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Morphophysiology of passion-fruit seedlings in different substrates under different strategies of irrigation with brackish water

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ABSTRACT

The presence of salts in the irrigation waters of the semiarid region negatively affects the production of seedlings. Thus, substrates based on carbonized rice husk are an alternative for production in saline environments, associated with irrigation management strategies with brackish water. This study evaluated the physiological responses of yellow passion-fruit seedlings as a function of irrigation strategies with brackish water in different substrates. The study was developed at the Universidade da Integração Internacional da Lusofonia Afro-Brasileira, Redenção, Ceará. The design was entirely randomized, in a 2 × 5 factorial scheme with two substrate compositions (S1 - soil; S2 – sandy soil, sand and carbonized rice husk - 1:1:1 – volume basis) and five irrigation strategies with brackish water (IS1 - low salinity water during the entire cycle 1-65 days after sowing (DAS); and salt stress initiation at: IS2 - 11 DAS; IS3 - 16 DAS; IS4 - 21 DAS; IS5- 26 DAS), with five repetitions. Soil as a substrate was efficient for photosynthesis and chlorophyll. It also provided better results for transpiration, stomatal conductance and leaf area regardless of the strategy. Internal carbon concentration and water use efficiency were higher in soil associated with saline stress at 11, 16 and 21 DAS. Under the same substrate, the stem diameter of the seedlings was greater when irrigated with lower salinity during the phase. The substrate composed of carbonized rice is associated with greater seedling height and increased leaf temperature.

Keywords: plant physiology, *Passiflora edulis*, salinity, substrate.

Morfofisiologia de mudas de maracujazeiro sob diferentes estratégias de irrigação com água salobra em diferentes substratos

RESUMO

A região semiárida sofre com a presença de sais nas águas disponíveis para irrigação, afetando a produção de mudas. Assim, substratos a base de casca de arroz carbonizada surgem como alternativa para produção em ambientes salinos, associados a estratégias de manejo da irrigação com águas salobras. Objetivou-se avaliar as respostas fisiológicas e o crescimento de mudas de maracujazeiro amarelo em função de estratégias de irrigação com água salobra em diferentes substratos. O estudo foi desenvolvido na Universidade da Integração Internacional da Lusofonia Afro-Brasileira, Redenção, Ceará. O delineamento foi inteiramente casualizado, em esquema fatorial 2 × 5 com duas composições de substratos (S1 - solo; S2 - areia, arisco e casca de arroz carbonizada - 1:1:1 – base de volume) e cinco estratégias de irrigação (IS1 - água de baixa salinidade durante todo o ciclo 1-60 dias após a semeadura (DAS); e início do estresse salino em: IS2 - 11 DAS; IS3 - 16 DAS; IS4 - 21 DAS; IS5- 26 DAS), com cinco repetições. O solo como substrato foi eficiente para a fotossíntese e clorofila. Também proporcionou melhores resultados para transpiração, condutância estomática e área foliar independente da estratégia. A concentração interna de carbono e a eficiência do uso da água foram superiores no solo associado ao estresse salino aos 11, 16 e 21 DAS. Sob o mesmo substrato, o diâmetro caulinar foi maior quando irrigadas com menor salinidade durante a fase. O número de folhas e clorofila foram maiores com o uso de água de menor salinidade. O substrato composto por arroz carbonizada possibilita maior desempenho em altura de mudas e aumentou a temperatura foliar.

Palavras-chave: fisiologia vegetal, *Passiflora edulis*, salinidade, substrato.

1.INTRODUCTION

Passion fruit (*Passiflora edulis*) is an important part of Brazilian fruticulture, grown in more than 90% of the backyards of Brazilian producers (Pinheiro *et al.*, 2022). It is widely cultivated in tropical and semi-tropical regions, and its fruits are used for consumption, either fresh or processed (Lima *et al.*, 2020).

Arid and semiarid regions suffer from problems related to the scarcity of water resources, both in terms of quantity and quality (Dias *et al.*, 2019). Most waters of these regions present large amounts of salts in their composition, and are often the only water source available for use in agriculture (Goes *et al.*, 2019; Freire *et al.*, 2021).

The salts present in irrigation water can cause accumulation of sodium (Na^+) and chloride (Cl^-) ions, and their excess affects the growth, development, and yield of crops (Bezerra *et al.*, 2018; Dias *et al.*, 2019; Sousa *et al.*, 2021). Irrigation with brackish water also causes nutritional and physiological disturbances in plants, including reduced nitrogen and potassium uptake, stomatal conductance, and photosynthesis (Rodrigues *et al.*, 2021; Sousa *et al.*, 2022a).

However, one of the alternatives to mitigate the effects of irrigation water salinity is the use of alternative substrates (Mendonça *et al.*, 2021), such as carbonized rice husk which is an important nutritional source to produce good quality seedlings. In other words, it can help to maintain production, improve quality and reduce costs (Silva *et al.*, 2019). Silva Junior *et al.* (2020) verified higher performance of watermelon seedlings containing carbonized rice husk as substrate under salt stress.

