Brazilian Journal of Biology®

ISSN 1519-6984 (Print)
ISSN 1678-4375 (Online)

Original Article

Salicylic acid concentrations and forms of application mitigate water stress in sour passion fruit seedlings

Concentrações e formas de aplicação de ácido salicílico mitiga o estresse hídrico em mudas de maracujazeiro-azedo

R. T. Fatima[a]*, G. S. Lima[a], L. A. A. Soares[b], L. L. S. A. Veloso[a], A. A. R. da Silva[a], C. N. Lacerda[a], F. A. Silva[a], J. S. Nobrega[c], J. T. A. Ferreira[a] and W. E. Pereira[c]

[a]Universidade Federal de Campina Grande – UFCG, Unidade Acadêmica de Engenharia Agrícola – UAEA, Campus Campina Grande, Campina Grande, PB, Brasil

[b]Universidade Federal de Campina Grande – UFCG, Centro de Ciência e Tecnologia Agroalimentar – CCTA, Campus Pombal, Pombal, PB, Brasil

[c]Universidade Federal da Paraíba – UFPB, Centro de Ciências Agrárias – CCA, Campus Areia, Areia, PB, Brasil

Abstract

The present study aimed to evaluate concentrations and forms of application of salicylic acid used for water stress mitigation on the gas exchange and growth of yellow passion fruit. The experimental design was arranged in randomized blocks in a 4 × 4 × 2 factorial scheme, with four concentrations of salicylic acid (SA) via foliar application (0.0, 0.7, 1.4, and 2.1 mM), four SA concentrations via fertigation (0.0, 0.7, 1.4, and 2.1 mM), and two irrigation depths estimated based on the actual evapotranspiration - ETr (50 and 100% of ETr), with three replications. Water stress negatively affected the physiology and growth of yellow passion fruit seedlings at 75 days after sowing (DAS). The application of salicylic acid, regardless of the form of application, attenuates the effects of water stress on gas exchange and growth of yellow passion fruit, with the best results obtained when applying a concentration of 1.30 mM via leaf or 0.90 mM via fertirrigation. The combination of foliar application of AS and fertigation contributed to improve photosynthetic and growth parameters under water conditions of 50 and 100% of ETr. The foliar application of AS presents superior responses to the application via fertigation. These results reinforce the hypothesis that the attenuation of water stress by salicylic acid is related to the maintenance of gas exchange, which depends on the concentration and form of application, and studies testing combinations throughout the crop cycle become promising for advances in knowledge from the action of this phytohormone on abiotic stress.

Keywords: *Passiflora edulis*, gas exchange, deficit irrigation, fitohormone.

Resumo

O presente estudo teve como objetivo avaliar as concentrações e formas de aplicação do ácido salicílico como mitigador do estresse hídrico sobre as trocas gasosas e parâmetros de crescimento do maracujazeiro amarelo. O delineamento experimental foi em blocos casualizados, em esquema fatorial 4 × 4 × 2, com tratamentos constituídos por quatro concentrações de ácido salicílico (SA) via aplicação foliar (0,0, 0,7, 1,4 e 2,1 mM), quatro concentrações de SA via fertirrigação (0,0, 0,7, 1,4 e 2,1 mM) e duas lâminas de irrigação estimadas com base na evapotranspiração real - ETr (50 e 100% da ETr), com três repetições. O estresse hídrico afetou negativamente a fisiologia e o crescimento das mudas de maracujazeiro amarelo aos 75 dias após o semeio (DAS). A aplicação de ácido salicílico, independente da forma de aplicação, atenua os efeitos do estresse hídrico nas trocas gasosas e crescimento do maracujazeiro amarelo, com os melhores resultados obtidos ao se aplicar a concentração de 1,30 mM via foliar ou 0,90 mM via fertirrigação. A combinação de aplicação de AS via foliar e fertirrigação contribuiu para melhorar os parâmetros fotossintéticos e de crescimento nas condições hídricas de 50 e 100% da ETr. A aplicação via foliar de AS apresenta respostas superiores a aplicação via fertirrigação. Esses resultados reforçam a hipótese de que a atenuação do estresse hídrico pelo ácido salicílico está relacionada a manutenção das trocas gasosas, a qual depende da concentração e forma de aplicada, sendo que estudos testando combinações ao longo do ciclo da cultura se torna promissores para avanços nos conhecimentos provenientes da atuação desse fitormônio sobre o estresse abiótico.

Palavras-chave: *Passiflora edulis*, trocas gasosas, irrigação deficitária, fitormônio.

*e-mail: reynaldo.t16@gmail.com
Received: January 04, 2023 – Accepted: April 13, 2023

1. Introduction

Passion fruit (*Passiflora edulis*) is a variety of tropical and subtropical fruit-bearing plants of the genus *Passiflora* (Vieira et al., 2022). In Brazil, their main representative is *Passiflora edulis* Sims, popularly known as yellow or sour passion fruit, corresponding to more than 95% of the passion fruit orchards in this country (Meletti, 2011). The relevance and high acceptance of these fruits are because of their high contents of carbohydrates, sugars, vitamins, minerals, and organic compounds, favoring their commercialization and industrial use (Biswas et al., 2021; Vieira et al., 2022).

The Northeast region of Brazil has stood out as the largest passion fruit producer, accounting for 69.6% of the 683,993 tons produced in 2021 (IBGE, 2022). However, this region is characterized by irregular rainfall and high evaporation rates, limiting water availability during the crop cycle and causing losses in production and fruit quality (Lima et al., 2015; Soares et al., 2018; Pinheiro et al., 2022a).

In this scenario, the effects of water stress on each crop may vary depending on the management of irrigation and fertilization, edaphoclimatic conditions, species, or even the different developmental stages (Deka et al., 2018), manifesting more intensely during early development through metabolic and physiological changes and resulting in growth inhibition (Morales et al., 2020; Zombardo et al., 2020). As already observed in passion fruit, the photosynthetic rate decreases through stomatal closure due to the lower CO_2 entry and electron transport limitations in the PSII, resulting in lower carbon fixation and forming reactive oxygen species (ROS), which affect the metabolic activity of the plant (Pinheiro et al., 2022b; Souza et al., 2018; Cavalcante et al., 2020; Lozano-Montaña et al., 2021).

Furthermore, the application of salicylic acid (SA) has shown promising responses in mitigating water stress in several crops (Andrade et al., 2021; Kaya, 2021; Kulak et al., 2021) by maintaining the photosynthetic apparatus while inducing greater enzymatic activity, also controlling ROS, contributing to the water status, and improving the cell turgor and nutrient assimilation (Shadmehri and Khatiby, 2020; Damalas and Koutroubas, 2022). With regard to gas exchange, SA contributes to stomatal opening and CO_2 entry into the substomatal chamber, thus favoring transpiration and increasing the carboxylation rate, as observed by Saheri et al. (2020) in common purslane (*Portulaca oleracea* L.), by Zafar et al. (2021) in Java plum (*Syzygium cumini* L), and by Galviz et al. (2021) in tomato (*Solanum lycopersicum* L.).

