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Scientific Article

Morphophysiology and production of guava trees under irrigation strategies using saline water and ascorbic acid

Morfofisiologia e produção de goiabeira sob estratégias de irrigação com água salina e ácido ascórbico

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ABSTRACT – Prolonged periods of drought in the Brazilian semiarid region have encouraged the use of water with high salt concentrations to meet the water requirements of agricultural crops. Therefore, this study aimed to evaluate the effects of ascorbic acid application on the morphophysiology and yield of guava plants (cv. Paluma) subjected to irrigation strategies using saline water at different phenological stages. The experimental design was a randomized block design in a split-plot arrangement, with three replicates and one plant per plot. The main plots consisted of four irrigation strategies: irrigation with low-electrical-conductivity water (0.9 dS m^{-1}) throughout the crop cycle; and irrigation with saline water (3.3 dS m^{-1}) during the vegetative, flowering, or fruiting stages. The subplots consisted of three ascorbic acid (AsA) concentrations (0, 200, and 400 mg L^{-1}). Irrigation with water at an electrical conductivity of 3.3 dS m^{-1} during the flowering stage resulted in higher leaf transpiration, number of fruits, and production per guava plant. The application of 200 and 400 mg L^{-1} AsA increased chlorophyll *a* synthesis and CO_2 assimilation rate in guava plants under salt stress during the vegetative and flowering stages, respectively. Furthermore, ascorbic acid enhanced guava plant growth under irrigation with water at an electrical conductivity of 3.3 dS m^{-1} during the different phenological stages. AsA application at a concentration of 400 mg L^{-1} also had beneficial effects on guava yield components.

Keywords: *Psidium guajava* L.. Salt stress. Non-enzymatic antioxidant.

RESUMO - Os prolongados períodos de seca no semiárido brasileiro têm impulsionado o uso de águas com elevados teores de sais para atender às necessidades hídricas das culturas agrícolas. Assim, objetivou-se avaliar os efeitos da aplicação de ácido ascórbico na morfofisiologia e na produção da goiabeira cv. Paluma submetidas as estratégias de irrigação com água salina em diferentes fases fenológicas. O delineamento experimental foi em blocos casualizados, em esquema de parcelas subdivididas, com três repetições e uma planta por parcela. As parcelas foram constituídas por quatro estratégias de irrigação: uso de irrigação com água de baixa condutividade elétrica (0,9 dS m^{-1}), durante todo o ciclo; e aplicação de água salina (3,3 dS m^{-1}) nas fases vegetativa; de floração; e de frutificação. As subparcelas corresponderam de três concentrações de ácido ascórbico - AsA (0, 200, e 400 mg L^{-1}). A irrigação com água de 3,3 dS m^{-1} na fase de floração resultou em maior transpiração foliar, número de frutos e produção por planta de goiabeira. A aplicação de 200 e 400 mg L^{-1} de AsA aumentou a síntese de clorofila *a* e a taxa de assimilação de CO_2 da goiabeira sob estresse salino nas fases vegetativa e de floração, respectivamente. Além disso, o ácido ascórbico proporcionou incremento no crescimento da goiabeira irrigada com água de 3,3 dS m^{-1} nas distintas fases fenológicas. A aplicação de AsA na concentração de 400 mg L^{-1} promoveu efeitos benéficos nos componentes de produção da goiabeira.

Palavras-chave: *Psidium guajava* L.. Estresse salino. Antioxidante não enzimático.

Conflict of interest: The authors declare no conflict of interest related to the publication of this manuscript.

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INTRODUCTION

Guava (*Psidium guajava* L.) is a fruit crop of high nutritional, social, and economic value, ranking among the main species cultivated in irrigated areas of northeastern Brazil (ANGULO-LÓPEZ et al., 2021). Guava fruits have high nutritional value, as they are rich in vitamin C, dietary fiber, and antioxidant compounds. In addition, they have considerable industrial versatility and can also be consumed fresh (SOUZA; FERRAREZI JUNIOR, 2022). In the 2024 growing season, Brazilian guava production reached 557,225 metric tons, harvested from an area of 22,987 hectares, with the Northeast region accounting for 275,736 metric tons of total production (IBGE, 2025). Under these conditions, the use of saline water has become a recurrent alternative for irrigation. However, excessive salt accumulation in the soil impairs essential physiological processes, thereby reducing crop growth and yield (BEZERRA et al., 2019).

Excessive salt concentrations in irrigation water and/or soil induce osmotic effects in plants, which hinder water and nutrient uptake by the roots, resulting in effects similar to those caused by water stress (LIMA et al., 2019). The accumulation of toxic ions, such as Na^+ and Cl^-, in plant tissues causes nutritional imbalances and oxidative disturbances (GHANI; AWANG; ISMAIL, 2018).

Consequently, photosynthetic pigments are degraded and carbon assimilation is reduced, leading to increased production of reactive oxygen species (ROS), which cause cellular damage (DIAS et al., 2021). In guava plants, these effects have been documented throughout the crop cycle, ranging from reduced vegetative growth (VELOSO et al., 2018) to yield losses and impaired postharvest quality (LACERDA et al., 2022), with the severity of damage varying according to the plant's developmental stage (SOARES et al., 2021a).

To enable crop production under these conditions, the adoption of irrigation strategies involving saline water is essential for expanding irrigated fruit production in the semiarid region of northeastern Brazil. Thus, the planned use of saline water, by applying it during phenological stages in which crops exhibit greater tolerance (SILVA et al., 2021), may minimize adverse effects and even trigger plant defense responses under stress conditions (SOARES et al., 2021a). Additionally, the application of elicitor compounds has proven effective, particularly ascorbic acid (AsA) (ALVES et al., 2022; CHEN; HAN, 2023). AsA is a non-enzymatic antioxidant compound involved in photoprotection, cellular regulation, and the activation of metabolic defense pathways, thereby helping plants mitigate the detrimental effects of stress (CELI et al., 2023).

Several studies have highlighted foliar application of AsA as an effective strategy to alleviate the adverse effects of salt stress in plants, as this compound enhances antioxidant enzyme activity and contributes to maintaining plant physiological homeostasis (AKRAM; SHAFIQ; ASHRAF, 2017). However, studies investigating the effects of foliar AsA application on grafted guava plants subjected to salt stress at different phenological stages remain scarce. Previous studies addressing the effects of salt stress on guava have primarily focused on plants propagated through stem cuttings (VELOSO et al., 2018; XAVIER et al., 2022), leaving a knowledge gap regarding grafted plants.

Caetano et al. (2024), when evaluating application methods and concentrations of ascorbic acid in sour passion fruit plants irrigated with saline water, found that foliar spraying with 0.8 mM AsA substantially alleviated the effects of salt stress, improving electrolyte leakage, leaf relative water content, photosynthetic pigment concentrations, and gas exchange. The hypothesis of this study is that foliar application of AsA mitigates the effects of salt stress according to the developmental stage of guava plants, owing to its protective action at the cellular level, thereby contributing to photosynthetic pigment synthesis, maintenance of cellular water content, and plant growth.

