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Growth and quality of passion fruit seedlings under salt stress and foliar application of H_2O_2

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Abstract

Passion fruit is a species with high production potential under the climatic conditions of the Brazilian Northeast region. However, water sources in this region show high salt concentrations that can compromise crop development. In this scenario, the application of small hydrogen peroxide concentrations reduces the deleterious effects caused by salt stress on the production yield of agricultural crops. From this perspective, this study aimed to evaluate the growth and quality of yellow passion fruit seedlings irrigated with saline water and foliar application of hydrogen peroxide. The experiment was conducted in a plant nursery at the Federal University of Campina Grande, PB. The experimental design was in randomized blocks in a 5 x 2 factorial. Treatments consisted of five levels of electrical conductivity of the irrigation water - ECw (0.6, 1.2, 1.8, 2.4, and 3.0 dS m^{-1}) and two concentrations of hydrogen peroxide - H_2O_2 (0 and 20 μM), with four replications and two plants per plot, totaling 80 plants. The exogenous application of 20 μM hydrogen peroxide favored plant growth in stem diameter, plant height, number of leaves, and the Dickson Quality Index of the passion fruit cv. BRS Rubi do Cerrado. The application of 20 μM hydrogen peroxide induced the acclimatization of the passion fruit cv. BRS Rubi do Cerrado irrigated with saline water.

Keywords: salinity, oxidative stress, *Passiflora edulis* Sims

Introduction

Passion fruit (*Passiflora edulis* Sims) is a fruit species from tropical America (Silva et al., 2015), and Brazil is its largest producer and consumer worldwide, producing 602 thousand tons with a mean yield of 14,103 kg ha^{-1}. The Northeast is the leading passion fruit producing region in Brazil, concentrating 62.32% of national production (IBGE, 2019).

Although the Northeast stands out in the national production, semi-arid areas are naturally limited with regard to water availability (Ferreira et al., 2018). From this perspective, the water used for irrigation in this region has high salt contents, limiting its use for agricultural production (Santos et al., 2016).

Plant growth reduction under salt stress conditions occurs due to the osmotic and toxic effects of salts, promoting changes in ionic homeostasis and affecting water uptake by plants (Carneiro et al., 2017; Bezerra et al., 2018). In addition, salinity can restrict gas exchange with the environment, usually improving the water-use efficiency of the plant and, therefore, reducing the amount of carbon dioxide that can be fixed by the plant and used for growth (Lima et al., 2020).

Plants can detoxify the excessive oxidation caused by salt stress conditions due to enzymatic and non-enzymatic mechanisms that protect their cells against oxidative damage, such as the function of superoxide dismutase, which dismutes the O^{-2} to hydrogen peroxide (H_2O_2) and, subsequently, H_2O_2 can be detoxified in the form of water due to the action of ascorbate peroxidase, catalase, glutathione peroxidase, guaiacol and thioredoxin peroxidase, which can act directly in the signaling processes of the plant and define the redox status of transcription factors related to the DNA promoter region. In turn, peroxiredoxins are responsible for detecting the stress and the redox status of the cell,

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acting in the plant signaling of abiotic stress (Alves et al., 2018; Shekari et al., 2017; Liebthal et al., 2018; Laxa et al., 2019).

Passion fruit cultivation in Northeastern Brazil can be favored by strategies that can mitigate the effects of salt stress (Silva et al., 2019a). Among these alternatives, the exogenous application of H_2O_2 is a promising method to mitigate the effects caused by salt stress on agricultural crops (Ashfaque et al., 2014).

At low concentrations, H_2O_2 promotes a moderate stress condition that results in the accumulation of latent signs in different parts of the plant. Thus, when the crop is subjected to more severe stress conditions, the stored or "memorized" signs lead to molecular adjustments, resulting in acclimatization mechanisms (Savvides et al., 2016).

Therefore, this study aimed to evaluate the growth and quality of passion fruit seedlings irrigated with saline water and hydrogen peroxide application.

Material and Methods

The experiment was conducted under plant nursery conditions from September to November 2019 using 3-dm^3 plastic bags in a structure belonging to the Agricultural Engineering Academic Unit (UAEA) of the Federal University of Campina Grande - UFCG, located in Campina Grande, PB (7°15'18'' S, 35°52'28'' W), at a mean elevation of 550 m above sea level.

The treatments resulted from the combination of five levels of electrical conductivity of the irrigation water - ECw (0.6, 1.2, 1.8, 2.4, and 3.0 dS m^{-1}) and two concentrations of hydrogen peroxide – H_2O_2 (0 and 20 μM) in a 5 x 2 factorial, distributed in a randomized block design with four replications and two plants per plot, totaling 80 plants.

Seeds of the passion fruit cultivar BRS Rubi do Cerrado were obtained from the orchard of the Federal Institute of Paraíba (IFPB), located in Sousa – PB. The plants were manually selected and standardized based on plant vigor and health. The cultivar BRS Rubi do Cerrado (BRS SC1) has large, yellow fruits with an oblong shape, weighing from 150 to 350 g and with a pulp yield of around 38% (EMBRAPA, 2012).

