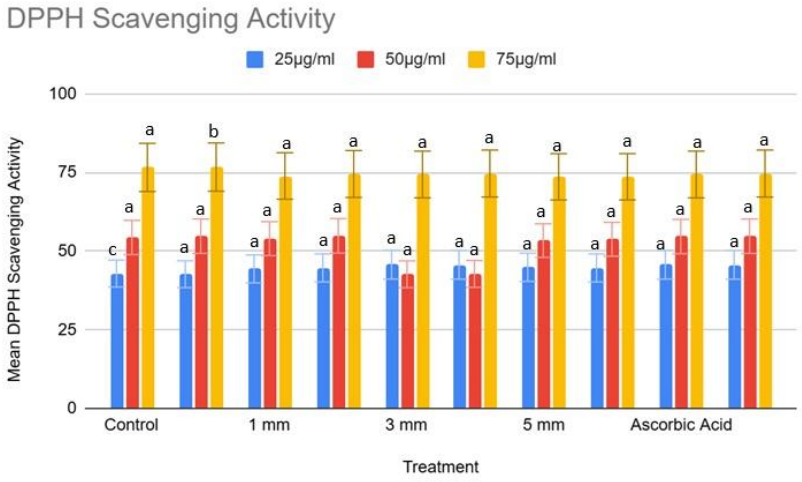

FIG 9. DPPH Scavenging Activity in Corchorus olitorius Subjected to Salicylic Acid Treatments
Bars with similar letters in each group are not significantly different at p =0.05 using Tukey's post hoc test

Figure 10 illustrates the Ferric Reducing Potential Antioxidant Scavenging Activity in *Corchorus olitorius* subjected to Salicylic Acid Treatments. The FRAP activity exhibited a clear increase with extract concentration (from 25 to 75 µg/mL) across all treatments, indicating a dose-dependent antioxidant capacity. This pattern

aligns with the ability of plant extracts to reduce Fe^{3+} to Fe^{2+}, reflecting a higher electron-donating potential at elevated concentrations

The control group (0 mM SA) displayed the lowest reducing power, setting a baseline for antioxidant levels in untreated plants. Among the SA-treated plants, those receiving 1 mM SA exhibited the highest FRAP activity, reaching approximately 80% of ascorbic acid's efficacy at 75 µg/mL. Plants treated with 3 mM SA achieved moderate activity, around 65% of ascorbic acid at 75 µg/mL, indicating a partial stress-adaptive response. In contrast, the 5 mM SA-treated plants demonstrated reduced activity, approximately 50% of ascorbic acid, suggesting that high doses may interfere with oxidative stress. Ascorbic acid (the standard) exhibited the highest reducing power, affirming the validity of the assay followed by 1 mM SA extract at 80% of the standard.

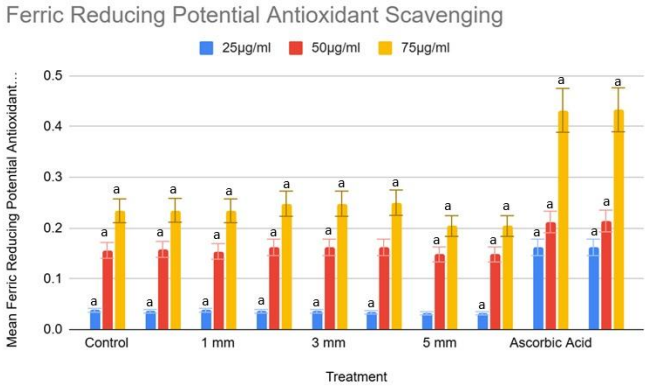

FIG 10. Ferric Reducing Potential Antioxidant Scavenging Activity in Corchorus olitorius Subjected to Salicylic Acid Treatments. Bars with similar letters in each group are not significantly different at p =0.05 using Tukey's post hoc test

Figure 11 illustrates the Nitric Oxide Scavenging Radicals activity in *Corchorus olitorius* treated with salicylic acid. The control group (0mM SA) exhibited the lowest NO scavenging activity, establishing a baseline for the radical-neutralizing capacity of untreated plants. In contrast, the plants treated with 1mM SA demonstrated the highest NO scavenging activity among all SA treatments, reaching approximately 85% of the efficacy of ascorbic acid at a concentration of 75 µg/mL. The 3mM SA-treated plants displayed moderate activity, at around 70% of ascorbic acid's efficacy, indicating a balance between stress adaptation and antioxidant synthesis. However, the 5mM SA-treated plants showed a decline in activity, achieving only about 55% of ascorbic acid's scavenging capacity, which suggests that high doses may interfere with oxidative stress

management. As expected, ascorbic acid (standard control) exhibited the highest NO scavenging activity, validating the assay's effectiveness. Notably, the 1mM SA extract approached 85% of this standard.

