Ciência Rural, Santa Maria, v.53:7, e20210447, 2023 http://doi.org/10.1590/0103-8478cr20210447

ISSNe 1678-4596
CROP PRODUCTION

Exogenous application of salicylic acid on the mitigation of salt stress in *Capsicum annuum* L.

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ABSTRACT: Salinity has limited conventional vegetable cultivation, especially in semi-arid regions. In this regard, the use of elicitors that act to induce tolerance to salt stress, such as salicylic acid, has emerged as a promising alternative. This study evaluated the effects of foliar spraying with salicylic acid on the mitigation of salt stress on the morphophysiology and production of bell pepper cv. All Big. The study was conducted in a greenhouse in Campina Grande - PB, Brazil, adopting a completely randomized design, in a 4 x 4 factorial arrangement, corresponding to four levels of electrical conductivity of irrigation water (0.8; 1.6; 2.4; and 3.2 dS m^{-1}) and four concentrations of salicylic acid (0; 1.2; 2.4 and 3.6 mM), with three replicates. The foliar application of salicylic acid at a concentration of 1.6 mM attenuated the effects of salt stress in gas exchange, growth, mean fruit weight, and total production per plant, and decreased the percentage of intercellular electrolyte leakage of sweet pepper cv. All Big plants, at 80 days after sowing.
Key words: bell pepper, saline waters, salt tolerance, elicitor.

Aplicação exógena de ácido salicílico na mitigação do estresse salino em *Capsicum annuum* L.

RESUMO: A salinidade tem afetado negativamente o cultivo de hortaliças, sobretudo em regiões semiáridas. Neste sentido, o uso de elicitores que atuem na indução de tolerância ao estresse salino, como ácido salicílico, tem se destacado como uma alternativa promissora. Objetivou-se com esse estudo, avaliar os efeitos da pulverização foliar do ácido salicílico na mitigação do estresse salino na morfofisiologia e produção do pimentão cv. All Big. O estudo foi conduzido em casa de vegetação em Campina Grande – PB, utilizando-se o delineamento inteiramente casualizado, em arranjo fatorial 4 x 4, sendo quatro níveis de condutividade elétrica da água de irrigação (0,8; 1,6; 2,4; e 3,2 dS m^{-1}) e quatro concentrações de ácido salicílico (0; 1,2; 2,4 e 3,6 mM), com três repetições. A aplicação foliar de ácido salicílico na concentração de 1,6 mM atenuou os efeitos do estresse salino nas trocas gasosas, no crescimento, no peso médio de fruto e na produção total por planta, e diminuiu a porcentagem de extravasamento de eletrólitos intercelulares das plantas de pimentão cv. All Big aos 80 dias após o semeio.
Palavras-chave: pimentão, águas salinas, tolerância a salinidade, elicitor.

INTRODUCTION

Bell pepper (*Capsicum annuum* L.) is one of the most consumed vegetables in Brazil, and its production has been increasing in recent years, due to its better adaptation to protected environments compared to other crops, making the country one of the largest producers in the world, standing out as a vegetable of great economic importance in the national market (SANTOS et al., 2020; SILVA et al., 2020a). According to the Anuário Brasileiro de Horti & Fruti, the estimated production of peppers in Brazil in 2019 was 350 thousand tons with a harvested area of 13 thousand hectares.

The Northeast region has favorable climatic conditions for bell pepper production. However, in the semi-arid area, the quality of water for irrigation is a limiting factor because the poor spatial and temporal distribution of rainfall, combined with high temperatures, has contributed to a gradual reduction in water availability, in terms of both quality and quantity, so high salt contents are commonly reported in surface water and groundwater (SILVA et al., 2014; SÁ et al., 2018).

The high concentration of salts in irrigation water can result in osmotic and ionic imbalance, due to the excess of Na^+ and Cl^- ions, affecting the availability and absorption of water and nutrients by plants (YAISH et al., 2016; SAMADDAR et al., 2019). Symptoms observed in plants under salt stress include delay in leaf

Received 06.08.21 Approved 08.03.22 Returned by the author 10.12.22
CR-2021-0447.R1
Editors: Leandro Souza da Silva Maria Jaislanny Medeiros

production, necrosis of shoots and roots, and reduction of leaf area and photosynthetic activity, thus affecting their growth (VELOSO et al., 2021).

Considering the importance of using saline water for the expansion of irrigated agriculture and guarantee of food production, it is necessary to take action on strategies that minimize the deleterious effects of this abiotic stress, enabling safe agricultural production in the semi-arid region and promoting socioeconomic and environmental development (LIMA et al., 2020). In this context, research has been conducted using salicylic acid to mitigate the effects of salt stress on common bean (ARAÚJO et al., 2018), watermelon (NÓBREGA et al., 2020), basil (SILVA et al., 2019), soursop (SILVA et al., 2020b) and West Indian cherry (DANTAS et al., 2021). However, information on its use in bell pepper crop irrigated with saline waters is still scarce in the literature.

Salicylic acid is a substance that can improve the efficiency of plant metabolic processes, which results in adaptation to abiotic stresses (AGOSTINI et al., 2013). The mechanisms by which salicylic acid generates these improvements are related to the protection of cell membranes, increases in carbon metabolism, antioxidant system and, the regulation of stress defense proteins (KANG et al., 2012; SHARMA et al., 2017).

However, the attenuating effects of salicylic acid on plants depend on the concentration used, the form of application, and the plant development stage (NÓBREGA et al., 2020). Because of the above, this study aimed to evaluate the effects of foliar application of salicylic acid on the mitigation of salt stress on the morphophysiology and production of bell pepper cv. All Big in a protected environment.

MATERIALS AND METHODS

The experiment was carried out between May and September 2020 in a protected environment (greenhouse) belonging to the Academic Unit of Agricultural Engineering - UAEA of the Federal University of Campina Grande - UFCG, in Campina Grande, Paraíba, Brazil, at the geographic coordinates 7°15’18” South latitude, 35°52’28” West longitude and an average altitude of 550 m. The data of temperature (maximum and minimum) and average relative air humidity of the experimental site are shown in figure 1.

