ISSN 1807-1929
Revista Brasileira de Engenharia Agrícola e Ambiental
Brazilian Journal of Agricultural and Environmental Engineering
v.26, n.11, p.855-862, 2022
Campina Grande, PB – http://www.agriambi.com.br – http://www.scielo.br/rbeaa

DOI: http://dx.doi.org/10.1590/1807-1929/agriambi.v26n11p855-862

Salicylic acid alleviates salt stress on guava plant physiology during rootstock formation[1]

O ácido salicílico ameniza o estresse salino na fisiologia de goiabeira na formação de porta-enxerto

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HIGHLIGHTS:

Salicylic acid increased the concentrations of chlorophyll b and total chlorophyll up to 1.60 and 1.50 mM L^{-1}, respectively.
Salicylic acid concentrations of up to 1.3 mM L^{-1} decreases leaf electrolyte leakage.
Water electrical conductivity of 4.2 dS m^{-1} and 3.2 mM L^{-1} of salicylic acid decreased relative water content and Fv/Fm fluorescence.

ABSTRACT: Guava is a fruit widely produced in Northeast Brazil, a region that has sources of water with high levels of salts, making it difficult for irrigated fruit production to expand. Thus, it is extremely important to search for techniques that allow the management of these waters in irrigated agriculture. Thus, the objective of present study was to evaluate the photosynthetic pigments, the quantum yield of photosystem II, and the intercellular leakage of electrolytes in the leaf blade of 'Paluma' guava seedlings as a function of irrigation with water of different salinities and foliar application of salicylic acid during rootstock formation phase. The experiment was conducted in a greenhouse, using the randomized block design in a 5 × 5 factorial arrangement, which consisted of five values of electrical conductivity of water - ECw (0.6, 1.5, 2.4, 3.3, and 4.2 dS m^{-1}) and five concentrations of salicylic acid (0, 0.8, 1.6, 2.4, and 3.2 mM L^{-1}), with four replicates, and two plants per plot. Salicylic acid up to 1.3 mM L^{-1} increased the relative water content and decreased electrolyte leakage in the leaf blade of 'Paluma' guava seedlings. Foliar application of salicylic acid at a concentration of 1.7 mM L^{-1} attenuated the effects of salt stress on chlorophyll a of guava seedlings irrigated with water of up to 1.6 dS m^{-1}. The deleterious effects of salt stress on carotenoid concentration and quantum efficiency of photosystem II were not attenuated by the application of salicylic acid.

Key words: *Psidium guajava* L., rootstock, acclimatization

RESUMO: A goiaba é uma fruta amplamente produzida no Nordeste do Brasil, região que possui fontes de água com altos teores de sais, dificultando a expansão da fruticultura irrigada. Assim, é de extrema importância a busca por técnicas que permitam o manejo dessas águas para a agricultura irrigada. Assim, objetivou-se avaliar os pigmentos fotossintéticos, o rendimento quântico do fotossistema II, e o extravasamento de eletrólitos celulares no limbo foliar de mudas de goiabeira 'Paluma' em função da irrigação com águas de diferentes salinidades e aplicação foliar de ácido salicílico durante a fase de formação de porta-enxerto. O experimento foi conduzido em casa de vegetação, utilizando-se o delineamento de blocos casualizados em arranjo fatorial 5 × 5, sendo cinco valores de condutividade elétrica da água - CEa (0,6, 1,5, 2,4, 3,3 e 4,2 dS m^{-1}) e cinco concentrações de ácido salicílico (0, 0,8, 1,6, 2,4 e 3,2 mM L^{-1}), com quatro repetições e duas plantas por parcela. O ácido salicílico até a concentração de 1,3 mM L^{-1} aumentou o teor relativo de água e diminuiu o extravasamento de eletrólitos no limbo foliar de mudas de goiaba 'Paluma'. A aplicação foliar do ácido salicílico na concentração de 1,7 mM L^{-1} amenizou os efeitos do estresse salino sobre a clorofila a das mudas de goiaba irrigadas com água de até 1,6 dS m^{-1}. Os efeitos deletérios do estresse salino sobre o teor de carotenoides e rendimento quântico do fotossistema II não foram atenuados pela aplicação do ácido salicílico.

Palavras-chave: *Psidium guajava* L., porta-enxerto, aclimatação

• Ref. 261077 – Received 15 Feb, 2022
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• Accepted 05 July, 2022 • Published 15 July, 2022
Editors: Rener Luciano de Souza Ferraz & Walter Esfrain Pereira

Introduction

Guava is a tropical fruit crop native to South and Central America and its fruit can be easily found in open markets and supermarkets throughout Brazil (Manica et al., 2001; Alencar et al., 2016). Guava is a fruit with characteristic flavor and aroma which are pleasant to the taste, with high food value and that can be consumed both fresh and through juices, jams, liqueurs, ice cream, and others (Bezerra et al., 2019).

The irregularity of rainfall in the Brazilian semi-arid region and the long periods of drought make irrigation indispensable for agricultural production in this region; however, the use of water sources with electrical conductivity above 1.5 dS m^{-1} limits the expansion of irrigated agriculture (Lima et al., 2016; Bezerra et al., 2019). High concentrations of salts in irrigation water can impair photosynthetic processes, restrict stomatal opening and CO_2 assimilation, and increase the degradation of chlorophyll synthesis and the production of reactive oxygen species (ROS), triggering oxidative stress (Silva et al., 2022) and changes in the functional state of chloroplast thylakoid membranes, which cause changes in the characteristics of fluorescence signals (Dias et al., 2018).

As most cultivated species are sensitive to the presence of excess salts, it is extremely important to search for alternatives capable of alleviating the effects of salt stress, since the use of saline water in agriculture is a necessity to ensure production (Dias et al., 2019; Lima et al., 2022). Among the alternatives capable of alleviating the deleterious effects of salt stress, foliar application of salicylic acid (SA) stands out. SA is a phenolic compound that plays an important role in plant tolerance to salt stress, by producing osmolytes and secondary metabolites (Khan et al., 2015). Salicylic acid is synthesized in plants due to abiotic sources and accumulates in the tissues, thus contributing to increasing their resistance to stress conditions through improvement in nutrient absorption, membrane protection, and photosynthesis maintenance, and may also interact with signaling pathways of reactive oxygen species, reducing oxidative stress (Lotfi et al., 2020; Souana et al., 2020).

