Research Square

Preprints are preliminary reports that have not undergone peer review.
They should not be considered conclusive, used to inform clinical practice,
or referenced by the media as validated information.

Nitrate-dominated nutrition delays bolting and improves yield of hydroponic lettuce: interactive roles of ammonium:nitrate ratios and salicylic acid

Srwa GHAFWR MUHAMMED MUHAMMED
Harran University
Selçuk SÖYLEMEZ
ssoylemez@harran.edu.tr

Harran University

Research Article

Keywords: Lactuca sativa L., Nitrogen forms, Ammonium–nitrate ratios, Flowering regulation, Salicylic acid, Hydroponic cultivation

Posted Date: April 13th, 2026

DOI: https://doi.org/10.21203/rs.3.rs-9202614/v1

License: This work is licensed under a Creative Commons Attribution 4.0 International License.
Read Full License

Additional Declarations: No competing interests reported.

Abstract

Optimizing the ammonium-to-nitrate ratio in the nutrient solution can enhance lettuce growth performance, yield, and quality by improving nitrogen uptake, assimilation, and utilization. Delayed bolting is highly desirable in lettuce production as it allows prolonged vegetative growth and improved marketable yield. Agronomic strategies that suppress or delay flowering may improve the efficiency of lettuce cultivation. Salicylic acid (SA) is a plant signaling molecule reported to stimulate growth and influence flowering behavior in various crops. This study evaluated the interactive effects of varying ammonium:nitrate (NH_4^+:NO_3^-) ratios and SA concentrations on growth characteristics and bolting time of hydroponically grown lettuce. The experiment consisted of three NH_4^+:NO_3^- ratios (100% NO_3^-:0% NH_4^+, 80% NO_3^-:20% NH_4^+, and 40% NO_3^-:60% NH_4^+) combined with four SA concentrations (0, 0.75, 1.5, and 3.0 mM). Data relating to head fresh weight and diameter, total plant biomass, stem length, diameter and biomass, root length and biomass, total soluble solids, chlorophyll index, and the number of days to bolting were recorded. All measured traits (except total soluble solids) were significantly influenced by nitrogen form, while SA effects were limited and trait dependent. Nitrate-dominated nutrition (100% NO_3^-) delayed bolting by 5–6 days and increased head fresh weight by ~ 42% compared to NH_4^+-containing treatments. A late-season heatwave likely accelerated floral initiation, potentially masking the full effect of SA on bolting regulation. For this reason, future experiments should be conducted under controlled conditions to more precisely assess the role of SA.

Introduction

Lettuce (*Lactuca sativa* L.) is an annual leafy vegetable of high economic importance, primarily cultivated for its edible leaves [1]. It belongs to the Asteraceae family, characterized by species bearing composite inflorescences [2, 3]. Lettuce ranks among the most consumed vegetables globally and is valued for its nutritional properties, although its nutrient composition varies considerably among cultivars. Romaine and leaf lettuce types are known to contain higher levels of vitamins, minerals, antioxidants, and vitamin C, contributing to their health-promoting and anti-carcinogenic properties [4–6].

Increasing demand for sustainable and efficient agricultural practices has encouraged the development of innovative cultivation techniques aimed at improving yield and quality [7]. Hydroponic production systems, in which plants are grown in nutrient solutions without soil, have gained significant attention due to their potential for higher productivity, reduced water consumption, and precise control of environmental and nutritional conditions [8, 9]. These systems also limit soil-borne diseases and allow year-round production, making them suitable for leafy vegetables such as lettuce [10–12].

Nitrogen form is a critical factor influencing plant growth and development in hydroponic systems[13]. The balance between two primary inorganic nitrogen sources available to plants, i.e., ammonium (NH_4^+) and nitrate (NO_3^-), plays a key role in regulating physiological processes, nutrient uptake, and biomass accumulation in lettuce [14, 15]. Premature bolting is undesirable in leafy vegetables such as lettuce and

spinach because it severely reduces marketable yield and compromises quality, leading to significant economic losses. Nitrates and other N-nutrients or their metabolites are known to alter flowering time in plants [16]. Nitrate can also function as a signaling molecule, directing genome-wide gene expression changes that affect plant metabolism, physiology, growth, and development [17]. Nitrate signaling controls plant growth and development using molecular mechanisms that include phytohormone signaling pathways [17, 18]. Nitrate has been reported to promote plant growth in *Arabidopsis* and wheat by modulating the accumulation of DELLA growth suppressors regulated by gibberellin (GA) through a signal transduction pathway [18]. Optimizing the NH_4^+:NO_3^- ratio is therefore essential for improving the performance of hydroponic lettuce [19].

The use of plant growth regulators has emerged as a promising strategy to further enhance crop productivity in addition to nutrient management [20]. Salicylic acid (SA), a naturally occurring phenolic compound, has been reported to influence plant growth, stress tolerance, and flowering behavior [19, 21, 22]. Although previous studies have separately examined the effects of NH_4^+:NO_3^- ratios and SA applications on plant growth and development, information on their combined effects in hydroponically grown lettuce remains limited. Furthermore, the interactive effects of nitrogen form and SA on bolting regulation under hydroponic conditions, particularly in semi-arid greenhouse environments, remain poorly understood.

Hydroponic systems provide an ideal experimental platform for investigating such interactions, as they allow precise manipulation of nutrient composition and environmental factors [23]. Therefore, the objective of this study was to evaluate the effects of different NH_4^+:NO_3^- ratios and SA treatments on growth characteristics and flowering behavior of lettuce grown under hydroponic conditions. The results are expected to contribute to a better understanding of nutrient–hormone interactions and to support the optimization of hydroponic lettuce production.

Materials and Methods

Plant Material and Experimental Conditions

The experiment was conducted during the spring growing season (March to May) 2024 in an unheated greenhouse at the Department of Horticulture, Harran University, Şanlıurfa, Türkiye. The ‘El Maria’ (curly leaf type) lettuce cultivar was used as plant material due to its late-bolting trait and suitability for greenhouse cultivation. Seeds were purchased from Ag Seed Company (Antalya, Türkiye). Seeds were germinated in peat moss, and seedlings were transplanted into the hydroponic system on March 13, 2024, at the 3–4 true leaf stage. The hydroponic setup used the Nutrient Film Technique (NFT), consisting of square PVC channels (100 mm wide × 40 mm high) mounted on 90 cm-high stands with a standard slope to ensure nutrient flow. The nutrient solution was recirculated from a reservoir tank to the root zone via a submersible pump controlled by a timer mechanism.