The levels of electrical conductivity of the irrigation water were prepared by dissolving the NaCl, $CaCl_2.2H_2O$, and $MgCl_2.6H_2O$ salts in the ratio of 7:2:1 in local supply water (ECw = 0.38 dS m^{-1}). This proportion is commonly found in the water sources used for irrigation in small northeastern properties (Medeiros, 1992) and was obtained based on the relationship between the ECw and the salt concentration ($mmol_c$ L^{-1} = 10 ECw dS m^{-1}) recommended by Richards (1954).

The plastic bags were filled with 3.0 kg of a substrate composed of soil (84%) + sand (15%) + earthworm humus (1%). The soil used in the experiment was a Regolithic Neosol with a sandy-loam texture collected from the 0-20 cm layer in the rural area of Alagoa Nova, PB. The soil was previously ground and sieved, and its physical and chemical characteristics (Table 1) were determined according to the methodology proposed by Teixeira et al. (2017).

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

Chemical characteristics									
pH (H_2O) (1:2. 5)	OM dag kg^{-1}	P(mg kg^{-1})	K^+	Na^+	Ca^{2+}	Mg^{2+}	$Al^{3+} + H^+$	ESP (%)	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-water characteristics									
Granulometric fraction (dag kg^{-1})			Textural class	Moisture (kPa)		AW	Total porosity %	AD	PD
Areia	Silte	Argila		33.42	1,519.5 dag kg^{-1}			(kg dm^{-3})	
73.29	14.21	12.50	SL	11.98	4.32	7.66	47.74	1.39	2.66

OM – Organic matter: Walkley-Black digestion; Ca^{2+} and Mg^{2+} extracted with 1 M KCl at pH 7.0; $Na^+$and K^+ extracted with NH_4OAc 1 M pH 7.0; $Al^{3+}$and H^+ were extracted using 0.5 M CaOAc at pH 7.0; SL – Sandy loam; AW – Availabe water; AD – Apparent density; PD – Particle density

Before sowing, the soil moisture content was increased until the maximum water retention capacity using the water from each treatment. Subsequently, sowing was performed by equidistantly placing four seeds at a 3 cm depth. The seedlings were thinned to one plant per bag eight days after emergence by choosing the one with the best vigor.

Irrigation was performed daily by directly applying, to each bag, a sufficient was volume to maintain soil moisture close to field capacity. The water volume applied was determined according to the water requirement of the plants, estimated by measuring the difference between the water volume applied in the previous irrigation and the water volume added with a 0.10 leaching fraction.

Fertilization with nitrogen (N), potassium (K), and phosphorus (P) was applied as topdressing according to the recommendation by Novais et al. (1991) using 0.67 g of urea, 0.71 g of potassium chloride, and 5.0 g of single superphosphate, the equivalent to 100, 150, and 300 mg kg^{-1} of the N, K_2O, and P_2O_5 substrates, respectively. Single superphosphate was applied only once as topdressing, while nitrogen and potassium were split into eight applications via fertigation in 7-day intervals, with the first application performed seven days after sowing (DAS).

The concentration of hydrogen peroxide (H_2O_2) was established according to a study developed by Andrade et al. (2019). The H_2O_2 dilutions were made in distilled water, and the container used to store the solution (H_2O_2 in distilled water) was sealed with dark adhesive tape to avoid hydrogen peroxide degradation by light. The solution was manually sprayed on the leaves at 5:00 p.m. at 15, 30, and 45 DAS using a sprayer. The abaxial and adaxial surfaces of the leaves were sprayed in order to wet the foliage thoroughly.

Plant growth was evaluated 30 and 60 days after sowing based on plant height (PH), stem diameter (SD), number of leaves (NL), relative growth in plant height (RGR_{PH}), and stem diameter (RGR_{SD}) from 30 to 60 days after sowing, in addition to the Dickson Quality Index (DQI) and the phytomass accumulation parameters of dry leaf phytomass (LDP), dry stem phytomass (StDP), and dry root phytomass (RDP).

Plant height (cm) was measured from the base of the plant to the insertion of the apical meristem; stem diameter (mm) was measured at two centimeters from the base of the plant; finally, the number of leaves was obtained by counting the fully expanded leaves with a minimum length of three centimeters.

The relative growth rate in plant height and stem diameter was determined according to the methodology described by Benincasa (2003) using equations 1 and 2:

$$RGR_{PH} = \frac{(\ln PH2 - \ln PH1)}{t_2 - t_1} \quad (1)$$

$$RGR_{SD} = \frac{(\ln SD2 - \ln SD1)}{t_2 - t_1} \quad (2)$$

Where:

RGR_{PH} – Relative growth rate of plant height (cm cm^{-1} day^{-1});

RGR_{SD} – Relative growth rate of stem diameter (mm mm^{-1} day^{-1});

PH1 – Plant height (cm) at time t1; SD1 – Stem diameter (mm) at time 1;

PH2 – Plant height (cm) at time t2; SD2 – Stem diameter (mm) at time 2;

T1 – Time 1 in days;

T2 – Time 2 in days.

Phytomass accumulation was measured 60 days after sowing by cutting the stem of each plant close to the soil, which was then separated into different parts (stem, leaves, and root) and stored in paper bags. Subsequently, the plant parts were oven-dried at 65 °C until constant weight. Finally, the material was weighed to obtain the dry phytomass of leaves, stem, and root.