However, the best form of SA application and its appropriate concentrations are reasons for discussion since there are different responses when the application is performed exclusively via the leaves or roots (Lopes et al., 2019; Sohag et al., 2020; Nóbrega et al., 2021). In this scenario, studies assessing plant responses to different forms of application and concentrations are still incipient in the literature (Gharbi et al., 2018; Souri and Tohidloo, 2019).

The hypothesis of the present work is that salicylic acid in low concentrations attenuates the effects of water stress, regardless of the form of application, by maintaining gas exchange and increasing carbon assimilation, with gains in the growth of sour passion fruit seedlings.

From this perspective, this study aimed to evaluate different concentrations and forms of application of salicylic acid used for water stress mitigation on the physiology and growth of sour passion fruit plants.

2. Material and Methods

2.1. Site description and experimental design

The experiment was performed from September to December 2020 in a greenhouse (7° 15' 18" S, 35° 52' 28" W) located at the Agricultural Engineering Academic Unit (UAEA) of the Federal University of Campina Grande (UFCG), Campina Grande, Paraíba, Brazil. Air temperature data (maximum and minimum) and average relative humidity at the experimental site are shown in Figure 1.

The experimental design was arranged in randomized blocks in a 4 × 4 × 2 factorial scheme, with four salicylic acid (SA) concentrations via foliar application (0.0, 0.7, 1.4, and 2.1 mM), four SA concentrations via fertigation (0.0, 0.7, 1.4, and 2.1 mM), and two irrigation depths estimated based on the actual evapotranspiration (ETr - 50 and 100% ETr), with three replications, totaling 96 experimental units.

2.2. Conducting the experiment

The actual evapotranspiration was determined using drainage lysimeters, as proposed by Bernardo et al. (2006). The water depths for replacement were determined by the difference between the volume applied and the volume drained in the previous irrigation event, thus resulting in the volume consumed by the plants, which was multiplied by 0.5 and 1.0 to obtain the water depths corresponding to 50 and 100% ETr.

The salicylic acid concentrations were obtained according to Silva et al. (2020) by dissolving 30% ethyl alcohol - C_2H_6O (95.5%) in distilled water due to its low solubility in water at room temperature. The Wil-fix adjuvant (adhesive spreader, Charmon Destyl Ltd., Valinhos, BR) was used at the concentration of 0.5 mL L^{-1} to reduce the surface tension of the droplets on the leaf surface.

Four seeds of the sour passion fruit cv. 'BRS GA1' were used in each experimental unit due to the high yield and great market acceptance of this cultivar. Sowing

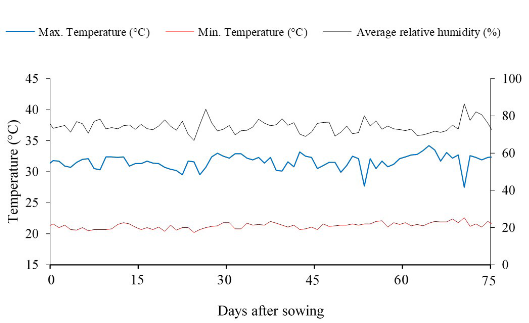

Figure 1. Temperature (maximum and minimum) and average relative humidity in the greenhouse area during the experimental period.

was performed in 10-L pots filled with Fluvic Neosol (Santos et al. 2018), classified as Fluvents by Soil Taxonomy (USDA, 2014), collected from the 0.0-0.40 m layer. The physical and chemical soil characteristics (Table 1) were analyzed according to Teixeira et al. (2017).

Each experimental unit corresponded to one pot containing an 18-mm wide drain covered with a geotextile fabric (Bidim type) and a 0.5-kg thick layer of crushed stone. The drained water was collected in a 2-L plastic bottle connected to the bottom of the lysimeters. The drained water volume was used to estimate the crop water balance. Soil moisture was maintained at maximum water retention capacity until seedling emergence. Then, the plants were thinned to one plant per plot when showing one pair of true leaves.

The applications of salicylic acid started 20 days after sowing (DAS), were performed every 15 days in the late afternoon and were split into four applications, totaling 200 mL in each plant at the end of the experiment. In the foliar application treatments, the product was applied on the abaxial and adaxial leaf surfaces using a manual sprayer with a capacity of 1 L, with the soil protected by a plastic barrier to prevent drift. In treatments via fertigation, the volume per application was 50 mL per plant, which was diluted until reaching the amount of water necessary to maintain the evapotranspiration defined per pot.

Fertilization with N (300 mg kg^{-1}), P_2O_5 (150 mg kg^{-1}), and K_2O (300 mg kg^{-1}) was performed according to Novais et al. (1991) using urea, potassium chloride, and monoammonium phosphate as the respective sources. A micronutrient solution with 1.0 g L^{-1} of Dripsol micro® (Dripsol SQM Vitas Ltd., Candeias, BR) containing Mg (1.4%), Zn (0.5%), B (0.05%), Fe (0.5%), Mn (0.05%), Cu (0.5%), and Mo (0.02%) was applied on the adaxial and abaxial leaf surfaces using a backpack sprayer. Fertilization with macro and micronutrients was performed every 20 days in the late afternoon.

All phytosanitary preparations and practices recommended for the crop were performed during the experiment by monitoring the emergence of pests and diseases and adopting control measures whenever necessary.

2.3. Gas exchange and growth analyses

The gas exchange variables were determined at 75 days after sowing from 7:00 to 9:00 a.m. using a portable infrared gas analyzer – (IRGA - ADC BioScientific Ltd., LCpro T model, Hoddesdon, UK) with an airflow of 300 mL min^{-1} and a light source of 1,200 µmol m^{-2} s^{-1} (Freire et al., 2014). The variables analyzed were net CO_2 assimilation rate (*A* - µmol CO_2 m^{-2} s^{-1}), stomatal conductance (*gs* - mol H_2O m^{-2} s^{-1}), intercellular CO_2 concentration (*Ci* - µmol CO_2 m^{-2} s^{-1}), and transpiration (*E* - mmol H_2O m^{-2} s^{-1}). The water use efficiency (WUE - µmol CO_2 m^{-2} s^{-1}/mmol H_2O m^{-2} s^{-1}), intrinsic water-use efficiency (iWUE - µmol CO_2 m^{-2} s^{-1}/ mol H_2O m^{-2} s^{-1}), and instantaneous carboxylation efficiency (iCEi - µmol CO_2 m^{-2} s^{-1}/ µmol CO_2 m^{-2} s^{-1}) were calculated.

