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Scientific Article

Foliar application of ascorbic acid in guava cultivation under water replacement levels

Aplicação foliar de ácido ascórbico em goiabeira sob níveis de reposição hídrica

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ABSTRACT - Ascorbic acid is a non-enzymatic compound with antioxidant action in the detoxification of reactive oxygen species in plants subjected to abiotic stress conditions. Therefore, the objective of this study was to evaluate gas exchange, photochemical efficiency, and growth of guava cv. Paluma under water replacement levels and foliar application of ascorbic acid in the post-grafting phase. The experiment was carried out under greenhouse conditions, at the Center of Technology and Natural Resources, at the Federal University of Campina Grande, in Paraíba, PB, Brazil. A randomized block experimental design was used, in a 2 × 4 factorial arrangement, whose treatments resulted from the combination of two factors: two levels of irrigation water (50 and 100% of actual evapotranspiration - ETr) and four concentrations of ascorbic acid - AsA (0, 30, 60, and 90 mM), with three repetitions. Irrigation with a 50% ETr depth reduced the growth in rootstock diameter, scion diameter, and scion volume, as well as gas exchange: stomatal conductance, transpiration, and maximum and variable fluorescence indices of guava cv. Paluma. Ascorbic acid concentration of 90 mM increases the CO_2 assimilation rate and instantaneous water use efficiency and reduces electrolyte leakage in the leaf blade of guava plants, 240 days after transplanting.

Keywords: *Psidium guajava* L.. Water scarcity. Non-enzymatic compound.

RESUMO - O ácido ascórbico é um composto não-enzimático com ação antioxidante na desintoxicação de espécies reativas de oxigênio em plantas submetidas a condições de estresses abióticos. Diante disso, objetivou-se com esse trabalho avaliar as trocas gasosas, a eficiência fotoquímica e o crescimento da goiabeira cv. Paluma sob diferentes níveis de reposição de água e aplicação foliar de ácido ascórbico na fase pós-enxertia. O experimento foi desenvolvido sob condições de casa de vegetação, no Centro de Tecnologia e Recursos Naturais, da Universidade Federal de Campina Grande, na Paraíba-PB. Foi utilizado o delineamento experimental de blocos casualizados, em arranjo fatorial 2 × 4, cujos tratamentos resultaram da combinação de dois fatores: duas lâminas de água de irrigação (50 e 100% da evapotranspiração real - ETr) e quatro concentrações de ácido ascórbico - AsA (0, 30, 60 e 90 mM), com três repetições. A irrigação com lâmina de 50% da ETr reduziu o crescimento em diâmetro do porta enxerto, do enxerto, da copa e volume de copa; assim como as trocas gasosas: condutância estomática e, a transpiração, e os índices de fluorescência máxima e variável da goiabeira cv. Paluma. A concentração de 90 mM de ácido ascórbico aumenta a taxa de assimilação de CO_2 e a eficiência instantânea do uso da água e diminuiu o extravasamento de eletrólitos no limbo foliar das plantas de goiabeira, aos 240 dias após o transplantio.

Palavras-chave: *Psidium guajava* L.. Escassez hídrica. Composto não-enzimático.

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INTRODUCTION

Fruit growing has been gaining prominence in agricultural production over the years, and guava (*Psidium guajava* L.) is among the most cultivated fruit crop, due to the diversity of uses of its fruit, which can be consumed fresh or in processed products, such as jams, jellies, sweets among others (ONIAS et al., 2018). Brazil stands out as the fourth largest producer of guava in the world, obtaining production of around 564,764 tons and an average yield of 24,956 kg ha^{-1} in 2022. The Northeast region is the main contributor to the significant increase in guava production in Brazil, with Pernambuco, Bahia and Ceará as its main producing states, with an average production of 196,381, 50,431, and 22,844 tons and mean yield of 35,087, 20,020 and 15,435 kg ha^{-1}, respectively (IBGE, 2023).

Despite the importance of guava for the northeastern semi-arid region, the shortage of water for irrigation becomes a limiting factor for year-round production, which is a consequence of irregular rainfall and high evapotranspiration rates in this region (NÓBREGA et al., 2024). The condition of low water availability is one of the main limiting factors for the growth and development of plants, affecting their morphological, physiological, and biochemical processes, including nutrient absorption (LACERDA et al., 2022).

Plants grown under water stress emit chemical signals that are transmitted from the roots to the leaves via the xylem, inducing partial closure of the stomata to avoid excessive loss of water to the environment, consequently leading to reduction in intracellular CO_2 levels (CHENG et al., 2018). Zhou et al. (2017), in a study carried out with peach crop under different irrigation depths (100, 75, 50, and 25% ETc), found significant reductions in stomatal conductance, transpiration, photosynthetic rate, and water use efficiency due to the reduction in water availability. Such effects have also been observed on the growth and development of guava plants, as reported by Usman et al. (2022).

Among the alternatives used to reduce the negative effects of water stress on plants, exogenous application of elicitors, such as ascorbic acid (AsA), stands out. AsA is a non-enzymatic antioxidant compound that promotes reductions in reactive oxygen species (ROS) due to water stress conditions (AKRAM et al., 2017). It acts in the protection of proteins and lipids, increasing the activity of enzymes such as catalase and superoxide dismutase (NAZ et al., 2016). It improves the synthesis of photosynthetic pigments and growth of plants (ALINIAEIFARD et al., 2016), being essential to maintain plant physiological processes, such as cell division and root growth, which are influenced by ROS (WASZCZAK et al., 2018).