In this sense, the objective of this study was to evaluate the physiological responses of yellow passion fruit seedlings as a function of irrigation strategies with brackish water in different substrates.

2. MATERIAL AND METHODS

The study was developed in the period from September to November 2020 in a protected environment with black shaded roof with 50% shading, in the experimental area of the Auroras Seedling Production Unit (UPMA), belonging to the Universidade da Integração Internacional da Lusofonia Afro-Brasileira (UNILAB), Redenção, Ceará.

The region's climate is of the BSh' type, predominating very hot temperatures and rainfall in the summer and autumn seasons (Alvares *et al.*, 2013). Figure 1 presents the climatic data during the conduct of the experiment.

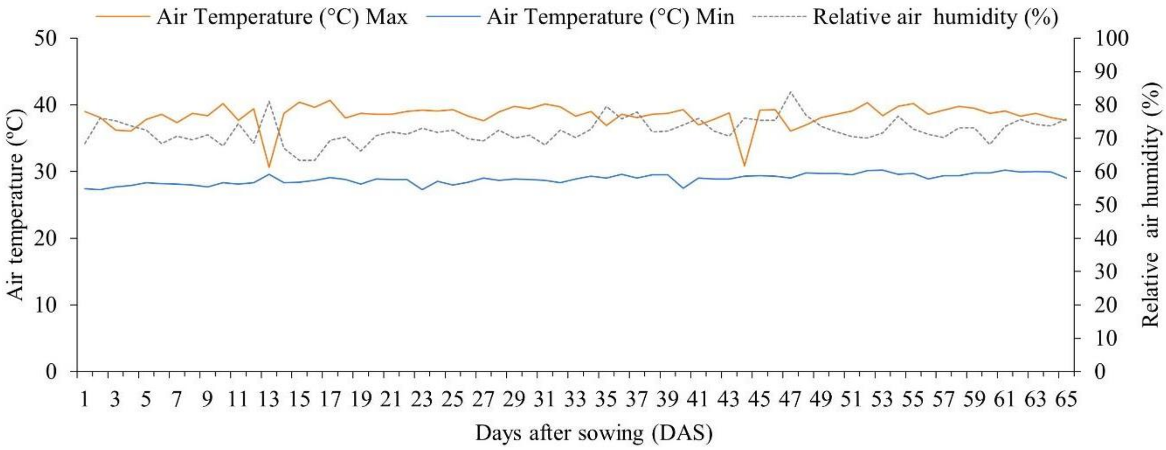

Figure 1. Average values of temperature and relative humidity during the experimental period.

The experimental design was entirely randomized, in a 2 × 5 factorial scheme, with five repetitions, corresponding to 10 treatments, totaling 50 experimental units. Referring to two substrate compositions (S1 - soil; S2 – sandy soil, sand and carbonized rice husk - 1:1:1 – volume basis) and five irrigation strategies with brackish water (3,0 dS m^{-1}) (IS1 - irrigation with lower salinity water throughout the seedlings establishment (65 days after sowing (DAS)); IS2 = beginning of salt stress at 11 DAS; IS3 = beginning of salt stress at 16 DAS; IS4 = beginning of salt stress at 21 DAS and IS5 = beginning of salt stress at 26 DAS), before the start of irrigation with brackish water in the respective treatments, the plants were irrigated with water of lower salinity (0.3 dS m^{-1}).

The seeds used were the Topssed® cv. yellow round, with 75% germination and 99% purity. Sowing was performed in black polyethylene bags (19 × 21 cm). Three seeds were sown per bag, two centimeters deep. Thinning was performed 10 DAS, leaving only one plant per bag.

The soil used as substrate 1 (S1) is classified as an Ultisol. The carbonized rice husk used for S2 came from farmers in the Maciço de Baturité region. Samples of the respective substrates were sent to the laboratory of the Universidade Federal do Ceará (UFC) to perform analyses and determine chemical attributes (Table 1), following the methodology of Teixeira *et al.* (2017).

Table 1. Chemical characteristics of the substrates used.

Substrates	OM	N	P	Ca	K	Mg	Na	H + Al	SB	CEC	V	ECse	pH
	(g kg^{-1})		(mg kg^{-1})	(cmol$_c$ kg^{-1})							(%)	(dS m^{-1})	H_2O
S1	4	0.24	2	2.50	0.06	0.30	0.57	0.33	3.43	3.76	91	0.37	7.6
S2	10	0.65	78	4.50	0.56	0.60	0.17	0.99	5.83	6.82	85	0.76	7.2

OM – organic matter; SB – Sum of bases (Ca^{2+} + Mg^{2+} + Na^+ + K^+); CEC – Cation exchange capacity - [Ca^{2+} + Mg^{2+} + Na^+ + K^+ + (H^+ + Al^{3+})]; V – Base saturation - (Ca^{2+} + Mg^{2+} + Na^+ + K+/ CEC) × 100; ECse – Electrical conductivity of the saturation extract of the substrate; S1 - soil; S2 – sandy soil, sand and carbonized rice husk - 1:1:1.