Studies conducted to evaluate the effect of SA application on the induction of tolerance to salt stress in soursop (Silva et al., 2020), West Indian cherry (Dantas et al., 2021), and guava (Lacerda et al., 2022a; Lacerda et al., 2022b) have reported improvements in plant physiology, growth and post-harvest fruit quality. Thus, the objective of present study was to evaluate the photosynthetic pigments, the quantum yield of photosystem II, and the intercellular leakage of electrolytes in the leaf blade of 'Paluma' guava seedlings as a function of irrigation with water of different salinities and foliar application of salicylic acid during rootstock formation phase.

Material and Methods

The experiment was conducted from October 2020 to April 2021 in a greenhouse (7° 15' 18" S latitude, 35° 52' 28" W longitude and an average altitude of 550 m) belonging to the Unidade Acadêmica de Engenharia Agrícola of the Universidade Federal de Campina Grande (UFCG), in Campina Grande, PB, Brazil. The air temperature (maximum and minimum) and relative humidity data during the experimental period were collected using a thermohygrometer and are shown in Figure 1.

The treatments consisted of the combination of five values of electrical conductivity of irrigation water - ECw (0.6, 1.5, 2.4, 3.3, and 4.2 dS m^{-1}) and five concentrations of salicylic acid - SA (0 - Control, 0.8, 1.6, 2.4, and 3.2 mM L^{-1}), distributed in randomized blocks in a 5 × 5 factorial arrangement with four replicates and two plants per plot. The values of electrical conductivity of irrigation water were based on the study conducted by Bezerra et al. (2019) in which they evaluated ECw ranging from 0.3 to 3.5 dS m^{-1}; whereas salicylic acid concentrations were adapted from a study conducted by Silva et al. (2020) with soursop.

In this study, the cultivar Paluma was evaluated as a rootstock. The seeds used in the experiment were obtained from a guava orchard in the experimental area of the Centro de Ciências e Tecnologia Agroalimentar (CCTA/UFCG), on the Campus of Pombal - PB (6° 48' 50" S; 37° 56' 31" W, 190 m), belonging to the UFCG, and were manually extracted by pulping the fruits and subsequently air-dried in an open environment.

Salicylic acid concentrations were obtained by dissolution of the acid in 30% of ethyl alcohol. The solution was freshly prepared in all events of biweekly applications, and the Wil fix spreader was added to assist in the fixation of SA on the leaves.

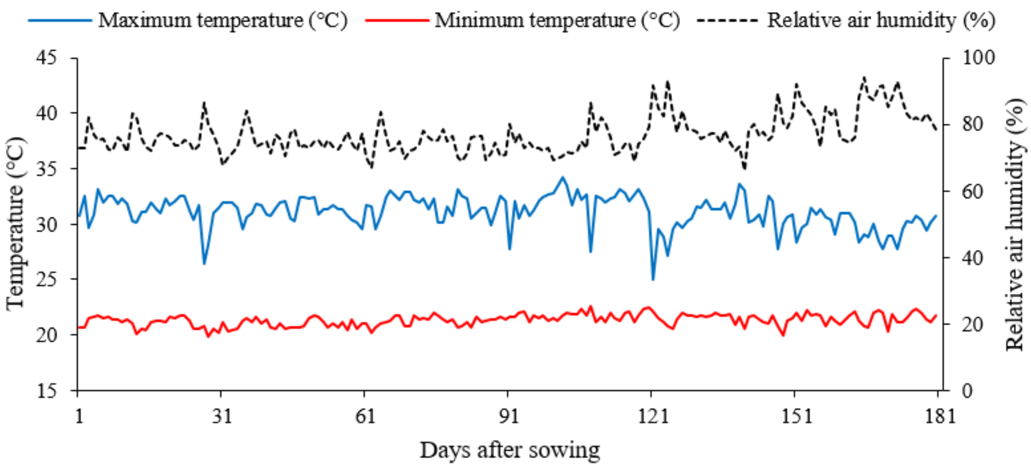


Figure 1. Mean values of air temperature (maximum and minimum) and relative air humidity observed in the internal area of the greenhouse during the experimental period

Table 1. Chemical and physical-hydraulic attributes of the soil used in the experiment, before the application of the treatments

Chemical attributes								
pH (H_2O) (1:2.5)	OM (g dm^{-3})	P (mg dm^{-3})	K^+	Na^+	Ca^{2+}	Mg^{2+}	Al^{3+}	H^+
			(cmol$_c$ kg^{-1})					
6.5	8.1	79	0.24	0.51	14.9	5.4	0	0.9

Chemical characteristics			Physical characteristics				
EC_{se} (dS m^{-1})	CEC (cmol$_c$ kg^{-1})	ESP (%)	Particle-size fraction (g kg^{-1})			Moisture (dag kg^{-1})	
			Sand	Silt	Clay	FC[1]	PWP[2]
2.15	21.95	2.32	572.7	100.7	326.6	25.91	12.96

OM - Organic matter: Walkley-Black Wet Digestion; Ca^{2+} and Mg^{2+} - Extracted with 1 M L^{-1} KCl at pH 7.0; Na^+ and K^+ - Extracted with 1 M L^{-1} NH_4OAc at pH 7.0; Al^{3+} and H^+ - Extracted with 0.5 M L^{-1} calcium acetate at pH 6.5; CEC - Cation exchange capacity; ESP - Exchangeable sodium percentage; ECse - Electrical conductivity of saturation extract; [1]Field capacity at tension of 33.42 kPa; [2]Permanent wilting point at tension of 1519.5 kPa

The seedlings were produced in plastic bags with dimensions of 10 × 20 cm, filled with 1.6 kg of a substrate in the proportion of 3:1 (soil:aged bovine manure, volume basis) of a soil classified as Entisol of sandy loam texture (0-20 cm, A horizon), from the municipality of Lagoa Seca, PB, whose chemical and physical-hydraulic characteristics are presented in Table 1.

Irrigation waters with different electrical conductivities were prepared by dissolving NaCl, $CaCl_2$, and $MgCl_2$ in local-supply water (ECw = 0.28 dS m^{-1}), following the equivalent ratio of 7:2:1 for Na^+, Ca^{2+}, and Mg^{2+}, commonly found in the water sources of Northeastern Brazil (Medeiros, 1992); the quantities of salts were determined considering the relationship between ECw and the salt concentration (Richards, 1954), according to Eq. 1. After preparation, the solutions were checked and, if necessary, adjustments were made.

$$Q \approx 10 \times ECw \tag{1}$$

where:

Q - sum of cations (mmol$_c$ L^{-1}); and,

ECw - electrical conductivity of water (dS m^{-1}).