Several studies have reported the beneficial effect of AsA on plants under water stress conditions, as observed by Sarkar et al. (2016) in mandarin orange (*Citrus reticulata*) and by El-Bially et al. (2018) in sunflower (*Helianthus annuus*). However, so far no records have been found in the literature on the application of this compound in mitigating the deleterious effects of water stress on guava, especially under the conditions of the semi-arid region of the Brazilian Northeast. Therefore, the objective of this study was to evaluate the gas exchange, photochemical efficiency, and growth of guava cv. Paluma under different water replacement levels and foliar application of ascorbic acid in the post-grafting phase.

MATERIAL AND METHODS

The experiment was carried out from January to August 2022 under greenhouse conditions, at the Center of Technology and Natural Resources, Federal University of Campina Grande, in Campina Grande, Paraíba, Brazil (07° 15'18" S, 35°52'28" W, and 550 m altitude). According to Köppen's classification, the site has an average annual rainfall of 700 mm, with a semi-arid tropical climate - Bsh (ALVARES et al., 2013). Maximum and minimum temperature and relative humidity data obtained during the experimental period are presented in Figure 1.

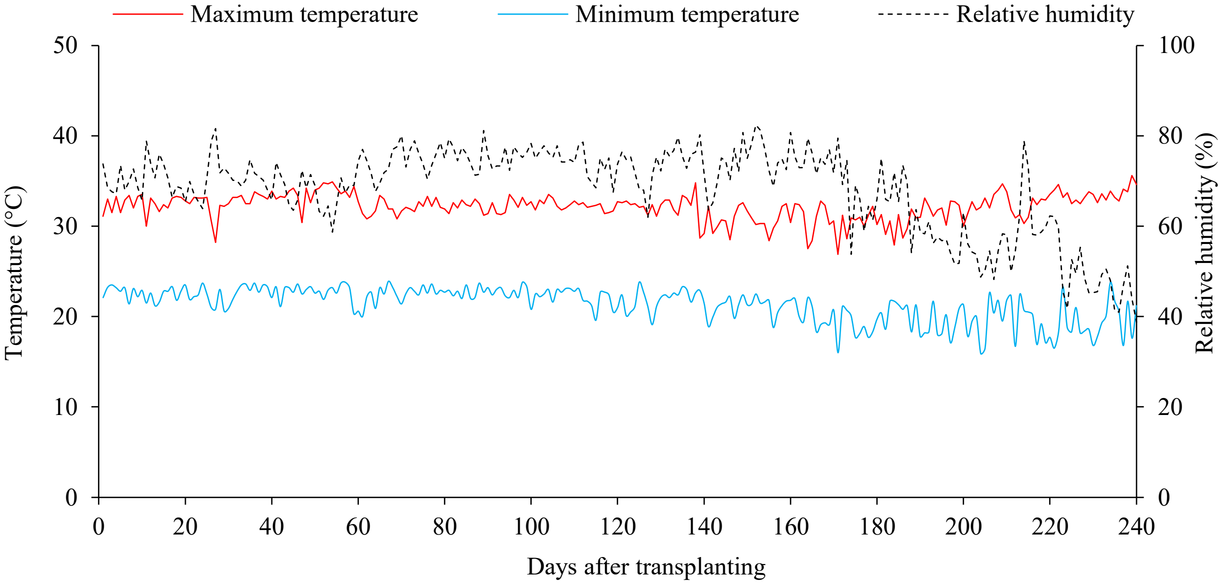


Figure 1. Data of maximum and minimum temperature (ºC) and relative humidity of air (%) during the experimental period.

A randomized block design was used, in a 2 × 4 factorial arrangement, whose treatments resulted from the combination of two factors: two irrigation water depths (50 and 100% of actual evapotranspiration - ETr) and four concentrations of ascorbic acid - AsA (0, 30, 60, and 90 mM), with three replicates. The depths were determined taking as references studies conducted by Ruiz-Sánchez et al. (2018) with peach (*Prunus persica*). AsA concentrations were based on a study carried out by Shafiq et al. (2014) with barley (*Hordeum vulgare*, L.). Recipients with 200 L capacity adapted as drainage lysimeters were used. Each lysimeter had a 16-mm-diameter drain at the bottom to eliminate excess water, connected to a container to collect the drained water, used to determine water consumded by the plants. To avoid clogging by the soil, the end of the drain inside the recepient was protected by a non-woven geotextile (Bidim OP 30).

The lysimeters were filled in layers, starting with 1 kg of crushed stone nº 0, followed by 250 kg of a *Neossolo Regolítico* soil (Entisol - Psamments) with a sandy clay loam texture, properly pounded to break up clods from the rural area of the municipality of Lagoa Seca, PB, at 0-20 cm depth, whose chemical and physical characteristics (Table 1) were obtained according to methodologies described by Teixeira et al. (2017).

In this study, ‘Crioula’ guava seedlings were used as rootstock and cv. Paluma as scion. The grafted seedlings were acquired 70 days after propagation by grafting, with rootstock diameter of 13.10 mm, scion diameter of 9.30 mm and average height of 38.21 cm at the time of transplanting. Guava transplanting into holes of 20 × 20 × 20 cm was performed 20 days after the acquisition of seedlings, which were previously checked for tangled roots. After transplanting, the seedlings were acclimatized for 50 days with full irrigation (100% ETr).

Table 1. Chemical and physical characteristics of the soil used in the experiment.