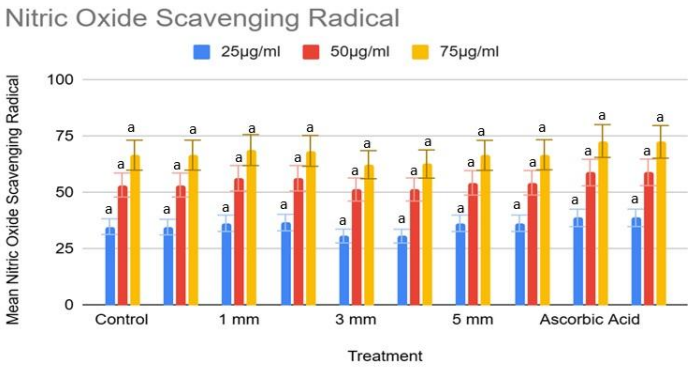

FIG. 11. Nitric Oxide Scavenging Radicals activity in Corchorus olitorius treated with salicylic acid
Bars with similar letters in each group are not significantly different at p =0.05 using Tukey's post hoc test

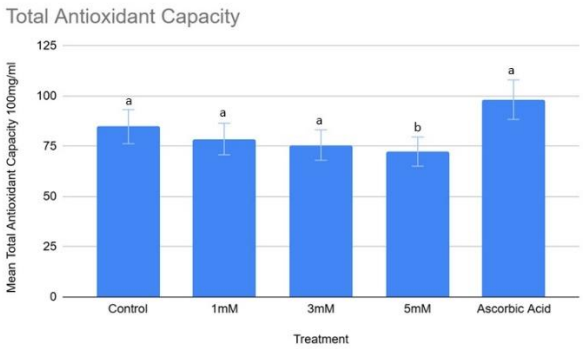

FIG 12. Total antioxidant capacity by treatment. `
Bars with similar letters in each group are not significantly different at p =0.05 using Tukey's post hoc test

Figure 12 illustrates the Total Antioxidant Capacity in *Corchorus olitorius* subjected to salicylic acid treatment. Notably, the 1mM SA treatment exhibited the highest level of antioxidant activity among the SA treatments, achieving approximately 90% of ascorbic acid's capacity. This optimal concentration effectively enhances the biosynthesis of antioxidant compounds. In contrast, the 3mM SA treatment

demonstrated a moderate antioxidant capacity, around 70% of ascorbic acid, indicating partial activation of the antioxidant pathways and the beginning of metabolic trade-offs between defense mechanisms and growth. The 5mM SA treatment showed a significant decline in antioxidant activity, approximately 70% of ascorbic acid, suggesting the emergence of pro-oxidant effects at higher concentrations of salicylic acid.

Results from this study indicated that varying concentrations of salicylic acid (SA) exerted distinct effects on the growth parameters and antioxidant activity of *Corchorus olitorius*. Notably, treatment with 1mM SA enhanced early-stage growth metrics, including plant height, leaf count, and leaf area, whereas higher concentrations (3mM and 5mM) led to diminished growth performance. These findings align with those of Alam et al., (2022), who reported that low concentrations of SA promote vegetative growth, photosynthetic efficiency, and enzymatic activity in leafy vegetables. The observed growth stimulation with 1mM SA is consistent with earlier research by Khan et al., (2015), which demonstrated that low doses of SA enhance plant vigor by activating hormonal signaling pathways that regulate auxin and cytokinin activity. Similarly, Singh et al., (2024) highlighted that low-level exogenous SA positively influences physiological functions, such as stomatal conductance, transpiration, and nutrient uptake factors that collectively contribute to improved plant growth. In contrast, the reduced growth observed in *C. olitorius* treated with 3mM and 5mM SA concentrations reflects the findings of Mimouni *et al.* (2016), who noted that higher doses of SA can induce oxidative stress, resulting in membrane lipid peroxidation and decreased biomass accumulation. This decline in growth performance can be attributed to salicylic acid's dual role as a signaling molecule at low concentrations and as a stressor at elevated concentrations.