Treatments and Experimental Design

The experiment was conducted as a randomized complete block design with split-plot arrangements. The NH_4:NO_3 ratios were assigned to main plots, whereas the SA to subplots. Each treatment had three replications, with five plants per replication. Two plants from each replicate were harvested at commercial maturity for yield and quality analyses, while the remaining three plants were maintained to determine the bolting time. The treatments consisted of interactions among three NH_4^+:NO_3^- ratios and four SA concentrations.

The nitrogen forms were adjusted to provide the following ratios while maintaining total nitrogen constant:

1. 100% NO : 0% NH

2. 80% NO_3^- : 20% NH_4^+

3. 40% NO_3^- : 60% NH_4^+

Target ratios were achieved by modifying the concentrations of ammonium sulphate, calcium nitrate, and calcium sulphate in the nutrient solution [24]. The pH and electrical conductivity (EC) of the nutrient solution were monitored daily. The pH was maintained between 5.8 and 6.5, and EC was regulated between 1.2 and 1.5 dS m^{-1} throughout the growing period. The nutrient solution was prepared using tap water having an initial electrical conductivity of 0.3 dS m^{-1}. The composition of the nutrient solution (mg/l) is shown in Table 1.

Table 1
Concentrations of essential macro- (N, P, K, Ca, Mg) and micronutrients (Fe, Mn, Zn, B, Cu, Mo) in the prepared nutrient solution (mg L^{-1}).

N	P	K	Ca	Mg	Fe	Mn	Zn	B	Cu	Mo
150	50	150	150	50	5	0.5	0.05	0.5	0.03	0.02

Salicylic acid (Sigma-Aldrich, Cas-No: 69-72-7) was applied as a foliar spray at concentrations of 0 (control), 0.75, 1.5, and 3.0 mM. The SA concentrations were selected based on a previous study reporting physiological responses in leafy vegetables [25]. The application was performed once on March 27, 2024 (14 days after transplanting), when plants reached the 5–6 leaf stage.

Data Collection

Plants were harvested at commercial maturity, and a subset was maintained to observe physiological bolting.

Morphological Measurements

Head fresh weight (g) and biomass partitioning (fresh and dry weights of leaves and roots) were determined using a precision balance. Dry weights were recorded after samples were dried in a forced-air oven at 65°C until constant weight. Head diameter (cm), head length (cm), stem diameter (mm), stem length (cm), and root length (cm) were measured using a digital caliper and ruler.

Physiological and Biochemical Analysis

The chlorophyll index was recorded from all plants in replicate using a SPAD-502 Plus meter (Konica Minolta, Japan), with measurements taken from 3 fully expanded leaves per plant. Total soluble solids (TSS, %) were determined from the juice of blended leaves (inner, middle, and outer) using a digital refractometer.

Phenological Observations

Bolting time was recorded as the number of days from transplanting until the visible elongation of the floral stem (bolting initiation).

Statistical Analysis

Data were tested for normality and homogeneity of variance prior to analysis. A two-way analysis of variance (ANOVA) was performed to evaluate the main effects of NH_4^+:NO_3^- ratios and SA doses, and their interactions, using SPSS software (Version 22.0, IBM Corp.). Means were compared using the Least Significant Difference (LSD) test at the $p < 0.05$ significance level.

Results

Head Characteristics and Yield Parameters

The ANOVA revealed significant interactive effects of NH_4^+:NO_3^- ratios and SA concentrations on head fresh weight ($p < 0.05$) (Fig. 1). The highest head weight (425.66 g) was recorded in plants cultivated with 100% NO_3^-: 0% NH_4^+ without SA application, whereas the lowest value (215.64 g) was observed in the 40% NO_3^-:60% NH_4^+ treatment combined with 1.5 mM SA. In general, reducing the NO_3^- proportion in the nutrient solution resulted in a significant decline in head weight. Yield decreased by approximately 27% and 30% when the NO_3^- ratio was lowered to 80% and 40%, respectively. Regarding head dimensions, 100% NO_3^-:0% NH_4^+ treatment combined with 3.0 mM SA yielded the greatest head diameter (40.33 cm). Conversely, head length showed a different trend, with the 80% NO_3^- ratio yielding the highest values, particularly when combined with 3.0 mM SA (25.00 cm) (Table 2).

Table 2
Effects of the interaction between NH_4^+:NO_3^- ratios and salicylic acid (SA) concentrations on head weight, head diameter, and head length

NH_4:NO_3 rates	Salicylic acid doses			
	0 mM	0.75 mM	1.5 mM	3 mM
Head weight				
40% NO_3 : 60% NH_4	276.76 de	264.52 de	215.64 f	273.56 de
80% NO_3 : 20% NH_4	247.61 e	283.57 cd	275.02 de	260.60 de
100% NO_3 : 0% NH_4	425.66 a	356.96 b	309.29 c	372.21 b
Head diameter				
40% NO_3 : 60% NH_4	34.83 def	37.00 bcd	32.50 f	35.00 de
80% NO_3 : 20% NH_4	38.50 ab	34.83 def	33.83 ef	35.50 cde
100% NO_3 : 0% NH_4	38.33 ab	35.83 cde	37.66 bc	40.33 a
Head Length				
40% NO_3 : 60% NH_4	22.33 cde	23.00 bcd	24.00 abc	24.66 ab
80% NO_3 : 20% NH_4	24.66 ab	24.66 ab	24.66 ab	25.00 a
100% NO_3 : 0% NH_4	21.66 def	21.33 def	21.00 ef	20.33 f

NH_4: Ammonium, NO_3: Nitrate, the means followed by different letters are statistically different from each other ($p < 0.05$)

Vegetative Growth and Biomass Production

The number of leaves per plant and stem characteristics were significantly influenced by the treatments (Fig. 2). The 100% NO_3^- treatment consistently promoted vegetative proliferation, yielding the highest number of leaves (56.66 leaves/plant) when combined with 0.75 mM SA. In contrast, increasing the ammonium proportion to 20% or 60% significantly reduced leaf number. Leaf biomass followed a similar pattern. The maximum leaf fresh and dry weights were obtained from the 100% NO_3^- treatments. Specifically, the highest leaf fresh weight (313.44 g) was observed in the 100% NO_3^- + 0 mM SA combination. Reducing the percentage to 40% resulted in a biomass reduction of nearly 29%. Nitrogen source and SA doses significantly affected the fresh:dry weight ratio ($p < 0.05$). Increasing NO_3 increased the fresh:dry weight ratio, while high NH_4 (especially 60%) decreased it. The greatest effect was observed at 1.5 mM SA, while values decreased at 3 mM SA. When the interactions were examined, the combination of 80% NO_3:20% NH_4 + 1.5 mM SA provided the highest fresh:dry weight ratio (26.74); the lowest values were recorded in high ammonium (40% NO_3) applications. Stem traits also varied; while stem diameter was maximized under 100% NO_3^-, stem length was greatest under the 80% NO_3^- regime

(13.00 cm with 0.75 mM SA), suggesting a differential allocation of resources to stem elongation versus thickening depending on the nitrogen form (Table 3).