Before sowing, the soil moisture content was increased to the level corresponding to the maximum retention capacity using water of the lowest level of electrical conductivity (ECw = 0.6 dS m^{-1}). Sowing was performed by placing four seeds per bag equidistantly at a depth of 2 cm. The thinning was performed only once, at 67 days after sowing (DAS), leaving only one plant per bag. After sowing, irrigation was carried out daily at 16:00 h, applying in each bag the volume corresponding to that obtained by the water balance, determined by Eq. 2:

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

where:

VI - volume of water to be used in the next irrigation event (mL);

Va - volume applied in the previous irrigation event (mL);

Vd - volume drained (mL); and,

LF - leaching fraction of 0.15.

Seedling emergence began at 20 DAS. After emergence, fertilization with nitrogen, phosphorus, and potassium was performed as recommended by Novais et al. (1991), increasing the levels of N, P_2O_5, and K_2O by 100, 300, and 150 mg kg^{-1}, respectively, via fertigation, with the solution prepared using public-supply water (0.28 dS m^{-1}). The fertilization was split into nine portions and applied from 40 DAS at intervals of 15 days. Foliar applications with micronutrients at the concentration of 2.5 g of Ubyfol® L^{-1} [(N (15%); P_2O_5 (15%); K_2O (15%); Ca (1%); Mg (1.4%); S (2.7%); Zn (0.5%); B (0.05%); Fe (0.5%); Mn (0.05%); Cu (0.5%) and Mo (0.02%)] were performed on the adaxial and abaxial surfaces, fortnightly, in order to meet the needs of micronutrients.

The application of SA at 67 DAS was initiated when the plants showed uniform growth, and the other applications were performed every two weeks until 165 DAS. To minimize the evaporation of the solution from the leaf surface, SA applications were carried out between 17:00 and 18:00 h. Before applying SA, the plants were removed from the proximity of the others, avoiding the cross-application of different concentrations in each plot, and later returned to their respective place after spraying. During the experiment, eight applications of SA were carried out, using an average volume of 50 mL of SA per plant in each event.

Irrigation with the different values of water salinity started 75 DAS, at a daily interval. At 180 DAS, electrolyte leakage, relative water content in the leaf blade, photosynthetic pigments, and quantum yield of photosystem II of guava seedlings were measured. Electrolyte leakage (%EL) in the leaf blade was determined using five discs of 1.54 cm^2 each, per experimental unit, which were washed, placed in an Erlenmeyer® flask containing 50 mL of distilled water and kept at room temperature (≈ 32 °C) for 90 min; after this time, the initial electrical conductivity (ECi) was measured. Subsequently, the Erlenmeyer® flasks were covered with aluminum foil and kept in a forced-air circulation oven for 90 min at 90 °C; after that time, they were removed and left at room temperature to cool to check the final electrical conductivity (ECf). Electrolyte leakage in the leaf blade was determined according to the methodology of Scotti-Campos et al. (2013), through Eq. 3:

$$\%EL = \frac{ECi}{ECf} \times 100 \tag{3}$$

where:

%EL - percentage of electrolyte leakage in the leaf blade;

ECi - initial electrical conductivity; and,

ECf - final electrical conductivity.

To determine relative water content (RWC), five discs of 12 mm in diameter were collected in leaves from the middle third of the plant. Immediately after collection, the discs were weighed, thus avoiding moisture loss, to obtain the fresh mass (FM); after weighing, the samples were immersed in 50 mL of distilled water for 24 hours; after this period, excess water on the discs was removed using paper towels and the samples were weighed to obtain the turgid mass (TM). The samples were dried in an oven at 65 ± 3 °C, until reaching constant mass, to obtain the dry mass (DM). RWC was determined according to Weatherley (1950), through Eq. 4:

$$RWC = \frac{FM - DM}{TM - DM} \times 100 \quad (4)$$

where:

RWC - relative water content (%);
FM - leaf fresh mass;
DM - leaf dry mass; and,
TM - leaf turgid mass.

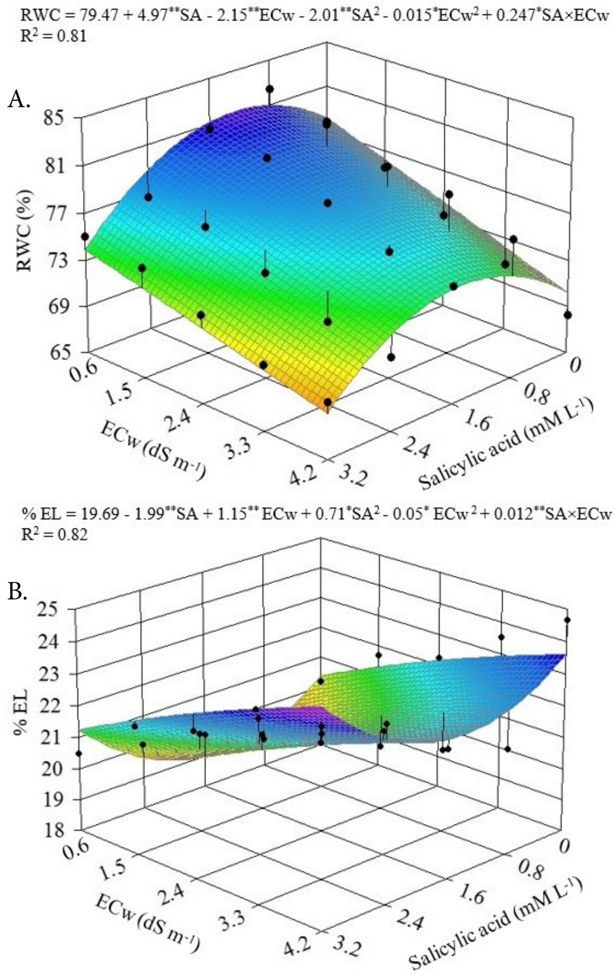
Photosynthetic pigments (chlorophyll a, chlorophyll b, total chlorophyll, and carotenoids) were determined according to the methodology of Arnon (1949). Quantum yield was determined by the initial fluorescence (F_0, electrons quantum^{-1}), maximum fluorescence (Fm, electrons quantum^{-1}), variable fluorescence (Fv, electrons quantum^{-1}), and quantum efficiency of photosystem II (Fv/Fm) in leaves pre-adapted to the dark using leaf clips for 30 min (ensuring that all reaction centers were open), between 07:00 and 09:30 a.m., using the pulse-modulated fluorometer Plant Efficiency Analyser - PEA II®.

The data collected were subjected to the normality and homogeneity test (Shapiro-Wilk test) followed by the analysis of variance. Subsequently, they were subjected to analysis of variance and polynomial regression analysis ($p \leq 0.05$), using the statistical program SISVAR version 5.6 (Ferreira, 2019). In cases where the interaction between the factors was significant, the TableCurve 3D software was used to obtain the response surface curves.