Moreover, the following growth variables were also determined in the same period: plant height (PH), measured from the base of the plant to the apical meristem; stem diameter (SD), measured with a digital caliper (mtx Ltd., 316119 model, CN) at 5 cm from the ground; and leaf area (LA), obtained by measuring the leaf length and width using the methodology proposed by Cavalcante et al. (2009), as seen in Equation 1:

$$LA = L \times W \times 0.74 \quad (1)$$

Where: LA= leaf area (cm^2); L= leaf length (cm); and W= leaf width (cm).

2.4. Statistical analyses

The data were subjected to analysis of variance and polynomial regression ($p \leq 0.05$ and $p \leq 0.01$) using the statistical software R (R Core Team, 2017).

3. Results

There were significant effects of the interaction irrigation depths x salicylic acid via foliar application x salicylic acid via fertigation on all sour passion fruit physiological variables at 75 DAS (Table 2).

In the plants subjected to water deficit (50% ETr), the *gs* was influenced by salicylic acid (Figure 2A), increasing by

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

Chemical characteristics									
pH (H_2O) (1:2.5)	**OM %**	**P (mg kg^{-1})**	**K^+**	**Na^+**	**Ca^{2+}**	**Mg^{2+}**	**Al^{3+} + H^+**	**ESP (%)**	**ECse (dS m^{-1})**
			...........(cmolc kg^{-1})						
5.90	1.36	6.80	0.22	0.16	2.60	3.66	1.93	1.87	1.00
Physical-hydraulic characteristics									
Particle-size fraction (dag kg^{-1})			Textural class	Moisture (kPa)		AW	Total porosity %	BD	PD
Sand	Silt	Clay		33.42	1519.5 g kg^{-1}			(g cm^{-3})	
732.90	142.10	125.00	SL	119.80	43.20	76.60	47.74	1.39	2.66

OM – Organic matter: Walkley-Black Wet Digestion; Ca^{2+} and Mg^{2+} extracted with 1 mol L^{-1} KCl at pH 7.0; Na^+ and K^+ extracted with 1 mol L^{-1} NH_4OAc at pH 7.0; Al^{3+} and H^+ extracted with 1 mol L^{-1} calcium acetate at pH 7.0; ESP - Exchangeable sodium percentage; ECse – Electrical conductivity of saturation extract; SL – Sandy loam; AW – Available water; BD - Bulk density; PD- Particle density.

Table 2. Summary of the analysis of variance for stomatal conductance (gs, mol H_2O m^{-2} s^{-1}), intercellular CO_2 concentration (Ci, µmol CO_2 m^{-2} s^{-1}), transpiration (E, µmol H_2O m^{-2} s^{-1}), net CO_2 assimilation rate (A, µmol CO_2 m^{-2} s^{-1}), water-use efficiency (WUE, µmol CO_2 m^{-2} s^{-1}/mmol H_2O m^{-2} s^{-1}), intrinsic water use efficiency (iWUE, µmol CO_2 m^{-2} s^{-1}/ mol H_2O m^{-2} s^{-1}), and instantaneous carboxylation efficiency (iCE, µmol CO_2 m^{-2} s^{-1}/ µmol CO_2 m^{-2} s^{-1}) of sour passion fruit plants subjected to different irrigation depths (I) and concentrations of salicylic acid applied via foliar application (L) and fertigation (F) 75 days after sowing.

Source of variation	DF	MS						
		gs	**Ci**	**E**	**A**	**WUE**	**WUEi**	**CEi**
Irrigation (I)	1	$1.0e^{-4ns}$	593.77^{*}	0.53^{**}	11.50^{**}	0.07^{ns}	270.91^{**}	$6.3e^{-5**}$
SA Leaves (L)	3	0.014^{**}	418.75^{*}	1.17^{**}	5.26^{**}	2.32^{**}	500.91^{**}	$6.3e^{-5**}$
SA Fertigation (F)	3	$5.5e^{-3**}$	547.88^{**}	0.32^{**}	7.88^{**}	0.83^{**}	229.27^{**}	$5.3e^{-5**}$
I × L	3	$5.5e^{-3**}$	1129.12^{**}	0.97^{**}	4.83^{**}	2.45^{**}	267.27^{**}	$8.4e^{-5**}$
I × F	3	$4.5e^{-3**}$	354.90^{*}	0.15^{*}	0.71^{ns}	0.10^{ns}	50.90^{ns}	$3.0e^{-6ns}$
L × F	9	$6.4e^{-3**}$	980.30^{**}	0.56^{**}	4.60^{**}	0.80^{**}	163.66^{**}	$7.2e^{-5**}$
I × L × F	9	$6.8e^{-3**}$	486.23^{**}	0.34^{**}	4.79^{**}	0.30^{**}	152.18^{**}	$6.3e^{-5**}$
Blocks	2	$4.0e^{-4ns}$	54.56^{ns}	0.26^{*}	0.57^{ns}	0.06^{ns}	14.44^{ns}	$9.0e^{-6ns}$
CV (%)		12.38	3.28	8.54	9.53	9.63	15.68	10.09
Mean		0.248	310.87	2.81	7.21	2.62	30.54	0.023

DF- degree of freedom; MS- mean square; R – regression; CV - coefficient of variation. ^{ns} not significant. * significant at p ≤0.05. ** significant at p ≤ 0.01.

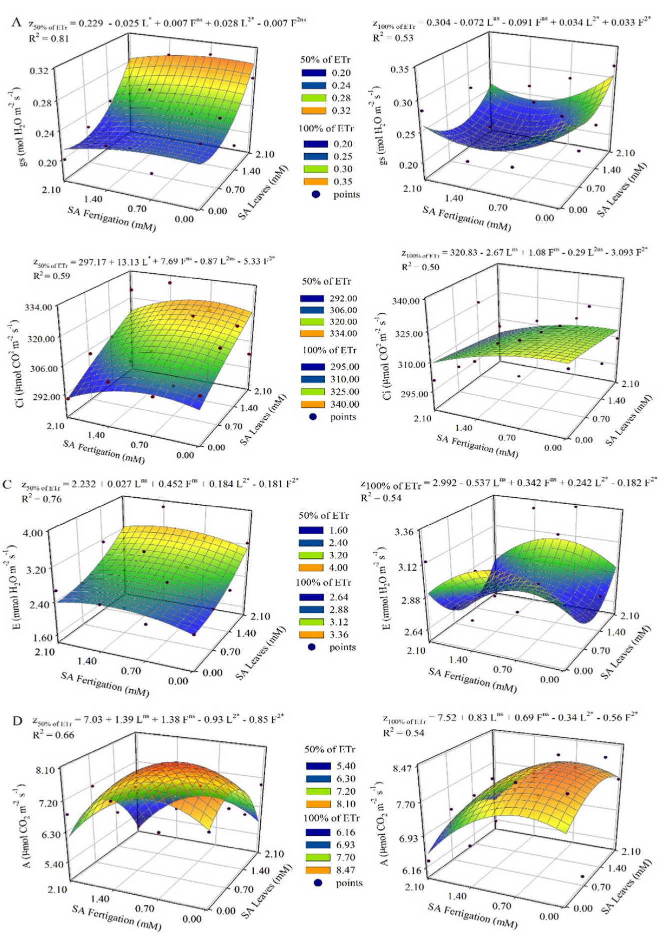

Figure 2. Stomatal conductance - gs (A), intercellular CO_2 concentration - Ci (B), transpiration - E (C), and net CO_2 assimilation rate – A (D) of sour passion fruit (*Passiflora edulis*) as a function of SA application via foliar application and fertigation under two water replacement depths 75 days after sowing. ^{ns}, *, **, respectively not significant, significant at p ≤ 0.05 and p ≤ 0.01.