Table 3
Effects of the interaction between NH_4^+:NO_3^- ratios and salicylic acid (SA) concentrations on leaf and stem biomass and morphological properties

Salicylic acid doses				
Number of leaves per plant				
NH_4:NO_3 rates	0 mM	0.75 mM	1.5 mM	3 mM
40% NO_3 : 60% NH_4	47.00 def	48.00 cde	50.00 bcd	47.33 def
80% NO_3 : 20% NH_4	39.00 g	44.66 ef	43.00 fg	44.33 ef
100% NO_3 : 0% NH_4	54.00 ab	56.66 a	52.33 abc	50.66 bcd
Leaf fresh weight				
40% NO_3 : 60% NH_4	224.48 d	201.51 ef	169.36 g	220.51 d
80% NO_3 : 20% NH_4	187.43 fg	224.54 d	223.46 d	206.40 de
100% NO_3 : 0% NH_4	313.44 a	288.74 b	249.00 c	298.90 ab
Leaf dry weight				
40% NO_3 : 60% NH_4	10.00 de	9.34 ef	7.48 i	10.47 cd
80% NO_3 : 20% NH_4	7.85 hi	8.84 fg	8.35 ghi	8.68 fgh
100% NO_3 : 0% NH_4	13.15 a	11.33 bc	9.49 ef	12.09 b
Fresh: dry weight rotios				
40% NO_3 : 60% NH_4	22.44 d	21.57 e	22.63 cd	21.04 e
80% NO_3 : 20% NH_4	23.87 bcd	25.46 ab	26.74 a	23.83 bcd
100% NO_3 : 0% NH_4	23.93 bcd	25.50 ab	26.21 a	24.71 abc
Stem length				
40% NO_3 : 60% NH_4	7.83 e	10.33 bcd	11.50 ab	10.33 bcd
80% NO_3 : 20% NH_4	8.16 e	13.00 a	10.50 bc	12.00 ab
100% NO_3 : 0% NH_4	9.16 cde	10.66 bc	8.33 de	7.66 e
Stem diameter				
40% NO_3 : 60% NH_4	3.79 ef	3.99 cde	3.48 f	3.88 de
80% NO_3 : 20% NH_4	3.95 cde	4.05 cde	4.04 cde	4.21 bcd
100% NO_3 : 0% NH_4	4.68 a	4.29 bc	4.23 bcd	4.54 ab

NH_4: Ammonium, NO_3: Nitrate, the means followed by different letters are statistically different from each other ($p < 0.05$)

Salicylic acid doses				
Stem fresh weight				
40% NO_3 : 60% NH_4	15.87 c	16.61 bc	12.02 d	16.86 bc
80% NO_3 : 20% NH_4	15.96 c	18.61 ab	15.79 c	20.93 a
100% NO_3 : 0% NH_4	19.98 a	20.18 a	19.34 a	20.69 a
Stem dry weight				
40% NO_3 : 60% NH_4	0.71 cd	0.61 de	0.56 e	0.60 de
80% NO_3 : 20% NH_4	0.81 bc	0.86 b	0.85 b	1.12 a
100% NO_3 : 0% NH_4	0.79 bc	0.84 b	0.51 e	0.88 b
NH_4: Ammonium, NO_3: Nitrate, the means followed by different letters are statistically different from each other ($p < 0.05$)				

Root System Architecture

Root development exhibited distinct responses to the nitrogen source (Fig. 3). Interestingly, while the 100% NO_3^- ratio maximized root fresh and dry weights (50.56 g and 1.70 g, respectively), the longest roots (43.00 cm) were observed in the 80% NO_3^- treatment supplemented with 3.0 mM SA. This indicates that a moderate ammonium supply (20%) may stimulate root elongation, whereas exclusive nitrate supply favors root biomass accumulation (Table 4).

Table 4
Effects of the interaction between NH_4^+:NO_3^- ratios and salicylic acid (SA) concentrations on root biomass and length of hydroponically grown lettuce

Salicylic acid doses				
Root fresh weight				
NH_4:NO_3 rates	0 mM	0.75 mM	1.5 mM	3 mM
40% NO_3 : 60% NH_4	19.44 de	22.16 d	18.66 e	26.96 c
80% NO_3 : 20% NH_4	13.51 f	19.03 de	13.36 f	13.12 f
100% NO_3 : 0% NH_4	40.18 a	31.21 b	34.46 b	34.07 b
Root dry weight				
40% NO_3 : 60% NH_4	1.03 cd	0.96 cde	0.91 de	0.81 ef
80% NO_3 : 20% NH_4	0.72 f	1.06 cd	1.10 c	1.07 cd
100% NO_3 : 0% NH_4	1.70 a	1.35 b	1.46 b	1.49 b
Root length				
40% NO_3 : 60% NH_4	43.67 a	32.00 de	33.33 cde	35.00 cde
80% NO_3 : 20% NH_4	38.33 abc	31.00 e	41.00 ab	43.00 a
100% NO_3 : 0% NH_4	33.66 cde	31.00 e	31.66 de	37.00 bcd

NH_4: Ammonium, NO_3: Nitrate, the means followed by different letters are statistically different from each other ($p < 0.05$)

Physiological and Biochemical Attributes

Chlorophyll index was significantly higher in plants supplied with 100% NO_3^- (9.60 with 3.0 mM SA) compared to those receiving ammonium-enriched solutions (Fig. 4; Table 5). The inclusion of ammonium (20–60%) resulted in a notable reduction in chlorophyll content. Total soluble solids (TSS), however, did not show significant variation among the treatments, indicating that the sugar accumulation in leaves was relatively stable across the tested nutrient regimes.