Results and Discussion

There was a significant effect of the interaction between the electrical conductivity of water and concentrations of salicylic acid (ECw × SA) on the relative water content (RWC), percentage of electrolyte leakage (%EL) in the leaf blade, and contents of chlorophyll a (Chl a) and carotenoids (Car) of 'Paluma' guava seedlings, at 180 days after sowing (Table 2). There was also a significant effect of ECw on Chl total of guava seedlings. On the other hand, SA concentrations significantly influenced Chl b and Chl total contents.

Plants subjected to the application of 1.3 mM L^{-1} of SA had the highest value (81.43%) of relative water content in the leaf blade (Figure 2A), equivalent to an increase of 4.0% when



*, ** - Significant at $p \leq 0.05$ and $p \leq 0.01$, respectively, by F test

Figure 2. Response surface for relative water content - RWC (A) and percentage of electrolyte leakage - %EL (B) in the leaf blade of 'Paluma' guava seedlings, as a function of the interaction between electrical conductivity of water - ECw and concentration of salicylic acid - SA, 180 days after sowing

Table 2. Summary of the analysis of variance for relative water content (RWC), percentage of electrolyte leakage (%EL), chlorophyll a (Chl a), chlorophyll b (Chl b), total chlorophyll (Chl total), and carotenoids (Car) of 'Paluma' guava seedlings as a function of irrigation water electrical conductivity and concentration of salicylic acid, 180 days after sowing

Source of variation	DF	Mean squares					
		RWC	%EL	Chl a	Chl b	Chl total	Car
Salinity levels (ECw)	4	33.98^{ns}	26.93**	32.6**	0.93^{ns}	35.19**	2.38**
Linear regression	1	16.72^{ns}	53.21**	88.30**	0.19^{ns}	80.28**	3.14**
Quadratic regression	1	33.33^{ns}	28.79**	0.17^{ns}	1.56^{ns}	2.80^{ns}	0.13^{ns}
Salicylic acid (SA)	4	172.78*	13.95**	7.08**	1.87**	15.39**	0.04^{ns}
Linear regression	1	46.62^{ns}	0.00^{ns}	0.93^{ns}	0.03^{ns}	1.31^{ns}	0.08^{ns}
Quadratic regression	1	535.26**	41.01**	24.29**	7.02**	57.40**	0.03^{ns}
Interaction (ECw × SA)	16	119.53**	12.57**	2.87*	0.51^{ns}	4.42^{ns}	0.61**
Blocks	3	119.47^{ns}	18.24**	3.53^{ns}	1.68*	0.86^{ns}	0.17^{ns}
Residual	72	48.90	4.16	1.58	0.52	2.79	0.27
CV (%)		9.07	9.70	23.92	37.35	23.19	25.52

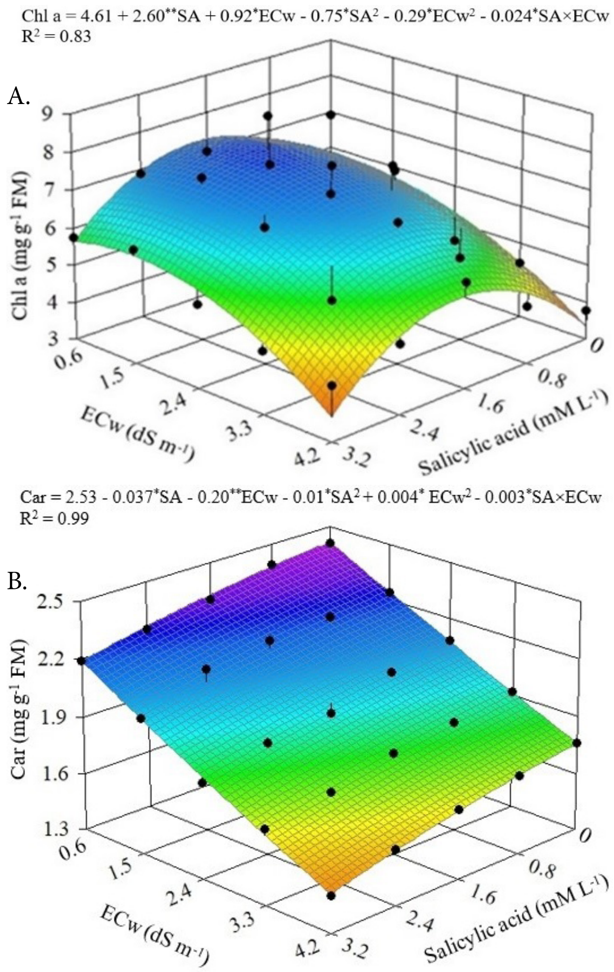
DF - Degrees of freedom; CV - Coefficient of variation; *, ** and ^{ns} - Significant at $p \leq 0.05$ and $p \leq 0.01$ and not significant, respectively compared to plants subjected to the same ECw (0.6 dS m^{-1}) without application of SA (0 mM L^{-1}), which showed RWC of 78.18%. The lowest RWC value (68.82%) was obtained in plants grown under SA concentration of 3.2 mM L^{-1} and ECw of 4.2 dS m^{-1}. This response may be associated with the effect exerted by the SA on the accumulation of osmolytes, contributing to the absorption and increase in the relative content of water in the tissues (Silva et al., 2021a). Farheen et al. (2018), evaluating the effect of SA application (50 μM) on mung bean (*Vigna radiata*) subjected to salt stress (50, 100, and 150 mM of NaCl), observed that plants subjected to SA maintained a higher relative water content, a prerequisite for plant survival and, consequently, biomass production.

For electrolyte leakage - %EL in the leaf blade (Figure 2B), the application of 1.3 mM L^{-1} of SA mitigated the effects of salt stress on plants subjected to ECw between 0.6 and 4.2 dS m^{-1}. However, the lowest value of %EL (18.98%) was obtained in plants subjected to ECw of 0.6 dS m^{-1} in the absence of foliar application of SA (0 mM L^{-1}). On the other hand, the highest value of %EL (24.70%) was reached in plants subjected to ECw (4.2 dS m^{-1}) and SA concentration of 3.2 mM L^{-1}. The excess of salts in irrigation water tends to increase electrolyte leakage, due to the toxic effects of Na^+ and Cl^- ions, which, when absorbed and accumulated in the leaves, cause structural changes in cell membranes and consequent rupture, leading to increase in reactive oxygen species (ROS), which cause lipid peroxidation and oxidation of cell membranes and organelles, also resulting in degradation and loss of the internal contents of the cell (Sachdev et al., 2021), as observed in Figure 2A, with the lowest RWC obtained in plants subjected to the highest level of water salinity (4.2 dS m^{-1}).