The lowest salinity water (0.3 dS m^{-1}) came from the local water supply. The 3.0 dS m^{-1} saline solution was prepared by adding sodium chloride (NaCl), calcium chloride ($CaCl_2.H_2O$)

and magnesium chloride ($MgCl_2.6H_2O$), in the proportion of 7:2:1, respectively (Medeiros, 1992), to the supply water, in order to obtain the desired electrical conductivity, according to the ratio between ECw and its concentration ($mmol_c L^{-1} = EC \times 10$), according to Richards (1954). The highest salt level of the water tested in this research is commonly observed in the semi-arid region of Northeast Brazil. It was therefore selected to evaluate the seedlings' development in different substrates under conditions above the threshold salinity, which is 1.3 dS m^{-1} for passion fruit (Ayers and Westcot, 1999).

Irrigation was performed daily, obeying the drainage lysimeter principle through water balance, keeping the soil at field capacity. A leaching blade of 15% was used every two days, according to Ayers and Westcot (1999). The volume of water to be applied was determined according to Equation 1:

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

Where:

VI - Volume of water to be applied in the irrigation event (mL);

Vp - Volume of water applied in the previous irrigation event (mL);

Vd - Volume of water drained (mL); and,

LF - Leaching fraction of 0.15;

At 65 DAS, the seedlings had a height between 15 and 30 centimeters, which was considered suitable for transplanting according to Gontijo (2017).

The physiological variables analyzed were: CO_2 assimilation rate (A, µmol CO_2 m^{-2} s^{-1}), stomatal conductance (gs, mol m^{-1} s^{-1}), transpiration (E, mmol m^{-2} s^{-1}), internal carbon concentration (Ci, µmol CO_2 mol^{-1}), instantaneous water use efficiency (WUEi - through the ratio between A/E, [(µmol CO_2 m^{-2} s^{-1}) (mol H_2O m^{-2} s^{-1})$^{-1}$]), leaf temperature (LT, ºC), using the IRGA infrared gas analyzer (LI 6400 XT from LICOR). The measurements were made in an open system, with air flow of 300 mL min^{-1} between 09h:00 and 11h:00 hours under natural conditions of air temperature and CO_2 concentration, on fully expanded leaves. On the same leaves, relative chlorophyll index (RCI, SPAD) measurements were performed using a non-destructive method with a portable meter (SPAD - 502 Plus, Minolta, Japan).

In the same period (65 DAS), the following biometric variables were evaluated: seedling height (PH, cm) measured from the base to the apex of the plant with a graduated ruler; stem diameter (SD, mm), measured 2 cm above the substrate with a digital caliper; number of leaves (NL) by direct counting of fully expanded leaves; and leaf area (LA, cm²), using an area integrator (Area meter, LI-3100, Li-Cor, Inc. Lincoln, NE, USA).

The data obtained were submitted to the Kolmogorov-Smirnov test ($p \leq 0.05$) to assess normality. After checking normality, the analysis of variance was performed using the F test, and when significant, the data were subjected to the Tukey test ($p \leq 0.05$), using Assistat 7.7 Beta software (Silva and Azevedo, 2016).

3. RESULTS AND DISCUSSION

According to Table 2, the variables CO_2 assimilation rate and relative chlorophyll index were influenced by the factors substrate and strategy, in isolation ($p \leq 0.01$). The interaction of the factors was significant for the variables of transpiration, stomatal conductance, internal carbon concentration, leaf temperature, and instantaneous water-use efficiency.

Table 2. Summary of analysis of variance for CO_2 assimilation rate (A), transpiration (E), stomatal conductance (gs), internal carbon concentration (Ci), relative chlorophyll index (RCI), leaf temperature (LT), instantaneous water use efficiency (WUEi) in passion fruit seedlings under different substrates (S) and irrigation strategies with brackish water (IS).

Source of variation	DF	Mean square						
		A	E	gs	Ci	RCI	LT	WUEi
Substrates (S)	1	31.43**	7.68**	0.063**	5964.30**	1858.89**	9.74**	0.0002^{ns}
Irrigation strategies (IS)	4	2.35*	1.08**	0.008**	3355.95**	47.34*	0.51**	0.11**
S × IS	4	0.73^{ns}	0.64**	0.004*	3865.05**	33.58^{ns}	0.77**	0.11**
Residue	20	0.74	0.11	0.001	150.1	14.07	0.08	0.01
CV (%)		24.63	21.52	22.69	4.04	10.99	0.86	23.05

DF – Degrees of freedom; CV – Coefficient of variation; *, **, ns -Significant at $p \leq 0.05$ and $p \leq 0.01$, and not significant, respectively, by F test.

It was observed that the substrate composed of sand, sandy soil and carbonized rice husk (S2) showed the lowest values of CO_2 assimilation rate (2.48 µmol CO_2 m^{-2} s^{-1}) differing statistically from the treatments with soil substrate (4.53 µmol CO_2 m^{-2} s^{-1}), with a reduction of 45.26% (Figure 2A).