Table 5
Effects of the interaction between NH_4^+:NO_3^- ratios and salicylic acid (SA) concentrations on Chlorophyll index and total soluble solid matter

Chlorophyll index.				
	Salicylic acid doses			
NH_4:NO_3 rates	0 mM	0.75 mM	1.5 mM	3 mM
40% NO_3 : 60% NH_4	8.35 bc	8.34 bc	8.33 bc	8.33 bc
80% NO_3 : 20% NH_4	8.31 bc	7.53 e	8.24 c	7.64 de
100% NO_3 : 0% NH_4	9.38 a	8.01 cd	8.66 b	9.60 a
Total soluble solid matter				
40% NO_3 : 60% NH_4	3.33 a	3.16 a	3.33 a	3.16 a
80% NO_3 : 20% NH_4	2.96 a	3.10 a	3.30 a	3.16 a
100% NO_3 : 0% NH_4	3.25 a	3.08 a	3.08 a	3.41 a
NH_4: Ammonium, NO_3: Nitrate, the means followed by different letters are statistically different from each other ($p < 0.05$)				

Bolting Behavior

Bolting time was significantly influenced by the nitrogen form (Fig. 5). Plants grown with 100% NO_3^- exhibited a delayed bolting response (50.50 days) compared to those grown with ammonium. The 80% NO_3^- ratio accelerated the onset of bolting, shortening the vegetative phase by approximately 5–6 days. Although the SA application showed a trend towards influencing physiological traits, its specific effect on delaying bolting was overshadowed by the dominant effect of the nitrogen source and likely environmental factors such as high temperatures during the late growing season. These results indicate that nitrate-dominated nutrition not only enhanced biomass accumulation but also postponed the transition to the reproductive phase (Table 6).

Table 6
Effects of the interaction between NH_4^+:NO_3^- ratios and salicylic acid (SA) concentrations on bolting time

Salicylic acid doses				
	Bolting time			
NH_4:NO_3 rates	0 mM	0.75 mM	1.5 mM	3 mM
40% NO_3 : 60% NH_4	46.33 bc	47.00 b	47.00 b	45.83 bc
80% NO_3 : 20% NH_4	45.16 bcd	44.83 cd	44.00 d	44.33 cd
100% NO_3 : 0% NH_4	50.50 a	50.33 a	50.50 a	50.50 a

Discussion

The results of this study demonstrate that the nitrogen form in the nutrient solution is a decisive factor regulating the growth, yield, and developmental timing of hydroponic lettuce. The superiority of the 100% NO_3^- regime in terms of head weight, biomass, and leaf number aligns with previous findings indicating that lettuce is highly sensitive to NH_4^+ toxicity [13, 14, 24]. Although lettuce can utilize both NH_4^+ and NO_3^-, high NH_4^+ ratios (≥ 20% in this study) likely induced metabolic costs associated with intracellular pH regulation and ammonium detoxification, thereby suppressing vegetative growth.

The beneficial effect of exclusive nitrate supply on the chlorophyll index contradict with some studies reporting enhanced leaf greenness under moderate ammonium nutrition due to improved nitrogen assimilation efficiency [26]. However, the present results indicate that NO_3^--dominated nutrition was more effective in maintaining higher chlorophyll index values than mixed nitrogen sources. This response may be associated with NO_3^--related signaling processes or a more stable chloroplast function, although further molecular evidence would be required to confirm the underlying mechanisms.

A key objective of this study was to explore agronomic strategies to delay bolting. The results clearly showed that maintaining a high NO_3^--to-NH_4^+ ratio (100:0) significantly prolonged the vegetative phase compared with NH_4^+-containing nutrient solutions. This finding supports previous evidence that NO_3^- may function not only as a nutrient but also as a signaling molecule involved in the regulation of flowering time, potentially through interactions with gibberellin-related pathways [16]. In contrast, the earlier bolting observed under the 80% NO_3^-:20% NH_4^+ treatment suggests that even moderate proportions of NH_4^+ may accelerate the transition to the reproductive phase, possibly by inducing mild physiological stress responses in lettuce.

The delayed bolting observed under NO_3^--dominated nutrition may be explained by the signaling role of NO_3^- in hormonal regulation of plant development. Nitrate has been shown to act as a signaling molecule that promotes plant growth by enhancing gibberellin (GA) biosynthesis and destabilizing GA-regulated DELLA growth repressors [18]. DELLA proteins are key negative regulators of growth and

developmental transitions, including stem elongation and floral initiation. Therefore, the sustained vegetative growth and delayed bolting observed in plants supplied with 100% NO_3^- in the present study may be associated with NO_3^--induced modulation of GA–DELLA signaling. In contrast, the inclusion of NH_4^+ in the nutrient solution may have disrupted this regulatory balance, leading to earlier reproductive transition and reduced vegetative performance.

While SA is well documented for its role in regulating stress responses and flowering processes [21], its interactive effect on bolting time in the present experiment was less pronounced than initially expected. This limited response may be attributed to a heatwave during the experimental period, which was likely masked by thermal induction overriding hormonal regulation. Elevated temperatures are recognized as a major environmental trigger for bolting in lettuce and can override hormonal or nutritional interventions [27]. Therefore, future studies conducted under strictly controlled temperature regimes would be required to disentangle the specific contribution of SA to flowering regulation, particularly with respect to its potential involvement in florigen-related signaling pathways in hydroponic lettuce.

In conclusion, for spring hydroponic production of ‘El Maria’ lettuce under semi-arid conditions, a nutrient solution containing 100% NO_3^- nitrogen can be recommended to maximize yield and quality while delaying bolting. Although SA applications resulted in marginal improvements in certain growth parameters, nitrogen form optimization emerged as the primary management strategy in this production system.

Conclusion

This study demonstrates that nitrogen form is a key determinant of growth performance, yield, quality attributes, and bolting behavior in hydroponically grown lettuce under semi-arid greenhouse conditions. Among the tested treatments, a NO_3^--dominated nutrient solution (100% NO_3^-) consistently enhanced head fresh weight, leaf biomass and chlorophyll index while significantly delaying bolting compared with ammonium-containing nutrient regimes. The inclusion of NH_4^+, even at moderate levels, reduced vegetative growth and accelerated the transition to the reproductive phase, highlighting lettuce’s sensitivity to NH_4^+ nutrition in hydroponic systems. Although foliar application of SA induced minor, parameter-dependent responses, its effect on bolting regulation was limited under the prevailing high-temperature conditions, suggesting that thermal induction may override hormonal modulation during late spring cultivation. Overall, the findings indicate that optimizing the NH_4^+:NO_3^- ratio is a more effective management strategy than exogenous SA application for delaying bolting and maximizing marketable yield in hydroponic lettuce production systems. From a practical perspective, NO_3^--based nutrition can be recommended for spring lettuce cultivation in semi-arid regions. Future research should focus on controlled-environment experiments to further elucidate the interaction between nitrogen form and SA under non-stressful temperature regimes and to explore the underlying physiological and molecular mechanisms governing bolting regulation.