Silva et al. (2021a) in a study evaluating the effect of foliar application of salicylic acid (0, 1.2, 2.4, and 3.6 mM L^{-1}) on soursop crop subjected to irrigation with saline water (0.8 - control; 1.6, 2.4, 3.2, and 4.0 dS m^{-1}), also observed that the concentration of 1.3 mM L^{-1} mitigated salt stress effects on the crop.

The application of SA at the concentration of 1.7 mM L^{-1} promoted an increment of 48.50% (2.45 mg g^{-1} FM) in the chlorophyll a contents (Figure 3A) of plants irrigated with ECw of 1.6 dS m^{-1} when compared to those subjected to the same salinity level (1.6 dS m^{-1}) and without application of SA (0 mM L^{-1}). The increase observed in Chl a contents (Figure 3A) may be related to the antioxidant action promoted by salicylic acid, which removes ROS under conditions of abiotic stress, improving photosynthetic synthesis (Dantas et al., 2021). Farheen et al. (2018) also observed that the application of salicylic acid (50 μM L^{-1}) in mung bean (*Vigna radiata*) subjected to salt stress (50, 100, and 150 mM L^{-1} of NaCl) significantly increased the chlorophyll a, chlorophyll b and carotenoid contents of the leaves.

For carotenoid concentrations (Figure 3B), it was verified that plants subjected to the highest salinity level (4.2 dS m^{-1}) and foliar application of 3.2 mM L^{-1} of SA had a reduction of 43.60% (1.05 mg g^{-1} FM) when compared to those grown under control treatment (0.6 dS m^{-1} and 0 mM L^{-1} of SA) and obtained the maximum estimated value of carotenoids (2.41 mg g^{-1} FM). Carotenoids are integrated components of



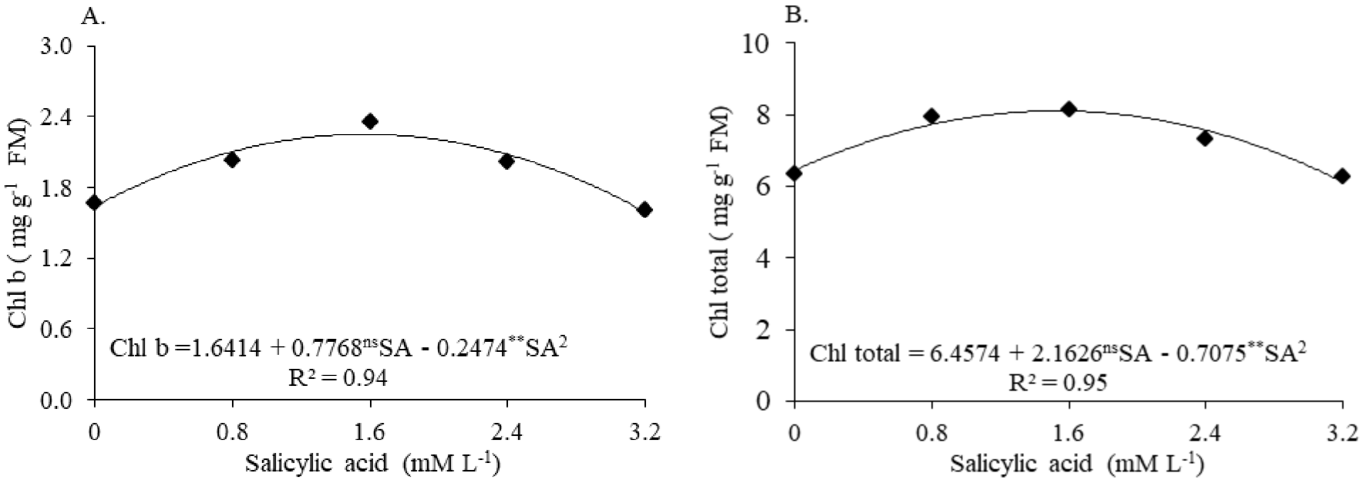
FM - Fresh matter; *, ** - Significant at $p \leq 0.05$ and $p \leq 0.01$, respectively, by F test

Figure 3. Response surface for chlorophyll a - Chl a (A) and carotenoids - Car (B) concentrations of 'Paluma' guava seedlings as a function of the interaction between electrical conductivity of water - ECw and concentration of salicylic acid - SA, 180 days after sowing

thylakoids and are responsible for the absorption and transfer of light to chlorophyll. Decrease in Car content may be related to the damage caused by salt stress to chloroplasts and the loss of activity of pigmentation proteins, besides stimulating the activity of the chlorophyll enzyme, which acts in the degradation of photosynthetic pigments (Dias et al., 2019; Silva et al., 2021b).

Salicylic acid (SA) concentrations significantly influenced the contents of Chl b and Chl total (Figures 4A and B) of 'Paluma' guava seedlings. Application of SA positively influenced Chl b contents up to the estimated concentration of 1.60 mM L^{-1}, where the maximum value of 2.25 mg g^{-1} FM was obtained, and the minimum value of 1.59 mg g^{-1} FM was found in plants under 3.2 mM L^{-1}. Salicylic acid also promoted a beneficial effect on the synthesis of Chl total, with the maximum estimated value of 8.10 mg g^{-1} FM obtained in plants grown under SA concentration of 1.50 mM L^{-1}.

The positive effect of salicylic acid on the synthesis of photosynthetic pigments varies with the applied concentration, mode of application, and stage of crop development and, as observed in Figures 4A and B, the SA concentrations of up to 1.5 mM L^{-1} mitigated the effect of salinity on Chl b and



FM - Fresh matter; **, ns - Significant at p ≤ 0.01 and not significant, respectively, by F test

Figure 4. Chlorophyll b - Chl b (A) and total chlorophyll - Chl total (B) of 'Paluma' guava seedlings as a function of salicylic acid concentrations - SA, 180 days after sowing

Chl total contents. Dantas et al. (2021) also observed a positive effect of SA application on the total chlorophyll contents of West Indian cherry under irrigation with waters of different salinity levels (0.8 to 4.0 dS m^{-1}) and concentrations of salicylic acid (0 to 4.0 mM L^{-1}). Despite the different ECw values having significantly influenced the total chlorophyll content of guava seedlings (Table 2), there was no satisfactory fit to the data for prognostic purposes and the regression model (Chl total = 8.4682 - 0.7921^{**}ECw) had R^2 value less than 0.60.

There was a significant effect of the interaction between the values of electrical conductivity of water and concentrations of salicylic acid (ECw × SA) on maximum fluorescence (Fm), variable fluorescence (Fv), and quantum efficiency of photosystem II (Fv/Fm) (Table 3). However, there was no significant effect of salinity levels (ECw) and salicylic acid concentrations (SA) on the initial fluorescence (F_0) of 'Paluma' guava seedlings.