The treatments consisted of four levels of electrical conductivity of irrigation water - ECw (0.8, 1.6, 2.4, and 3.2 dS m^{-1}) and four concentrations of salicylic acid - SA (0, 1.2, 2.4, and 3.6 mM) applied via foliar spraying, in a 4 x 4 factorial arrangement, distributed in a completely randomized design, with three replicates, totaling 48 experimental units. Salicylic acid (SA) concentrations were based on a study conducted by SILVA et al. (2020b), and water salinity levels were defined based on the study of LIMA et al. (2016), according to the tolerance of the crop to salinity and quality of the water available for irrigation in the Brazilian semi-arid region.

The bell pepper cultivar used in the experiment was All Big, belonging to the Cascadura group. This material has an upright growth habit, small size, firm, and thick pulp, sweet taste, high yield, and a cycle of about 110 days, besides being tolerant to stem rot (*Phytophthora capsici*) and to the tomato mosaic virus (ToMV) (SILVA et al., 2020c).

For the experiment, 8-dm^3 polyethylene pots (Citropote®) were lined at the bottom with a nonwoven geotextile (Bidim OP 30) and filled with a 0.3-kg layer of crushed stone (n° 0), followed by 8 kg of soil classified as *Neossolo Regolítico* (*Entisol* - United States, 2014), collected at a depth of 0-30 cm, from the municipality of Lagoa Seca-PB, whose physical and chemical attributes (Table 1) were determined according to TEIXEIRA et al. (2017).

The irrigation waters with different levels of electrical conductivity were prepared by dissolving the salts NaCl, $CaCl_2.2H_2O$, and $MgCl_2.6H_2O$, in the equivalent proportion of 7:2:1, respectively, in water from the local supply system (ECw = 0.38 dS m^{-1}). This proportion is commonly reported in sources of water used for irrigation in small properties in Northeast Brazil (MEDEIROS et al., 2003). The irrigation waters were prepared considering the relationship between ECw and salt concentration according to RICHARDS (1954), as shown in equation 1:

$$Q = 10 \times ECw \quad (1)$$

Where:

Q = amount of salts to be added ($mmol_c\ L^{-1}$)

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

At 30 days after sowing (DAS), irrigation with saline waters started, at intervals of 3 days, applying the water according to each treatment to maintain soil moisture close to field capacity and to avoid high concentration of salts in the soil solution. The volume applied was determined according to the water requirement of the plants, estimated by the water balance, as shown in equation 2:

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

where:

VI = volume of water to be applied in irrigation (mL);

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

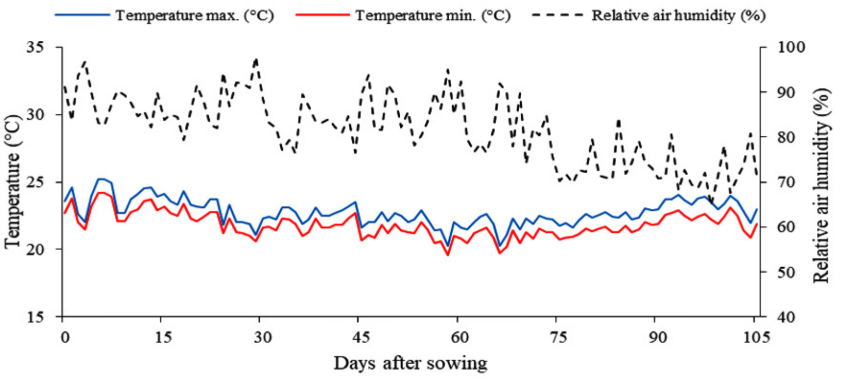

Figure 1 - Temperature (maximum and minimum) and average relative air humidity observed in the internal area of the greenhouse during the experimental period.

Vd = volume drained after the previous irrigation event (mL);
LF = leaching fraction of 0.15, applied every 15 days.

Salicylic acid concentrations were obtained by dissolution in 30% of ethyl alcohol (95.5%), as it is a substance of low solubility in water at room temperature. To reduce the surface tension of the drops on the leaf surface, the adjuvant Wil fix at the concentration of 0.5 mL L^{-1} of the solution was added in the preparation of the solution.

Foliar applications began five days before the application of saline water, i.e., at 25 DAS, by spraying on the abaxial and adaxial sides. Subsequent applications were made at intervals of 15 days, up to 70 DAS, using a backpack sprayer between 17:00 and 17:45 h. The sprayer had a working pressure (maximum) of 88 psi (6 bar) and JD 12P nozzle, and the average volume applied per plant was 250 mL.

Nitrogen, phosphorus, and potassium fertilization were based on the recommendations of NOVAIS et al. (1991), applying 100 mg of N, 300 mg of P_2O_5, and 150 mg of K_2O kg^{-1} of soil. Urea, monoammonium phosphate, and potassium chloride were used as sources of nitrogen, phosphorus, and potassium, respectively.

To meet the requirement for micronutrients, a backpack sprayer was used to spray the leaves, on the adaxial and abaxial sides, every two weeks, a solution at a concentration of 1.0 g L^{-1} of commercial product (Dripsol micro) containing: Mg (1.1%), Zn (4.2%), B (0.85%), Fe (3.4%), Mn (3.2%), Cu (0.5%) and Mo (0.05%).

During the experiment all the cultural practices and phytosanitary treatments recommended for the crop were carried out, monitoring the emergence of pests and diseases and adopting control measures when necessary.

Percentage of intercellular electrolyte leakage and gas exchange: stomatal conductance (*gs*), transpiration (*E*), CO_2 assimilation rate (*A*), and internal CO_2 concentration (*Ci*) were evaluated at 80 DAS. These data were then used to quantify water use efficiency (*WUE*) (*A/E*) and carboxylation efficiency (*CE*) (*A/Ci*). At the same time, the growth was evaluated by monitoring plant height (PH), stem diameter (SD), number of leaves (NL), and leaf area (LA).

At 110 DAS, dry mass of leaf (LDM), stem (SDM), root (RDM), and specific leaf area, leaf succulence, average fruit weight, and total production per plant were determined.

Gas exchange was measured in the third leaf, counted from the apex of the main branch of the plant, using irradiation of 1200 μmol photons m^{-2} s^{-1} and airflow of 200 mL min^{-1} by the portable photosynthesis meter “LCPro+” from ADC BioScientific Ltda.

PH (cm) was measured by taking as reference the distance from the plant collar to the insertion of the apical meristem, SD (mm) was measured two centimeters above the plant collar, and NL was obtained by counting fully expanded leaves with a minimum length of 3 cm.

Leaf area (cm^2) was determined, according to the recommendation of TIVELLI et al.