Declarations

Acknowledgements

This study was produced from Srwa GHAFWR MUHAMMED MUHAMMED's master's thesis. We gratefully acknowledge the support provided by the HÜBAP unit within the scope of the project.

Author contributions

S.G.M.M.: Conceptualization, Methodology, Investigation, Formal analysis, Writing – original draft. **S.S.:** Conceptualization, Validation, Investigation, Supervision, Funding acquisition. All authors have read and agreed to the published version of the manuscript.

Availability of data and materials

Correspondence and requests for materials should be addressed to S.S.

Funding

This study was supported by Harran University Scientific Research Projects Unit under project number 24008.

Data availability

The data supporting the findings of this study are available from the corresponding author.

Ethics approval and consent to participate

Not applicable.

Consent for publication

Not applicable.

Competing interests

The authors declare no competing interests.

Clinical trial number

Not applicable.

Author details

1 Harran University, Graduate Education Institute, 63050, Şanlıurfa/Türkiye

2 Harran University, Faculty of Agriculture, Department of Horticulture, 63050, Şanlıurfa/Türkiye

References

1. Mohammed AA, Söylemez S, Sarhan TZ. Effect of biofertilizers, seaweed extract and inorganic fertilizer on growth and yield of lettuce (Lactuca sativa var. longifolia L). Harran Tarım ve Gıda Bilimleri Dergisi. 2022;26(1):60–71.
2. Lebeda A, et al. Unraveling the variation, phylogeny, and taxonomy of Lactuca spp. for germplasm management and breeding. Planta. 2025;262(6):1–46.
3. Abdel Bar FM et al. Genus Lactuca (Asteraceae): A Comprehensive Review. Records Nat Prod, 2023. 17(2).
4. Mou B. Nutrient content of lettuce and its improvement. Curr Nutr Food Sci. 2009;5(4):242–8.
5. Shi M, et al. Phytochemicals, nutrition, metabolism, bioavailability, and health benefits in lettuce—A comprehensive review. Antioxidants. 2022;11(6):1158.
6. Kim MJ, et al. Nutritional value, bioactive compounds and health benefits of lettuce (Lactuca sativa L). J Food Compos Anal. 2016;49:19–34.
7. Tutar FK, Abukalloub A, Çat M. Geleneksel Tarımdan Akıllı Tarım Uygulamalarına Dönüşüm Süreci: Türkiye Örneği. Volume 5. MTÜ Sosyal ve Beşeri Bilimler Dergisi; 2025. pp. 46–62. 1.
8. Rajaseger G, et al. Hydroponics: current trends in sustainable crop production. Bioinformation. 2023;19(9):925.
9. Sousa Rd, et al. Challenges and solutions for sustainable food systems: The potential of home hydroponics. Sustainability. 2024;16(2):817.
10. Waiba KM, et al. Soil-less vegetable cultivation: A review. J Pharmacognosy Phytochemistry. 2020;9(1):631–6.
11. Fussy A, Papenbrock J. An overview of soil and soilless cultivation techniques—chances, challenges and the neglected question of sustainability. Plants. 2022;11(9):1153.
12. Lakhiar IA, et al. Soilless agricultural systems: Opportunities, challenges, and applications for enhancing horticultural resilience to climate change and urbanization. Horticulturae. 2025;11(6):568.
13. Abd-Elmoniem E, et al. Effect of nitrogen form on lettuce plant grown in hydroponic system. Strategies Market Oriented Greenh Prod. 1995;434:47–52.
14. Song J, Yang J, Jeong BR. Growth, Quality, and Nitrogen Assimilation in Response to High Ammonium or Nitrate Supply in Cabbage (Brassica campestris L.) and Lettuce (Lactuca sativa L). Agronomy. 2021;11(12):2556.
15. Savvas D, et al. Effects of ammonium nitrogen on lettuce grown on pumice in a closed hydroponic system. HortScience. 2006;41(7):1667–73.
16. Gras DE, et al. SMZ/SNZ and gibberellin signaling are required for nitrate-elicited delay of flowering time in Arabidopsis thaliana. J Exp Bot. 2018;69(3):619–31.
17. Vega A, O'Brien JA, Gutiérrez RA. Nitrate and hormonal signaling crosstalk for plant growth and development. Curr Opin Plant Biol. 2019;52:155–63.

18. Camut L, et al. Nitrate signaling promotes plant growth by upregulating gibberellin biosynthesis and destabilization of DELLA proteins. Curr Biol. 2021;31(22):4971–82. e4.
19. Xu J et al. Optimizing nitrogen form ratio for enhanced nutrient uptake and lettuce performance in hydroponic systems. Bioresource Technol Rep, 2025: p. 102384.
20. Zhumanova N, et al. A comprehensive review of new generation plant growth regulators. Volume 17. ES Food & Agroforestry; 2024. p. 1190.
21. Skrypnik L, Golovin A, Savina T. Effect of salicylic acid on phenolic compounds, antioxidant and antihyperglycemic activity of Lamiaceae plants grown in a temperate climate. Front Bioscience-Elite. 2022;14(1):3.
22. Kumar K, et al. An overview of plant phenolics and their involvement in abiotic stress tolerance. Stresses. 2023;3(3):570–85.
23. Reza MN, et al. Trends of soil and solution nutrient sensing for open field and hydroponic cultivation in facilitated smart agriculture. Sensors. 2025;25(2):453.
24. Söylemez S. The impact of different growth media and ammonium-nitrate ratio on yield and nitrate accumulation in lettuce (Lactuca sativa var. longifolia). Volume 49. Notulae Botanicae Horti Agrobotanici Cluj-Napoca; 2021. pp. 12540–12540. 4.
25. Talabany NK, Albarzinji IM. Effects of Salicylic Acid on some Growth and Physiological Characteristics of Lettuce (Lactuca sativa L.) under Cadmium Stress Conditions. Sci J Univ Zakho. 2023;11(1):37–44.
26. Zhu X, et al. Effects of different NO3–: NH4 + ratios on the photosynthesis and ultrastructure of lettuce seedlings. Environ Biotechnol. 2020;61(3):459–72. Horticulture.
27. Han R, et al. A composite analysis of flowering time regulation in lettuce. Front Plant Sci. 2021;12:632708.