The data on plant height (PH), stem diameter (SD), shoot dry phytomass (StDP), root dry phytomass (RDP), and total dry phytomass (TDP) were used to calculate the Dickson Quality Index (DQI) according to equation 3:

$$DQI = \frac{TDP}{\left(\frac{PH}{SD}\right) / \left(\frac{StDP}{RDP}\right)} \quad (3)$$

The data obtained were evaluated by analysis of variance at the 0.05 level of probability. In cases of significance, linear and quadratic polynomial regression analyses were used for the salinity levels, while the Tukey test was applied for the H_2O_2 concentrations (0.05 probability). All analyses were performed with the statistical software SISVAR-ESAL, version 5.6 (Ferreira, 2019).

Results and Discussion

Based on the summary of the analysis of variance (Table 2), the interaction between salinity levels and hydrogen peroxide concentrations significantly influenced ($p \leq 0.01$) plant height (PH), stem diameter (SD), and the number of leaves (NL) 60 days after sowing. With regard to the salinity levels, a significant effect ($p \leq 0.01$) was verified for all variables analyzed, except stem diameter at 30 DAS. The hydrogen peroxide concentrations significantly influenced ($p \leq 0.01$) all variables analyzed at 30 and 60 DAS.

The increased electrical conductivity of the irrigation water negatively affected the plant height of the passion fruit cv. BRS Rubi do Cerrado 30 days after sowing and, according to the regression equation (Figure 1A), a maximum value (16.93 cm) was observed when the seedlings were irrigated with the salinity level of 1.50 dS m^{-1}, decreasing from this ECw level down to a minimum value of 14.23 cm in plants irrigated with the ECw level of 3.0 dS m^{-1}. In relative terms, plant height is decreased by 11.02% (1.76 cm) when the plants irrigated with the highest water salinity (3.0 dS m^{-1}) are compared with the lowest salinity level (0.6 dS m^{-1}). Growth reduction stands out among the most noticeable effects of salt stress on agricultural crops (Nan et al., 2018).

Plants are known to modify their morpho physiological characteristics when under salt stress conditions. One of these changes is osmotic adjustment, which occurs due to the increased synthesis of organic solutes by translocating organic and inorganic solutes across plant tissues, reducing the osmotic potential and increasing the cell turgor potential, thus facilitating water and nutrient uptake and the maintenance of cell growth (Lima et al., 2016).

Table 2. Summary of the analysis of variance referring to plant height (PH), stem diameter (SD), and number of leaves (NL) of yellow passion fruit seedlings irrigated with saline water and exogenous application of hydrogen peroxide 30 and 60 days after sowing (DAS).

Source of variation	DF	Mean squares					
		PH		SD		NL	
		30	60	30	60	30	60
		Days after sowing					
Salinity levels (SL)	4	9.09**	12.73**	0.14ns	1.22**	13.71**	16.97**
Linear regression	1	15.48**	26.91**	0.18ns	4.43**	45.00**	56.11**
Quadratic regression	1	20.06**	23.22**	0.37ns	0.41**	1.28**	2.58**
Peróxido de hidrogênio (H_2O_2)	1	4.22**	2.41**	5.35**	1.65**	21.02**	15.62**
Linear regression	1	3.13*	3.25*	0.31*	0.61**	16.90**	27.22 **
Quadratic regression	1	14.20**	10.63**	0.16ns	0.15ns	0.64ns	2.16**
Interaction (SL × H_2O_2)	4	0.84ns	4.83**	0.06ns	0.27**	0.34ns	0.25*
Residual	27	0.55	0.63	0.07	0.06	0.19	0.09
CV		4.65	4.65	8.47	7.15	6.96	4.08

ns - Non-significant; DF – Degrees of freedom; CV – Coefficient of variation; *,** significant at 0.05 and 0.01, respectively.

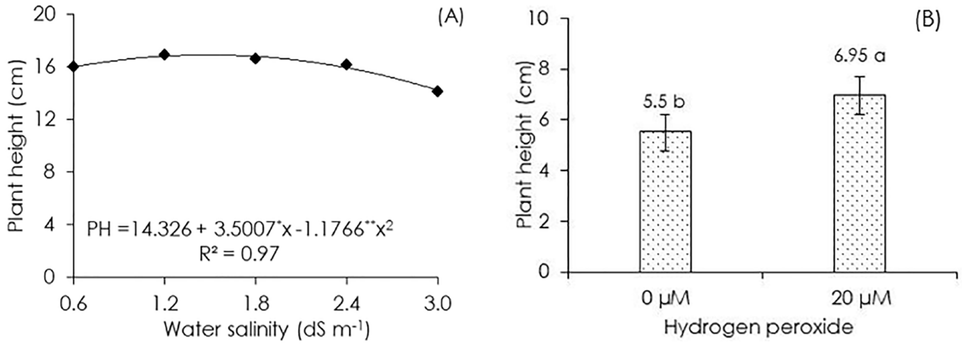

Figure 1. Plant height of the yellow passion fruit cv. Rubi do Cerrado as a function of irrigation water salinity - ECw (A) and different concentrations of hydrogen peroxide (B) 30 days after sowing.

Means followed by different letters show a significant difference between treatments by the Tukey test p≤5%.