Chemical characteristics							
pH H_2O	OM	P	K^+	Na^+	Ca^{2+}	Mg^{2+}	$Al^{3+} + H^+$
1:2.5	g dm^{-3}	mg dm^{-3}	cmol$_c$ kg^{-1}				
6.5	8.1	79	0.24	0.51	14.90	5.40	0.90

Chemical characteristics						Physical characteristics				
EC_{se}	CEC	SAR_{se}	ESP	SB	V	Particle-size fraction (g kg^{-1})			Moisture (dag kg^{-1})	
dS m^{-1}	cmol$_c$ kg^{-1}	(mmol L^{-1})$^{0.5}$	%	cmol$_c$ kg^{-1}	%	Sand	Silt	Clay	33.42 kPa^1	1519.5 kPa^2
2.15	21.95	0.16	3.08	21.05	95.89	572.7	100.7	326.6	25.91	12.96

pH – Hydrogen potential, OM – Organic matter: Walkley-Black wet digestion; Ca^{2+} and Mg^{2+} extracted with 1 M KCl at pH 7.0; Na^+ and K^+ extracted with 1 M NH_4OAc at pH 7.0; Al^{3+}+H^+ extracted with 0.5 M CaOAc at 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} Referring to field capacity and permanent wilting point, respectively.

Before transplanting the seedlings, the soil moisture content was increased until reaching the maximum water retention capacity. After transplanting, irrigation was carried out daily at 5 p.m., and the volume of water to be applied to each lysimeter was determined by Equation 1:

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

Where:
VI – volume of water to be used in the irrigation event (mL);
Va - volume applied in the previous irrigation event (mL);
Vd - volume drained (mL); and,
LF - leaching fraction of 0.10 every 15 days.

Water replacement depth was determined based on the water consumed by plants under 100% ETr, using the drainage lysimetry method. The water depth of plants subjected to 50% ETr was determined by multiplying the value of the ETr obtained in 100% ETr treatment by 0.5.

Foliar application of AsA began 72 h before the beginning of the application of the different irrigation depths. Subsequently, the applications were carried out from 5 p.m., at 30-day intervals until the plants reached the full flowering stage. Ascorbic acid concentrations were obtained by dissolution in distilled water, which was prepared on the day of each application.

During the applications, the leaves were sprayed on both sides (abaxial and adaxial) to ensure complete wetting. A surfactant (Tween 20 - 0.025%) was added to the spray solutions to reduce the surface tension of the water, facilitating the absorption of AsA by the leaves. During the applications, the plants were isolated with a structure made of plastic tarpaulin to avoid the drift of the solutions between different treatments. Each plant received an average of 1.016 L of solution in a total of 12 applications.

Formative pruning was carried out when the plants reached a height of 50 cm, by cutting the main branch to stimulate the production of lateral branches and reduce apical dominance. With the emergence of new branches, 4 well-distributed branches were selected, and these lateral branches were later cut when they reached 40 cm in length, according to the methodology described by Lacerda et al. (2022).

Fertilization with nitrogen (N), potassium (K), and phosphorus (P) was performed according to the recommendation of Cavalcanti (2008), applying 100, 100, and 60 g per plant of N, P_2O_5, and K_2O. Urea (45% N), potassium chloride (60% K_2O) and monoammonium phosphate (50% P_2O_5 and 11% N) were used as sources of nitrogen, potassium, and phosphorus, respectively. Fertilization began at 15 days after transplanting (DAT) and was carried out in fortnightly applications via fertigation.

Fertilization with micronutrients was also carried out fortnightly through the leaves, starting at 30 DAT, on the adaxial and abaxial surfaces, applying Dripsol Micro® at a concentration of 1 g L^{-1} (0.85% boron, 3.4% iron, 4.2% zinc, 3.2% manganese, 0.5% copper, and 0.06% molybdenum).

Phytosanitary management was carried out preventively to avoid the emergence of pests such as psyllid (*Triozoida limbata*), fruit fly (*Anastrepha* spp., *Ceratitis capitata*), passionvine bug (*Leptoglossus gonagra*) and Florida wax scale (*Ceroplastes floridensis*), through selective chemicals based on Imidacloprid and Acetamiprid, recommended especially for guava crop, using, respectively 5 and 2.5 mL for 10 L, in the preparation of the mixture.

At 240 DAT, growth was evaluated by: rootstock diameter (RSD) and scion diameter (SCD), measured using a digital caliper; crown diameter (DCrown), obtained by the average crown diameter observed in the row direction (DR) and interrow direction (DIR); crown volume (VCrown), calculated from plant height (H), crown diameter in the row direction (DR), and crown diameter in the interrow direction (DIR); and vegetative vigor index (VVI), calculated according to the methodology described by Portella et al. (2016), as shown in Equations 2 and 3:

$$\text{VCrown} = \left(\frac{\pi}{6}\right) \times \text{H} \times \text{DR} \times \text{DIR} \tag{2}$$

Where:
VCrown – crown volume (m^3);
H – plant height (m);
DR – crown diameter in the row direction (m); and
DIR – crown diameter in the interrow direction (m).

$$\text{VVI} = \frac{[\text{H} + \text{DCrown} + (\text{SD} \times 10)]}{100} \tag{3}$$

Where:
VVI – vegetative vigor index;
H – plant height (m);
DCrown – crown diameter (m); and
SD – stem diameter (m).