In this context, this study aimed to evaluate the effects of irrigation with saline water at different phenological stages, combined with foliar application of ascorbic acid, on the physiological responses, growth, and yield of guava plants.

MATERIALS AND METHODS

The experiment was conducted from July 2023 to August 2024 using 100-L pots adapted as drainage lysimeters under field conditions in the experimental area of the 'Rolando Enrique Rivas Castellón' farm, located at the Center for Science and Agri-Food Technology (CCTA), Federal University of Campina Grande (UFCG), in the municipality of São Domingos, Paraíba State, Brazil (06°48′50″ S and 37°56′31″ W; altitude of 190 m). According to the Köppen climate classification, the region is classified as BSh, corresponding to a hot semiarid climate characterized by high temperatures and low humidity (ALVARES et al., 2013). Annual rainfall is approximately 750 mm, whereas mean annual evaporation reaches approximately 2,000 mm, indicating a negative water balance typical of dry environments. Maximum and minimum air temperatures, relative humidity, and rainfall data were obtained from the meteorological station located in the São Gonçalo district, municipality of Sousa, Paraíba State, Brazil, as presented in Figure 1.

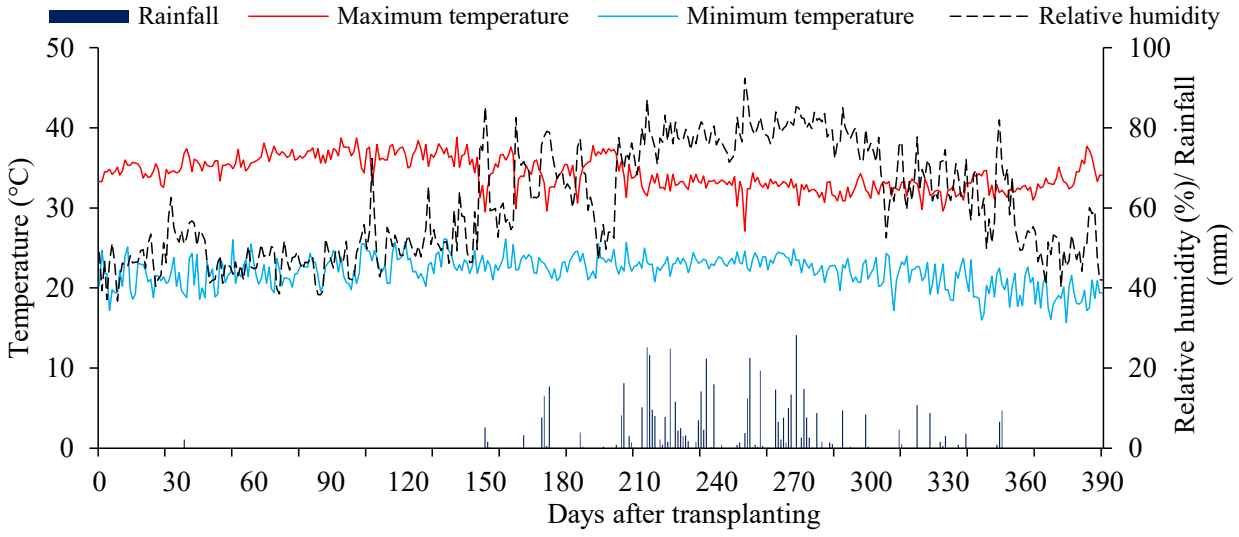


Figure 1. Maximum and minimum air temperatures, relative humidity, and rainfall during the period from July 18, 2023, to August 10, 2024.

The experiment was conducted in a randomized block design with a split-plot arrangement, comprising three replicates and one plant per plot. The main plots consisted of four irrigation strategies: irrigation with low-electrical-conductivity water (0.9 dS m^{-1}) throughout the crop cycle (SE); and irrigation with saline water (3.3 dS m^{-1}) during the vegetative (VE), flowering (FLO), and fruiting (FR) stages. The subplots consisted of three ascorbic acid (AsA) concentrations (0, 200, and 400 mg L^{-1}). In the experimental area, each irrigation strategy was assigned to a row of plants, and the different AsA concentrations were distributed within each row. The AsA concentrations were based on Gaafar et al. (2020), whereas the electrical conductivity levels of the irrigation water (ECw) were established according to Bezerra et al. (2019).

In this study, seedlings of the Paluma cultivar were used as scions. This cultivar is characterized by large fruits (weighing more than 200 g), with a pyriform shape, a short neck, and smooth skin. Its pulp is thick, firm, deep red, and pleasant in flavor. The BRS Guaraçá cultivar was used as the rootstock, as it is a proven option for controlling root-knot nematodes in guava plants and is also characterized by vigorous growth. After transplanting, the plants were irrigated with low-electrical-conductivity water until 99 days after transplanting (DAT). At 100 DAT, irrigation with water having an electrical conductivity of 3.3 dS m^{-1} was initiated according to the respective treatments.

Drainage lysimeters were filled with a 0.5-kg layer of gravel and 110 kg of a sandy loam Eutrophic Fluvent soil collected from the 0–40 cm layer. The soil's physicochemical properties (Table 1) were determined according to Teixeira et al. (2017). The end of the drainage pipe inside the lysimeter was wrapped with nonwoven geotextile fabric (Bidim OP 30) to prevent clogging by soil particles. A 2-L plastic bottle was connected to the drainage outlet to collect the drained water, allowing plant water consumption to be estimated. To prevent rainwater from entering the lysimeters during the period from 5:00 p.m. to 6:00 a.m., the lysimeters were partially covered with lids, leaving only an opening for the plant stem, thereby preventing interference with the applied treatments. Whenever rainfall occurred during the daytime, the lysimeters were kept covered.

Table 1. Chemical and physical properties of the soil used in the experiment before treatment application.

Chemical characteristics								
pH H_2O	OM	P	K^+	Na^+	Ca^{2+}	Mg^{2+}	Al^{3+}	H^+
(1:2.5)	g kg^{-1}	(mg kg^{-1})	$cmol_c$ kg^{-1}					
8.53	3.10	77.30	0.56	0.20	5.08	5.11	0	0

Chemical characteristics			Physical characteristics				
			Particle-size fractions			Soil moisture	
EC_{se}	CEC	SAR_{se}	ESP	Sand	Silt	Clay	33.42 kPa[1]	1519.5 kPa[2]
(dS m^{-1})	$cmol_c$ kg^{-1}	(mmol L^{-1})$^{0.5}$	%	(g kg^{-1})			(dag kg^{-1})	
0.46	10.95	1.02	1.83	775.70	180.90	43.40	12.45	5.00

pH – hydrogen potential; OM – organic matter: determined by the Walkley - Black wet digestion method; Ca^{2+} and Mg^{2+} extracted with 1 M KCl (pH 7.0); Na^+ and K^+ extracted with 1 M NH_4OAc (pH 7.0); Al^{3+} + H^+ extracted with 0.5 M CaOAc (pH 7.0); EC_{se} – electrical conductivity of the saturation extract; CEC – cation exchange capacity; SAR_{se} – sodium adsorption ratio of the saturation extract; ESP – exchangeable sodium percentage; [1],[2] refer to soil moisture contents corresponding to field capacity and permanent wilting point, respectively.