The exogenous application of hydrogen peroxide also affected the growth of passion fruit plants 30 days after sowing. The means comparison test (Figure 1B) revealed that the plants that received 20 µM H_2O_2 had a statistically higher PH than those without H_2O_2. Also, the plants that received 20 µM H_2O_2 showed a 20.86% increase in plant height (1.44 cm) compared to those that did not receive H_2O_2 (0 µM). The increase in the PH of passion fruit plants may be related to the characteristics of hydrogen peroxide since, when applied at low concentrations, this reactive oxygen species can induce plant tolerance to biotic and abiotic stress for being an adaptive signaling molecule that acts in kinase transduction, in the modification of transcription factors, and in gene expression (Ellouzi et al., 2017; Engineer et al., 2016).

When observing the effect of salinity on the number of leaves of the passion fruit cv. Rubi do Cerrado at 30 DAS (Figure 2A), a quadratic reduction is observed in response to the increase in irrigation water salinity. The regression equation (Figure 2A) shows a maximum estimated value of 7.51 leaves in the plants irrigated with the lowest ECw level (0.6 dS m^{-1}) and a minimum of 4.51 leaves in the plants cultivated under the ECw of 3.0 dS m^{-1}. In relative terms, the plants irrigated with the highest salinity level (3.0 dS m^{-1}) showed a 39.95% NL reduction (3.00 leaves) compared to those grown under the lowest water salinity level (0.6 dS m^{-1}). The reduction in the number of leaves may be an alternative to minimize water losses to the atmosphere and maintain the high water potential. Also, it could be associated with the sensitivity of passion fruit to salt stress, which directly affects the photosynthetic apparatus of plants and compromises the production of photosynthetic pigments, thus reducing plant growth (Cavalcante et al., 2018).

Therefore, high salt concentrations in the root zone could cause changes to plant physiology due to osmotic imbalance and the inhibition of cell division and expansion, thus reducing the number of leaves, including the production of young leaves and, consequently, the transpiration capacity of plants, which directly depend on the extensibility process of the cell wall (Taiz & Zeiger, 2013).

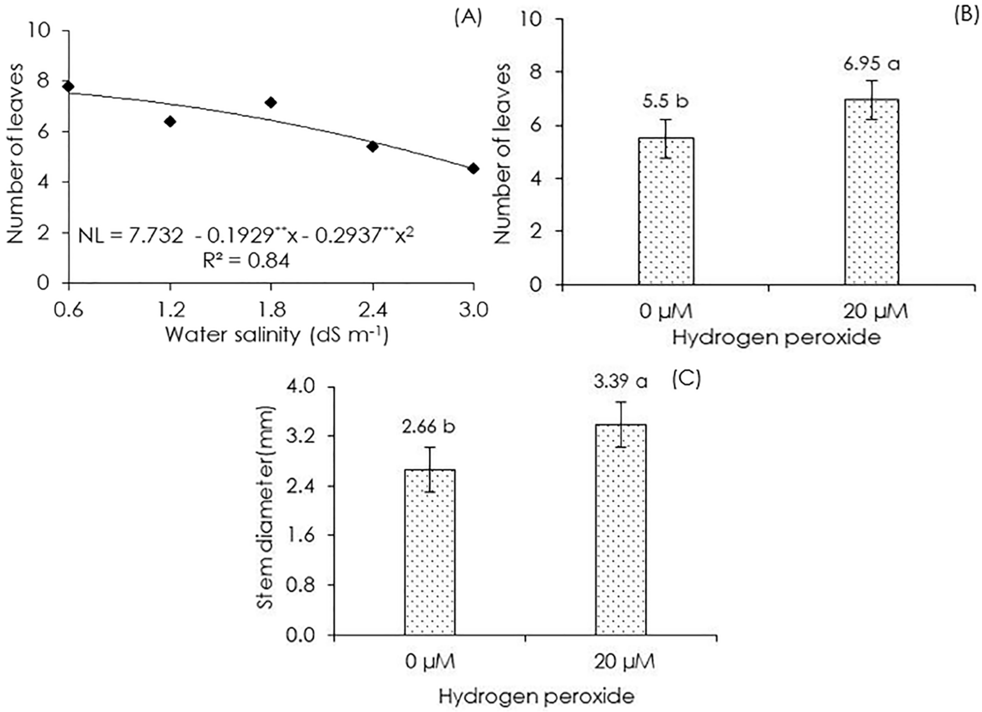

Figure 2. Number of leaves of the yellow passion fruit cv. Rubi do Cerrado grown under water salinity – ECw (A) 30 days after sowing and number of leaves (B) and stem diameter (C) as a function of hydrogen peroxide concentrations - H_2O_2 60 days after sowing.
Means followed by different letters show a significant difference between treatments by the Tukey test p≤5%..

The foliar application of hydrogen peroxide also increased the number of leaves of the passion fruit cv. BRS Rubi do Cerrado (Figure 2B) 30 days after sowing. The means comparison test (Figure 2B) shows that the plants that received 20 µM H_2O_2 produced a statistically higher number of leaves than the plants under the control treatment (0 µM). The higher leaf production could be associated with the capacity of the hydrogen peroxide molecule to stimulate plant growth and yield by promoting cell division and elongation as well as forming a secondary cell wall in the attempt to maintain the integrity of the cell and prevent osmotic lysis (Orabi et al., 2015).

The analysis of the effect of H_2O_2 application revealed that the plants that received 20 µM H_2O_2 differed statistically from the control treatment (0 µM H_2O_2) (Figure 2C). When comparing the plants that received 20 µM H_2O_2, there was a 21.53% increase (0.73 mm) in stem diameter in relation to the plants that did not receive H_2O_2 (0 µM), showing that the application of low hydrogen peroxide concentrations can benefit the early seedling growth of the passion fruit cv. BRS Rubi do Cerrado.