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

---Chemical characteristics---									
pH (H_2O) (1:2.5)	OM dag kg^{-1}	P (mg kg^{-1})	K^+	Na^+	Ca^{2+}	Mg^{2+}	$Al^{3+} + H^+$	PST (%)	ECse (dS m^{-1})
			---($cmol_c$ kg^{-1})---						
5.90	1.36	6.80	0.22	0.16	2.60	3.66	1.93	1.87	1.0
---Physical characteristics---									
---Size fraction (g kg^{-1})---			Textural class	Water content (kPa)		AW	Total porosity %	BD	PD
Sand	Silt	Clay		33.42*	1519.5** dag kg^{-1}			(kg dm^{-3})	
732.9	142.1	125.0	SL	11.98	4.32	7.66	47.74	1.39	2,66

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+} and H^+ extracted with 0.5 M CaOAc at pH 7.0; ESP - Exchangeable sodium percentage; ECse - Electrical conductivity of saturation extract; SL - Sandy loam; AW - Available water; AD - Apparent density; PD - Particle density; * - Field capacity; ** - Wilting point.

(1997), considering equation 3:

$$LA=0.81\times X \quad (3)$$

where:

LA - leaf area (cm^2); and

X - product of length and width (cm^2).

To determine the percentage of intercellular electrolyte leakage (% IEL), a copper hole puncher was used to obtain five-leaf discs of known area, per experimental unit, which were washed and placed in an Erlenmeyer flask containing 50 mL of distilled water. After being closed with aluminum foil, the Erlenmeyer flasks were kept at a temperature of 25 °C for 90 minutes, and then the initial electrical conductivity of the medium (Xi) was measured using a benchtop conductivity meter (MB11, MS Techonopon®). Subsequently, the Erlenmeyer flasks were subjected to a temperature of 90 °C for 90 minutes in a drying oven (SL100/336, SOLAB®) and, after their contents cooled, the final electrical conductivity (Xf) was measured. The percentage of intercellular electrolyte leakage was estimated expressing electrical conductivity after treatment for 90 min at 90 °C, as the percentage of initial electrical conductivity according to the methodology proposed by SCOTTI-CAMPOS et al. (2013), considering equation 4:

$$\%\,IEL=\left(\frac{Xi}{Xf}\right)\times 100 \quad (4)$$

where:

% IEL - percentage of intercellular electrolyte leakage;

Xi - initial conductivity;

Xf - final conductivity.

Leaf succulence (SUC) was determined according to the methodology proposed by MANTOVANI (1999). Specific leaf area (SLA) was determined following the recommendation of BENINCASA (2003). To obtain the dry mass, the stem of each plant was cut close to the soil and then separated into different parts (stem, leaf, and root). The material was placed in a paper bag, and subsequently dried in a forced-air ventilation oven at 65 °C until reaching constant weight. After that, the material was weighed to obtain the dry mass of leaves, stems, and roots. Ripe fruits were harvested at weekly intervals from 90 DAS, extending up to 110 DAS, when the average fruit weight was evaluated.

The data were subjected to the normality test of the distribution (Shapiro-Wilk test) at a 0.05 probability level. Subsequent analysis of variance was performed at a 0.05 probability level and, in cases of significance, linear and quadratic regression analysis was performed using the statistical program SISVAR (FERREIRA, 2019). The model was chosen based on the significance of the coefficients. In case of the significance of the interaction between factors, TableCurve 3D software was used to create response surfaces.

RESULTS AND DISCUSSION

According to the summary of the analysis of variance (Table 2), the interaction between salinity levels (SL) and salicylic acid concentrations (SA) significantly influenced all variables analyzed, except the internal CO_2 concentration and transpiration. Salinity levels, on the other hand, significantly affected (P≤0.01) *Ci*, *A*, *CE,* and *WUE*. Salicylic acid concentrations caused a significant effect (P≤0.01) on *A* and *WUE*. The transpiration (*E*) was not influenced by the application of treatments.

The increase in the electrical conductivity of irrigation water caused a positive linear effect on *Ci* (Figure 2), with a 10.53% increment per unit increase in ECw. In relative terms, there was an increase of 23.32% (33.8 μmol CO_2 m^{-2} s^{-1}) in the *Ci* between plants irrigated using water with the highest salinity (3.2 dS m^{-1}) and those cultivated with the lowest salinity level (0.8 dS m^{-1}).

The increase in *Ci* indicates that CO_2 was not used in the synthesis of carbohydrates by photosynthesis, suggesting that some non-stomatal factors negatively interfered in this process (LARCHER, 2006) and may be related to the degradation of the photosynthetic apparatus in response to leaf tissue senescence resulting from stress caused by the excess of salts (SILVA et al., 2013).

For the *gs* of bell pepper (Figure 3A), plants irrigated with water of 1.9 dS m^{-1} and subjected to a concentration of 1.6 mM obtained the highest value (0.47 mol H_2O m^{-2} s^{-1}), corresponding to an increase of 17.5% (0.07 mol H_2O m^{-2} s^{-1}) compared to plants of the control treatment (0.8 dS m^{-1}).

Conversely, SA concentrations above 1.6 mM associated with the increase in ECw cause reduction in *gs*, with the lowest value (0.36 mol H_2O m^{-2} s^{-1}) obtained in plants subjected to SA concentration of 3.6 mM and irrigated with water of 3.2 dS m^{-1}. The reduction in stomatal conductance stands out as a mechanism of tolerance to salinity of this species, aiming to reduce water loss, and consequently the absorption of water and salts from the soil solution, without compromising photosynthetic activity (DIAS et al., 2019).

For the CO_2 assimilation rate of bell pepper (Figure 3B), plants irrigated with water of 1.6 dS m^{-1} and subjected to SA concentration of 1.6 mM obtained the highest *A* (42.04 mmol CO_2 m^{-2} s^{-1}), corresponding to an increase of 10.92% (4.14 mmol CO_2 m^{-2} s^{-1}) in comparison to plants grown under the same ECw level and without application of SA (0 mM). However, it can be observed that SA concentrations above 1.7 mM reduce the CO_2 assimilation rate, regardless of salinity level, and the lowest *A* (26.7 mmol CO_2 m^{-2} s^{-1}) was observed in plants subjected to SA concentration of 3.6 mM and irrigated with water of 3.2 dS m^{-1}.

