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Scientific Article

Morphophysiological aspects of eggplant grown under irrigation with brackish water and foliar application of chitosan

Aspectos morfofisiológicos da berinjela cultivada sob irrigação com águas salobras e aplicação foliar de quitosana

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ABSTRACT - The aim of this study was to evaluate the morphophysiological aspects of eggplant cv. Preta Comprida irrigated with brackish water and subjected to foliar application of chitosan. Treatments consisted of five levels of electrical conductivity of irrigation water - ECw (0.4; 1.4; 2.4; 3.4 and 4.4 dS m^{-1}) and two concentrations of chitosan (0 and 0.50 g L^{-1}), arranged in randomized blocks, in a 5 × 2 factorial scheme with four replicates. Relative water content, electrolyte leakage, chlorophyll *a*, chlorophyll *b* and total chlorophyll contents and carotenoid contents, initial fluorescence, maximum fluorescence, variable fluorescence and quantum efficiency of photosystem II were evaluated. Growth was evaluated by relative growth rates in plant height and stem diameter. Chitosan application attenuated the effects of salt stress on relative water content up to the estimated ECw of 1.9 dS m^{-1}. Foliar application of chitosan at a concentration of 0.50 g L^{-1} promoted beneficial effects on the synthesis of chlorophyll *a*, total chlorophyll and carotenoids in eggplant grown under water salinity of 0.4 dS m^{-1}. Irrigation water salinity above 0.8 dS m^{-1} increased electrolyte leakage and inhibited the synthesis of photosynthetic pigments and chlorophyll *a* fluorescence in eggplant, regardless of foliar application of chitosan. Chitosan promoted a higher growth rate in height of eggplant in the period of 58-85 days after sowing.

Keywords: *Solanum melongena* L.. Salt stress. Biopolymers. Elicitors.

RESUMO - Objetivou-se com este trabalho avaliar os aspectos morfofisiológicos da berinjela cv. Preta Comprida irrigada com águas salobras submetidas a aplicação foliar de quitosana. Os tratamentos constituíram em cinco níveis de condutividade elétrica da água de irrigação – CEa (0,4; 1,4; 2,4; 3,4 e 4,4 dS m^{-1}) e duas concentrações de quitosana (0 e 0,50 g L^{-1}), dispostos em blocos casualizados, em esquema fatorial 5 × 2 com quatro repetições. Avaliaram-se o conteúdo relativo de água, o extravasamento de eletrólitos, os teores de clorofila a, *b*, e total, carotenoides, a fluorescência inicial, a fluorescência máxima, a fluorescência variável e a eficiência quântica do fotossistema II. O crescimento, foi avaliado pelas taxas de crescimento relativo em altura de plantas e diâmetro do caule. A aplicação de quitosana atenuou os efeitos do estresse salino sobre o conteúdo relativo de água até a CEa estimada de 1,9 dS m^{-1}. A aplicação foliar da quitosana na concentração de 0,50 g L^{-1} proporcionou efeitos benéficos na síntese clorofila *a*, clorofila total e carotenoides da berinjela cultivada sob salinidade da água de 0,4 dS m^{-1}. A salinidade na água de irrigação acima de 0,8 dS m^{-1} elevou o extravasamento de eletrólitos e inibiu a síntese de pigmentos fotossintéticos e a fluorescência da clorofila *a* da berinjela, independentemente da aplicação foliar de quitosana. A quitosana proporcionou maior taxa de crescimento em altura de plantas da berinjela no período de 58-85 dias após a semeadura.

Palavras-chave: *Solanum melongena* L.. Biopolímeros. Elicitores.

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INTRODUCTION

Eggplant (*Solanum melongena* L.) is a vegetable crop that has fruits rich in vitamins and antioxidant substances such as phenols and anthocyanins, making it widely appreciated (SONG et al., 2023). In 2017, Brazil produced 71,914 tons of eggplant, led by São Paulo and Rio de Janeiro, with 27,961 tons and 11,116 tons, respectively. In the Northeast, Bahia and Ceará stood out with production of 2,432.1 t and 1,482 t, respectively (IBGE, 2022). However, the temporal and spatial irregularity in the distribution of rainfall, combined with high evapotranspiration rates and newly formed young soils with low water retention capacity, limits and/or hinders agricultural yield in this region (RAMOS et al., 2024).