The soil used in this study is representative of the semiarid region. Given its alkaline pH (8.53), nitrogen fertilization was performed using urea [$CO(NH_2)_2$], an amide-based nitrogen source. Following the action of urease and water, urea undergoes hydrolysis, producing ammonia (NH_3), which is subsequently converted into ammonium (NH_4^+). In the soil, ammonium is then subjected to nitrification, resulting in its conversion into nitrate (NO_3^-). It is important to emphasize that this biochemical transformation releases H^+ ions into the soil solution, thereby contributing to soil acidification. Thus, despite the initially alkaline soil pH, the application of nitrogen as urea throughout the crop cycle naturally contributes to reducing soil pH. This process was not accompanied by any visually detectable symptoms of nutritional deficiencies in the plants.

The irrigation water with an electrical conductivity (ECw) of 0.9 dS m^{-1} was collected from a deep well located within the experimental area of the CCTA/UFCG Experimental Farm. The ECw level of 3.3 dS m^{-1} was obtained by adding non-iodized NaCl to water with an initial ECw of 2.6 dS m^{-1}, collected from a second well located on the same experimental farm. The salt concentration was determined based on the relationship between ECw and salt concentration, as described by Richards (1954), according to Equation 1:

$$C = 640 \times ECw \quad (1)$$

Where:
C = Amount of salts to be added (mg L^{-1}); and,
ECw = Electrical conductivity of water (dS m^{-1}) discounting the initial value of the water used (2.66 dS m^{-1}).

Irrigation strategies involving saline water were initiated at 100 days after transplanting (DAT) using a drip irrigation system. The system consisted of 32-mm PVC pipes in the main line and 16-mm low-density polyethylene pipes in the lateral lines, equipped with emitters with a flow rate of 10 L h^{-1}. Two pressure-compensating emitters (GA 10 Grapa model) were installed for each plant, each positioned 15 cm from the stem. The plants were irrigated daily at 7:00 a.m., with the volume of water applied to each container corresponding to the amount determined by the water balance,

according to Equation 2:

$$VI = \frac{(Va - Vd)}{(1 - LF)} \quad (2)$$

Where:
VI = Volume of water to be applied (mL);
Va = volume of water applied in the previous irrigation event (mL);
Vd = volume of water drained (mL); and,
LF = leaching fraction of 0.10 applied every 20 days.

The seedlings were transplanted into the lysimeters when they reached approximately 50 cm in height, with the root collar positioned slightly above the soil surface to prevent excessive moisture and preserve root development. After transplanting, the plants were staked using support structures to maintain upright stems, guide lateral branch growth, and promote the development of a well-structured crown with an architecture suitable for crop management.

Nitrogen (N), phosphorus (P_2O_5), and potassium (K_2O) fertilization was performed according to Cavalcanti (2008), considering the plant's nutritional requirements at different developmental stages and the initial soil fertility. Urea (45% N) and monoammonium phosphate (MAP; 50% P_2O_5 and 11% N) were used to meet the nitrogen requirements and supply phosphorus. Potassium sulfate (50% K_2O) was used as the potassium source. Fertilizers were applied weekly through fertigation. Micronutrients were applied every 15 days via foliar spraying, starting at 10 days after transplanting (DAT). During the vegetative stage, a concentration of 0.5 g L^{-1} was applied, whereas 1.0 g L^{-1} was used during the subsequent stages, using Dripsol Micro® solution (composition: 1.2% magnesium, 0.85% boron, 3.4% iron, 4.2% zinc, 3.2% manganese, 0.5% copper, and 0.06% molybdenum).

The plants were trained to a single stem until reaching 50 cm in height, at which point the terminal bud was removed. From the emerging shoots, four well-distributed primary branches were selected, oriented toward the four cardinal directions. Once these branches had matured, they were pruned to maintain a length of 40 cm. This management practice, combined with the careful selection of properly positioned branches, promotes a balanced crown architecture, ensuring adequate light penetration into the inner crown and greater photosynthetic efficiency. After the first fruiting cycle, a sanitation pruning was performed to remove dry, diseased, poorly positioned, or unproductive branches.

The different ascorbic acid (AsA) concentrations were applied by foliar spraying using a Jacto® backpack sprayer (Pompéia, São Paulo, Brazil), model Jacto XP, with a 12-L capacity, a maximum operating pressure of 88 psi, and a JD 12P nozzle. Applications were performed at 20-day intervals, starting at 87 days after transplanting (DAT) and continuing until full flowering. The spray solution was prepared by dissolving analytical-grade AsA (PA) in low-salinity water (0.3 dS m^{-1}), with the addition of a spreader-sticker (Wil fix®, 0.5 mL L^{-1}) to improve foliar coverage. To prevent spray drift between treatments, the plants were isolated using a plastic barrier. Applications were performed in the late afternoon (after 5:00 p.m.). The average spray volume applied per plant was 1,500 mL (pH = 4.0), according to the established treatments.

At 324 DAT, crown diameter, crown volume, and vegetative vigor index were evaluated in guava plants. Crown diameter (CD) was determined as the average of measurements taken along the planting row (DR) and between rows (DBR). Crown volume (CV), in turn, was calculated based on plant height (H), DR, and DBR, as described in Equation 3.

$$CrV = \pi\ 6 \times PH \times DR \times DBR \quad (3)$$

Where:
CrV – Crown volume (m^3);
PH – Plant height (m);
DR – Crown diameter in the direction of the row (m);
DBR – Crown diameter in the direction of the row spacing (m).

Finally, the vegetative vigor index (VVI) was determined according to the methodology described by Portella et al. (2016), as presented in Equation 4.

$$VVI = [PH + CD_1 + (SD \times 10)] / 100 \quad (4)$$

Where:
VVI – Vegetative Vigor Index;
PH – Plant height (m);
CD_1 – Crown diameter (m);
SD – Stem diameter (m).

Gas exchange measurements were also performed at 324 days after transplanting (DAT) using an infrared gas analyzer (IRGA, LCPro+ model) between 7:00 and 10:00 a.m. The following variables were determined: CO_2 assimilation rate (*A*, µmol CO_2 m^{-2} s^{-1}), stomatal conductance (*gs*, mmol H_2O m^{-2} s^{-1}), transpiration rate (*E*, mmol H_2O m^{-2} s^{-1}), and intercellular CO_2 concentration (*Ci*, µmol CO_2 mol^{-1}). Measurements were performed under natural air temperature and CO_2 concentration conditions, with one reading taken per leaf from the middle third of each plant. An artificial photosynthetically active radiation source of 1,200 µmol m^{-2} s^{-1} was used, based on the photosynthetic light-saturation curve.