For *CE* (Figure 3C), it was verified that SA concentrations above 0.8 mM associated with irrigation water salinity from 0.8 dS m^{-1} negatively affected the *CE* of bell pepper plants, with the lowest *CE* value [0.197 (μmol m^{-2} s^{-1}) (μmol mol^{-1})$^{-1}$] obtained in plants of the control treatment irrigated with water of 3.2 dS m^{-1}.

The reductions observed in *A* and *CE* coincided with the increase in the internal CO_2 concentration of plants grown under high salinity (Figure 2A; Figure 3B and 3C). According to SOUSA et al. (2016), reduction in carboxylation efficiency is

Table 2 - Summary of the analysis of variance of the internal CO_2 concentration (Ci), stomatal conductance (gs), transpiration (E), CO_2 assimilation rate (A), carboxylation efficiency (CE), and water use efficiency (WUE) of bell pepper plants cv. All Big irrigated with saline waters and subjected to foliar application of salicylic acid, at 80 days after sowing.

Source of variation	DF	--Mean squares--					
		Ci	*gs*	*E*	*A*	*CE*	*WUE*
Salinity levels (SL)	3	2640.55^{**}	0.002^{ns}	0.13^{ns}	270.64^{**}	0.02^{**}	5.49^{**}
Linear regression	1	7616.26^{**}	0.003^{ns}	0.19^{ns}	772.21^{**}	0.05^{**}	16.05^{**}
Quadratic regression	1	96.33^{ns}	0.002^{ns}	0.08^{ns}	26.41^{ns}	0.009^{ns}	0.29^{ns}
Salicylic acid (SA)	3	269.55^{ns}	0.005^{ns}	0.07^{ns}	107.08^{**}	0.002^{ns}	3.97^{**}
Linear regression	1	112.06^{ns}	0.0013^{ns}	0.12^{ns}	11.82^{ns}	0.001^{ns}	0.81^{ns}
Quadratic regression	1	341.33^{ns}	0.0011^{ns}	0.07^{ns}	206.51^{**}	0.005^{ns}	7.59^{**}
Interaction (SL × SA)	9	484.08^{ns}	0.007^{*}	0.36^{ns}	32.30^{*}	0.01^{**}	2.24^{**}
Residue	30	163.59	0.093	0.22	11.88	0.002	0.59
CV (%)		7.90	12.41	7.91	9.69	17.26	12.90

^{ns}, *, ** not significant, significant at P ≤ 0.05 and P ≤ 0.01, respectively. CV: Coefficient of variation.

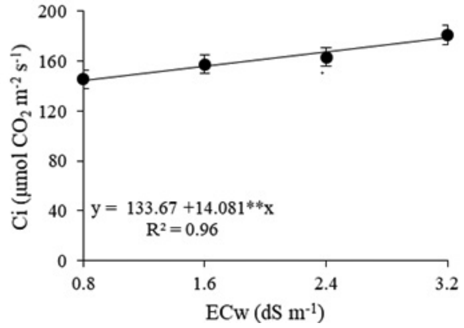

Figure 2 - Internal CO_2 concentration - *Ci* of bell pepper plants cv. All Big as a function of the electrical conductivity of the water - ECw, at 80 days after sowing.

**Significant at P ≤ 0.01. Vertical bars represent the standard error of mean (n=3).

related to metabolic restrictions in the Calvin cycle, where the carbon received was not being fixed in the carboxylation phase in mesophyll cells.

As observed for *gs* and *A* (Figure 3A and 3B), the *WUE* (Figure 3D) was also influenced by the interaction (SL × SA) and its highest value [6.85 (μmol m^{-2} s^{-1}) (μmol mol^{-1})$^{-1}$] was obtained in bell pepper plants subjected to SA concentration of 1.6 mM and irrigated with water of 1.6 dS m^{-1}, which corresponds to an increase of 9.95% [0.62 (μmol m^{-2} s^{-1}) (μmol mol^{-1})$^{-1}$] in comparison to plants of the control treatment (0 mM) irrigated with water of 1.6 dS m^{-1}.

It is also observed that SA concentrations above 1.6 mM combined with the increase in ECw from 1.7 dS m^{-1} intensify the deleterious effects on *WUE*, which reached its lowest value [4.48 (μmol m^{-2} s^{-1}) (μmol mol^{-1})$^{-1}$] in plants sprayed with a concentration of 3.6 mM and irrigated with water of 3.2 dS m^{-1}.

Corroborating the present study, SILVA et al. (2020b), evaluating the effect of foliar application of SA in soursop plants under saline stress (0.8 to 4.0 dS m^{-1}), reported that the application of SA at a concentration of 1.4 mM promoted an increase in *gs*, *A,* and *WUE*, such a response has been attributed to the ability of salicylic acid to induce changes in carbohydrate metabolism in plants and, thus, soluble sugars, especially non-reducing ones, accumulate to work as osmotic regulators. VELOSO et al. (2021) also observed a beneficial effect of foliar application of SA (1.7 mM) on gas exchange in sweet pepper plants irrigated with saline water (0.8 to 3.2 dS m^{-1}). The beneficial effect of salicylic acid concentrations between 1.6 and 1.7 mM observed on *gs, A* and *WUE* may be related to the capacity of salicylic acid to improve enzymatic and photosynthetic activities, also maintaining the balance between the production and elimination of reactive oxygen species - ROS (BATISTA et al., 2019).

The summary of the analysis of variance (Table 3) revealed that the SL × SA interaction significantly influenced the stem diameter, leaf area, and dry mass of the leaf, stem, and root. Salinity levels, on the other hand, significantly affected (P≤0.01) all variables. Salicylic acid concentrations caused a significant effect on plant height, leaf área, number of leaves and dry mass of the leaf, stem, and root.

The increase in the electrical conductivity of irrigation water reduced the PH of bell pepper (Figure 4A) by a 4.81% per unit increase in ECw. When comparing in relative terms the PH of plants irrigated with water of highest salinity (3.2 dS m^{-1}) with the PH of those cultivated with the lowest salinity level (0.8 dS m^{-1}), there was a reduction of 12.0% (5.88 cm).

A decrease in the height of bell pepper plants was also observed by LIMA et al. (2016), evaluating growth and production of All Big bell pepper, as a function of irrigation with saline water. These authors found PH reduction of 18.01%, at 90 days after transplanting, in plants irrigated with water of the highest salinity (3.0 dS m^{-1}), compared to those that received 0.6 dS m^{-1}.