Excess of salts in the water and/or soil solution causes osmotic effects on plants, acting mainly to reduce the absorption of water and nutrients, such as potassium, leading to an imbalance in the K^+/Na^+ ratio, which directly affects the osmoregulation capacity and consequently the morphophysiological aspects of the plants (LIMA et al., 2023). Excessive accumulation of salt ions also triggers ionic stress in cells, causing changes in physiological and biochemical processes, directly affecting the functioning of photosystem II (PSII) and regulation of gene expression, besides causing partial closure of stomata, leading to reduced plant yield (ALKHATIB et al., 2021).

In this context, understanding and developing strategies to mitigate the effects of salt stress on plants is of paramount importance and, among them, elicitors stand out for performing several functions as signaling and osmoprotective molecules (GOHARI et al., 2023). Among these substances, there is chitosan, which is a linear unbranched polymer of β-1,4-d-glucosamine considered a non-toxic and biodegradable stimulant, obtained from the deacetylation process of chitin, a co-polymer of N-acetyl-d-glucosamine ed-glucosamine (HASSAN et al., 2021).

This biopolymer has been standing out in several areas, including agriculture, where its use has shown a positive effect on plant growth and development, particularly under stress conditions, such as salt stress (ELSHARKAWY et al., 2022). Chitosan is related to increasing crop tolerance to various abiotic stresses, preventing chlorophyll degradation, increasing the content of flavonoids and phenolic compounds, in addition to eliminating reactive oxygen species (ROS) and inducing antioxidant systems (BAKHOUM, SADAK; BADR, 2020).

In this context, several studies have highlighted the beneficial effects of chitosan on plants grown under salt stress, as observed by Gohari et al. (2023) in spearmint, Ullah et al. (2020) in tomato, and Bakhoum, Sadak, and Badr (2020) in sunflower. However, its beneficial effect may be associated with its form of application and also its concentration. In this context, the objective of this study was to evaluate the effects of foliar application of chitosan on the morphophysiological aspects of eggplant cv. Preta Comprida irrigated with brackish water.

MATERIAL AND METHODS

The experiment was carried out from September 2023 to January 2024 under greenhouse conditions at the Academic Unit of Agricultural Engineering (UAEA) of the Federal University of Campina Grande (UFCG), in Campina Grande, Paraíba, located by the geographic coordinates 07° 15’ 18” S latitude, 35° 52’ 28” W longitude and average altitude of 550 m. Air temperature (maximum and minimum) and relative humidity data during the experimental period are presented in Figure 1.

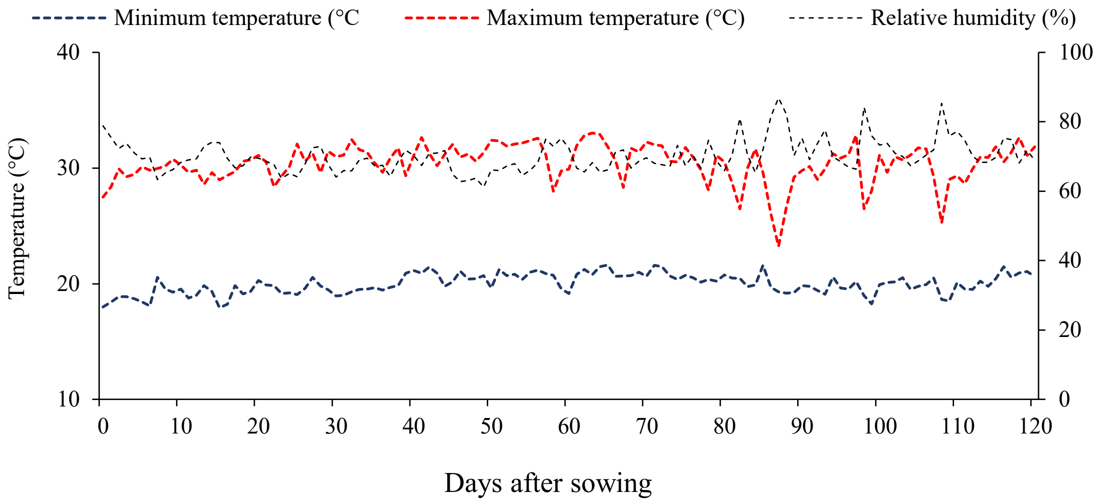


Figure 1. Maximum and minimum temperatures and relative humidity inside the greenhouse during the experimental period (September 18, 2023 to January 15, 2024).