Figures

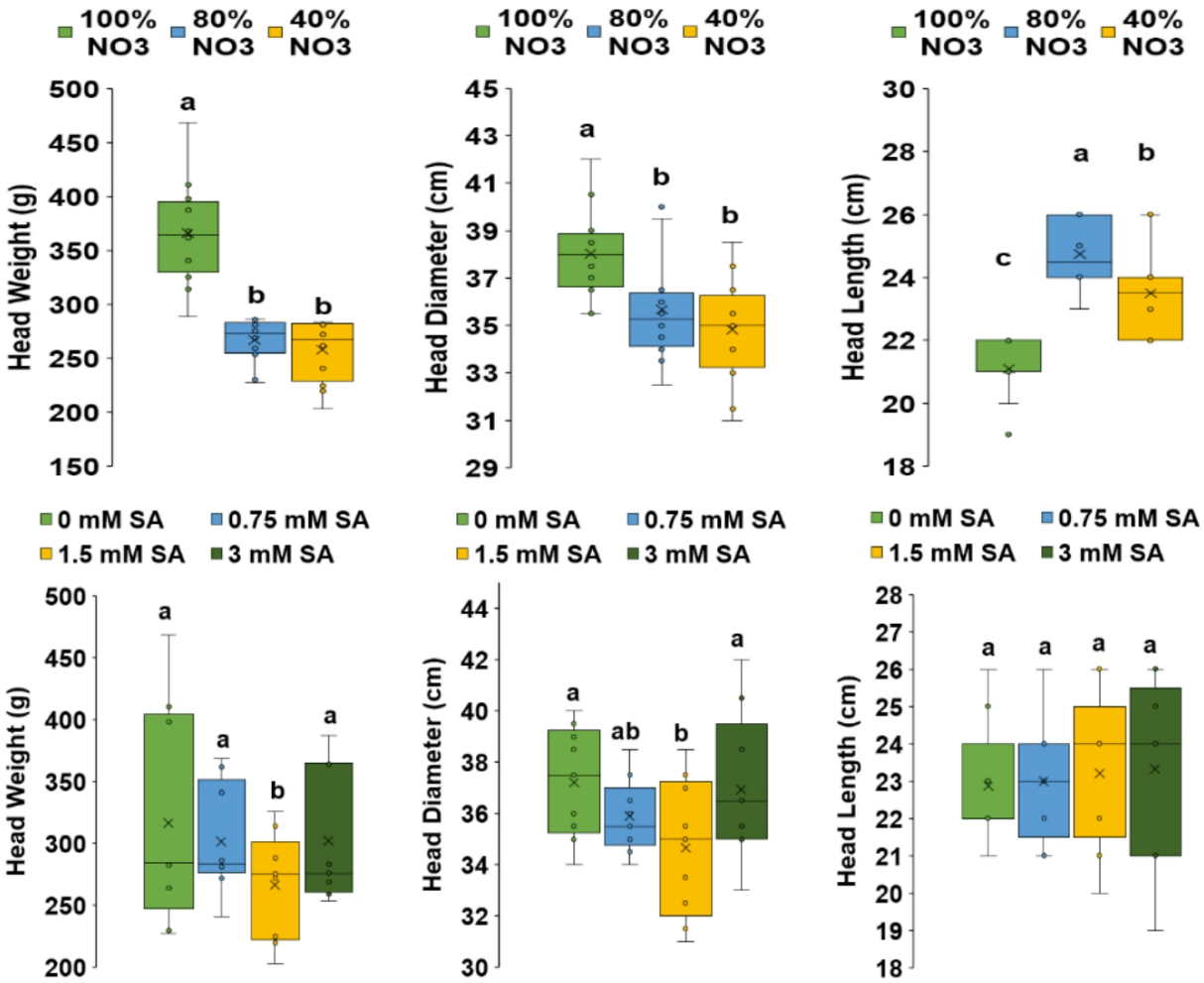


Figure 1

Individual effects of NH_4^+:NO_3^- ratios (upper panel) and salicylic acid (SA) concentrations (lower panel) on head weight, head diameter, and head length of hydroponically grown lettuce. Box plots show median, interquartile range, and minimum–maximum values; × indicates the mean. Means followed by different letters within each factor indicate significant differences ($P < 0.05$).