Photosynthetic pigment concentrations were determined using leaf discs (3.14 cm²) collected from the middle third of the plants. The samples were immediately immersed in 5 mL of dimethyl sulfoxide (DMSO) and kept in the dark for 48 hours to ensure complete pigment extraction. Subsequently, the absorbance of the extracts was measured using a spectrophotometer at wavelengths of 480, 649, and 665 nm. Chlorophyll *a* (Chl *a*), chlorophyll *b* (Chl *b*), and carotenoid (Car) concentrations were determined according to Wellburn (1994), using Equations 5, 6, and 7, respectively.

$$Chl\ a = 12.19\ ABS665 - 3.45\ ABS649 \quad (5)$$

$$Chl\ b = 21.99 \times ABS649 - 5.32 \times ABS665 \quad (6)$$

$$Car = (1000\ ABS480 - 2.14\ Chl\ a - 70.16\ Chl\ b)/220 \quad (7)$$

Harvesting was performed manually, starting at 360 days after transplanting (DAT) and continuing until 390 DAT. The following variables were evaluated: number of fruits per plant (NF), production per plant (PROD), and mean fruit weight (MFW). Fruits were harvested based on skin color, with the transition from green to yellow being considered the appropriate harvest maturity stage. NF was determined by counting all fruits harvested from each plant. MFW was

calculated as the ratio between total production per plant and the total number of harvested fruits.

The data were subjected to normality (Shapiro - Wilk test) and homoscedasticity (Bartlett's test) assessments, followed by analysis of variance (ANOVA) using the F-test at the 0.05 probability level. When significant effects were detected ($p \leq 0.05$), the treatment means for both experimental factors were compared using Tukey's test. Statistical analyses were performed using SISVAR statistical software, version 5.7 (FERREIRA, 2019).

RESULTS AND DISCUSSION

The interaction between saline water irrigation strategies and ascorbic acid concentrations significantly affected intercellular CO_2 concentration, CO_2 assimilation rate, and chlorophyll *a*, chlorophyll *b*, and carotenoid concentrations in guava plants (Table 2). Irrigation strategies involving saline water also significantly affected stomatal conductance.

Table 2. Summary of the analysis of variance for stomatal conductance (*gs*), transpiration rate (*E*), intercellular CO_2 concentration (*Ci*), CO_2 assimilation rate (*A*), and chlorophyll *a* (Chl *a*), chlorophyll *b* (Chl *b*), and carotenoid (Car) concentrations in guava plants under saline water irrigation strategies applied at different phenological stages and foliar application of ascorbic acid.

Sources of variation	DF	Mean squares						
		gs	*E*	*Ci*	*A*	Chl *a*	Chl *b*	Car
Irrigation strategies (IS)	3	10880.37**	1.68**	7098.49**	20.05**	0.46^{ns}	0.43^{ns}	0.09^{ns}
Ascorbic acid (AsA)	2	815.94^{ns}	0.30^{ns}	387.41^{ns}	6.41*	7.20*	0.68^{ns}	0.07^{ns}
Interaction (IS × AsA)	6	1450.35^{ns}	0.13^{ns}	1751.05**	5.50*	10.36**	2.71**	0.23*
Blocks	2	1049.91^{ns}	0.13^{ns}	8.96^{ns}	1.24^{ns}	0.44^{ns}	0.36^{ns}	0.02^{ns}
Error 1	6	572.22	0.05	270.01	1.24	3.21	0.37	0.04
Error 2	16	544.26	0.10	290.85	1.59	1.85	0.31	0.06
CV1%		14.21	7.17	6.26	12.68	11.13	9.25	9.91
CV2%		13.85	9.74	6.50	14.34	8.45	8.57	12.23

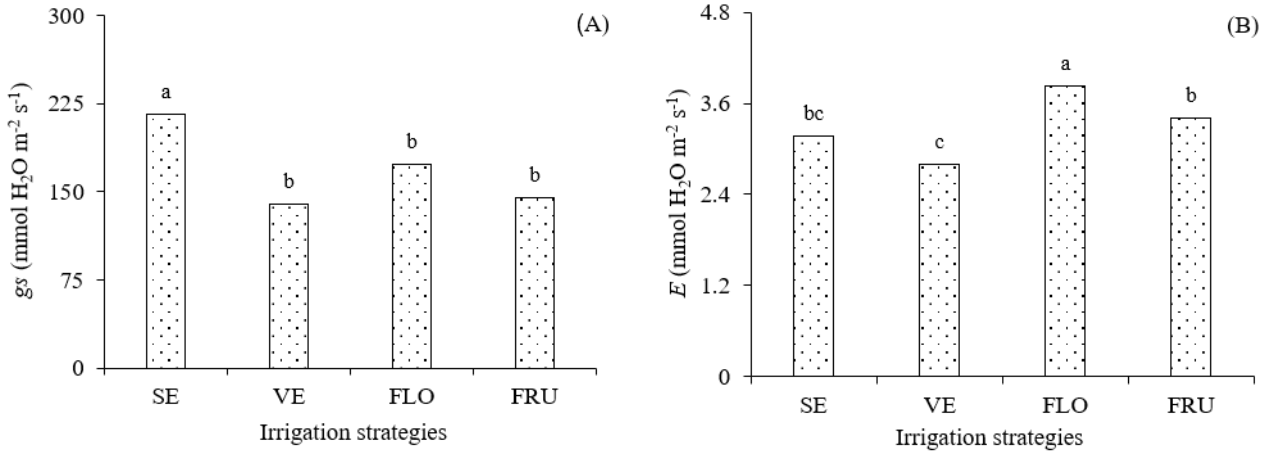
DF – Degrees of freedom; ** significant at $p \leq 0.01$; ns – not significant; CV – coefficient of variation.

Stomatal conductance reached its highest value (215.37 mmol H_2O m^{-2} s^{-1}) in plants irrigated with low-electrical-conductivity water (0.9 dS m^{-1}) throughout the crop cycle (Figure 2A), being significantly higher than the values observed in plants irrigated with water at an electrical conductivity (ECw) of 3.3 dS m^{-1} during the vegetative, flowering, and fruiting stages. In contrast, leaf transpiration (Figure 2B) was highest in plants irrigated with water at an ECw of 3.3 dS m^{-1} during the flowering stage, showing a 20.82% increase compared with plants irrigated with water at an ECw of 0.9 dS m^{-1} throughout the crop cycle (SE).

It is noteworthy that the other saline water irrigation strategies did not differ significantly from SE. This response is particularly relevant because reduced stomatal opening is a common plant response and represents one of the mechanisms adopted to restrict water loss under conditions of altered osmotic potential, which limits water uptake and consequently leads to stomatal closure (GHANI; AWANG; ISMAIL, 2018). However, the maintenance or increase in transpiration may be associated with enhanced metabolic activity in plants exposed to salinity during the vegetative stage, which influences water and nutrient uptake (VELOSO et al., 2018; SOARES et al., 2021a).