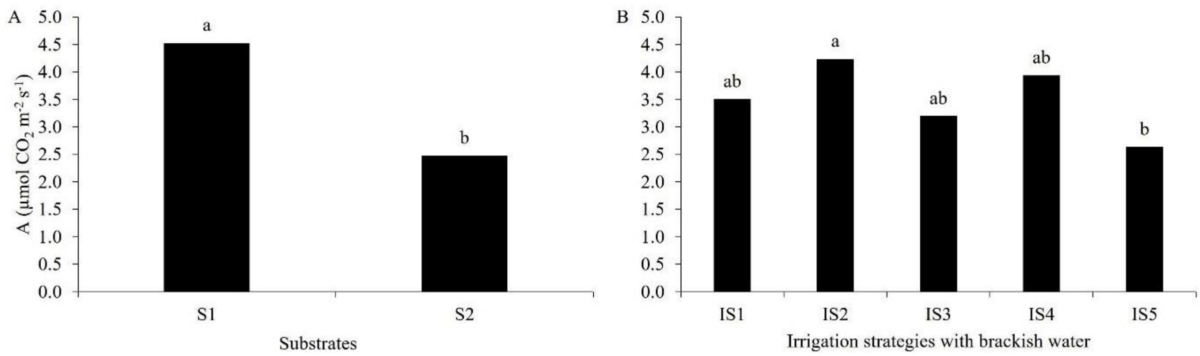

Figure 2. CO2 assimilation rate of yellow passion-fruit seedlings as a function of substrates (A) and irrigation strategies with brackish water (B). In Figure A, S1- soil; S2- sandy soil, sand and carbonized rice husk (1:1:1 – volume basis). In Figure B, IS1- irrigation with lower salinity water throughout the seedling establishment (65 days after sowing (DAS)); IS2 = onset of salt stress at 11 DAS; IS3 = onset of salt stress at 16 DAS; IS4 = onset of salt stress at 21 DAS and IS5 = onset of salt stress at 26 DAS. Lowercase letters compare means by Tukey test ($p \leq 0.05$).

The reduction in CO_2 assimilation rate presented in S2 may be related to the higher CEes (Table 1). This result may be associated with osmotic and toxic effects that affect net CO_2 assimilation, inhibiting leaf expansion and consequently the area destined to the photosynthetic process. However, these results are divergent from those found by Sousa *et al.* (2021), working with the use of substrates in the culture of cowpea, in which the treatments with carbonized rice husk showed higher values of photosynthesis than the treatment with soil.

For the irrigation strategies, except IS5, the others were statistically superior and similar (Figure 1B). That is, the seedlings irrigated in strategies IS1, IS2, IS3 and IS4, possibly induced an acclimation adopted by the plants as an adaptation mechanism to salt stress (Guimarães *et al.*, 2019).

Lima *et al.* (2020) at 200 days after transplanting observed discordant results to the present study working with passion fruit plants: the authors observed higher photosynthetic rates in plants without salt stress (9.8 µmol m^{-2} s^{-1}). Dantas *et al.* (2021) also observed different results in acerola plants; the authors obtained a reduction of 43.05% in photosynthetic rate in plants that were irrigated with water of 4.0 dS m^{-1}.

As shown in Table 3, regarding transpiration in passion fruit seedlings, Substrate 1 associated with strategies IS1, IS2, IS3 and IS4 were statistically superior, where the highest mean values were observed in IS1 and IS4 (2.46 and 2.45 mmol m^{-2} s^{-1}, respectively).

Table 3. Mean values of transpiration (E), stomatal conductance (gs) and internal carbon concentration (Ci) as a function of the interaction between factors, substrates and irrigation strategies with brackish water.

Irrigation strategies with brackish water	E (mmol m^{-2} s^{-1})		gs (mol m^{-1} s^{-1})		Ci (µmol CO_2 mol^{-1})	
	S1	S2	S1	S2	S1	S2
IS1	2.46 aA	1.68 aB	0.18 aA	0.12 aB	327.00 aA	342.50 aA
IS2	2.23 aA	0.65 bB	0.17 aA	0.03 bB	312.00 abA	247.50 cB
IS3	1.84 abA	0.28 cB	0.13 abA	0.01 bB	326.50 aA	234.00 cB
IS4	2.45 aA	1.31 abA	0.20 aA	0.07 abB	328.50 aA	303.00 bB
IS5	1.25 bA	1.25 abA	0.07 bA	0.06 abA	293.00 bB	319.00 abA

S1- soil; S2- sandy soil, sand and carbonized rice husk (1:1:1 – volume basis); IS1- irrigation with lower salinity water throughout the seedling establishment (65 days after sowing (DAS)); IS2 = onset of salt stress at 11 DAS; IS3 = onset of salt stress at 16 DAS; IS4 = onset of salt stress at 21 DAS and IS5 = onset of salt stress at 26 DAS. Uppercase letters compare the means in between the substrates in each irrigations strategies, and lowercase letters compare the means of different irrigations strategies in each substrate by the Tukey test ($p \leq 0.05$).