Gas exchange was evaluated at 240 DAT, based on stomatal conductance – (*gs*, mol H_2O m^{-2} s^{-1}), transpiration – (*E*, mmol H_2O m^{-2} s^{-1}), CO_2 assimilation rate – (*A*, µmol CO_2 m^{-2} s^{-1}), internal CO_2 concentration – (*Ci*, µmol CO_2 m^{-2} s^{-1}), and instantaneous water use efficiency – (*WUEi*, [(µmol CO_2 m^{-2} s^{-1}) (mmol H_2O m^{-2} s^{-1})$^{-1}$], determined using an infrared gas analyzer – IRGA (Model LCPro – SD, from ADC BioScientific, UK). Readings were taken between 7:00 and 10:00 a.m., on the third fully expanded leaf counted from the apical bud under natural conditions of air temperature and CO_2 concentration, using an artificial radiation source of 1,200 µmol m^{-2} s^{-1} established through the light response curve of photosynthesis.

Chlorophyll *a* fluorescence was measured by initial fluorescence (F_0), maximum fluorescence (Fm), variable fluorescence (Fv), and effective quantum yield of PSII (Fv/Fm) on leaves pre-adapted to the dark by using leaf clips for 30 minutes, between 07:00 and 10:00 a.m., on the leaf of the middle third of the plant in order to ensure that all first acceptors were oxidized, i.e., the reaction centers were opened, using an OS5p pulse-modulated fluorometer from Opti Science. During the same period, electrolyte leakage (EL%) was determined after using a copper hole punch to obtain five leaf discs with an area of 1.54 cm^2 each, per experimental unit, which were washed and stored in an Erlenmeyer® flask containing 50 mL of distilled water. After being closed with aluminum foil, the Erlenmeyer® flasks were stored at temperature of 25 °C for 90 min, the initial electrical conductivity of the medium (ECi) was measured using a benchtop conductivity meter (MB11, MS Techonopon®). Soon after, the Erlenmeyer® flasks were subjected to a temperature of 90 °C for 90 min in a drying oven (SL100/336, SOLAB®) and, after cooling of the contents, the final electrical conductivity (ECf) was determined. The electrolyte leakage in the leaf blade was expressed as the percentage, according to the methodology proposed by Scotti-Campos et al. (2013), as shown in Equation 4:

$$\text{EL\%} = \frac{\text{ECi}}{\text{ECf}} \times 100 \tag{4}$$

Where:
EL% - electrolyte leakage in the leaf blade (%);
ECi - initial electrical conductivity (dS m^{-1}); and,
ECf - final electrical conductivity (dS m^{-1}).

The data collected in this study were initially tested for normality using the Shapiro-Wilk test and then subjected to analysis of variance using the F test; when there was significance, linear and quadratic regression analysis was performed for ascorbic acid concentrations, using the statistical software SISVAR-ESAL version 5.7.

RESULT AND DISCUSSION

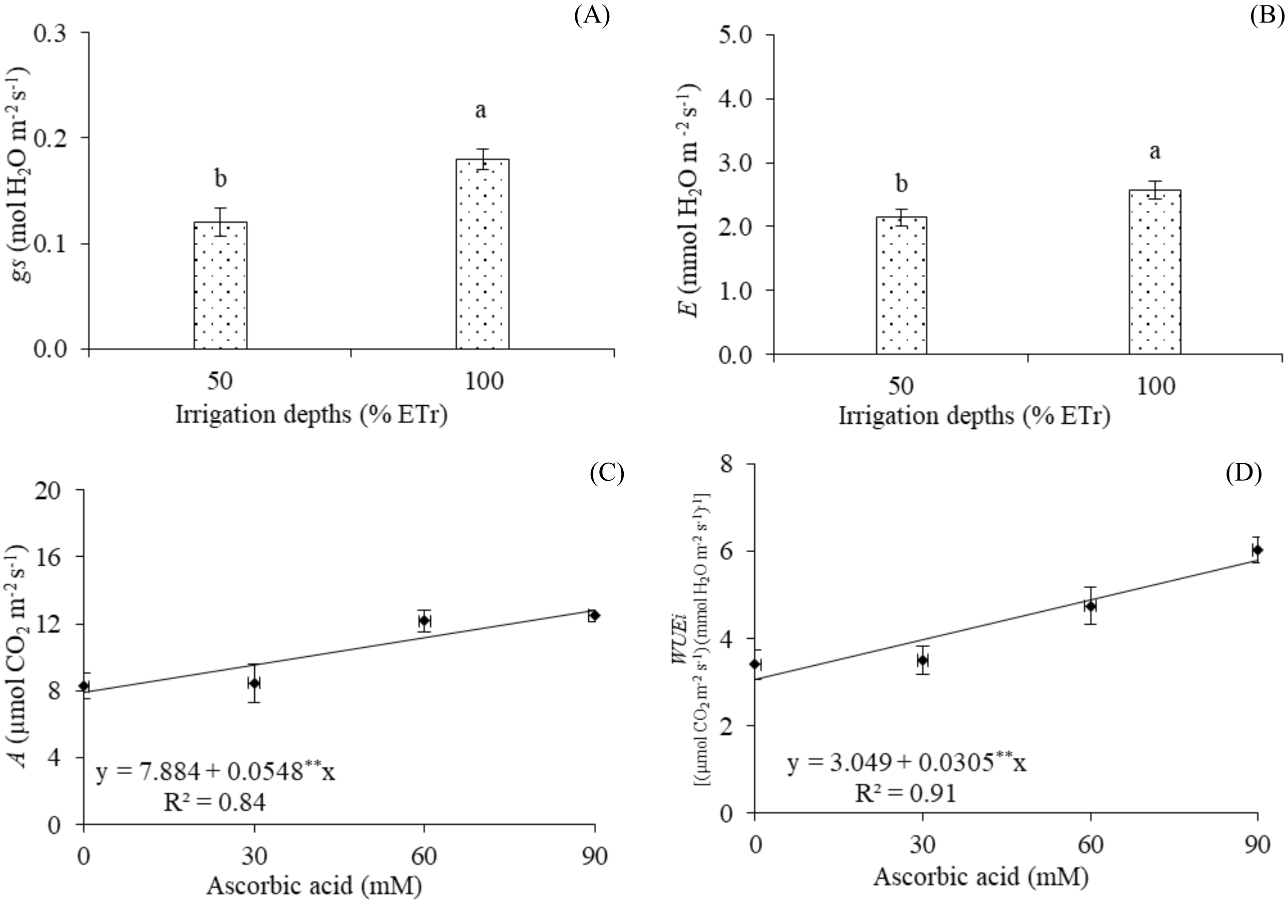
There was a significant effect of the irrigation depths on the stomatal conductance (*gs*) and transpiration (*E*) of guava plants. Ascorbic acid concentrations significantly influenced the CO_2 assimilation rate (*A*) and instantaneous water use efficiency (*WUEi*) of guava cv. Paluma, at 240 DAT (Table 2). The interaction between the factors (ID × AsA) did not significantly influence any of the variables analyzed in guava cv. Paluma, at 240 DAT.