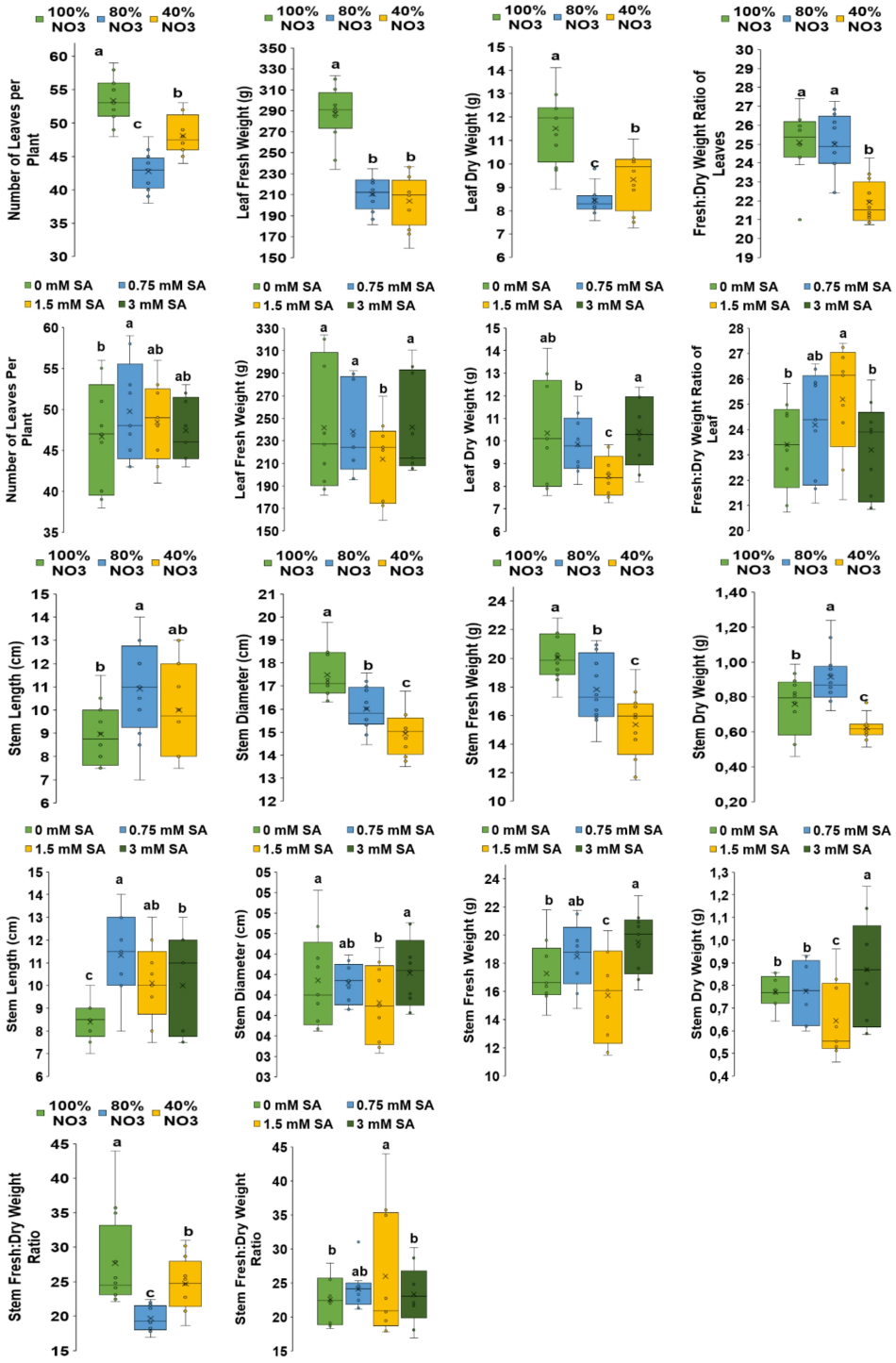


Figure 2

Individual effects of NH_4^+:NO_3^- ratios (upper panel) and salicylic acid (SA) concentrations (lower panel) on leaf and stem biomass and morphologic properties. Box plots show median, interquartile range, and minimum–maximum values; × indicates the mean. Means followed by different letters within each factor indicate significant differences ($P < 0.05$).

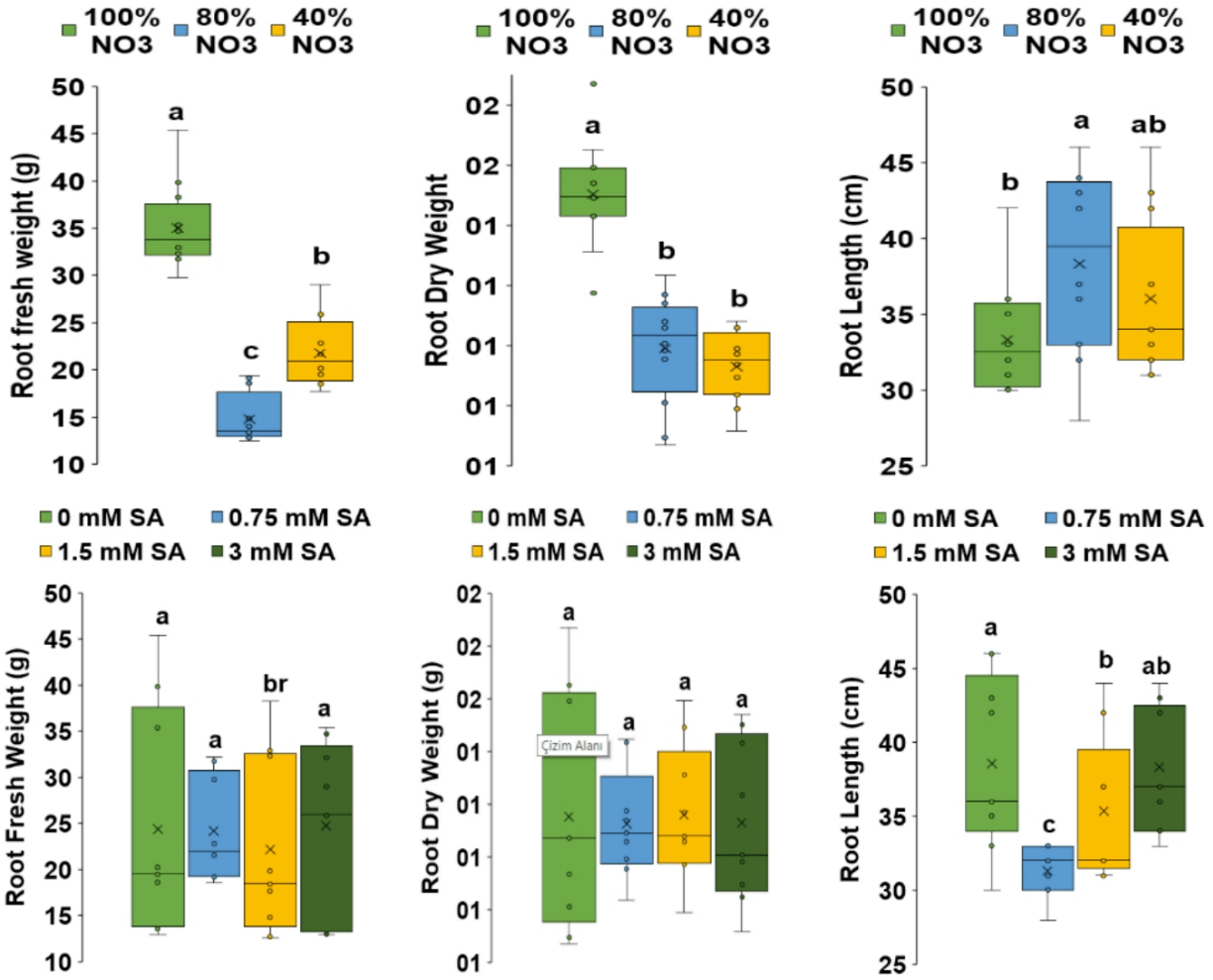


Figure 3

Individual effects of NH_4^+:NO_3^- ratios (upper panel) and salicylic acid (SA) concentrations (lower panel) on root biomass and length. . Box plots show median, interquartile range, and minimum–maximum values; × indicates the mean. Means followed by different letters within each factor indicate significant differences ($P < 0.05$).