The reduction of transpiration in the treatments with substrate composed of carbonized rice husk and subjected to salinity conditions for a longer period may be related to the increase in the electrical conductivity of the soil saturation extract. This reduction in transpiration rate in the treatments submitted to a longer period of stress can be justified by the reduction in stomatal conductance, as presented in Table 4, which reduces transpiration and increases the internal temperature of the leaf (Table 5) as a response to water loss (Freire *et al.*, 2021).

Table 4. Mean values of leaf internal temperature (LT) and water use efficiency (WUEi) as a function of the interaction between factors, substrates and irrigation strategies with brackish water.

Irrigation strategies with brackish water	LT (ºC)		WUEi [(µmol CO_2 m^{-2} s^{-1}) (mol H_2O m^{-2} s^{-1})$^{-1}$])	
	S1	S2	S1	S2
IS1	32.30 bB	33.45 aA	0.53 aB	0.72 aA
IS2	32.30 bB	33.65 aA	0.39 aA	0.23 cdA
IS3	32.00 bB	33.60 aA	0.49 aA	0.12 dB
IS4	31.90 bB	33.50 aA	0.49 aA	0.44 bcA
IS5	33.50 aA	33.40 aA	0.35 aB	0.70 abA

S1- soil; S2- sandy soil, sand and carbonized rice husk (1:1:1 – volume basis); IS1- irrigation with lower salinity water throughout the seedling establishment (65 days after sowing (DAS)); IS2 = onset of salt stress at 11 DAS; IS3 = onset of salt stress at 16 DAS; IS4 = onset of salt stress at 21 DAS and IS5 = onset of salt stress at 26 DAS. Uppercase letters compare the means in between the substrates in each irrigations strategies, and lowercase letters compare the means of different irrigations strategies in each substrate by the Tukey test ($p \leq 0.05$).

Table 5. Summary of analysis of variance for seedling height (SH), leaf area (LA), number leaves (NL), and stem diameter (SD) in passion fruit seedlings under different substrates (S) and irrigation strategies with brackish water (IS).

Source of variation	DF	Mean square			
		SH	LA	SD	NL
Substrates (S)	1	46.27*	8774.59**	0.40^{ns}	2.42^{ns}
Irrigation strategies (IS)	4	27.30^{ns}	1.92^{ns}	0.86**	6.55**
S × IE	4	15.63^{ns}	3.95**	0.62*	0.57^{ns}
Residue	20	10.78	421.49	0.21	1.34
CV (%)		23.47	22.51	17.30	15.86

DF – Degrees of freedom; CV – Coefficient of variation; *, **, ns -Significant at $p \leq 0.05$ and $p \leq 0.01$, and not significant, respectively, by F test.

A similar trend was observed by Lima *et al.* (2020), who verified reductions in transpiration values in yellow passion fruit plants irrigated with higher conductivity water compared to plants irrigated with lower conductivity water. Similarly, Lessa *et al.* (2022) at 60 DAS, observed that passion-fruit seedlings irrigated with water of 0.3 dS m^{-1} obtained the highest values of transpiration, differing statistically from water of 3.0 dS m^{-1}.

The results found for transpiration in this study differ from the results obtained by Sousa *et al.* (2021) working with the use of substrates in the culture of cowpea bean, where they observed that the treatments that had carbonized rice husk in their composition provided higher values of transpiration compared to the control treatment (soil).

For stomatal conductance (Table 3), strategies IS1, IS2, IS3 and IS4 together with S1 were statistically superior to S2. The lower stomatal conductance values observed for the substrate treatments may be related to the higher ECse compared to Substrate 2 composed of soil (Table 1). Gomes *et al.* (2018), describe that the high content of Na can cause a reduction in the osmotic potential of the soil causing the plants to have difficulty in absorbing water and, consequently, decreases the gs.

Pinheiro *et al.* (2022) working with passion-fruit culture, observed that irrigation with water of higher electrical conductivity (4.0 dS m^{-1}) negatively affected the stomatal conductance of the plants. Sousa *et al.* (2021) evaluating the use of substrates in the culture of cowpea, obtained results contrary to the present study, where the treatments with soil substrate showed lower values of stomatal conductance compared to treatments with rice husk in their composition.

The results obtained for internal carbon concentration (Table 3) show that there was no statistical difference in IS1, but in IS2, 3 and 4 in S1 were statistically superior to S2, while in IS5, S2 was superior to S1. The superiority in IS2, IS3 and IS4 may be related to lower ECse and consequently greater osmotic adjustment provided by S1, the plants in these treatments reduced to a lesser extent the stomatal opening and subsequently the internal CO_2 concentration (Oliveira *et al.*, 2017; Pereira Filho *et al.*, 2017; Park *et al.*, 2021). Similar results to the present study for the reduction of internal CO_2 concentration, were observed by Bezerra *et al.* (2018), in guava plants under salt stress.

The results obtained in relation to the relative chlorophyll index for the substrates evaluated (Figure 3A) showed that the substrate that had carbonized rice husk in its composition showed a lower index compared to the substrate composed only by soil, with a reduction of 37.47%.