40.48% (0.300 mol H_2O m^{-2} s^{-1}) when the SA concentration of 2.1 mM was applied via foliar application. On the other hand, SA application via fertigation reduced stomatal opening, reaching the lowest values at the SA concentration of 2.1 mM (0.209 mol H_2O m^{-2} s^{-1}). With 100% ETr water replacement, the highest stomatal opening value (0.304 mol H_2O m^{-2} s^{-1}) was also observed with 2.1 mM SA via foliar application. Under this condition, the lowest value of this variable was observed with the combination of 1.05 mM SA via foliar application and 1.31 mM via fertigation (0.204 mol H_2O m^{-2} s^{-1}).

With regard to *gs* (Figure 2A), the intercellular CO_2 concentration also increased with SA application (Figure 2B), and the association between the highest SA concentration via foliar application and 0.656 mM via fertigation resulted in the highest values of this variable (323.65 µmol CO_2 m^{-2} s^{-1}), decreasing to 289.78 µmol CO_2 m^{-2} s^{-1} when plants were subjected only to 2.1 mM SA via fertigation. There was no positive influence of salicylic acid on the intercellular CO_2 concentration under the water depth corresponding to 100% ETr, with the highest value of this variable occurring without SA application (320.92 µmol CO_2 m^{-2} s^{-1}) and the lowest value occurring with 2.1 mM SA for both forms of application (302.57 µmol CO_2 m^{-2} s^{-1}), showing that the highest CO_2 entry resulted from stomatal opening in plants under water stress.

Salicylic acid increased the transpiration rate of sour passion fruit plants grown under irrigation with 50% ETr (Figure 2C), ranging from 2.23 mmol H_2O m^{-2} s^{-1} in plants without SA to 3.38 mmol H_2O m^{-2} s^{-1} in plants that received 2.1 mM via foliar application and 1.18 mM via fertigation, increasing the *E* variable by 51.57%. In plants irrigated with 100% ETr, the application of 0.92 mM salicylic acid via fertigation led to the highest transpiration value (3.15 mmol H_2O m^{-2} s^{-1}), whereas the lowest value of this parameter was observed in the combination between 1.05 mM via foliar application and 2.1 mM via fertigation (2.61 mmol H_2O m^{-2} s^{-1}), decreasing by 17.14% compared to the highest concentration.

The combination of the SA concentrations of 0.79 mM via fertigation and foliar application resulted in the highest net CO_2 assimilation rate (Figure 2D) in plants subjected to water deficit (8.10 µmol CO_2 m^{-2} s^{-1}). In contrast, the lowest value of this variable was obtained with 2.1 mM SA in both forms of application (4.96 µmol CO_2 m^{-2} s^{-1}). On the other hand, the plants grown under irrigation with 100% ETr showed the lowest net CO_2 assimilation rate (6.49 µmol CO_2 m^{-2} s^{-1}) with 2.1 mM SA via fertigation, decreasing by 13.70% compared to plants that did not receive salicylic acid. The highest *A* value was obtained by associating the concentrations of 1.18 mM via foliar application and 0.65 mM via fertigation (8.23 µmol CO_2 m^{-2} s^{-1}).

In plants under water deficit, the lowest values of water use efficiency (1.44 µmol CO_2 m^{-2} s^{-1}/mmol H_2O m^{-2} s^{-1}), intrinsic water use efficiency (17.25 µmol CO_2 m^{-2} s^{-1}/mol m^{-2} s^{-1}), and instantaneous carboxylation efficiency (0.0163 µmol CO_2 m^{-2} s^{-1}/µmol CO_2 m^{-2} s^{-1}) were obtained with 2.1 mM in both forms of application (Figure 3), resulting in respective losses of 55.28, 52.29, and 38.26%. On the other hand, the highest mean values were obtained with the combination of 0.39 mM via foliar application and 0.52 mM via fertigation for WUE (3.22 µmol CO_2 m^{-2} s^{-1}/mmol H_2O m^{-2} s^{-1}), 0.65 mM via foliar application and 0.92 mM via fertigation for iWUE (36.16 µmol CO_2 m^{-2} s^{-1}/mol H_2O m^{-2} s^{-1}), and 0.52 mM via foliar application and 0.79 mM via fertigation for iCE (0.0264 µmol CO_2 m^{-2} s^{-1}/ µmol CO_2 m^{-2} s^{-1}).

Under water replacement conditions corresponding to 100% ETr, the highest water use efficiency was achieved with 1.18 mM SA via foliar application (2.96 µmol CO_2 m^{-2} s^{-1}/mmol H_2O m^{-2} s^{-1}), increasing by 30.97% compared to the lowest mean (2.26 µmol CO_2 m^{-2} s^{-1}/mmol H_2O m^{-2} s^{-1}) obtained with 2.1 mM via fertigation (Figure 3A). The highest intrinsic water use efficiency (39.70 µmol CO_2 m^{-2} s^{-1}/mol m^{-2} s^{-1}) was achieved using 1.05 mM SA in both forms of application (Figure 3B), increasing by 57.29% compared to plants that did not receive SA (25.24 µmol CO_2 m^{-2} s^{-1}/mol H_2O m^{-2} s^{-1}). The combination using 1.18 mM via foliar application and 0.39 mM via fertigation resulted in the highest instantaneous carboxylation efficiency (0.026 µmol CO_2 m^{-2} s^{-1}/ µmol CO_2 m^{-2} s^{-1}) (Figure 3C).

As observed for gas exchange, the interaction between factors had significant effects on the growth variables of plant height, leaf area, and stem diameter of sour passion fruit 75 DAS (Table 3).

Salicylic acid application in plants irrigated with 50% ETr increased plant height and the leaf area, the first variable ranging from 15.43 cm in plants without SA application to 53.35 cm in plants grown under 1.31 mM via foliar application and 1.05 mM via fertigation (Figure 4A), and the second ranging from 232.56 cm^2 to 643.88 cm^2 in plants grown under the combination with 1.57 mM via foliar application and 1.05 mM via fertigation (Figure 4B). Salicylic acid application also increased the stem diameter of plants grown under In plants under water deficit (Figure 4C), achieving the highest value (4.38 mm) with 1.44 mM via foliar application and 0.92 mM via fertigation.