Table 2. Summary of the analysis of variance for stomatal conductance (*gs*), transpiration (*E*), internal CO_2 concentration (*Ci*), CO_2 assimilation rate (*A*) and instantaneous water use efficiency (*WUEi*) of guava cv. Paluma under water replacement levels and foliar application of ascorbic acid (AsA) at 240 days after transplanting.

Sources of variation	DF	Mean squares				
		gs	*E*	*Ci*	*A*	*WUEi*
Irrigation depths (ID)	1	247.041^{*}	235.626^{*}	2.788^{ns}	4.175^{ns}	2.561^{ns}
Ascorbic acid (AsA)	3	247.041^{ns}	0.550^{ns}	0.045^{ns}	0.108^{*}	0.079^{*}
Linear Regression	1	0.108^{ns}	0.867^{ns}	0.083^{ns}	0.172^{*}	0.000^{*}
Quadratic Regression	1	22.815^{ns}	0.540^{ns}	0.001^{ns}	0.047^{ns}	0.232^{ns}
Interaction (ID × AsA)	3	2.451^{ns}	2.430^{ns}	0.052^{ns}	0.014^{ns}	0.024^{ns}
Blocks	2	2.373^{ns}	0.050^{ns}	0.322^{ns}	0.100^{ns}	0.024^{ns}
Residual	14	0.000	0.088	611.863	2.587	0.465
CV (%)		5.87	1.12	0.02	0.04	0.05

*and **significant at 0.05 and 0.01 probability level by the F test, respectively; ^{ns} not significant by the F test; CV= coefficient of variation; DF – Degrees of freedom.

The limitation in water availability caused reductions in stomatal conductance - *gs* and transpiration - *E* of guava (Figures 2A and 2B). Plants cultivated under full irrigation (100% ETr) differed significantly from those subjected to a depth of 50% ETr. When comparing the *gs* and *E* of plants irrigated with depths of 50 and 100%, reductions of 33.3 and 16.7% were observed, respectively. The reduction of *gs* in plants under water restriction is a defense mechanism, which prevents excessive loss of water to the atmosphere, also causing reductions in plant transpiration (LIMA et al., 2020). Usman et al. (2022), in a study carried out with guava under irrigation depths of 100, 75, and 50% of field capacity, found reductions in physiological variables (chlorophyll contents, transpiration, and CO_2 assimilation rate), as well as morphological variables (leaf area and leaf fresh weight) due to the restriction in water availability, corroborating the results observed in the present study.



Means followed by different letters indicate a significant difference between the irrigation depths by the F test ($p \leq 0.05$. Vertical bar represents the standard error of the mean (n=3)

Figure 2. Stomatal conductance – *gs* (A) and transpiration – *E* (B) of guava cv. Paluma, as a function of the irrigation depths – ETr and CO_2 assimilation rate – *A* (C) and instantaneous water use efficiency – *WUEi* (D), as a function of the concentrations of ascorbic acid – AsA, at 240 days after transplanting.

CO_2 assimilation rate (*A*) and water use efficiency (*WUEi*) increased linearly with the increase of ascorbic acid concentrations (Figures 2C and 2D). Increments of 0.0548 µmol CO_2 m^{-2} s^{-1} and 0.0305 [(µmol CO_2 m^{-2} s^{-1}) (mmol H_2O m^{-2} s^{-1})$^{-1}$] for *A* and *WUEi*, respectively, were observed per unit increase in AsA concentration. When comparing the *A* and *WUEi* of plants grown under foliar application of 90 mM to those of plants that received 0 mM, increments of 62.55 and 90.02% were observed, respectively. This increase can be attributed to the antioxidant properties of ascorbic acid, which help in the elimination of ROS and in the increase in the synthesis of photosynthetic pigments; these characteristics are fundamental for the control of physiological processes in plants (WASZCZAK et al., 2018). El-Bially et al. (2018), in a study with sunflower crop under irrigation depths (100, 80, and 70% ETr) and 400 ppm of ascorbic acid, also observed increase in water use efficiency in treatments that received foliar application of AsA.