The superiority of S1 for CO_2 assimilation rate (Figure 1A) was reflected in the chlorophyll index. That is, the lower EC provided greater absorption and transmission of light energy. Sousa *et al.* (2022b) working with the culture of zucchini under salt stress, observed linear reductions caused by increasing electrical conductivity of irrigation water, with 14.23% of difference between the lowest and highest conductivity.

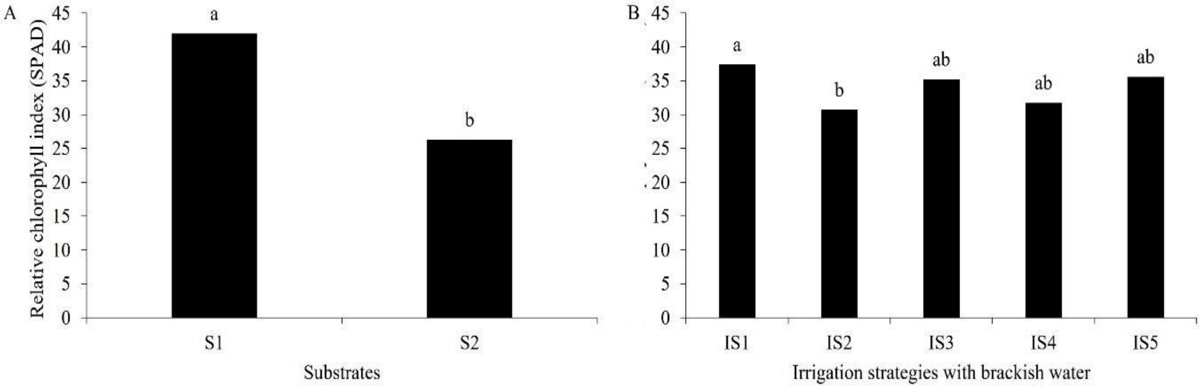

Figure 3. Relative chlorophyll index in yellow passion-fruit seedlings as a function of substrates (A) and irrigation strategies with brackish water (B). In Figure A, S1- soil; S2- sandy soil, sand and carbonized rice husk (1:1:1 – volume basis). In Figure B, IS1- irrigation with lower salinity water throughout the seedling establishment (65 days after sowing (DAS)); IS2 = onset of salt stress at 11 DAS; IS3 = onset of salt stress at 16 DAS; IS4 = onset of salt stress at 21 DAS and IS5 = onset of salt stress at 26 DAS. Lowercase letters compare means by Tukey test ($p \leq 0.05$).

For irrigation strategies (Figure 3B), the results indicate that strategies IS1, IS3, IS4 and IS5 were statistically superior to IS2. When plants are subjected to salt stress conditions, salts usually impair photosynthetic activity and consequently affect chlorophyll (Figueiredo *et al.*, 2019). Oliveira *et al.* (2018) observed reductions in chlorophyll values for cowpea crop as a function of water salinity.

According to Table 4, the mean internal leaf temperatures were higher for treatments IS1, IS2, IS3, IS4 and IS5 in S1. The substrate S2 presented higher ECse than S1, and when associated with irrigation water of higher electrical conductivity it provided higher values of internal temperature of the leaf. This behavior can be explained by water stress potentiated by the presence of salts in the water, causing stomatal closure and reduction of transpiration, culminating in increased temperature inside the plant, as observed by Rodrigues *et al.* (2021), in studies on corn cultivation, where they state that salt stress can reduce the amount of water transpired or absorbed.

Evaluating the bean crop irrigated with saline water grown in soil, Freire *et al.* (2021) found similar results to the present study, where the increase in salt concentration in irrigation water contributed to the increase in leaf temperature of the varieties.

In Table 4, the substrate containing sand, sandy soil and carbonized rice husk (S2) associated with IS1 (irrigation with low salinity water during the entire cycle) and IS5 (beginning of salt stress at 26 DAS) presented the highest values of water-use efficiency, (0.72 and 0.70 µmol CO_2 m^{-2} s^{-1}(µmol m^{-2} $s^{-1})^{-1}$), respectively). The same substrate associated with irrigation Strategy 2 (onset of salt stress at 11 DAS) and irrigation Strategy 3 (onset of salt stress at 16 DAS) showed the lowest values, corresponding to 0.23 and 0.12 µmol m^{-2} s^{-1}(mol H_2O m^{-2} $s^{-1})^{-1}$, respectively.

Under saline conditions, the electrical conductivity of the substrate composed of sand, sandy soil and carbonized rice husk may have been increased due to the use of the strategies used for a longer period of time, which would increase the salts in the substrate (IS2 to IS4), implying the reduction of the osmotic potential of the soil solution and consequently, in the reduction of water uptake by the roots, resulting in low efficiencies, precisely due to the reduction of stomata opening, with a consequent decrease in CO_2 uptake from the environment and in the photosynthesis rate in these same treatments, which implies less efficient use of water for seedlings under stress (Taiz *et al.*, 2017; Figueiredo *et al.*, 2019).