Under 100% ETr, the growth variables showed the lowest mean values under 2.1 mM SA via fertigation, with 19.42 cm for PH (Figure 4A), 336.45 cm^2 for LA (Figure 4B), and 2.96 mm for SD (Figure 4C), decreasing by 72.52, 59.89, and 40.44% compared to the highest means obtained for the three variables with 1.31 mM via foliar application and with fertigation at 0.79 mM for PH (70.66 cm), 0.39 mM for LA (838.77 cm^2), and 0.52 mM for SD (4.97 mm).

4. Discussion

The reduction in the gas exchange of sour passion fruit plants due to low water availability results from plant signaling under stress conditions, ultimately resulting in stomatal closure. Hsu et al. (2021) associated this plant signaling situation with increased abscisic acid production and the potassium efflux from guard cells, decreasing CO_2 entry into the substomatal chambers and decreasing water loss. This behavior was observed in the present experiment through the lower stomatal conductance, transpiration, and net CO_2 assimilation rate in plants grown under stress. Moreover, this effect is recurrent in water deficit studies, as observed by Galviz et al. (2021) in tomato (*Solanum lycopersicum* L.), by Kaya (2021) in pepper

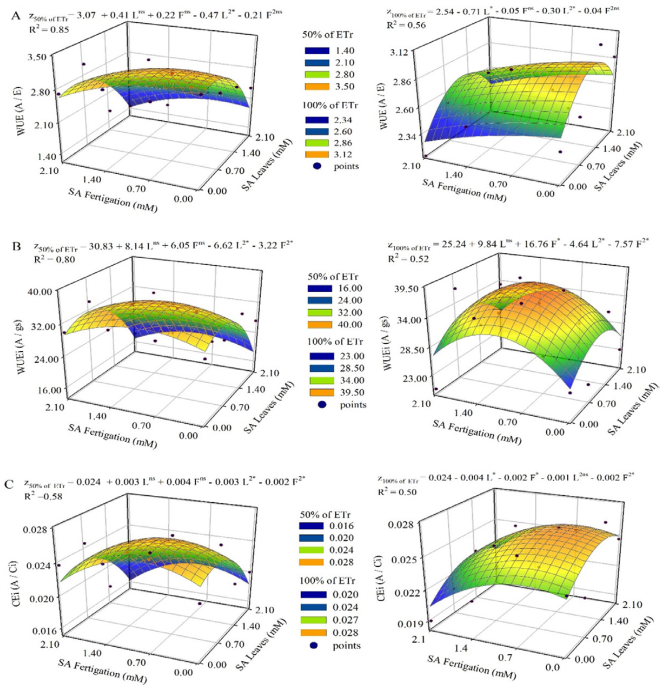

Figure 3. Water-use efficiency – WUE (A), intrinsic water-use efficiency - WUEi (B), and instantaneous carboxylation efficiency - CEi (C) of sour passion fruit (*Passiflora edulis*) as a function of SA application via foliar application and fertigation for two water replacement depths 75 days after sowing. ^{ns}, *, **, respectively not significant, significant at $p \leq 0.05$ and $p \leq 0.01$.

Table 3. Summary of the analysis of variance for plant height (PH, cm), stem diameter (SD, mm), and leaf area (LA, cm^2) of sour passion fruit plants subjected to different irrigation depths (I) and concentrations of salicylic acid applied via foliar application (L) and fertigation (F) 75 DAS.

Source of variation	DF	MS		
		PH	SD	LA
Irrigation (I)	1	4589.20**	9.66**	816683**
SA Leaves (L)	3	2798.32**	5.44**	289183**
SA Fertigation (F)	3	1608.50**	3.94**	213877**
I × L	3	69.41^{ns}	0.25^{ns}	25697**
I × F	3	483.63**	0.24**	79107**
L × F	9	213.51**	0.66**	71094**
I × L × F	9	209.21**	0.32*	16344**
Blocks	2	7.52^{ns}	0.058^{ns}	18896**
CV (%)		17.14	10.14	6.98
Mean		43.57	3.87	555.37

DF- degree of freedom; MS- mean square; R – regression; CV - coefficient of variation. ^{ns} not significant. * significant at $p \leq 0.05$. ** significant at $p \leq 0.01$.

(*Capsicum annuum* L.), and by Lozano-Montaña et al. (2021) in purple passion fruit (*Passiflora edulis*).

Salicylic acid mitigated the effects of stress on passion fruit. The initially greater stomatal opening is related to the action of SA in forming secondary metabolites, such as proline and glycine betaine, that act by reducing the root osmotic potential, favoring water and nutrient uptake from the soil (Silva et al., 2020; Ali, 2021). As a result, transpiration is also increased, corroborating the results obtained by Saheri et al. (2020), who evaluated the gas exchange of common purslane (*Portulaca oleracea* L.) and observed that the transpiration flux increased with the

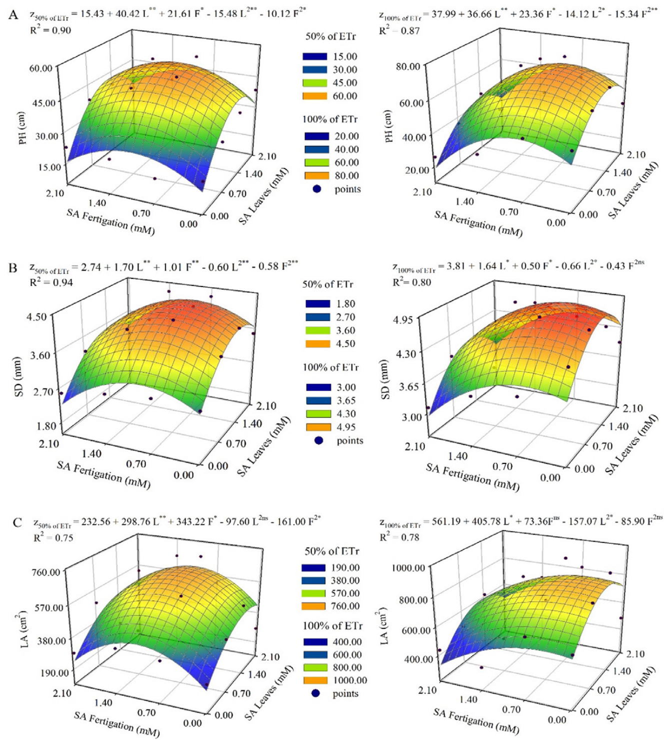

Figure 4. Plant height – PH (A), stem diameter - SD (B), and leaf area – LA (C) of passion fruit plants (*Passiflora edulis*) as a function of SA application via foliar application and fertigation for two water replacement depths 75 days after sowing. ^{ns}, *, **, respectively not significant, significant at p ≤0.05 and p ≤0.01.

greater stomatal opening in plants subjected to salicylic acid under water deficit conditions.