According to SOUSA et al. (2016), excess salts in water and/or soil cause deleterious effects

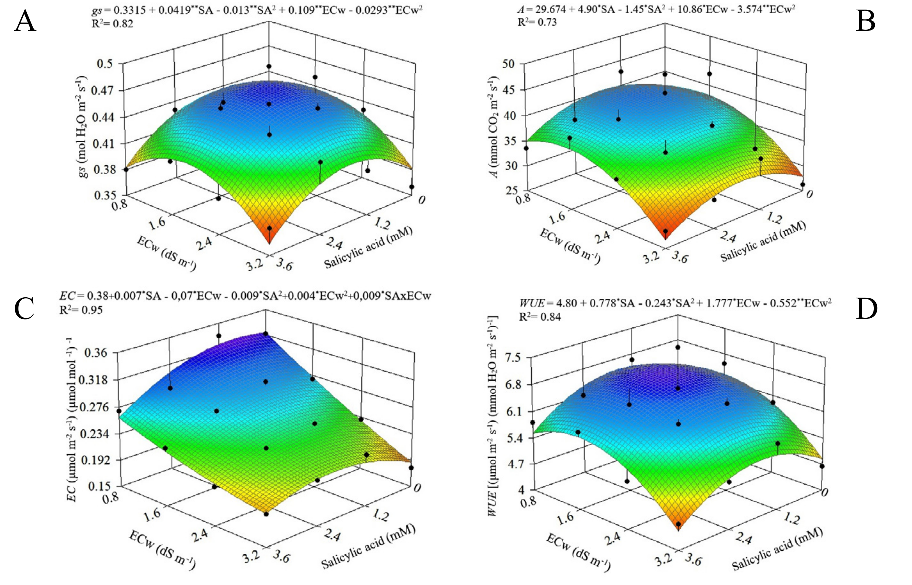

Figure 3 - Response surface for stomatal conductance - *gs* (A), CO_2 assimilation rate - *A* (B), carboxylation efficiency - *CE* (C), and water use efficiency - *WUE* (D) of bell pepper plants as a function of the interaction between the electrical conductivity of the water - ECw and salicylic acid concentrations, at 80 days after sowing.

* and **Significant at $P \leq 0.05$ and $P \leq 0.01$, respectively.

on plants due to the reduction in water potential and low osmotic adjustment capacity of the crop, thus affecting its growth.

Foliar application of SA up to the concentration of 1.65 mM promoted an increase in PH (Figure 4B). It is possible to observe that plants subjected to 1.65 mM of SA stood out with the highest PH value (50.88 cm). Comparing the PH of plants subjected to an estimated concentration of SA of 1.65 mM with the PH of those subjected to 0 mM of SA, there was an increase of 8.24% (4.19 cm). This response may be related to the fact that salicylic acid under adequate conditions acts in the regularization of physiological and biochemical processes, reducing oxidative damage in plants, which leads to greater growth (FARHADI & GHASSEMI-GOLEZANI, 2020).

For SD (Figure 4C), bell pepper plants that were not subjected to the salicylic acid application had a reduction in SD when irrigated with ECw water greater than 1.6 dS m^{-1}, with the lowest SD value (7.73 mm) observed with water of 3.2 dS m^{-1}.

However, the application of salicylic acid up to a concentration of 1.6 mM promoted an increase in stem diameter even when plants were exposed to salinity, and the highest SD (8.91 mm) was obtained in plants subjected to SA concentration of 1.6 mM and irrigated with water of 1.6 dS m^{-1}, corresponding to an increase of 6.45% (0.54 mm) compared to plants grown under ECw of 1.6 dS m^{-1} without application of SA.

As observed for SD (Figure 4C), the SA concentration of 1.6 mM favored the higher growth in LA of bell pepper plants (Figure 4D). It was verified that plants irrigated with water of 1.7 dS m^{-1} and subjected to SA concentration of 1.6 mM obtained the highest value of LA (2184.3 cm^2), corresponding to an increase of 21.06% (380.04 cm^2) in comparison to those of the control treatment (0 mM) irrigated with water of 0.8 dS m^{-1}.

The NL of bell pepper plants was negatively affected by the increase in the electrical conductivity of irrigation water (Figure 4E), with a

Table 3 - Summary of the analysis of variance for plant height (PH), stem diameter (SD), leaf area (LA), number of leaves (NL), dry mass of leaf (LDM), stem (SDM), and root (RDM) of bell pepper plants cv. All Big irrigated with saline water and subjected to foliar application of salicylic acid, at 80 days after sowing.

Source of variation	DF	---	------	------	Mean squares	------	---	
		PH	SD	LA	NL	LDM	SDM	RDM
Salinity levels (SL)	3	80.55**	1.28**	516337.1**	412.14**	26.97**	18.72**	7.91**
Linear regression	1	231.08**	0.03ns	792858.4*	1188.01**	75.25**	50.79**	5.30ns
Quadratic regression	1	4.38ns	2.18*	701340.9**	48.01ns	2.14ns	0.43ns	10.01**
Salicylic acid (SA)	3	35.13*	0.24ns	1059816**	325.80**	13.47**	5.15**	2.32*
Linear regression	1	9.01ns	0.12ns	202695.8*	201.66ns	2.73ns	4.12ns	0.61ns
Quadratic regression	1	76.25**	0.38ns	1930325.1**	705.33**	23.53**	11.22**	3.91*
Interaction (SL × SA)	9	2.06ns	0.69*	134214**	59.49ns	2.01*	3.87**	1.47*
Residue	30	9.71	0.31	24338	44.87	0.93	0.63	0.69
CV (%)		6.77	6.57	8.94	13.61	7.71	9.48	14.44

ns, *, ** not significant, significant at $P \leq 0.05$ and $P \leq 0.01$, respectively. CV: Coefficient of variation.

reduction of 10.04% per unit increase in ECw. When comparing the NL of plants irrigated with water of the highest salinity (3.2 dS m^{-1}) with that of plants cultivated with the lowest salinity level (0.8 dS m^{-1}), there was a reduction of 26.21%.

A study conducted by SÁ et al. (2019), evaluating the initial development and tolerance of pepper species to salt stress (0.6 to 3.0 dS m^{-1}), also found a reduction in NL as a function of the increase in salinity. High concentrations of salts in the soil negatively affect the physiological aspects of the plant, causing ionic, osmotic, hormonal, and nutritional changes, consequently affecting the growth and development of crops (DIAS et al., 2019).