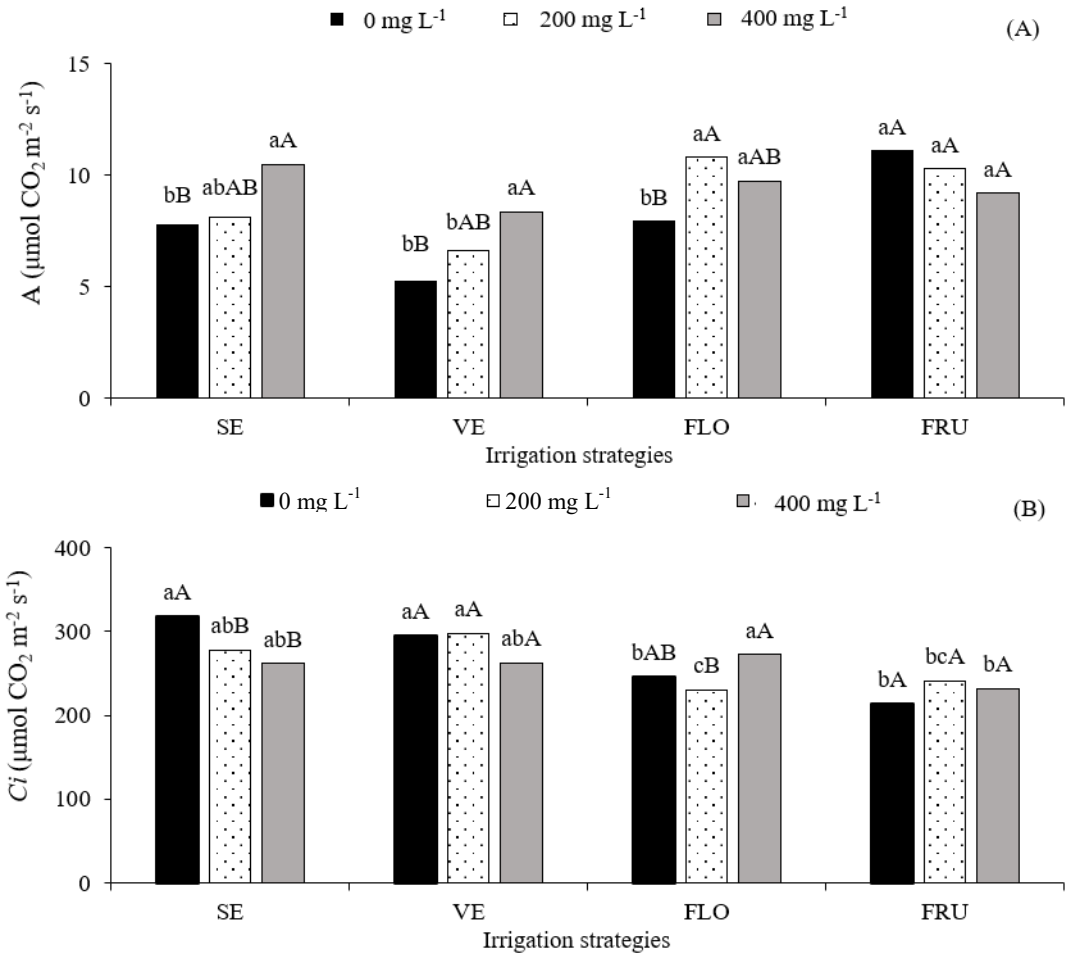
The CO_2 assimilation rate (Figure 3A) reached its highest value (11.07 µmol CO_2 m^{-2} s^{-1}) in plants that did not receive AsA application and were irrigated with water at an electrical conductivity of 3.3 dS m^{-1} during the fruiting stage (FRU). Regarding the effects of AsA application, compared with untreated plants, the application of 400 mg L^{-1} increased CO_2 assimilation rates under the SE and VE irrigation strategies, whereas the 200 mg L^{-1} concentration increased these values under the FLO strategy. No significant differences were observed among plants subjected to salt stress during the FRU stage. The increase in *A* in plants exposed to salt stress during the fruiting stage suggests a possible physiological acclimation response, potentially associated with osmotic adjustment and antioxidant protection mechanisms (DIAS et al., 2021). This response may be related to the functions of AsA, which participates in the activation of enzymes involved in secondary metabolite production and contributes to maintaining the plant's antioxidant defense system (CELI et al., 2023).

Guava plants subjected to salt stress during the FRU stage without AsA application exhibited the lowest intercellular CO_2 concentration (Figure 3B), indicating that the maintenance of photosynthetic activity resulted in CO_2 consumption within the substomatal cavity. Following AsA application, the pattern of *Ci* consumption was consistent with that observed for *A*, potentially indicating the maintenance of carboxylation efficiency and photosynthetic metabolism (CHEN; HAN, 2023).



Bars with the same letter indicate no significant differences among treatments according to Tukey's test ($p \leq 0.05$). SE – irrigation with moderately saline water throughout the crop cycle (1 - 390 days after transplanting - DAT); VE – salt stress during the vegetative stage (90 - 217 DAT); FLO – salt stress during the flowering stage (218 - 325 DAT); FRU - salt stress during the fruiting stage (326 - 390 DAT)

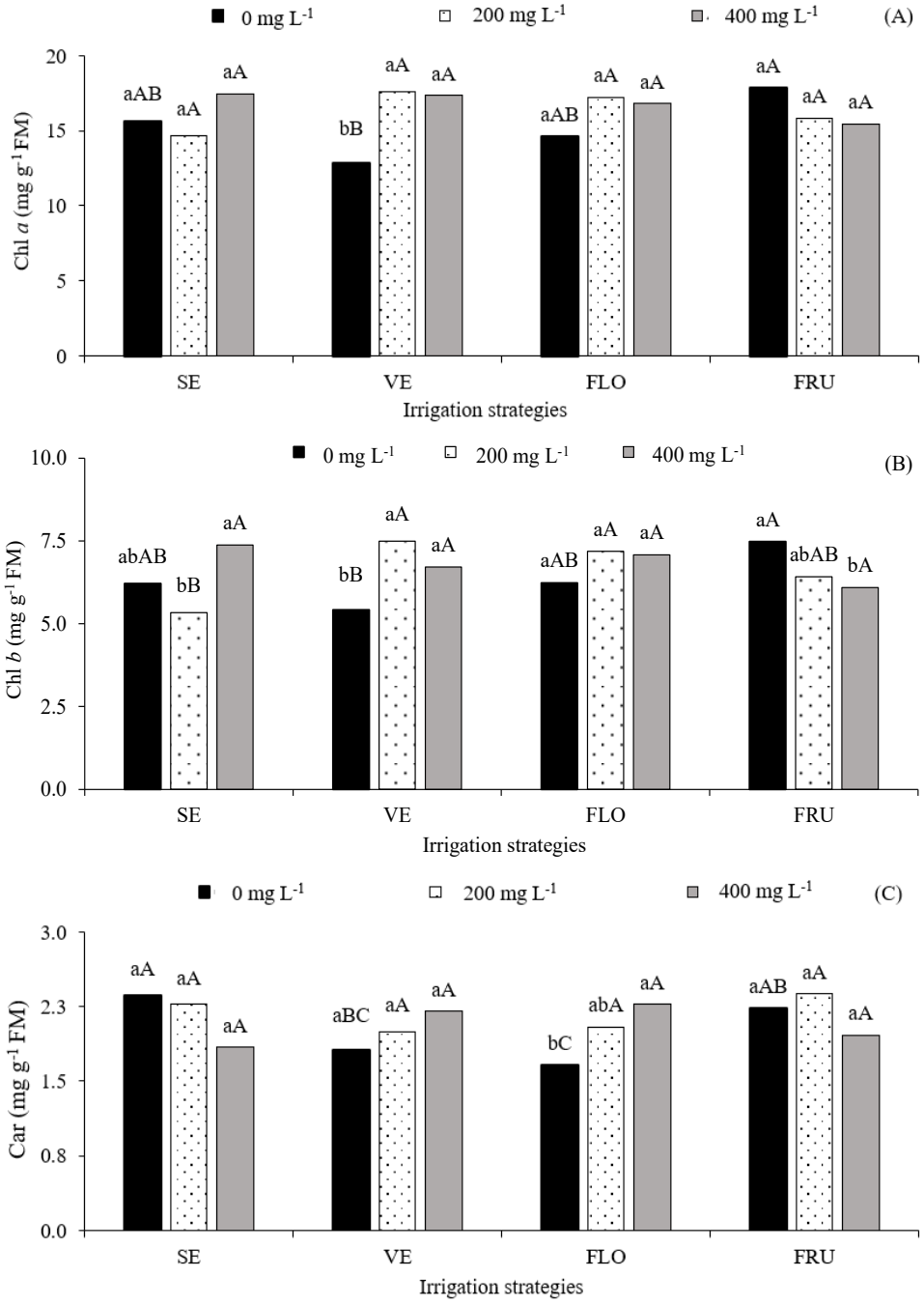
Figure 2. Stomatal conductance - *gs* (A) and transpiration - *E* (B) of guava plants as affected by saline water irrigation strategies applied at different phenological stages.