On the other hand, the increase in the CO_2 assimilation rate caused by SA application highlights the role of the studied phytohormone in maintaining the photosynthetic apparatus, probably due to the plant's photochemical activity and the CO_2 entry induced by stomatal opening (Kumara et al., 2021; Zafar et al., 2021). These findings were also observed by Lobato et al. (2021), who evaluated tomato plants under water deficit and found a positive correlation between the increase in photosynthesis and the increase in the antioxidant defense of plants after SA application, especially superoxide dismutase, catalase, peroxidase, and ascorbate peroxidase.

The photosynthetic efficiency of passion fruit is high under the water deficit condition caused by SA application. However, under high SA concentrations, the excessive accumulation of this metabolite decreases the photosynthetic efficiency, which is related to the pro-oxidant function of the phytohormone (Shadmehri and Khatiby, 2020). These effects were noticed when applying the highest SA concentration via fertigation, with the greater water loss through transpiration (Figure 1C), which, associated with the behavior of the CO_2 concentration in the intercellular spaces (Figure 1B), tends to explain the preference of the oxygenase function for RuBisCO enzyme sites to the detriment of carboxylase, leading to the loss of photosynthetic efficiency due to low leaf capacitance (Xiong and Nadal, 2020; Morales et al., 2020).

The benefits of SA application were also observed in plants subjected to irrigation with 100% ETr, promoting improvements ranging from stomatal opening to photosynthesis. However, the internal CO_2 concentration did not differ from plants without SA application, which could be related to carbon consumption during the photosynthetic activity, stabilizing CO_2 entry and exit in the substomatal chamber (Morales et al., 2020), which was confirmed by the increased carboxylation efficiency of passion fruit plants. In this case, SA probably acts early in maintaining the photosynthetic apparatus by maximizing the use of energy during the photochemical phase of photosynthesis (Chen et al., 2020).

Passion fruit growth is favored by SA application, corroborating photosynthetic gains and increasing meristematic activity and leaf expansion. These findings highlight the importance of salicylic acid in reducing the root osmotic potential, contributing to water and nutrient uptake and cell turgor, thus facilitating stem growth in diameter (Andrade et al., 2021; Zafar et al., 2021).

As observed for gas exchange, salicylic acid application at high concentrations via fertigation reduces the growth of sour passion fruit under ideal irrigation conditions, which is related to the phytotoxic effect of SA due to its low translocation to the shoots, unbalancing the enzymatic activity, damaging the photosystem, and affecting plant growth (Souri and Tohidloo, 2019; Shadmehri and Khatiby, 2020).

With regard to the highest values obtained by the growth variables under the two water conditions, the PH, LA, and SD variables decreased by 24.50, 23.23, and 11.87% when plants were irrigated with 50% ETr compared to the values obtained under 100% ETr. However, when comparing the highest mean values under the stress condition (50% ETr) to those obtained in plants without SA application and under ideal water conditions, there were increments of 40.43% in PH, 15.22% in SD, and 14.73% in LA. Therefore, these results highlight the positive influence of salicylic acid in mitigating water stress in passion fruit plants.

With regard to the forms of application, the highest values were observed when associating the application via foliar application and via fertigation. However, gains were also observed with the individual application of SA through one of the forms of application, with values 10% higher than those obtained in plants of the control treatment for stomatal opening, internal CO_2 concentration, net CO_2 assimilation rate, water-use efficiency, and growth of passion fruit plants. In this growth stage, the foliar application of SA led to the greatest differences compared to fertigation, with gains 55.02 and 31.78% higher for PH, 10.94 and 42.72% higher for LA, and 24.29 and 22.02% higher for SD under the respective conditions of 50 and 100% ETr. In addition, foliar application led to a greater tolerance margin to SA application, with favorable effects up to the concentration of 1.30 mM. On the other hand, for fertigation, this limit was 0.90 mM. These results corroborated those found by Souri and Tohidloo (2019) when studying tomato, whose best results were obtained when applying SA via foliar application compared to fertigation.

5. Conclusions

Water stress negatively affected the physiology and growth of sour passion fruit seedlings at 75 days after sowing. The application of salicylic acid, regardless of the form of application, attenuates the effects of water stress on gas exchange and growth of sour passion fruit, with the best results obtained when applying a concentration of 1.30 mM via leaf or 0.90 mM via fertirrigation. The combination of foliar application of AS and fertigation contributed to improve photosynthetic and growth parameters under water conditions of 50 and 100% of ETr. The foliar application of AS presents superior responses to the application via fertigation. These results reinforce the hypothesis that the attenuation of water stress by salicylic acid is related to the maintenance of gas exchange, which depends on the concentration and form of application, and studies testing combinations throughout the crop cycle become promising for advances in knowledge from the action of this phytohormone on abiotic stress.

Acknowledgements

The authors gratefully acknowledge the Post-Graduate Agricultural Engineering Program at Universidade Federal de Campina Grande. To the National Council for Scientific Development and Technology – CNPq (Proc. CNPq 429732/2018-0)) and Coordination of Improvement of Higher Education Personnel (CAPES, Code 001).

Reference

ALI, B., 2021. Salicylic acid: an efficient elicitor of secondary metabolite production in plants. *Biocatalysis and Agricultural Biotechnology*, vol. 31, pp. 101884. http://dx.doi.org/10.1016/j.bcab.2020.101884.

ANDRADE, W.L., MELO, A.S., MELO, Y.L., SÁ, F.V.S., ROCHA, M.M., OLIVEIRA, A.P.S. and FERNANDES JÚNIOR, P.I., 2021. *Bradyrhizobium* inoculation plus foliar application of salicylic acid mitigates water deficit effects on cowpea. *Journal of Plant Growth Regulation*, vol. 40, no. 2, pp. 656-667. http://dx.doi.org/10.1007/s00344-020-10130-3.

BERNARDO, S., SOARES, A.A., and MANTOVANI, E.C., 2006. *Manual de irrigação*. 8. ed. Viçosa: UFV. 625 p.

BISWAS, S., MISHRA, R. and BIST, A.S., 2021. Passion to profession: a review of Passion fruit processing. *Aptisi Transactions on Technopreneurship*, vol. 3, no. 1, pp. 48-57. http://dx.doi.org/10.34306/att.v3i1.143.

CAVALCANTE, L.F., SOUSA, G.G., GONDIM, S.C., FIGUEIREDO, F.L., CAVALCANTE, Í.H.L. and DINIZ, A.A., 2009. Crescimento inicial do maracujazeiro amarelo manejado em dois substratos irrigados com água salina. *Irriga*, vol. 14, no. 4, pp. 504-517. http://dx.doi.org/10.15809/irriga.2009v14n4p504-517.