The positive effect of hydrogen peroxide on stem diameter (Figure 2C) can be attributed to the modulation of physiological and biochemical processes, such as photosynthesis, proline accumulation, and the detoxification of reactive oxygen species (Hossain et al., 2015). Therefore, the exogenous application of hydrogen peroxide acts directly to minimize the negative effects of salt stress on plants, increasing the antioxidant activity and with reflections on plant growth (Sun et al., 2016).

There was a significant effect ($p<0.05$) of the interaction between water salinity levels and H_2O_2 concentrations on the height of passion fruit plants at 60 DAS (Figure 3A). The means comparison test shows that the different H_2O_2 concentrations did not significantly influence the PH when the plants were subjected to the water salinity levels of 0.6, 1.2, and 1.8 dS m^{-1}. However, at 2.4 and 3.0 dS m^{-1}, the application of 20 µM H_2O_2 resulted in higher plant growth in height compared to the plants that did not receive H_2O_2.

Therefore, small hydrogen peroxide concentrations favor plant development when these are subjected to abiotic stress conditions by stimulating a higher accumulation of soluble proteins and carbohydrates, which act as organic solutes and assist in the osmotic adjustment of plants to salt stress, resulting in higher water and nutrient uptake (Carvalho et al., 2011).

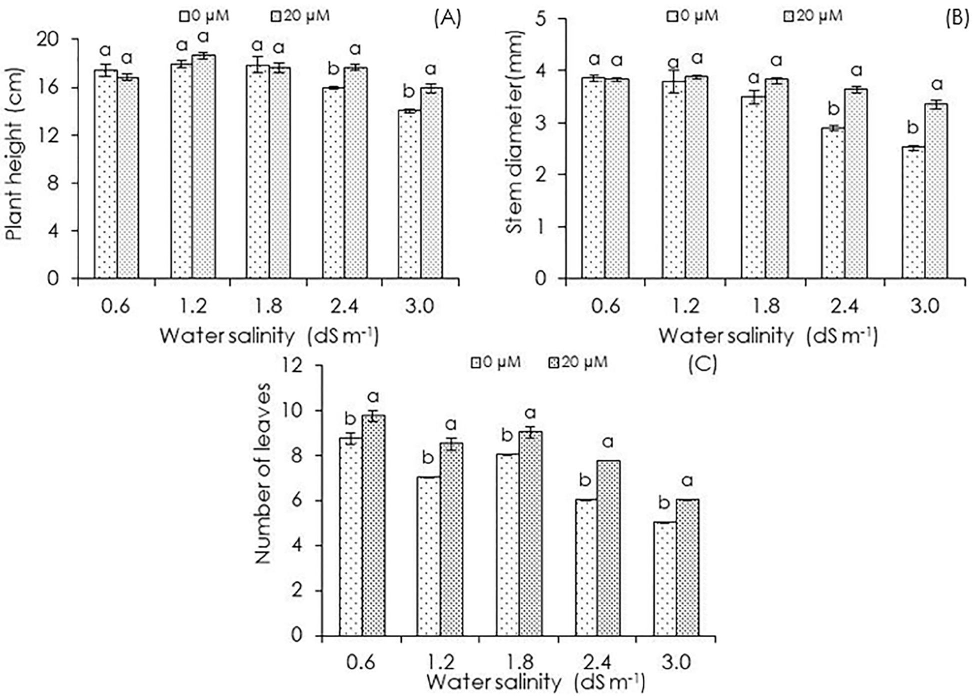

Figure 3. Plant height (A), stem diameter (B), and number of leaves (C) as a function of the interaction between water salinity levels – ECw and hydrogen peroxide concentrations 60 days after sowing.
Means followed by different letters show a significant difference between treatments by the Tukey test p≤5%..

The interaction between the salinity levels of the irrigation water and hydrogen peroxide concentrations significantly affected the stem diameter of passion fruit plants 60 days after sowing. The means comparison test (Figure 3B) reveals that the plants grown under the ECw levels of 2.4 and 3.0 dS m^{-1} had the highest growth in stem diameter when receiving the exogenous application of 20 μM H_2O_2. In relative terms, the SD was reduced by 34.71% (1.34 mm) when comparing the plants irrigated with the lowest salinity level (0.6 dS m^{-1}) with those under the ECw of 3.0 dS m^{-1} and those without hydrogen peroxide (0 μM) at 60 DAS. The positive effect of H_2O_2 at low concentrations could be associated with its function as a signaling molecule, acting in the regulation of several physiological mechanisms, including responses to salt stress, by promoting the production of antioxidant enzymes that act to mitigate the deleterious effects of abiotic stresses (Baxter et al., 2014).

For the number of passion fruit leaves, the means comparison test (Figure 3C) shows that the plants that received 20 μM H_2O_2 had the highest leaf production regardless of the level of water electrical conductivity. The higher leaf production in these plants is possibly associated with the fact that this reactive oxygen species, when applied at moderate concentrations, works as a transmitting mechanism of stress signs through signaling pathways and acts synergistically with kinase proteins and transcription factors to finally regulate plant metabolism, development, and the expression of genes related to resistance to stress (Noctor et al., 2018).