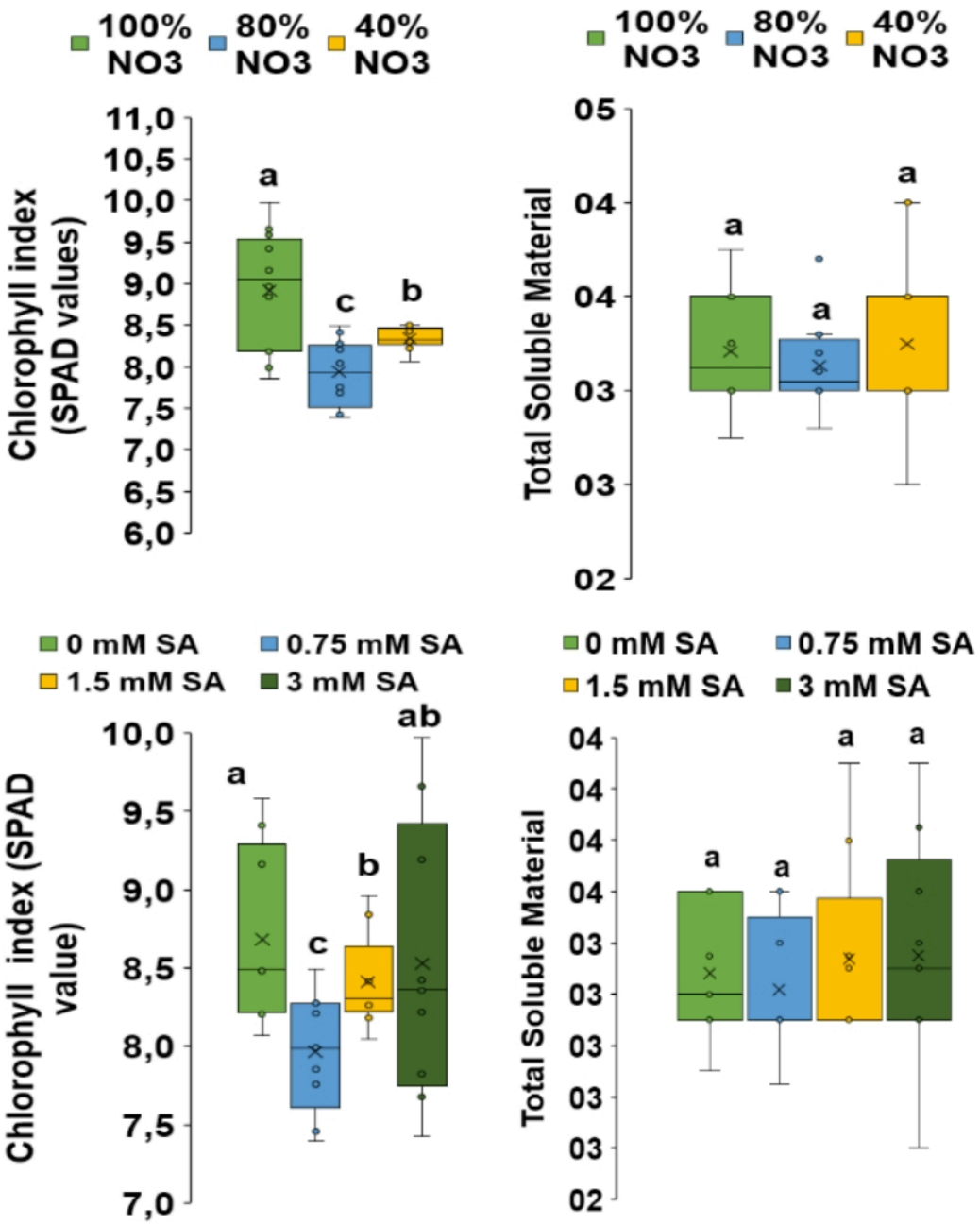


Figure 4

Individual effects of NH_4^+:NO_3^- ratios (upper panel) and salicylic acid (SA) concentrations (lower panel) on Chlorophyll index and total soluble solid matter. Box plots show median, interquartile range, and minimum–maximum values; × indicates the mean. Means followed by different letters within each factor indicate significant differences ($P < 0.05$).

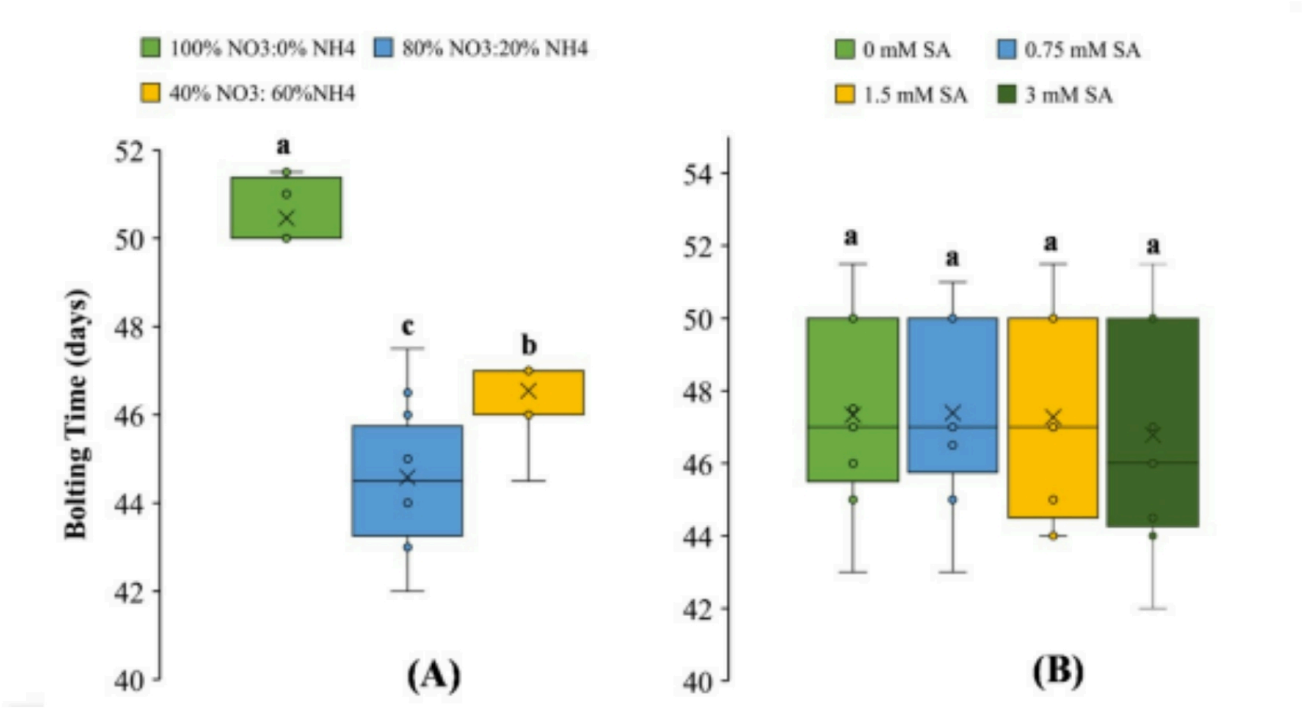


Figure 5

Individual effects of NH_4^+:NO_3^- ratios (upper panel) and salicylic acid (SA) concentrations (lower panel) on bolting time. Box plots represent the distribution of treatment values (median, interquartile range, and minimum–maximum). Means followed by different letters within each factor indicate significant differences ($P < 0.05$).