CAVALCANTE, A.G., CAVALCANTE, L.F., SOUTO, A.G.D.L., CAVALCANTE, A.C.P., ARAÚJO, D.L.D., NASCIMENTO, A.P.P.D. and ZANUNCIO, J.C., 2020. Physiology and production of yellow passion fruit with hydroabsorbent polymer and different irrigation depths. *Revista Ceres*, vol. 67, no. 5, pp. 365-373. http://dx.doi.org/10.1590/0034-737x202067050004.

CHEN, Y.E., MAO, H.T., WU, N., DIN, M.U., KHAN, A., ZHANG, H.Y. and YUAN, S., 2020. Salicylic acid protects photosystem II by alleviating photoinhibition in *Arabidopsis thaliana* under high light. *International Journal of Molecular Sciences*, vol. 21, no. 4, pp. 1229. http://dx.doi.org/10.3390/ijms21041229. PMid:32059402.

DAMALAS, C.A. and KOUTROUBAS, S.D., 2022. Exogenous application of salicylic acid for regulation of sunflower growth under abiotic stress: a systematic review. *Biologia*, vol. 77, pp. 1685-1697. http://dx.doi.org/10.1007/s11756-022-01020-y.

DEKA, D., SINGH, A.K. and SINGH, A.K., 2018. Effect of drought stress on crop plants with special reference to drought avoidance and tolerance mechanisms: a review. *International Journal of Current Microbiology and Applied Sciences*, vol. 7, no. 9, pp. 2703-2721. http://dx.doi.org/10.20546/ijcmas.2018.709.336.

FREIRE, J.L.D.O., DIAS, T.J., CAVALCANTE, L.F., FERNANDES, P.D. and LIMA NETO, A.J.D., 2014. Rendimento quântico e trocas gasosas em maracujazeiro amarelo sob salinidade hídrica, biofertilização e cobertura morta. *Revista Ciência Agronômica*, vol. 45, no. 1, pp. 82-91. http://dx.doi.org/10.1590/S1806-66902014000100011.

GALVIZ, Y.C., BORTOLIN, G.S., GUIDORIZI, K.A., DEUNER, S., REOLON, F. and MORAES, D.M., 2021. Effectiveness of seed priming and soil drench with salicylic acid on tomato growth, physiological and biochemical responses to severe water deficit. *Journal of Soil Science and Plant Nutrition*, vol. 21, no. 3, pp. 2364-2377. http://dx.doi.org/10.1007/s42729-021-00528-7.

GHARBI, E., LUTTS, S., DAILLY, H. and QUINET, M., 2018. Comparison between the impacts of two different modes of salicylic acid application on tomato (*Solanum lycopersicum*) responses to salinity. *Plant Signaling & Behavior*, vol. 13, no. 5, pp.

e1469361. http://dx.doi.org/10.1080/15592324.2018.1469361. PMid:29944448.

HSU, P.K., DUBEAUX, G., TAKAHASHI, Y. and SCHROEDER, J.I., 2021. Signaling mechanisms in abscisic acid-mediated stomatal closure. *The Plant Journal*, vol. 105, no. 2, pp. 307-321. http://dx.doi.org/10.1111/tpj.15067. PMid:33145840.

INSTITUTO BRASILEIRO DE GEOGRAFIA E ESTATÍSTICA – IBGE, 2022 [viewed 4 April 2023]. *Sidra - Produção das lavouras temporárias e permanentes - Maracujá 2021* [online]. SIDRA/IBGE. Available from: https://sidra.ibge.gov.br/tabela/5457#resultado

KAYA, C., 2021. Nitrate reductase is required for salicylic acid-induced water stress tolerance of pepper by upraising the AsA-GSH pathway and glyoxalase system. *Physiologia Plantarum*, vol. 172, no. 2, pp. 351-370. http://dx.doi.org/10.1111/ppl.13153. PMid:32542778.

KULAK, M., JORRÍN-NOVO, J.V., ROMERO-RODRIGUEZ, M.C., YILDIRIM, E.D., GUL, F. and KARAMAN, S., 2021. Seed priming with salicylic acid on plant growth and essential oil composition in basil (*Ocimum basilicum* L.) plants grown under water stress conditions. *Industrial Crops and Products*, vol. 161, pp. 113235. http://dx.doi.org/10.1016/j.indcrop.2020.113235.

KUMARA, K., AMPITIYAWATTA, A.D., PADMAPERUMA, A., BENERAGAMA, C. and PING, X.Y., 2021. Alleviation of salt-induced adverse effects on gas exchange, photosynthetic pigments content and chloroplast ultrastructure in gerbera jamesonii l. by exogenous salicylic acid application. *Asian Journal of Research in Agriculture and Forestry*, vol. 7, no. 2, pp. 1-13. http://dx.doi.org/10.9734/ajraf/2021/v7i230123.

LIMA, G.S., NOBRE, R.G., GHEYI, H.R., SOARES, L.A.A. and SILVA, A.O., 2015. Produção da mamoneira cultivada com águas salinas e doses de nitrogênio. *Revista Ciência Agronômica*, vol. 46, no. 1, pp. 1-10. http://dx.doi.org/10.1590/S1806-66902015000100001.

LOBATO, A.K.D.S., BARBOSA, M.A.M., ALSAHLI, A.A., LIMA, E.J.A. and SILVA, B.R.S.D., 2021. Exogenous salicylic acid alleviates the negative impacts on production components, biomass and gas exchange in tomato plants under water deficit improving redox status and anatomical responses. *Physiologia Plantarum*, vol. 172, no. 2, pp. 869-884. http://dx.doi.org/10.1111/ppl.13329. PMid:33421143.

LOPES, M.F.Q., SILVA, T.I., NÓBREGA, J.S., SILVA, R.T., FIGUEIREDO, F.R.A. and BRUNO, R.D.L.A., 2019. Crescimento de *Erythrina velutina* Willd. submetida a estresse salino e aplicação de ácido salicílico. *Colloquium Agrariae*, vol. 15, no. 4, pp. 31-38. http://dx.doi.org/10.5747/ca.2019.v15.n4.a309.

LOZANO-MONTAÑA, P.A., SARMIENTO, F., MEJÍA-SEQUERA, L.M., ÁLVAREZ-FLÓREZ, F. and MELGAREJO, L.M., 2021. Physiological, biochemical and transcriptional responses of *Passiflora edulis* Sims f. *edulis* under progressive drought stress. *Scientia Horticulturae*, vol. 275, pp. 109655. http://dx.doi.org/10.1016/j.scienta.2020.109655.

MELETTI, L.M.M., 2011. Avanços na cultura do maracujá no Brasil. *Revista Brasileira de Fruticultura*, vol. 33, no. spe1, pp. 83-91. http://dx.doi.org/10.1590/S0100-29452011000500012.