According to the summary of the analysis of variance (Table 3), the relative growth rate in plant height (RGR_{PH}), stem diameter (RGR_{SD}), and leaf dry biomass, stem, and root, in addition to the Dickson Quality Index (DQI), were all significantly affected (p<0.01) by the interaction between salinity levels and hydrogen peroxide concentrations. The water salinity levels significantly influenced all growth variables analyzed. Except for RGR_{PH}, the H_2O_2 concentrations significantly influenced all variables analyzed.

The interaction between salinity levels and hydrogen peroxide concentrations significantly influenced the relative growth rate in plant height (RGR_{PH}) of the passion fruit cv. BRS Rubi do Cerrado. The means comparison test (Figure 4A) reveals that the application of 20 μM H_2O_2 decreased the RGR_{PH}. However, irrigation with the highest salinity level and H_2O_2 application resulted in higher plant growth. When comparing the RGR_{PH} of passion fruit plants irrigated with the lowest salinity level (0.6 dS m^{-1}) with those irrigated with the highest salinity level (3.0 dS m^{-1}) and sprayed with 20 μM H_2O_2, it is possible to observe an increase of 18.75% (0.0006 cm cm^{-1} day^{-1}).

The reduction in plant growth observed through the RGR_{PH} may have been caused by the excess salts in the substrate. According to Nobre et al. (2014), glycophytic plants, such as passion fruit, show restricted water and nutrient uptake when subjected to salt concentrations above a threshold level, decreasing the CO_2 assimilation rate and, consequently, inhibiting growth.

The plants cultivated under the salinity level of 1.8 dS m^{-1} and without hydrogen peroxide spraying (0 µM) showed the highest RGR_{PH}. Thus, the exogenous application of hydrogen peroxide at high concentrations in situations where the plant is still not under stress conditions results in a toxic effect, reducing plant growth, as stated by Yamauchi et al. (2013), who stressed that hydrogen peroxide (H_2O_2) could reduce enzymatic complexes and lead to cell death when applied at high concentrations.

Table 3. Summary of the analysis of variance referring to the relative growth rate in plant height (RGR_{PH}) and stem diameter (RGR_{SD}) in the period from 30 to 60 DAS, and leaf dry phytomass (LDP), stem (StDP), and root (RDP) and Dickson Quality Index (DQI) of passion fruit plants irrigated with saline water and exogenous hydrogen peroxide application 60 days after sowing.

Source of variation	DF	Mean squares					
		RGR_{PH}	RGR_{SD}	LDP	StDP	RDP	DQI
Salinity levels (SL)	5	$4\times10^{-6**}$	$7\times10^{-4**}$	$0,932^{**}$	0.149^{**}	0.020^{**}	0.018^{**}
Linear regression	1	$7\times10^{-7**}$	$2\times10^{-3**}$	$3,54^{**}$	0.456^{**}	0.073^{**}	0.078^{**}
Quadratic regression	1	1×10^{-7ns}	$1\times10^{-4**}$	$0,128^{ns}$	$1.4x10^{-4ns}$	$7x10^{-3ns}$	0.038^{**}
Peróxido de hidrogênio (H_2O_2)	1	$2.72x10^{-7ns}$	$1,3\times10^{-4**}$	$0,696^{**}$	0.595^{**}	0.201^{**}	0.071^{**}
Linear regression	1	$1\times10^{-7**}$	1×10^{-6ns}	$4,33^{**}$	0.668^{**}	0.098^{**}	0.078^{**}
Quadratic regression	1	$8\times10^{-5**}$	1×10^{-6ns}	$0,64^{**}$	0.212^{**}	0.111^{**}	0.038^{**}
Interaction (SL × H_2O_2)	4	$6\times10^{-6**}$	$75\times10^{-4**}$	$0,733^{**}$	0.183^{**}	0.077^{**}	0.028^{**}
Residual		$4.04x10^{-7}$	$5.73x10^{-7}$	0,025	0.002	0.001	0.0006
CV		28.31	15.73	15.20	7.07	8.27	10.45

^{ns} - Non-significant; DF – Degrees of freedom; CV – coefficient of variation; *,** significant at 5 and 1%, respectively.

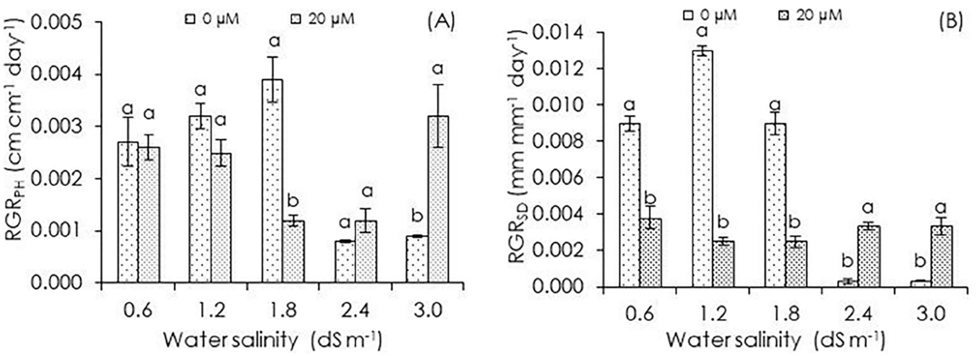

Figure 4. Relative growth rate in plant height – RGR_{PH} (A) and stem diameter – RGR_{SD} (B) of the passion fruit cv. Rubi do Cerrado as a function of the interaction between salinity levels - ECw and hydrogen peroxide concentrations in the period from 30 to 60 days after sowing.
Means followed by different letters show a significant difference between treatments by the Tukey test p≤5%.