Salicylic acid concentrations significantly influenced the number of leaves of bell pepper plants and, according to the regression equation (Figure 4F), plants subjected to a concentration of 1.6 mM stood out with the highest value of NL (53.8 leaves). When comparing the NL of plants subjected to the salicylic acid concentration of 1.6 mM with that of plants subjected to SA concentration of 0 mM, there was an increase of 9.8% (4.8 leaves).

Spraying with salicylic acid up to a concentration of 1.6 mM promoted increments in LDM, SDM, and RDM, regardless of the electrical conductivity of irrigation water and, according to the regression equations (Figure 5A, 5B, and 5C), plants subjected to a concentration of 1.6 mM and irrigated with water of 1.5 dS m^{-1} obtained the highest values of LDM (14.37 g per plant), SDM (9.86 g per plant), and RDM (6.6 g per plant). When comparing in relative terms the LDM, SDM, and RDM of plants irrigated with water of 1.5 dS m^{-1} and subjected to SA concentration of 1.6 mM with that of plants cultivated with the same salinity level and without application of SA (0 mM), there were increments of 10.1% (1.32 g per plant), 10.9% (0.97 g per plant), and 9.1% (0.55 g per plant), respectively.

The increase in biomass accumulation observed in plants subjected to SA concentration of 1.6 mM, even when exposed to the salinity of irrigation water, is probably because salicylic acid increases the production of proteins, lignin, and carbohydrates, producing photoassimilates, thus promoting greater tolerance to stress (SHAO et al., 2018). In addition, the increase in biomass may be related to the increase in the number of leaves and leaf area (Figure 4D and 4F) observed in the present study.

It is possible to observe in this study that the application of salicylic acid at concentrations between 1.6 and 1.7 mM mitigated the deleterious effects of salinity on *gs*, *A*, and *WUE* (Figure 3A, 3B and 3C), and this response led to increased growth in SD, LA, and NL (Figure 4C, 4D, and 4F) and, consequently, to higher accumulation in LDM, SDM, and RDM (Figure 5A, 5B and 5C).

There was a significant interaction between salinity levels and salicylic acid concentrations for the percentage of intercellular electrolyte leakage, average fruit weight, and total production per plant (Table 4). The salinity levels of irrigation water negatively and significantly ($P \leq 0.01$) affected all variables analyzed. Salicylic acid concentrations

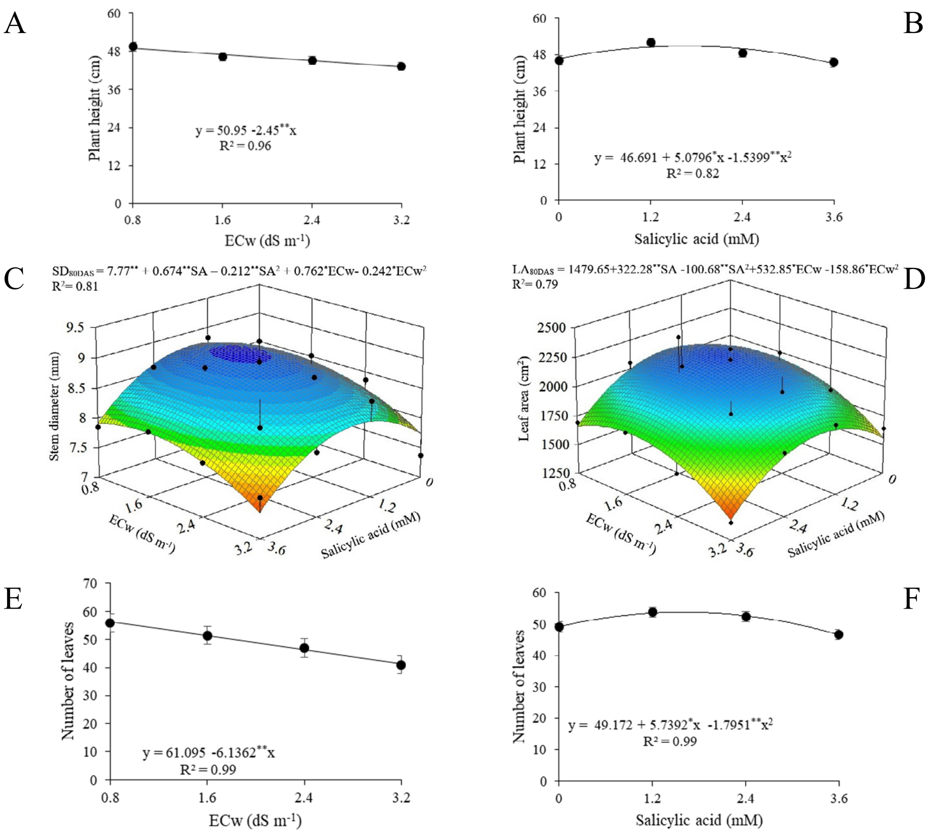

Figure 4 - Plant height (A) and the number of leaves (E) as a function of the electrical conductivity of the water - ECw and salicylic acid concentrations – SA (B and F); and response surface for stem diameter (C) and leaf area (D) of bell pepper plants as a function of the interaction between ECw and SA concentrations, at 80 days after sowing.

* and **Significant at $P \leq 0.05$ and $P \leq 0.01$, respectively. Vertical bars represent the standard error of mean (n=3).

significantly influenced the specific leaf area, average fruit weight, and total production per plant.

The increase in the electrical conductivity of irrigation water reduced the SLA of bell pepper (Figure 6A) by 8.24% per unit increase in ECw. When comparing the SLA of plants irrigated with water of the highest salinity (3.2 dS m^{-1}) with the value of plants cultivated with ECw of 0.8 dS m^{-1}, there was a reduction of 21.1% (33 cm^2 g^{-1}). Under conditions of high water salinity, the reduction in plant growth occurs as previously explained, due to the decrease in the osmotic potential of the soil leading to a reduction in the availability of water to plants, affecting cell division and turgor, inhibiting the expansion of the cell wall (ACOSTA-MOTOS et al., 2017).