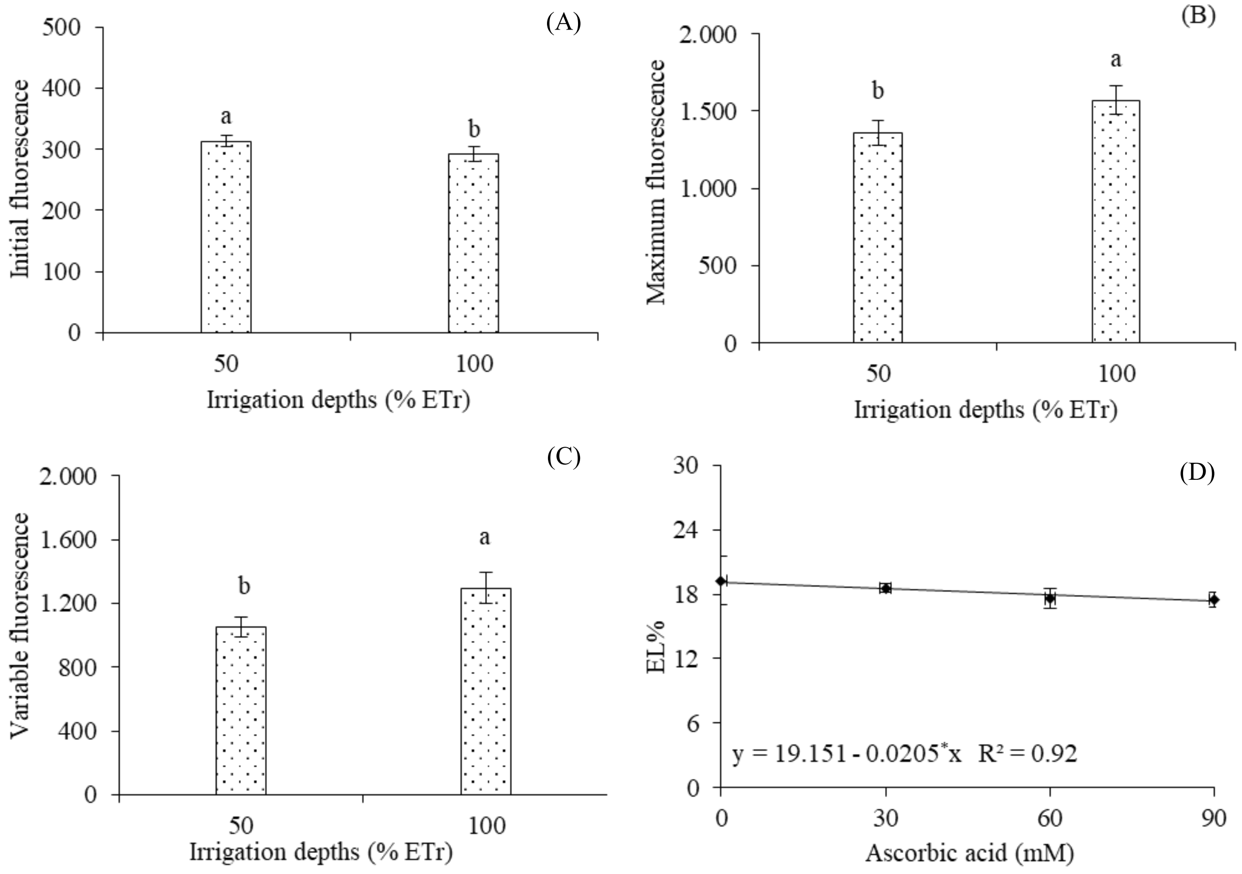
Irrigation depths had a significant effect on initial fluorescence (F_0), maximum fluorescence (Fm), and variable fluorescence (Fv). Ascorbic acid concentrations significantly influenced the electrolyte leakage (EL) of guava cv. Paluma, at 240 DAT (Table 3). The interaction between the factors (ID × AsA) did not significantly influence any of the variables analyzed, at 240 DAT.

Table 3. Summary of the analysis of variance for initial fluorescence (F_0), maximum fluorescence (Fm), variable fluorescence (Fv), quantum efficiency of photosystem II (Fv/Fm), and electrolyte leakage (EL%) in the leaf blade of guava cv. Paluma under water replacement levels and foliar application of ascorbic acid (AsA) at 240 days after transplanting.

Sources of variation	Mean squares					
	DF	F_0	Fm	Fv	Fv/Fm	EL%
Irrigation depths (ID)	1	2604.166*	265440.666*	362112.666*	0.022^{ns}	15.456^{ns}
Ascorbic acid (AsA)	3	1093.611^{ns}	20895.11^{ns}	28513.000^{ns}	0.011^{ns}	40.219*
Linear Regression	1	730.133^{ns}	10304.533	51170.700^{ns}	0.014^{ns}	0.726*
Quadratic Regression	1	48.166^{ns}	130.666^{ns}	22448.133^{ns}	0.017^{ns}	58.468^{ns}
Interaction (ID × AsA)	3	2145.833^{ns}	150127.111^{ns}	104341.000^{ns}	0.005^{ns}	11.775^{ns}
Blocks	2	80.79^{ns}	37189.541^{ns}	30318.166^{ns}	0.004^{ns}	3.480^{ns}
Residual	14	409.125	27000.065	30849.309	0.003	2.947
CV (%)		6.68	11.23	14.99	7.74	9.33

* and **significant at 0.05 and 0.01 probability level by the F test, respectively; ^{ns} not significant by the F test; CV= coefficient of variation; DF – Degrees of freedom.