The relative growth rate in stem diameter of the passion fruit cv. BRS Rubi do Cerrado (Figure 4B) was also significantly influenced by the interaction between salinity levels and hydrogen peroxide concentrations. The increase in the salinity levels reduced the RGR_{SD} by 96.38% (0.0086 mm day^{-1}) in seedlings irrigated with the lowest salinity level (0.6 dS m^{-1}) compared to those under the highest salinity level (3.0 dS m^{-1}) without H_2O_2.

The deleterious effects on the RGR_{SD} of the passion fruit cv. Rubi do Cerrado under salt stress reduced the soil water potential by inducing important changes in the hormonal balance of the plant, affecting plant growth and stimulating the production of specific plant hormones, such as abscisic acid (ABA), which plays a key role in stomatal conductance and plant growth (Santos Júnior et al., 2016). The positive effect of the exogenous application of hydrogen peroxide is related to its role as a signaling messenger and the induction of tolerance to several types of stress as long as it is applied at low concentrations. Otherwise, hydrogen peroxide acts as an inducer of oxidative stress (Fedina et al., 2009).

The application of hydrogen peroxide (20 µM) promoted a negative effect up to the water electrical conductivity level of 1.8 dS m^{-1}, with the most expressive value at the salinity level of 1.2 dS m^{-1}. In relative terms, H_2O_2 promoted an 80.76% reduction (0.0104 mm $plant^{-1}$ day^{-1}) in relation to the seedlings that did not receive hydrogen peroxide via foliar application. For the plants subjected to the highest salinity level (3.0 dS m^{-1}) and spraying with 20 µM H_2O_2, the RGR_{SD} was increased by

0.00302 mm mm^{-1} day^{-1} in relation to the control treatment (0 μM). Nadeem et al. (2013) stated that plant growth reduction under salt stress conditions occurs due to the osmotic effects of salinity and the toxic effects of specific ions, reducing water uptake and promoting a nutritional unbalance that affects plant development.

Therefore, the use of small hydrogen peroxide concentrations acts directly in the production of antioxidant enzymes by fighting free radicals that cause oxidative stress in plants and, consequently, their early senescence (Gondim et al., 2012).

A significant effect of the interaction between factors (SL × H_2O_2) is observed when studying the dry phytomass of leaves (Figure 5A). The plants irrigated with the ECw of 0.6 dS m^{-1} and foliar application of 20 μM H_2O_2 showed higher LDP accumulation. For the plants that received the electrical conductivity of 1.8 dS m^{-}, H_2O_2 caused a negative effect when applied to the yellow passion fruit cv. Rubi do Cerrado. When relating the ECw level of 3.0 dS m^{-1} to plants irrigated with the lowest salinity level (0.6 dS m^{-1}), it is possible to observe a 65.70% reduction (1.36 g $plant^{-1}$) when the plants received 20 μM H_2O_2.

When applied at high concentrations, hydrogen peroxide can cause negative effects to plants by damaging the photosystem II (PSII), decreasing the chlorophyll content, hindering chloroplast development, changing the composition of the thylakoid membrane, and, consequently, affecting phytomass production (Dey et al., 2014).