Treatments consisted of five levels of electrical conductivity of irrigation water – ECw (0.4, 1.4, 2.4, 3.4 and 4.4 dS m^{-1}) and two concentrations of chitosan (0 and 0.50 g L^{-1}), arranged in randomized blocks, in a 5 × 2 factorial scheme, with four replicates. The levels of electrical conductivity of the water were based on a study carried out by Roque et al. (2022), with cultivation of cherry tomato (*Solanum lycopersicum* L.), while the concentrations of chitosan were based on the study conducted by Almeida et al. (2020) with cultivation of maize (*Zea mays* L.).

Plants were grown in 10 L plastic pots, adapted as lysimeters. The base of the pots was perforated and connected to a 16-mm-diameter hose, which served as a drain, directing the drained water to a 2 L plastic container. Inside the lysimeters, a non-woven geotextile (Bidim) was placed over the drain outlets, followed by a 0.5-kg layer of crushed stone to avoid clogging.

Then, the lysimeters were filled with 10 kg of a soil classified as *Neossolo Regolítico* (Entisol), with sandy loam texture, collected in the municipality of Riachão do Bacamarte, PB, at 0-30 cm depth, located at the geographic coordinates 7° 10’ 8” S and 35° 51’ 20” W. Chemical and physical-hydraulic attributes of the soil (Table 1) were determined according to Teixeira et al. (2017).

The cultivar selected for the study was cv. Preta Comprida. Five seeds were sown per pot, at 2 cm depth, distributed equidistantly. At 30 days after sowing (DAS), the plants were thinned, keeping only the most vigorous in each container.

Table 1. Chemical and physical-hydraulic characteristics of the soil used in the experiment.

Chemical characteristics								
pH (H_2O)	OM	P	K^+	Na^+	Ca^+	Mg^{2+}	Al^{3+}	H^+
1:2.5	g dm^{-3}	mg dm^{-3}	cmol$_c$ kg^{-1}					
5.40	17.62	2.92	0.28	0.04	1.87	1.70	0.20	2.85
Chemical characteristics				Physical characteristics				
EC_{se}	CEC	SAR_{se}	ESP	Particle-size fraction (g kg^{-1})			Moisture (dag kg^{-1})	
dS m^{-1}	cmol$_c$ kg^{-1}	(mmol L^{-1})$^{0.5}$	%	Sand	Silt	Clay	33.42 kPa[1]	1519.5 kPa[2]
0.72	6.94	0.03	0.58	675.2	221.8	103	12.94	5.32

pH – Hydrogen potential; OM – Organic matter: Walkley-Black wet digestion; Ca^{2+} and Mg^{2+} extracted with 1 M KCl at pH 7.0; Na^+ and K^+ extracted with 1 M NH_4OAc at pH 7.0; Al^{3+}+H^+ extracted with 0.5 M CaOAc at pH 7.0; EC_{se} - Electrical conductivity of the saturation extract; CEC - Cation exchange capacity; SAR_{se} - Sodium adsorption ratio of the saturation extract; ESP - Exchangeable sodium percentage; [1,2] Referring to field capacity and permanent wilting point, respectively.

The levels of electrical conductivity of irrigation water were prepared to reach a proportion of 7:2:1 in the ratio of Na:Ca:Mg, using NaCl, $CaCl_2.2H_2O$ and $MgCl_2.6H_2O$ salts, respectively, by adjusting the concentrations of the available local-supply water (ECw = 0.4 dS m^{-1}), as this is the proportion of salts commonly found in the water bodies of the semi-arid region. Irrigation waters were prepared considering the relationship between ECw and salt concentration, as described by Richards (1954), according to Equation 1:

$$C \approx 10 \times ECw \qquad (1)$$

where: C - Concentration of salts to be added (mmol$_c$ L^{-1}); and ECw - Electrical conductivity of water (dS m^{-1}).

Prior to sowing, soil moisture content was brought to the level corresponding to the maximum water retention capacity, through the application of water in the lysimeter. After sowing, irrigation was carried out daily in the afternoon (5:00 p.m.), applying to each container the volume corresponding to that obtained by the water balance, determined by Equation 2.