Guava seedlings irrigated with water of low electrical conductivity (0.6 dS m^{-1}) and subjected to an estimated salicylic acid concentration of 0.9 mM L^{-1}, obtained the highest value of maximum fluorescence (1,806.6 electrons $quantum^{-1}$) (Figure 5A), which corresponded to an increase of 1.56% (27.7 electrons $quantum^{-1}$) compared to those under control treatment (0 mM L^{-1}) and irrigated with the same electrical conductivity. However, in plants grown under the highest concentration of SA (3.2 mM L^{-1}), the increase in salinity led to a reduction in Fm, with the lowest value (1,346.5 electrons $quantum^{-1}$) obtained in those subjected to ECw of 4.2 dS m^{-1}. The reduction in Fm under ECw of 4.2 dS m^{-1} may indicate the ability of the plant to activate photoprotection mechanisms associated with heat dissipation via the xanthophyll cycle (Silva et al., 2021b).

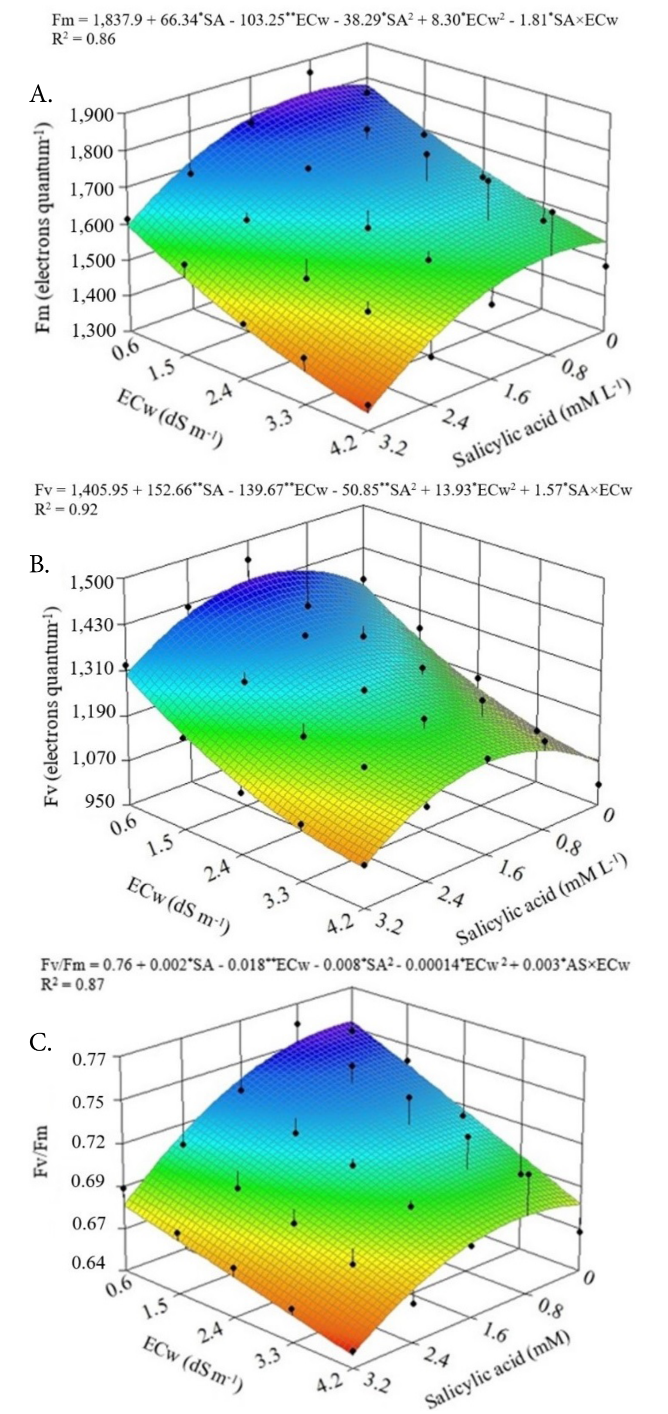
Foliar application of salicylic acid increased the variable fluorescence (Figure 5B) of plants subjected to the lowest electrical conductivity (0.6 dS m^{-1}) under the concentration of 1.5 mM L^{-1}, an increase of 8.74% (115.99 electrons $quantum^{-1}$) when compared to the control treatment (0 mM L^{-1}) under the same water salinity level. The lowest estimated value of Fv (1,053.97 electrons $quantum^{-1}$) was obtained in plants subjected to the highest ECw (4.2 dS m^{-1}) and SA concentration (3.2 mM L^{-1}).

Maximum fluorescence directly influences variable fluorescence, which is the potentially active energy in photosystem II (Sá et al., 2018), in addition to representing the plant's ability to transfer electrons from pigment molecules to energy storage (NADPH, ATP, and reduced ferredoxin - Fdr), which implies a greater CO_2 assimilation capacity in the biochemical phase of photosynthesis (Dias et al., 2019). Thus, the reduction in Fv is related to maximum fluorescence losses and may also be related to the decrease observed in

Table 3. Summary of the analysis of variance for the initial fluorescence (F_0), maximum fluorescence (Fm), variable fluorescence (Fv), and quantum efficiency of photosystem II (Fv/Fm) of 'Paluma' guava seedlings irrigated with different salinity levels as a function of irrigation water electrical conductivity and concentration of salicylic acid, 180 days after sowing

Source of variation	DF	Mean squares			
		F_0	Fm	Fv	Fv/Fm
Salinity levels (ECw)	4	$1,955.63^{ns}$	$255,446.58^{**}$	$79,951.91^{ns}$	$1.37\times10^{-3\ ns}$
Linear regression	1	256.85^{ns}	$1,832.42^{ns}$	$6,813.65^{ns}$	$2.00\times10^{-6\ ns}$
Quadratic regression	1	123.25^{ns}	$348,813.42^{**}$	$123,675.80^{ns}$	$2.52\times10^{-3\ ns}$
Salicylic acid (SA)	4	683.43^{ns}	$82,177.91^{*}$	$97,448.59^{ns}$	$4.28\times10^{-3*}$
Linear regression	1	245.97^{ns}	$348,813.43^{**}$	$228,217.00^{*}$	$1.17\times10^{-2**}$
Quadratic regression	1	$2,050.82^{ns}$	$335,570.24^{**}$	$145,373.31^{ns}$	$4.8\times10^{-3\ ns}$
Interaction (ECw × SA)	16	$1,532.20^{ns}$	$109,749.63^{**}$	$90,427.50^{*}$	$3.89\times10^{-3**}$
Blocks	3	$3,447.69^{*}$	$15,026.01^{ns}$	$2,164.67^{ns}$	$1.72\times10^{-3\ ns}$
Residual	72	956.53	33,348.76	47,775.90	1.51×10^{-3}
CV (%)		6.96	10.58	17.05	5.30