The SLA of bell pepper was also significantly influenced by salicylic acid concentrations (Figure 6B). It was observed that the plants subjected to a concentration of 1.6 mM obtained a higher SLA value (149.3 cm^2 g^{-1}). When comparing with that of plants subjected to SA concentration of 3.6 mM, there was an increase of 12.9% (17.1 cm^2 g^{-1}).

For the SUC of bell pepper, the regression equation (Figure 6C) pointed to an increase of 16.9% per unit increment in ECw. When comparing the SUC of plants irrigated with water of the highest

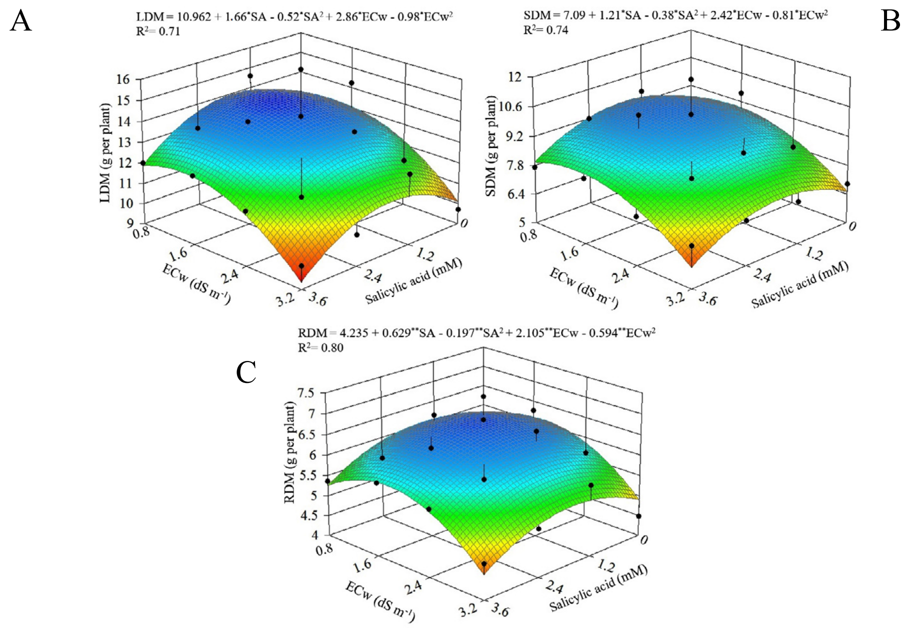

Figure 5 - Response surface for the dry mass of leaf - LDM (A), stem - SDM (B), and root - RDM (C) of bell pepper plants as a function of the interaction between the electrical conductivity of the water - ECw and salicylic acid concentrations, at 105 days after sowing.

* and **Significant at $P \leq 0.05$ and $P \leq 0.01$, respectively.

salinity (3.2 dS m^{-1}) with that of plants cultivated with the lowest salinity level (0.8 dS m^{-1}), there was an increase of 35.6% (0.012 g H_2O cm^2).

Similar results were obtained by LIMA et al. (2019) in a study conducted with colored fiber cotton cv. BRS Rubi under salt stress (ECw between 5.1 and 9.1 dS m^{-1}), where a 10% increase in SUC was observed per unit increment in ECw, and the authors attributed this increase in SUC to a possible osmotic adjustment of the plants, thus allowing the regulation of salt concentration in leaf tissues.

Considering the effect of the interaction between irrigation water salinity and salicylic acid concentrations on % IEL (Figure 6D), it was verified that the SA concentration of 1.6 mM was able to reduce the % IEL in bell pepper plants, with the lowest % IEL value (23.21%) obtained in plants irrigated with water of 1.6 dS m^{-1} and subjected to SA concentration of 1.6 mM.

Conversely, plants in the control treatment (0 mM) when subjected to irrigation water salinity of 1.6 dS m^{-1} showed a % IEL of 27.3%, i.e., an increase of 4.1% in comparison to plants subjected to the concentration of 1.6 mM. The positive effect of SA in reducing the % IEL in pepper plants can be attributed to the improvement in nutrient absorption, membrane protection, and increased photosynthetic activity (SILVA et al., 2021). In addition, SA can interact with ROS signaling pathways and reduce oxidative stress (BATISTA et al., 2019).

The interaction between the electrical conductivity of irrigation water and salicylic acid concentrations significantly influenced the AFW of bell pepper (Figure 7A). It can be noted that plants irrigated with water of 1.8 dS m^{-1} and subjected to SA concentration of 1.6 mM obtained the highest AFW (38.75 g per fruit), which corresponded to an increase of 20.8% (6.7 g per fruit) in comparison to plants of the control treatment.

There was a decrease in AFW under SA concentrations above 1.6 mM, regardless of the electrical conductivity of the irrigation water used; the lowest AFW (26.9 g per fruit) was obtained in plants irrigated with water of 3.2 dS m^{-1} and subjected to SA concentration of 3.6 mM, which

Table 4 - Summary of the analysis of variance for the specific leaf area (SLA), leaf succulence (SUC), percentage of intercellular electrolyte leakage (% IEL), average fruit weight (AFW), and total production per plant (TPP) of bell pepper irrigated with saline waters and subjected to foliar application of salicylic acid.

Source of variation	DF	---Mean squares--				
		SLA	SUC	% IEL	AFW	TPP
Salinity levels (SL)	3	2420.36**	0.0011**	994.24**	67.41**	21870.3**
Linear regression	1	5741.22**	$6.7x10^{-4}$**	2754.99**	180.18**	64402.2**
Quadratic regression	1	373.12ns	$1.7x10^{-5}$ns	0.15ns	22.07ns	75.0ns
Salicylic acid (SA)	3	2526.26**	$2.3x10^{-4}$ns	95.53ns	66.06**	8773.9**
Linear regression	1	3500.87*	$1.3x10^{-3}$ns	21.80ns	127.22*	2130.5ns
Quadratic regression	1	2913.61**	$1.1x10^{-3}$ns	241.53ns	70.27**	18246.6**
Interaction (SL × SA)	9	382.14ns	$5.7x10^{-5}$ns	168.81**	108.42**	4433.2**
Residue	30	198.61	$7.8x10^{-5}$ns	36.49	13.52	729.9
CV (%)		10.11	22.11	19.20	10.67	11.36

ns, *, ** not significant, significant at P≤0.05 and P≤0.01, respectively. CV: Coefficient of variation.

corresponded to a reduction of 30.6% (11.9 g per fruit) when compared to plants with highest AFW.