MORALES, F., ANCÍN, M., FAKHET, D., GONZÁLEZ-TORRALBA, J., GÁMEZ, A.L., SEMINARIO, A., SOBA, D., MARIEM, S.B., GARRIGA, M. and ARANJUELO, I., 2020. Photosynthetic metabolism under stressful growth conditions as a bases for crop breeding and yield improvement. *Plants*, vol. 9, no. 1, pp. 88. http://dx.doi.org/10.3390/plants9010088. PMid:31936732.

NÓBREGA, J.S., FIGUEIREDO, F.R.A., SILVA, T.I., RIBEIRO, J.E.S., FÁTIMA, R.T., FERREIRA, J.T.A. and BRUNO, R.D.L.A., 2021. Salinidade da água e ácido salicílico no crescimento de plantas de tomate. *Research. Social Development*, vol. 10, no. 7, pp. 31-38. http://dx.doi.org/10.33448/rsd-v10i7.16630.

NOVAIS, R.F., NEVES, J.C.L. and BARROS, N.F., 1991. Ensaio em ambiente controlado. In: A.J. Oliveira, ed. *Métodos de pesquisa em fertilidade do solo*. Brasília: Embrapa-SEA, pp. 189-253.

PINHEIRO, F.W.A., LIMA, G.S., GHEYI, H.R., SOARES, L.A.A., OLIVEIRA, S.G. and SILVA, F.A., 2022a. Gas exchange and yellow passion fruit production under irrigation strategies using brackish water and potassium. *Revista Ciência Agronômica*, vol. 53, pp. 1-11. http://dx.doi.org/10.5935/1806-6690.20220009.

PINHEIRO, F.W.A., LIMA, G.S., GHEYI, H.R., SOARES, L.A.A., NOBRE, R.G., SILVA, L.A., LACERDA, C.F. and FERNANDES, P.D., 2022b. Quantum yield, chlorophyll, and cell damage in yellow passion fruit under irrigation strategies with brackish water and potassium. *Brazilian Journal of Biology = Revista Brasileira de Biologia*, vol. 82, pp. e265519. http://dx.doi.org/10.1590/1519-6984.265519.

R CORE TEAM, 2017 [viewed 4 January 2023]. *R: A language and environment for statistical computing* [online]. Vienna: R Foundation for Statistical Computing. Available from: https://www.R-project.org.

SAHERI, F., BARZIN, G., PISHKAR, L., BOOJAR, M.M.A., and BABAEEKHOU, L., 2020. Foliar spray of salicylic acid induces physiological and biochemical changes in purslane (Portulaca oleracea L.) under drought stress. *Biologia*, v. 75, no. 12, pp. 2189-2200. https://doi.org/10.2478/s11756-020-00571-2.

SANTOS, H.G., JACOMINE, P.K.T., ANJOS, L.H.C., OLIVEIRA, V.A., LUMBRERAS, J.F., COELHO, M.R. and CUNHA, T.J.F., 2018. *Sistema brasileiro de classificação de solos*. Brasília: Embrapa.

SHADMEHRI, A.A., and KHATIBY, A., 2020. The effect of salicylic acid on different plant processes–a review. *Cercetări Agronomice* în *Moldova*, vol. 53, no. 2, pp. 233-242. http://dx.doi.org/10.46909/cerce-2020-020.

SILVA, A.A.R., LIMA, G.S., AZEVEDO, C.A.V., VELOSO, L.L.S. and GHEYI, H.R., 2020. Salicylic acid as an attenuator of salt stress in soursop. *Revista Caatinga*, vol. 33, no. 4, pp. 1092-1101. http://dx.doi.org/10.1590/1983-21252020v33n424rc.

SOARES, L.A.A., FERNANDES, P.D., LIMA, G.S., SUASSUNA, J.F., BRITO, M.E.B. and SÁ, F.V.S., 2018. Growth and fiber quality of colored cotton under salinity management strategies. *Revista Brasileira de Engenharia Agrícola e Ambiental*, vol. 22, no. 5, pp. 332-337. http://dx.doi.org/10.1590/1807-1929/agriambi.v22n5p332-337.

SOHAG, A.A.M., TAHJIB-UL-ARIF, M., BRESTIC, M., AFRIN, S., SAKIL, M.A., HOSSAIN, M.T., HOSSAIN, M.A. and HOSSAIN, M.A., 2020. Exogenous salicylic acid and hydrogen peroxide attenuate drought stress in rice. *Plant, Soil and Environment*, vol. 66, no. 1, pp. 7-13. http://dx.doi.org/10.17221/472/2019-PSE.

SOURI, M.K. and TOHIDLOO, G., 2019. Effectiveness of different methods of salicylic acid application on growth characteristics of tomato seedlings under salinity. *Chemical and Biological Technologies in Agriculture*, vol. 6, no. 1, pp. 26. http://dx.doi.org/10.1186/s40538-019-0169-9.

SOUZA, P.U., LIMA, L.K.S., SOARES, T.L., JESUS, O.N., COELHO FILHO, M.A. and GIRARDI, E.A., 2018. Biometric, physiological and anatomical responses of *Passiflora* spp. to controlled water deficit. *Scientia Horticulturae*, vol. 229, pp. 77-90. http://dx.doi.org/10.1016/j.scienta.2017.10.019.

TEIXEIRA, P.C., DONAGEMMA, G.K., FONTANA, A. and TEIXEIRA, W.G., 2017. *Manual de métodos de análise de solo*. 3. ed. Brasília: Embrapa, 573p.

UNITED STATES DEPARTMENT OF AGRICULTURE – USDA, 2014. *Keys to soil taxonomy*. 12th ed. Washington: USDA, 360 p.

VIEIRA, M.L.C., COSTA, Z.P., VARANI, A.M., SADER, M.A., CAUZ-SANTOS, L.A., GIOPATTO, H.A. and PEDROSA-HARAND, A., 2022.

The passion fruit genome. In: M.A. CHAPMAN, ed. *Underutilised crop genomes*. Cham: Springer, pp. 131-148. http://dx.doi.org/10.1007/978-3-031-00848-1_8.

XIONG, D. and NADAL, M., 2020. Linking water relations and hydraulics with photosynthesis. *The Plant Journal*, vol. 101, no. 4, pp. 800-815. http://dx.doi.org/10.1111/tpj.14595. PMid:31677190.

ZAFAR, Z., RASHEED, F., ATIF, R.M., MAQSOOD, M. and GAILING, O., 2021. Salicylic acid-induced morpho-physiological and biochemical changes triggered water deficit tolerance in *Syzygium cumini* L saplings. *Forests*, vol. 12, no. 4, pp. 491. http://dx.doi.org/10.3390/f12040491.

ZOMBARDO, A., MICA, E., PUCCIONI, S., PERRIA, R., VALENTINI, P., MATTII, G.B. and STORCHI, P., 2020. Berry quality of grapevine under water stress as affected by rootstock–Scion interactions through gene expression regulation. *Agronomy*, vol. 10, no. 5, pp. 1-25. http://dx.doi.org/10.3390/agronomy10050680.