DF - Degrees of freedom; CV - Coefficient of variation; *,** and ns - Significant at p ≤ 0.05 and p ≤ 0.01 and not significant, respectively



*, ** - Significant at $p \leq 0.05$ and $p \leq 0.01$, respectively, by F test

Figure 5. Response surface for maximum fluorescence - Fm (A), variable fluorescence - Fv (B), and quantum efficiency of photosystem II - Fv/Fm (C) of 'Paluma' guava seedlings as a function of the interaction between electrical conductivity of water - ECw and concentrations of salicylic acid - SA, 180 days after sowing

chlorophyll a and total chlorophyll contents (Figures 3A and 4C) with the increase in salinity. Dias et al. (2021) verified that values of electrical conductivity of water above 2.6 dS m^{-1} led to reductions in chlorophyll a fluorescence parameters of West Indian cherry.

According to these authors, the decrease in fluorescence may be related to the degradation of chlorophyll pigments due to ionic toxicity, and decreases in Fv and Fm result in great damage to the photosynthetic apparatus caused by water deficit due to excess salinity.

Considering that Fv/Fm values between 0.75 and 0.85 are indicative that the photosynthetic apparatus is intact (Bolhàr-Nordenkampf, 1989), it can be said that the quantum efficiency of photosystem II - Fv/Fm (Figure 5C) was negatively affected by salinity and SA since plants subjected to the highest salinity level (4.2 dS m^{-1}) and SA concentration of 3.2 mM L^{-1} presented the lowest value (0.65) of Fv/Fm, a reduction of 13.67% (0.10) when compared to those under the control treatment (0 mM L^{-1} and 0.6 dS m^{-1}), and the highest Fv/Fm value (0.75) was obtained in plants irrigated with ECw of 0.6 dS m^{-1} and under foliar application of SA at the estimated concentration of 0.2 mM L^{-1}. The decrease in the quantum efficiency of photosystem II with the increase in salinity and SA is probably due to photoinhibition damage caused to the reaction centers of photosystem II (PSII), promoting the formation of ROS (Oliveira et al., 2018; Dias et al., 2019).

Conclusions

1. Salicylic acid up to a concentration of 1.3 mM L^{-1} increases the relative water content and decreases electrolyte leakage in the leaf blade of 'Paluma' guava seedlings.

2. Foliar application of 1.7 mM L^{-1} of salicylic acid mitigates the effects of salt stress on chlorophyll a of 'Paluma' guava seedlings irrigated with water of up to 1.6 dS m^{-1}. The concentrations of 1.6 and 1.5 mM L^{-1} of salicylic acid increase the concentrations of chlorophyll b and total chlorophyll, respectively.

3. The deleterious effects of salt stress on carotenoid concentration and photosystem II quantum efficiency are not attenuated by the application of salicylic acid.

Literature Cited

Alencar, R. D.; Leite, G. A.; Mendonça, V.; Lima, F. V. de; Pereira, G. A.; Farias, W. C. Adubação potássica na produção e qualidade pós-colheita de goiaba 'Paluma' no semiárido potiguar. Comunicata Scientiae, v.7, p.139-148, 2016. http://dx.doi.org/10.14295/CS.v7i1.1332

Arnon, D. I. Copper enzymes in isolated chloroplasts: Polyphenoloxidase in *Beta vulgaris*. Plant Physiology, v.24, p.1-15, 1949. https://doi.org/10.1104/pp.24.1.1

Bezerra, I. L.; Gheyi, H. R.; Nobre, R. G.; Lima, G. S. de; Lacerda, C. F. de; Lima, B. G. F.; Bonifácio, B. F. Water salinity and nitrogen fertilization in the production and quality of guava fruits. Bioscience Journal, v.35, p.837-848, 2019. http://dx.doi.org/10.14393/BJ-v35n3a2019-42005

Bolhàr-Nordenkampf, H. R.; Long, S. P.; Baker, N. R.; Oquist, G.; Schreiber, U.; Lechner, E. G. Chlorophyll fluorescence as probe of the photosynthetic competence of leaves in the field: a review of current instrumentation. Functional Ecology, v.3, p.497-514, 1989. https://doi.org/10.2307/2389624

Dantas, M. V.; Lima, G. S. de; Gheyi, H. R.; Silva, A. A. R. da; Melo, A. S. de; Medeiros, L. C. Gas exchange and photosynthetic pigments of West Indian cherry under salinity stress and salicylic acid. Comunicata Scientiae, v.12, p.1-9, 2021.

Dias, A. S.; Lima, G. S. de; Gheyi, H. R.; Furtado, G. F.; Soares, L. A. A.; Nobre, R. G.; Fernandes, P. D. Choroplast pigments and photochemical efficiency of West Indian cherry under salt stress and potassium-phosphorus fertilization. Semina: Ciencias Agrárias, v.42, p.87-104, 2021. http://dx.doi.org/10.5433/1679-0359.2021v42n1p87

Dias, A. S.; Lima, G. S. de; Pinheiro, F. W. A.; Gheyi, H. R.; Soares, L. A. A. Gas exchanges, quantum yield and photosynthetic pigments of West Indian cherry under salt stress and potassium fertilization. Revista Caatinga, v.32, p.429-439, 2019. https://doi.org/10.1590/1983-21252019v32n216rc

Dias, A. S.; Lima, G. S. de; Sá, F. V. da S.; Gheyi, H. R.; Soares, L. A. dos A.; Fernandes, P. D. Gas exchanges and photochemical efficiency of West Indian cherry cultivated with saline water and potassium fertilization. Revista Brasileira de Engenharia Agrícola e Ambiental, v.22, p.628-633, 2018. http://dx.doi.org/10.1590/1807-1929/agriambi.v22n9p628-633

Farheen, J.; Mansoor, S.; Abideen, Z. Exogenously applied salicylic acid improved growth, photosynthetic pigments and oxidative stability in mungbean seedlings (*Vigna radiata*) at salt stress. Pakistan Journal of Botany, v.50, p.901-912, 2018.