Concerning the TPP of bell pepper (Figure 7B), plants subjected to SA concentration of 1.8 mM and irrigated with water of 1.6 dS m^{-1} reached the highest TPP (280.7 g per plant), corresponding to an increase of 23.1% (52.7 g per plant) in comparison to the control plants. Conversely, the lowest TPP (169 g

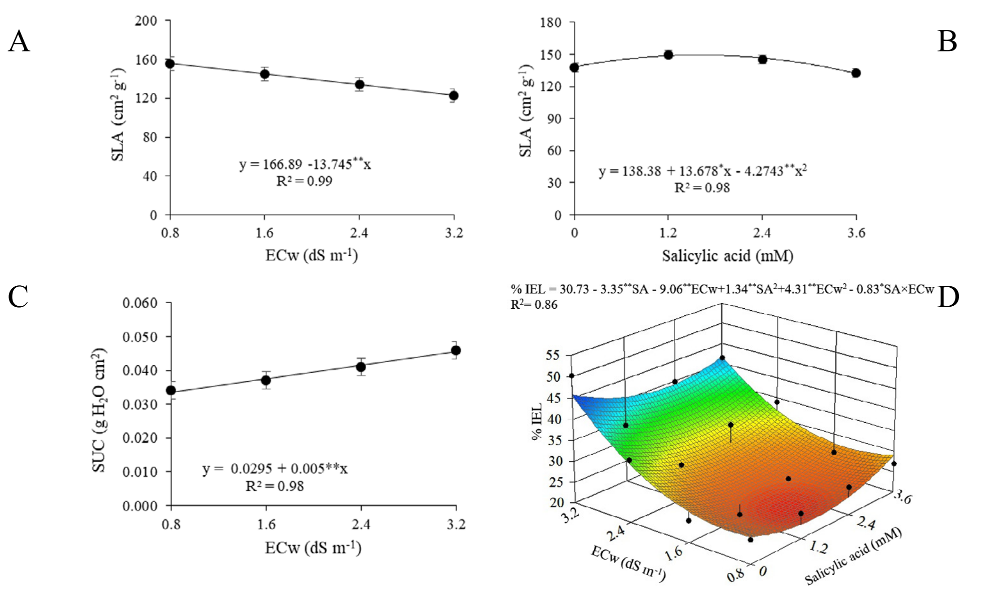

Figure 6 - Specific leaf area - SLA (A) and leaf succulence - SUC (C) as a function of the electrical conductivity of the water - ECw and SLA as a function of salicylic acid - SA concentrations (B); and response surface for the percentage of intercellular electrolyte leakage - % IEL (D) as a function of the interaction between ECw and SA concentrations, at 80 days after sowing.

* and **Significant at P ≤ 0.05 and P ≤ 0.01, respectively. Vertical bars represent the standard error of mean (n=3).

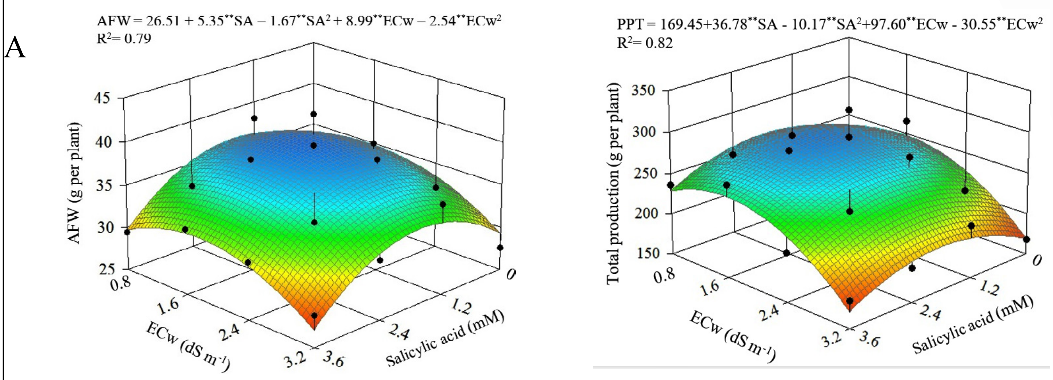

Figure 7 - Response surface for average fruit weight - AFW (A) and total production per plant - TPP (B) as a function of the interaction between the electrical conductivity of the water - ECw and salicylic acid concentrations.

**Significant at $P \leq 0.01$.

per plant) was obtained in plants irrigated with water of 3.2 dS m^{-1} and without the application of SA (0 mM).

The increase in AFW and TPP observed in plants subjected to SA concentration of 1.6 mM may be related to the fact that in this treatment there was a higher CO_2 assimilation rate (Figure 3B); in addition, there was an increase in the carboxylation efficiency and water use efficiency (Figure 3C and 3D) of bell pepper plants.

ELWAN & EL-HAMAHMY (2009), evaluating the effect of foliar application of salicylic acid on the yield and quality of pepper fruits, also reported increases in average fruit weight and total production, due to the foliar spraying with salicylic acid. Under conditions of salt stress, the foliar application of salicylic acid can play a considerable role in plant defense mechanisms and impacts on the physiological and molecular processes of plant cells, which lead to improved plant growth and development through the reduction in the harmful consequences of salinity (ELHINDI et al., 2017).

In general, it was observed that the foliar application of salicylic acid at concentrations between 1.6 and 1.8 mM induced acclimatization of bell pepper plants cv. All Big to salt stress. However, with applications of salicylic acid at higher concentrations, a negative effect was observed on the variables analyzed. According to POÓR et al. (2019), the beneficial effect of SA application depends on several factors, including concentration, plant species, plant development stage, and mode of application.

CONCLUSION

Foliar application of salicylic acid at a concentration of 1.6 mM attenuates salt stress in gas exchange, growth, average fruit weight, and total production per plant, and reduces the percentage of intercellular electrolyte leakage of sweet pepper plants cv. All Big at 80 days after sowing.

ACKNOWLEDGEMENTS

The authors thank the Graduate Program in Agricultural Engineering at the Universidade Federal de Campina Grande, the Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq) and the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES) for their support in carrying out this research.

DECLARATION OF CONFLICT OF INTEREST

The authors declare no conflict of interest.

AUTHORS' CONTRIBUTIONS

All authors conceived and designed experiment. All authors participated in preparation of the draft of the manuscript. All authors critically revised the manuscript and approved the final version.

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