SE – irrigation with moderately saline water throughout the crop cycle (1 - 390 days after transplanting - DAT); VE – salt stress during the vegetative stage (90 - 217 DAT); FLO – salt stress during the flowering stage (218 - 325 DAT); FRU - salt stress during the fruiting stage (326 - 390 DAT). Bars with the same lowercase letters indicate no significant differences among irrigation strategies, whereas bars with the same uppercase letters indicate no significant differences among AsA concentrations (Tukey's test, $p \leq 0.05$)

Figure 3. CO_2 assimilation rate - *A* (A) and intercellular CO_2 concentration - *Ci* (B) in guava plants as affected by the interaction between saline water irrigation strategies applied at different phenological stages and ascorbic acid concentrations.

Regarding chlorophyll *a* concentrations (Figure 4A), no significant differences were observed among the irrigation strategies involving moderately saline water and the control treatment (SE) in plants that did not receive AsA. AsA application, however, affected only plants irrigated with moderately saline water during the VE stage, with both concentrations increasing chlorophyll *a* levels. This indicates that salt stress imposed during the different phenological stages was not sufficient to substantially impair energy capture by the photosynthetic apparatus. This response may reflect the ability of AsA to alleviate oxidative stress during the biosynthesis of pigments, particularly chlorophyll *a*, thereby contributing to the maintenance of photochemical energy conversion and photosynthesis (HAMIDI et al., 2024).



SE – irrigation with moderately saline water throughout the crop cycle (1–390 days after transplanting - DAT); VE - salt stress during the vegetative stage (90 - 217 DAT); FLO - salt stress during the flowering stage (218 - 325 DAT); FRU – salt stress during the fruiting stage (326 - 390 DAT). Bars with the same lowercase letters indicate no significant differences among irrigation strategies, whereas bars with the same uppercase letters indicate no significant differences among AsA concentrations (Tukey's test, $p \leq 0.05$)

Figure 4. Chlorophyll *a* - Chl *a* (A), chlorophyll *b* - Chl *b* (B), and carotenoid - Car (C) concentrations in guava plants as affected by the interaction between saline water irrigation strategies applied at different phenological stages and ascorbic acid concentrations.

Chlorophyll *b* concentrations (Figure 4B) remained similar in plants irrigated with water at an electrical conductivity of 0.9 dS m^{-1} throughout the crop cycle (SE), regardless of the AsA concentration applied. When plants were subjected to saline water irrigation during the vegetative stage, foliar application of AsA at concentrations of 200 and 400 mg L^{-1} resulted in significant effects compared with the treatment without AsA (0 mg L^{-1}). A reduction in chlorophyll *b* concentration relative to the control was observed with the application of 400 mg L^{-1} AsA during the fruiting stage. It is worth noting that chlorophyll *b* contributes to expanding the light absorption spectrum of the photosynthetic apparatus. Thus, the maintenance of photosynthetic pigments under the different irrigation strategies suggests that guava plants preserved their photosynthetic apparatus, indicating metabolic efficiency and the maintenance of antioxidant activity even under high light energy capture (GHANI; AWANG; ISMAIL, 2018; CELI et al., 2023). This response may be associated with the role of AsA in scavenging reactive oxygen species (ROS), preventing chlorophyll degradation and preserving RuBisCO enzyme activity, which may consequently contribute to increased CO_2 assimilation rates.

Regarding carotenoid concentrations (Figure 4C), plants irrigated with low-electrical-conductivity water throughout the crop cycle exhibited the highest estimated values at an AsA concentration of 0 mg L^{-1}. Conversely, the lowest carotenoid concentrations were observed in plants subjected to salt stress during the flowering stage (FLO) without AsA application. This response was mitigated by the application of 400 mg L^{-1} AsA, which resulted in values similar to those observed in the control treatment (SE). The reduction in carotenoid concentrations may be associated with the allocation of energy to other acclimation processes, potentially limiting carotenoid biosynthesis. This response may be alleviated by AsA application (SOARES et al., 2021b).

A significant interaction between saline water irrigation strategies and ascorbic acid concentrations was observed for crown volume, crown diameter, and vegetative vigor index (Table 3). Irrigation strategies significantly affected the number of fruits per plant and yield per plant. In contrast, AsA application alone had a significant effect on mean fruit weight.