Irrigation with a depth of 50% ETr promoted an increase in F_0 of guava plants cv. Paluma (Figure 3A), being 7.13% higher than that obtained in plants cultivated under 100% ETr. The increase in F_0 may be an indication of damage caused by water stress in the components of the PSII antenna and reaction center system, which are oxidized, thus losing the ability to transport electrons (LOPES, 2016). When studying coffee seedlings under water stress with different depths (25, 50, 75, 100, 125, 150, and 200% of available water in the soil), Cintra et al. (2020) found an increase in F_0 as a function of the reduction in the available water depth.



Means followed by different letters indicate a significant difference between the irrigation depths by the F test ($p \leq 0.05$). Vertical bar represents the standard error of the mean (n=3)

Figure 3. Initial fluorescence - F_0 (A), maximum fluorescence - Fm (B), variable fluorescence - Fv (C) of guava cv. Paluma, as a function of irrigation depths and electrolyte leakage – EL% (D), as a function of ascorbic acid concentrations – AsA, at 240 days after transplanting.

Maximum (Fm) and variable (Fv) fluorescence (Figures 3B and 3C) reached their highest values in plants irrigated with 100% ETr. When compared to plants irrigated with 50% ETr, there were reductions of 13.41 and 18.97%, respectively. The reduction in Fm and Fv may be related to a deficiency in the photoreduction of quinone A, a consequence of the inactivation of PSII, which reduces the flow of electrons between the photosystems and the photochemical activity in the leaves (DIAS et al., 2019). Souza et al. (2020) also observed decreases in maximum and variable fluorescence in sugarcane plants grown under different irrigation depths (100, 80, 60, and 40% of field capacity) as water availability was restricted.

The electrolyte leakage (EL%) in the leaf blade of guava decreased with the increase in the concentration of ascorbic acid (Figure 3D), by 0.107% per unit increase in AsA. When comparing plants subjected to a concentration of 90 mM with those without treatment (0 mM), a reduction of 8.99% in EL% was observed. Damage to the cell membrane in plants under water deficit may be a consequence of the action of ROS that cause peroxidation of lipids and proteins (SHAFIQ et al., 2014). The decrease in EL% observed in the present study may be related to the role of ascorbic acid, which acts in the protection of proteins and lipids in plants (AKRAM et al., 2017). Similar results were reported by Dantas et al. (2023), who studied mini-watermelon under irrigation depths (50, 75, 100, and 125% ETr) and observed decreases in EL% according to the reduction in water availability.

There was a significant effect of the irrigation depths on the rootstock diameter (RSD), scion diameter (SCD), crown volume (VCrown), and crown diameter (DCrown) of guava cv. Paluma, at 240 DAT (Table 4). Ascorbic acid concentrations and the interaction between the factors (ID × AsA) did not significantly influence any of the variables analyzed at 240 DAT.

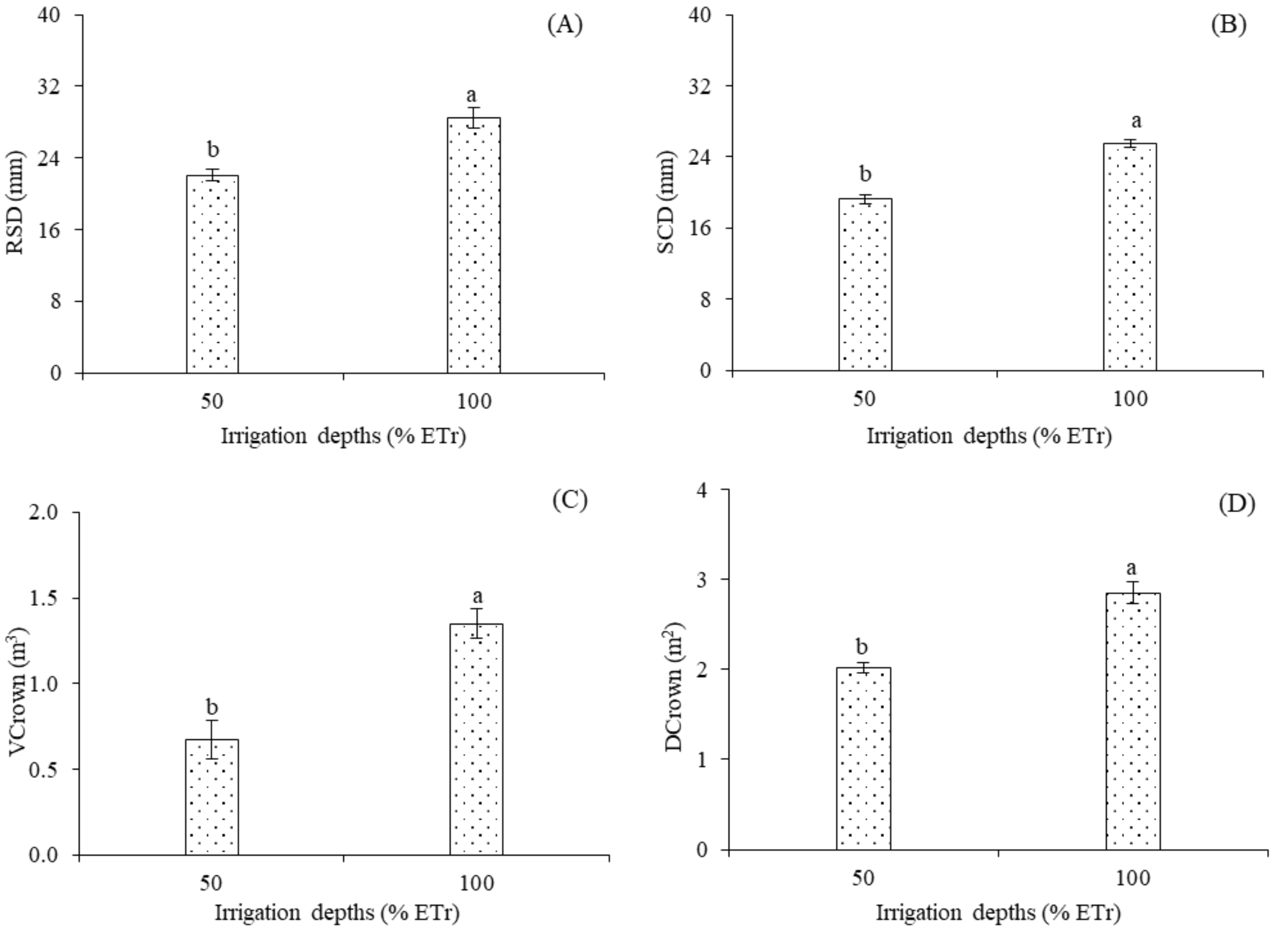
Table 4. Summary of the analysis of variance for rootstock diameter (RSD), scion diameter (SCD), crown volume (VCrown), crown diameter (DCrown), and vegetative vigor index (VVI) of guava cv. Paluma under water replacement levels and foliar application of ascorbic acid (AsA) at 240 days after transplanting.