Figueiredo *et al.* (2019) found reductions in instantaneous water-use efficiency in mulungu culture as the electrical conductivity of the irrigation water to which the plants were subjected increased. Similarly, Pinheiro *et al.* (2022) obtained reductions in the water-use efficiency of passion fruit plants under salt stress.

According to Table 5, the variables seedling height and number leaves were influenced by the substrate factors and irrigation strategy with brackish water separately, respectively ($p \leq 0.05$). The interaction of factors was significant for the variables of leaf area and stem diameter $p \leq 0.01$.

The seedlings grown under the substrate composed of sandy soil, sand and carbonized rice husk showed higher height (14.96 cm), statistically different from the plants with the soil, with an increase of 14.81% (Figure 4). Higher performance by this substrate is possibly related to the higher input of nutrients such as nitrogen (Table 1), which positively influences plant growth (Prado, 2020).

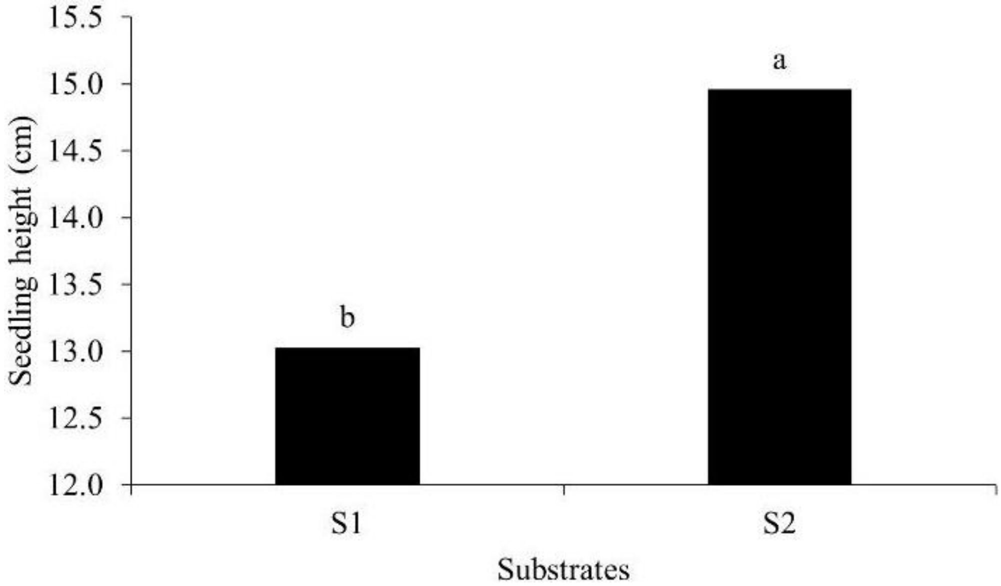

Figure 4. Seedling height in yellow passion-fruit as a function of substrates. S1- soil; S2- sandy soil, sand and carbonized rice husk (1:1:1 – volume basis); Lowercase letters compare means by Tukey test ($p \leq 0.05$).

Similar results to those found in present study were observed by Mendonça *et al.* (2021) working with bagana-carnauba based substrate on passion fruit seedlings. Similarly, Siqueira *et al.* (2020), evaluating alternative substrate (PuroHumus® + 25% sawdust + 50% soil), also detected an effect similar to that of the present study at 60 DAS.

The highest averages of leaf area in passion fruit seedlings were obtained using soilsubstrate, regardless of the irrigation strategies used (Table 6). The highest values were found in treatments with soil associated with IS4 and IS1 strategies (82.51 and 82.12 cm^2, respectively). The lowest value was found in the treatment composed of sand, skim and carbonized rice husk together with the IS4 strategy (16.89 cm^2).

The lower value of the leaf area present in the S2IS4 treatment can be explained by the period of exposure of the plants to irrigation with brackish water, that is, the plants may have developed an adaptation mechanism through the reduction of the leaf area, aiming to minimize transpiration and consequently maintain a high water potential inside the plants. In addition, the substrate composed of carbonized rice husk presents higher electrical conductivity of the saturation extract compared to the substrate with soil. These results corroborate the results found by Lessa *et al.* (2022) at 60 DAS, where irrigation with water of 3.0 dS m^{-1} in relation to the control (0.3 dS m^{-1}) negatively affected the leaf area of passion fruit seedlings.

Table 6. Mean values of leaf area (LA) and stem diameter (SD) as a function of the interaction between factors, substrates and irrigation strategies with brackish water.

Irrigation strategies with brackish water	LA (cm^2)		SD (mm)	
	S1	S2	S1	S2
IS1	82.12 aA	44.53 abB	2.86 aA	2.86 abA
IS2	62.78 aA	56.02 aA	2.28 bA	2.58 abA
IS3	76.57 aA	71.33 aA	2.32 bB	2.98 abA
IS4	82.51 aA	16.89 bB	2.46 abA	2.20 bA
IS5	74.63 aA	57.37 aA	2.48 abA	3.10 aA

S1- soil; S2- sandy soil, sand and carbonized rice husk (1:1:1 – volume basis); IS1- irrigation with lower salinity water throughout the seedling establishment (65 days after sowing (DAS)); IS2 = onset of salt stress at 11 DAS; IS3 = onset of salt stress at 16 DAS; IS4 = onset of salt stress at 21 DAS and IS5 = onset of salt stress at 26 DAS. Uppercase letters compare the means in between the substrates in each irrigations strategies, and lowercase letters compare the means of different irrigations strategies in each substrate by the Tukey test ($p \leq 0.05$).