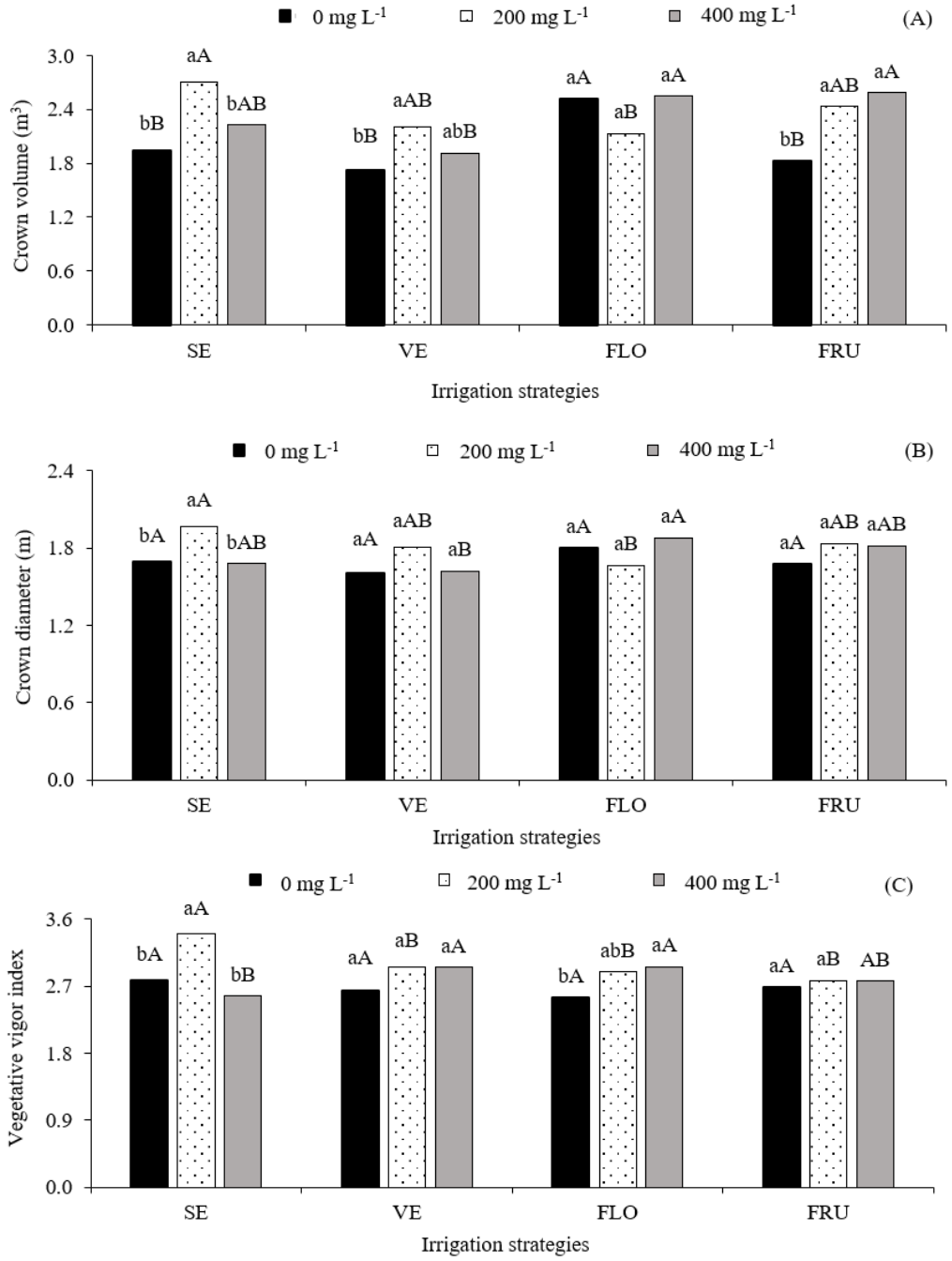
Table 3. Summary of the analysis of variance for crown volume (CV), crown diameter (CD), vegetative vigor index (VVI), mean fruit weight (MFW), number of fruits per plant (NF), and production per plant (PROD) in guava plants under saline water irrigation strategies applied at different phenological stages and foliar application of ascorbic acid.

Sources of variation	DF	Mean squares					
		CV	CD	VVI	MFW	NF	PROD
Irrigation strategies (IS)	3	0.33*	0.02^{ns}	0.04^{ns}	195.72^{ns}	116.91**	5288674.35**
Ascorbic acid (AsA)	2	0.48**	0.04*	0.35*	1771.32*	18.77*	21815333.89**
Interaction (IS × AsA)	6	0.26**	0.04*	0.15*	430.53^{ns}	9.55^{ns}	543706.20^{ns}
Blocks	2	0.01^{ns}	0.00^{ns}	0.00^{ns}	499.53^{ns}	11.36^{ns}	847523.25^{ns}
Error 1	6	0.05	0.00	0.00	520.84	4.13	284179.13
Error 2	16	0.04	0.18	0.18	472.32	4.56	236653.79
CV1%		10.46	4.63	4.63	10.21	15.48	18.12
CV2%		9.87	6.09	6.66	9.72	16.27	16.53

DF - Degrees of freedom; ** significant at $p \leq 0.01$; ^{ns} - not significant; CV - coefficient of variation.

Crown volume (Figure 5A) in plants irrigated with water at an ECw of 3.3 dS m^{-1} during the flowering stage differed significantly from that of plants receiving no foliar AsA application under the SE, VE, and FRU irrigation strategies. AsA application had beneficial effects, particularly at a concentration of 200 mg L^{-1} under the SE, VE, and FRU strategies, compared with the control. Similar effects were observed with the application of 400 mg L^{-1} AsA during the fruiting stage. These responses may reflect the regulation of photosynthetic activity, which helps balance photoassimilate translocation to support plant growth (LACERDA et al., 2022).

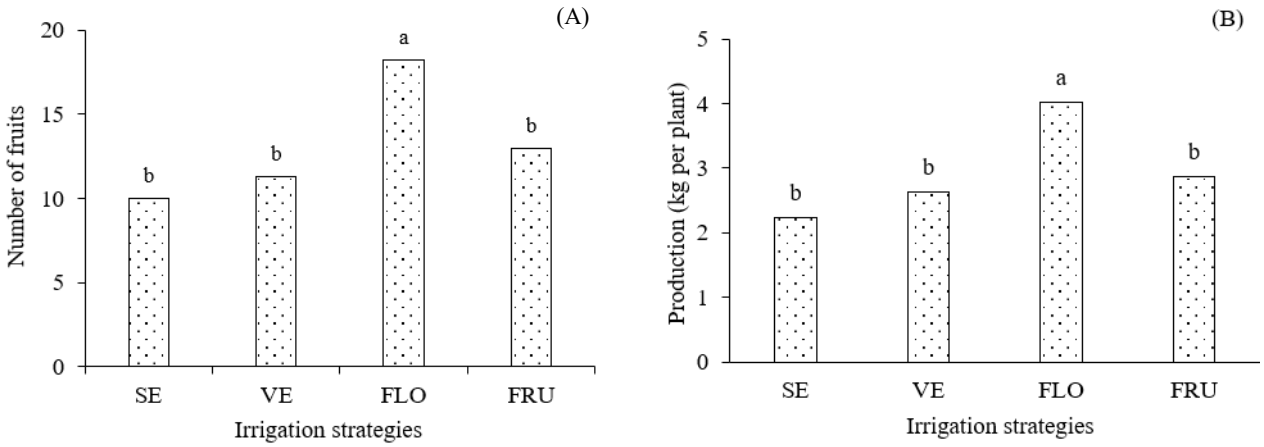
In plants receiving no AsA application, crown diameter was not affected by the irrigation strategy (Figure 5B). Among the AsA concentrations, the application of 200 mg L^{-1} stood out in plants grown without salt stress (SE). Thus, the maintenance of crown diameter indicates that plant growth was sustained by AsA application. A similar response was observed for the vegetative vigor index (Figure 5C), which remained at values comparable to those of the control across all irrigation strategies in the absence of AsA application. In contrast, under the control irrigation strategy (SE), the application of 200 mg L^{-1} AsA was the only treatment that resulted in a distinct response. This may be explained by the role of AsA in plant stress signaling, as it can help mitigate oxidative stress and regulate several biochemical processes, acting both as an enzymatic cofactor and as a modulator of cell division, thereby contributing to the maintenance of plant growth (CELI et al., 2023).