Ferreira, D. F. SISVAR: A computer analysis system to fixed effects split plot type designs. Revista Brasileira de Biometria, v.37, p.529-535, 2019. https://doi.org/10.28951/rbb.v37i4.450

Khan, M. I. R.; Fatma, M.; Per, T. S.; Anjum, N. A.; Khan, N. A. Salicylic acid-induced abiotic stress tolerance and underlying mechanisms in plants. Frontiers in Plant Science, v.6, p.1-17, 2015. https://doi.org/10.3389/fpls.2015.00462

Lacerda, C. N. de; Lima, G. S. de; Soares, L. A. dos A.; Fátima, R.T. de; Gheyi, H. R.; Azevedo, C. A. V. de. Morphophysiology and production of guava as a function of water salinity and salicylic acid. Revista Brasileira de Engenharia Agrícola e Ambiental, v.26, p.451-458, 2022a. http://dx.doi.org/10.1590/1807-1929/agriambi.v26n6p451-458

Lacerda, C. N. de; Lima, G. S. de; Soares, L. A. dos A.; Silva, A. A. R. da; Gheyi, H. R.; Silva, F. de A. da. Post-harvest fruit quality of grafted guava grown under salt stress and salicylic acid application. Revista Brasileira de Engenharia Agrícola e Ambiental, v.26, p.713-721, 2022b.

Lima, G. S. de; Gheyi, H. R.; Nobre, R. G.; Xavier, D. A.; Soares, L. A. dos A. Castor bean production and chemical attributes of soil irrigated with water with various cationic compositions. Revista Caatinga, v.29, p.54-65, 2016. http://dx.doi.org/10.1590/1983-21252016v29n107rc

Lima, G. S. de; Pinheiro, F. W. A.; Gheyi, H. R.; Soares, L. A. A.; Sousa, P. F. N.; Fernandes, P. D. Saline water irrigation strategies and potassium fertilization on physiology and fruit production of yellow passion fruit. Revista Brasileira de Engenharia Agrícola e Ambiental, v.26, p.180-189, 2022. https://doi.org/10.1590/1807-1929/agriambi.v26n3p180-189

Lotfi, R.; Ghassemi-Golezani, K.; Pessarakli, M. Salicylic acid regulates photosynthetic electron transfer and stomatal conductance of mung bean (*Vigna radiata* L.) under salinity stress. Biocatalysis and Agricultural Biotechnology, v.26, p.1-11, 2020. https://doi.org/10.1016/j.bcab.2020.101635

Manica, I.; Icuma, M.; Junqueira, N. T. V.; Salvador, J. O.: Moreira, A.; Malava, E. Goiaba: Do plantio ao consumidor: Tecnologia de produção, pós-colheita e comercialização. Porto Alegre: Cinco Continentes, 2001. 124p.

Medeiros, J. F. de. Qualidade da água de irrigação utilizada nas propriedades assistidas pelo "GAT" nos Estados do RN, PB, CE e avaliação da salinidade dos solos. Campina Grande: UFPB, 1992. 173p. Dissertação Mestrado

Novais, R. F.; Neves, J. C. L.; Barros, N. F. Ensaio em ambiente controlado. In: Oliveira, A. J. (ed.). Métodos de pesquisa em fertilidade do solo. Brasília: EMBRAPA, 1991. p.189-253.

Oliveira, W. J.; Souza, E. R. de; Santos, H. R. B.; Silva, E. F. de F.; Duarte, H. H. F.; Melo, D. V. M. de. Fluorescência da clorofila como indicador de estresse salino em feijão caupi. Revista Brasileira de Agricultura Irrigada, v.12, p.2592-2603, 2018. https://doi.org/10.7127/rbai.v12n300700

Richards, L. A. Diagnosis and improvement of saline and alkali soils. Washington: U. S. Department of Agriculture, 1954. 160p. USDA, Agriculture Handbook, 60

Sá, F. V. S.; Gheyi, H. R.; Lima, G. S. de; Paiva, E. P. de; Moreira, R. C. L.; Silva, L. A. Water salinity, nitrogen and phosphorus on photochemical efficiency and growth of West Indian cherry. Revista Brasileira de Engenharia Agrícola e Ambiental, v.22, p.158-163, 2018. https://doi.org/10.1590/1807-1929/agriambi.v22n3p158-163

Sachdev, S.; Ansari, S. A.; Ansari, M. I.; Fujita, M.; Hasanuzzaman, M. Abiotic stress and reactive oxygen species: generation, signaling, and defense mechanisms. Antioxidants, v.10, p.1-37, 2021. https://doi.org/10.3390/antiox10020277

Scotti-Campos, P.; Pham-Thi, A. T.; Semedo, J. N.; Pais, I. P.; Ramalho, J. C.; Matos, M. do C. Physiological responses and membrane integrity in three Vigna genotypes with contrasting drought tolerance. Emirates Journal of Food and Agriculture, v.25, p.1002-1013, 2013. https://doi.org/10.9755/ejfa.v25i12.16733

Silva, A. A. R. da; Lima, G. S. de; Azevedo, C. A. V. de; Gheyi, H. R.; Souza, A. R. de; Fernandes, P. D. Salicylic acid relieves the effect of salt stress on soursop morphophysiology. Ciência e Agrotecnologia, v.45, p.1-12, 2021a. https://doi.org/10.1590/1413-7054202145007021

Silva, A. A. R. da; Lima, G. S. de; Azevedo, C. A. V. de; Veloso, L. L. S. A.; Gheyi, H. R. Salicylic acid as an attenuator of salt stress in soursop. Revista Caatinga, v.33, p.1092-1101, 2020. https://doi.org/10.1590/1983-21252020v33n424rc

Silva, S. S. da; Lima, G. S. de; Lima, V. L. A. de; Soares, L. A. dos A.; Gheyi, H. R.; Fernandes, P. D. Quantum yield, photosynthetic pigments and biomass of mini watermelon under irrigation strategies and potassium. Revista Caatinga, v.34, p.659-669, 2021b. https://doi.org/10.1590/1983-21252021v34n318rc

Silva, T. I. da; Silva, J. de S.; Dias, M. G.; Martins, J. V. da S.; Ribeiro, W. S.; Dias, T. J. Salicylic acid attenuates the harmful effects of salt stress on basil. Revista Brasileira de Engenharia Agrícola e Ambiental, v.26, p.399-406, 2022. https://doi.org/10.1590/1807-1929/agriambi.v26n6p399-406

Souana, K.; Taïbi, K.; Abderrahim, L. A.; Amirat, M.; Achir, M.; Boussaid, M.; Mulet, J. M. Salt-tolerance in *Vicia faba* L. is mitigated by the capacity of salicylic acid to improve photosynthesis and antioxidant response. Scientia Horticulturae, v.273, p.1-7, 2020. https://doi.org/10.1016/j.scienta.2020.109641

Weatherley, P. E. Studies in the water relations of the cotton plant. I - The field measurementes of water deficits in leaves. New Phytologist, v.49, p.81-87, 1950.