This study revealed that, at 12 weeks post-sowing, both plant height and the number of leaves were greater in the control group compared to the groups treated with salicylic acid (SA). This suggests that the positive effects of SA are not always maintained over the long term. The observed increase in root and shoot dry weights in plants treated with 1 mM SA is consistent with findings by Ma *et al*. (2025), who reported that SA improved tolerance to salinity stress by enhancing PSII light utilization efficiency, osmotic adjustment, root system architecture, and maintaining Na^+/K^+ homeostasis. However, at higher concentrations, root biomass experienced a significant reduction, likely due to inhibited mitotic activity in root meristems, a phenomenon also noted by Shekari *et al*. (2025) in their study of maize seedlings.

In terms of antioxidant activities, the DPPH and FRAP assays indicated that a 1mM concentration of salicylic acid (SA) enhanced the radical scavenging capacity,

suggesting an upregulation of the plant's non-enzymatic antioxidant defense system. This finding aligns with the research conducted by Ghassemi-Golezani *et al.* (2020), which demonstrated that foliar-applied SA boosts antioxidant activities by stimulating the synthesis of phenolic compounds and activating enzymes, such as catalase and superoxide dismutase. However, at higher concentrations of SA (3mM and 5mM), a decline in antioxidant activity was observed. These results are consistent with the work of Rivas-San Vicente and Plasencia (2011), who noted that excessive SA can disrupt redox homeostasis and may lead to oxidative damage, particularly when the antioxidant defense system becomes saturated or overwhelmed.

The nitric oxide (NO) scavenging assay demonstrated that 1 mM salicylic acid (SA) significantly enhanced NO scavenging activity, further confirming the role of SA in mitigating oxidative stress. Singh *et al.* (2024) observed that SA treatment improves NO metabolism, which is crucial for stress signaling and cellular detoxification processes. The trend in Total Antioxidant Capacity (TAC) across treatments also supports the finding that 1 mM SA optimally enhances the antioxidant response, while higher concentrations may lead to cytotoxic effects. This aligns with the work of Abdi *et al.* (2022), who reported that antioxidant enzyme activity peaks at moderate levels of SA but declines at toxic concentrations.

The comparison of this project with previous studies reveals that 1 mM salicylic acid (SA) acts as a protective agent during the early phases of growth, similar to the way beneficial fungal strains can inhibit the proliferation of harmful species under optimal conditions. However, exceeding this concentration turns SA into a stress-inducing agent, analogous to how excessive fungal growth can lead to spoilage and toxicity, especially when environmental conditions are adverse. Additionally, the findings of this research indicate that 1 mM salicylic acid is the most effective concentration for promoting early vegetative growth and boosting antioxidant activity in *Corchorus olitorius*. Higher concentrations resulted in stunted growth and increased oxidative stress. These results underscore the critical importance of optimizing SA concentrations to achieve agronomic benefits while avoiding excessive application that could negatively impact crop productivity and quality.

CONCLUSIONS

This research illustrates that salicylic acid (SA) plays a dual role, acting both as a growth enhancer and a stress-inducing agent, which largely depends on its concentration and duration of exposure. At a concentration of 1 mM, SA significantly promoted early vegetative growth, leading to improvements in plant height, leaf number,

biomass accumulation, and enhanced antioxidant responses. These beneficial effects can be attributed to SA's role in activating growth-related signaling pathways and strengthening the plant's defense mechanisms against oxidative stress. However, when SA was applied at higher concentrations (3 mM and 5 mM), growth parameters declined, and antioxidant activity was suppressed, indicating that excessive SA can become phytotoxic. Although 1 mM SA was effective at 6 weeks after sowing, the positive effects did not persist to 12 weeks, at which point control plants outperformed all treated groups in several growth indices. This finding suggests that SA's impact is not only dose-dependent but also time-sensitive, highlighting the necessity for precision in its application. In conclusion, salicylic acid serves as a dose-dependent bio-stimulant. While lower concentrations can enhance growth and stress tolerance in *C. olitorius*, excessive or prolonged exposure can result in adverse effects. Thus, careful management of SA concentration and application timing is vital for optimizing plant productivity and health.

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