It can be seen in Table 6 that the largest stem diameters were obtained in treatments with soil associated with the IS1 strategy and in the substrate composed of sand, sand and carbonized rice husk associated with the IS5 strategy (3.28 and 3.10 mm), respectively. However, the lowest values were found in treatments with soil associated with the IS2 strategy and in the substrate composed of sand, sand and carbonized rice husk associated with IS4 strategy (2.28 and 2.20 mm), respectively.

The large amount of salts absorbed by plants, as a result of progressive irrigation with brackish water, results in the inhibition of photoassimilates and consequently lower SD. Deleterious effects caused by salinity on the stem diameter were also found by Lima *et al.* (2021) working with sour passion fruit seedlings. These same authors recorded a decrease of 28.80% from the highest to the lowest saline level (3.2 dS m^{-1}).

Plants under the IS1 strategy (irrigation with water of lower salinity throughout the seedling formation period) presented the highest value (8.5) in the number of leaves, statistically different from the IS3 and IS4 strategies, which presented the lowest values (6.6) (Figure 5).

The reduction in the number of leaves in treatments with IS3 and IS4 strategies can be related to the period of days that these treatments received irrigation with brackish water, with greater accumulation of salts. It should also be noted that leaves are sensitive to salts, causing their reduction in plants as a form of adaptation. Likewise, Lessa *et al.* (2022), working with irrigation with brackish water during the passion fruit seedling phase (60 DAS), also recorded fewer leaves.

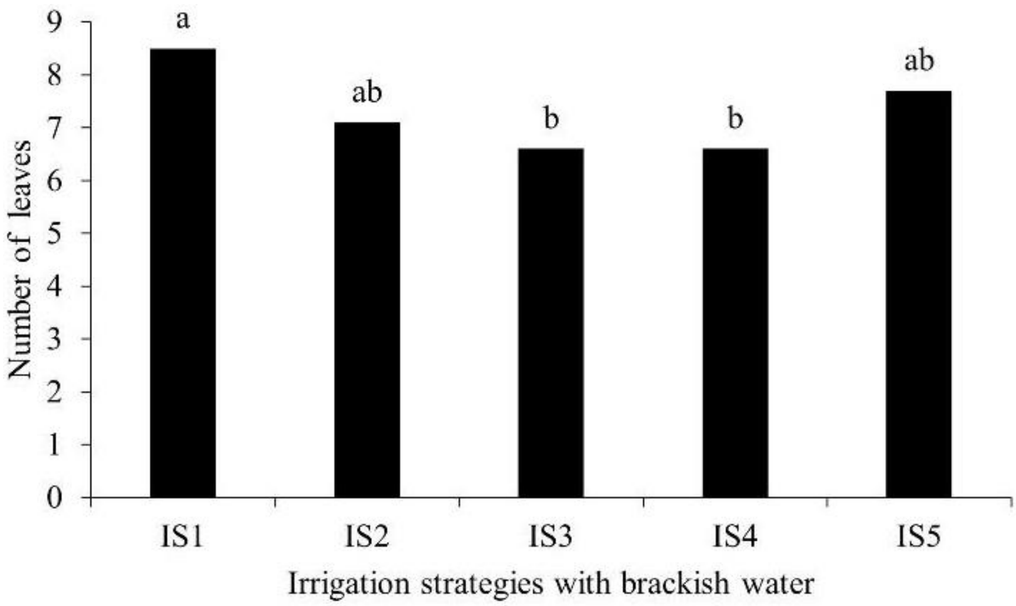

Figure 5. Number of leaves in yellow passion-fruit seedlings as a function of irrigation strategies with brackish water. IS1- irrigation with lower salinity water throughout the seedling establishment (65 days after sowing (DAS)); IS2 = onset of salt stress at 11 DAS; IS3 = onset of salt stress at 16 DAS; IS4 = onset of salt stress at 21 DAS and IS5 = onset of salt stress at 26 DAS; Lowercase letters compare means by Tukey test ($p \leq 0.05$).

4. CONCLUSIONS

The substrate composed of soil was more efficient for photosynthesis and chlorophyll. It also provided the best results for transpiration, stomatal conductance, and leaf area.

Internal carbon concentration and water use efficiency were higher in the substrate composed of soil associated with the onset of saline stress at 11, 16 and 21 DAS. Under the same substrate, the stem diameter of the seedlings was greater when irrigated with lower salinity throughout the formation of passion fruit seedlings.

Irrigation with water of lower salinity during the formation of passion fruit seedlings showed higher values of chlorophyll and number of leaves.

The substrate based on sand, arid and carbonized rice husk allows greater performance in seedling height and increased leaf temperature.

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