Sources of variation	DF	Mean squares				
		RSD	SCD	VCrown	DCrown	VVI
Irrigation depths (ID)	1	247.041*	235.626*	2.788*	4.175*	2.561^{ns}
Ascorbic acid (AsA)	3	247.041^{ns}	0.550^{ns}	0.045^{ns}	0.108^{ns}	0.079^{ns}
Linear Regression	1	0.108^{ns}	0.867^{ns}	0.083^{ns}	0.172^{ns}	0.000^{ns}
Quadratic Regression	1	22.815^{ns}	0.540^{ns}	0.001^{ns}	0.047^{ns}	0.232^{ns}
Interaction (ID × AsA)	3	2.451^{ns}	2.430^{ns}	0.052^{ns}	0.014^{ns}	0.024^{ns}
Blocks	2	2.373 ^{ns}	0.050 ^{ns}	0.322 ^{ns}	0.100 ^{ns}	0.024 ^{ns}
Residual	14	5.879	1.128	0.026	0.040	0.059
CV (%)		9.59	4.75	16.12	8.24	9.49

*and **significant at 0.05 and 0.01 probability level by the F test, respectively; ^{ns} not significant by the F test; CV= coefficient of variation; DF – Degrees of freedom.

The reduction in the irrigation depth inhibited the growth in rootstock diameter (RSD) and scion diameter (SCD) of guava cv. Paluma (Figures 4A and 4B). Plants subjected to irrigation depth with 100% ETr had statistically higher RSD and SCD than those that received 50% ETr. When comparing the RSD and SCD of plants irrigated with a replacement depth of 100% ETr to those of plants cultivated under 50% ETr, increments of 22.54 and 14.53% were observed, respectively. Low water availability in plants under water deficit conditions reduces turgor pressure, limiting cell division and expansion, and consequently affecting their growth (ANJUM et al., 2016). These results corroborate those reported by Fátima et al. (2023), who studied sour passion fruit under water stress conditions (depths of 50 and 100% ETr) and also found reduction in the growth in stem diameter of 23.2% when plants received 50% ETr, compared to those under 100% ETr.

The crown volume (VCrown) and crown diameter (DCrown) of plants irrigated with a depth of 100% ETr were statistically higher than those of plants irrigated with a depth of 50% ETr (Figures 4C and 4D). When comparing the VCrown and DCrown of plants irrigated with 100% ETr to those of plants irrigated with 50% ETr, decreases of 50.37 and 29.12% were observed, respectively. The reduction of crown growth in plants under water stress is probably a defense mechanism to reduce the transpiring area and prevent water loss to the atmosphere (SOARES et al., 2023). This effect can also be attributed to the partial closure of stomata in plants under conditions of water deficit, which reduces the production of photoassimilates and consequently reduces reserves for metabolic activities, growth, and production (RICKES et al., 2017). The results found here corroborate those reported by Ruiz-Sánchez et al. (2018), who evaluated peach under water stress (replacement levels ranging from 50 to 120% ETr) and observed that deficit irrigation negatively affected plant growth; however, this condition was reversed after rehydration.



Means followed by different letters indicate a significant difference between the irrigation depths by the F test ($p \leq 0.05$). Vertical bar represents the standard error of the mean (n=3)

Figure 4. Rootstock diameter – RSD (A), scion diameter – SCD (B), crown volume – VCrown (C), and crown diameter – DCrown (D) of guava cv. Paluma, as a function of irrigation depths, at 240 days after transplanting.

CONCLUSION

Irrigation with a depth of 50% ETr reduced growth in rootstock diameter, scion diameter, crown diameter, and crown volume, as well as gas exchange: stomatal conductance and transpiration, and the maximum and variable fluorescence indices of guava cv. Paluma.

Ascorbic acid concentration of 90 mM increases the CO_2 assimilation rate and instantaneous water use efficiency and reduces the electrolyte leakage in the leaf blade of guava plants, at 240 days after transplanting.

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