Bars with the same lowercase letters indicate no significant differences among irrigation strategies, whereas bars with the same uppercase letters indicate no significant differences among AsA concentrations (Tukey's test, $p \leq 0.05$)

Figure 5. Crown volume (A), crown diameter (B), and vegetative vigor index (C) of guava plants as affected by saline water irrigation strategies applied at different phenological stages and foliar application of ascorbic acid at different concentrations.

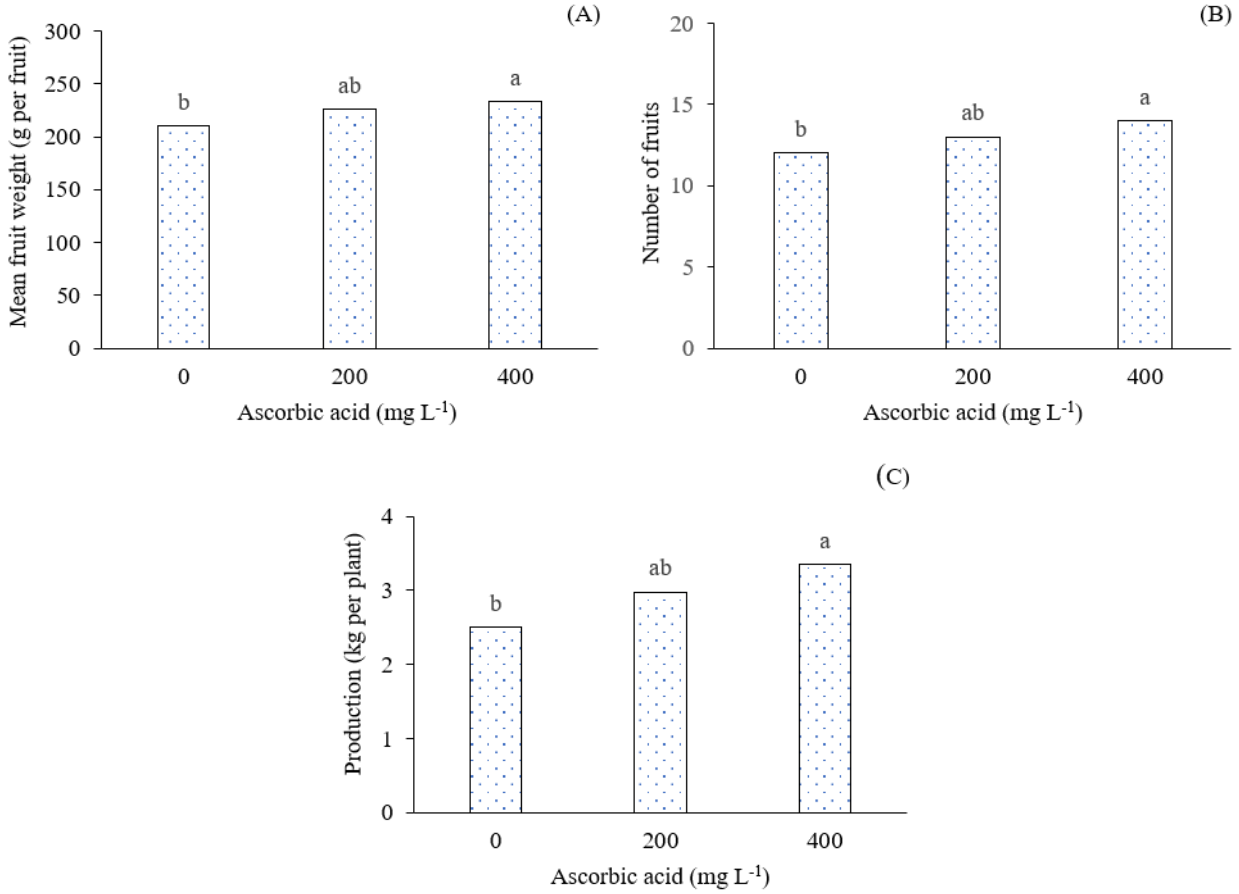
The number of fruits (Figure 6A) and production per plant (Figure 6B) were higher in plants irrigated with moderately saline water (3.3 dS m^{-1}) during the flowering stage (FLO) than in those irrigated with water at an ECw of 0.9 dS m^{-1} throughout the crop cycle (SE) or with water at an ECw of 3.3 dS m^{-1} during the vegetative (VE) and fruiting (FRU) stages. This response suggests a possible plant signaling process during flowering, potentially promoting flower formation and, following the resumption of irrigation with low-salinity water, maintaining fruit set (GHANI; AWANG; ISMAIL, 2018; SOARES et al., 2021a). The higher yield observed in plants irrigated with water at an ECw of 3.3 dS m^{-1} during flowering, as reflected by the greater number and mass of fruits, indicates that this may be the phenological stage at which guava plants are most tolerant to salt stress. This finding is particularly relevant because irrigation with low-electrical-conductivity water during the other phenological stages appears necessary to prevent reductions in yield components.



Bars with the same letter do not differ significantly according to Tukey's test ($p \leq 0.05$)

Figure 6. Number of fruits (A) and production per plant (B) of guava plants as affected by saline water irrigation strategies applied at different phenological stages.

The application of 400 mg L^{-1} AsA resulted in the highest mean fruit weight (Figure 7A), number of fruits (Figure 7B), and production per plant (Figure 7C) compared with plants receiving no AsA (0 mg L^{-1}). These findings highlight the beneficial role of AsA in regulating plant metabolism and potentially reducing energy losses commonly associated with semiarid environments. They also reinforce its role in energy dissipation, which may help prevent damage caused by reactive oxygen species under the climatic conditions prevailing during this study (HASSAN et al., 2021).



Bars with the same letter do not differ significantly according to Tukey's test ($p \leq 0.05$)

Figure 7. Mean fruit weight (A), number of fruits (B), and production per plant (C) of guava plants as affected by ascorbic acid concentrations.

These findings are consistent with those reported by Torres et al. (2025), who observed that foliar application of ascorbic acid at a concentration of 90 mM stimulated the synthesis of chlorophyll *a*, chlorophyll *b*, and total chlorophyll in guava plants irrigated with saline water (2.8 dS m^{-1}). This response may be associated with the activity of carbonic anhydrase and changes in the concentrations of phenolic compounds, flavonoids, and tannins, as well as reduced malondialdehyde levels, which are directly or indirectly related to antioxidant mechanisms.

CONCLUSIONS

Irrigation with water at an electrical conductivity of 3.3 dS m^{-1} during the flowering stage enhances leaf transpiration, number of fruits, and production per plant in grafted guava plants. Foliar application of ascorbic acid at concentrations of 200 and 400 mg L^{-1} helps maintain photosynthetic metabolism, increasing chlorophyll a concentrations and the CO_2 assimilation rate in guava plants subjected to salt stress during the vegetative and flowering stages, respectively. Ascorbic acid, particularly at a concentration of 400 mg L^{-1}, mitigates the effects of salt stress and promotes vegetative growth and yield components in guava plants irrigated with water at an electrical conductivity of 3.3 dS m^{-1} during the different phenological stages. Further studies are recommended to investigate the effects of saline water irrigation strategies and ascorbic acid application on the nutritional composition and postharvest quality of guava fruits.

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