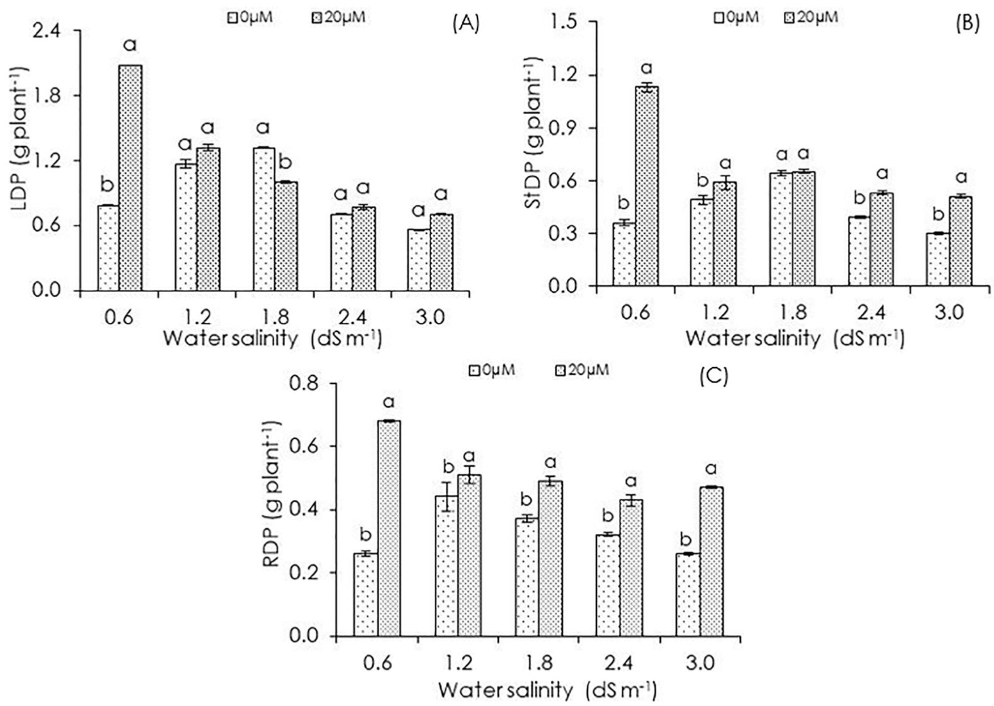

Figure 5. Leaf dry phytomass (A), stem (B), and root (C) of the yellow passion fruit cv. Rubi do Cerrado as a function of the interaction between salinity levels – ECw and hydrogen peroxide concentrations - H_2O_2 60 days after sowing.
Means followed by different letters show a significant difference between treatments by the Tukey test p≤5%.

The dry stem phytomass of passion fruit plants was also significantly affected by the interaction between the salinity levels of the irrigation water and hydrogen peroxide concentrations (Figure 5B). When the plants were irrigated with the lowest salinity level (0.6 dS m^{-1}) and without hydrogen peroxide, there was a 16.6% reduction (0.06 g $plant^{-1}$) in relation to those cultivated under the ECw of 3.0 dS m^{-1} and without hydrogen peroxide.

H_2O_2 is a versatile molecule involved in growth regulation and plant development when at lower concentrations in stress conditions, increasing cell wall resistance, strengthening the cell wall, stomatal movement, and cell growth and development. Reactive oxygen species are related to a series of enzymes that stimulate the expression and activation of stress tolerance genes through the physiological adjustment to withstand continuous environmental changes (Wojtyla et al., 2016; Gao et al., 2010; Khan et al., 2016).

Irrigation with different water electrical conductivity levels significantly affected the dry root phytomass of the yellow passion fruit cv. BRS Rubi do Cerrado 60 days after sowing (Figure 5C). The unfolding of the interaction reveals that the foliar application of H_2O_2 promoted a higher RDP accumulation regardless of the ECw level. This effect is possibly related to the inability of osmoregulation by the plant since, in saline soils, the osmotic gradient is reduced or reversed, and water does not effectively move to the roots, hindering the uptake of water and, consequently, of nutrients (Deinlein et al., 2014). Furthermore, hydrogen peroxide positively affected root development by the action of auxins, which is a key regulating hormone of root growth with sensitivity to environmental stimuli (Sun et al., 2018).

The hydrogen peroxide concentrations favored the accumulation of dry stem phytomass, and the highest value was observed when the plants were irrigated with the lowest salinity level (0.6 dS m^{-1}), increasing by 61.76% (0.42 g $plant^{-1}$).

The increase in the salinity levels of the irrigation water positively affected seedling quality at the levels from 0.6 to 1.8 dS m^{-1} without hydrogen peroxide, promoting, in relative terms, a 44.44% increase (0.12). For the lowest salinity level (0.6 dS m^{-1}), there was, in relative terms, a 64.28% increase (0.27), whereas, for the highest salinity level, the Dickson Quality Index increased by 47.82% (0.11) when the plants received H_2O_2 compared to those that did not receive the exogenous application of this reactive oxygen species. Associated with this, the increase in the H_2O_2 concentrations also increased stress tolerance, which can be possibly explained by the action of defense mechanisms developed by the plants by inducing the production of antioxidant enzymes and minimizing the deleterious effects of salinity (Silva et al., 2016).

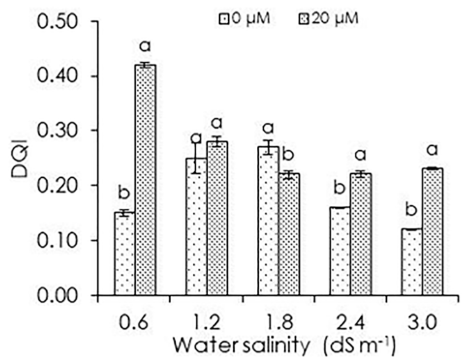

Figure 6. Dickson Quality Index -DQI of the passion fruit cv. Rubi do Cerrado as a function of the interaction between salinity levels - ECw and hydrogen peroxide concentrations - H_2O_2 60 days after sowing (DAS).
Means followed by different letters show a significant difference between treatments by the Tukey test p≤5%.

Thus, hydrogen peroxide improves seedling quality when applied at low concentrations via foliar spraying to the passion fruit cv. Sol do Cerrado. Silva et al. (2019), when cultivating seedlings of the soursop cv. Morada Nova at different water salinity levels (0.7, 1.4. 2.1, 2.8, and 3.5 dS m^{-1}) and small concentrations of hydrogen peroxide (0, 25, 50, 75, and 100 μM), observed that H_2O_2 mitigated the deleterious effects of salts on the seedling quality of the soursop cv. 'Morada Nova'.

Conclusions

The exogenous application of 20 μM hydrogen peroxide results in higher plant growth in stem diameter, plant height, and number of leaves 30 and 60 days after sowing, in addition to the Dickson Quality Index 60 days after sowing the passion fruit cv. BRS Rubi do Cerrado. The application of 20 μM hydrogen peroxide induces the acclimatization of the passion fruit cv. BRS Rubi do Cerrado irrigated with saline water.

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