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

Where: VI - volume of water to be applied in irrigation (mL); Va - volume applied in the previous irrigation (mL); Vd - volume drained (mL); LF - leaching fraction of 0.10, applied every 15 days.

Chitosan powder from Originalis® Biotech was used, with the following characteristics: appearance of whitish powder, 1% acetic acid solution forming a translucent gel, 40 mesh particle size, desiccation loss of 9.41%, total ash of 1.31%, pH of 7.4%, solubility in acetic acid solution of 11 minutes and degree of deacetylation of 86.12% according to the manufacturer. The chitosan solution was prepared for each application by dissolving 20 g L^{-1} of chitosan in 0.1 M acetic acid with the aid of a Centauro shaker and then diluted to 0.50 g L^{-1} using distilled water. For the control treatments (0 g L^{-1}), the biopolymer was not applied. To reduce the surface tension of the droplets on the leaf surface (adaxial and abaxial sides), Wil fix® adjuvant was used at a concentration of 0.5 mL L^{-1} at the time of application.

Applications were carried out at 5 p.m. with a manual sprayer coupled to a PET bottle, with a capacity of 2L; for application of the solution, each plant was isolated using plastic curtains to prevent the solution from drifting. In addition, an adhesive adjuvant was used to improve the fixation of the product. Applications began at 40 DAS with a spraying frequency of 15 days, in a total of three. During the experiment, a total of 223 mL of chitosan was applied in each plant at 40, 60 and 75 DAS.

Mineral fertilization with NPK was carried out according to Novais, Neves and Barros (1991), applying 100 mg of N, 300 mg of P_2O_5 and 150 mg K_2O per kg of soil, using urea (45.5% N), monoammonium phosphate (60% P_2O_5) and potassium chloride (60% K_2O), split into four portions and applied by fertigation, starting at 30 DAS. Every two weeks, plants received a solution of Dripsol micro® at a concentration of 1.0 g L^{-1}, containing: Mg (1.1%); Zn (4.2%); B (0.85%); Fe (3.4%); Mn (3.2%); Cu (0.5%); Mo (0.05%), on the leaves, covering their adaxial and abaxial faces.

Phytosanitary management was carried out using chemical pesticides with the active ingredient of Abamectin and Chlorfenapyr for the control of aphids (*Aphis gossypii*), mealybugs (*Phenacoccus Solenopsis*) and whiteflies (*Bemisia tabaci*) applied via spraying whenever necessary.

At 85 days after sowing (DAS), relative water content (RWC) and electrolyte leakage (EL) in the leaf blade, contents of chlorophyll a (Chl *a*), chlorophyll *b* (Chl *b*), total chlorophyll (Chl t) and carotenoids (Car), initial fluorescence (F_0), maximum fluorescence (Fm), variable fluorescence (Fv) and quantum efficiency of photosystem II (Fv/Fm) were evaluated. Growth was evaluated at 58 and 85 DAS, through relative growth rates in plant height (RGR_{PH}) and stem diameter (RGR_{SD}).

RWC was determined according to the method described by Weatherley (1950), using the fresh mass (FM) of five 12 mm diameter discs taken from leaves of the middle third of the main branch. Then, the discs were immersed in 50 mL of distilled water in beakers for 24 hours at ambient temperature. After this period, excess water was removed from the discs with paper towels, in order to obtain the turgid mass (TM) of the samples, and then the samples were stored at a temperature of 65 ± 3 °C until reaching constant mass to obtain the dry mass (DM). All measurements were made using a digital scale. RWC was then calculated according to Equation 3.

$$\mathrm{RWC} = \frac{(\mathrm{FM} - \mathrm{DM})}{(\mathrm{TM} - \mathrm{DM})} \times 100 \quad (3)$$

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

EL was measured using a copper hole punch to obtain five 12-mm-diameter leaf discs per experimental unit, which were washed and stored in an Erlenmeyer® flask containing 50 mL of deionized water. After being closed with aluminum foil, the Erlenmeyer® flasks were kept at 25 °C for 24 hours, and then the initial electrical conductivity of the medium (EC1) was measured using a benchtop conductivity meter. Subsequently, the Erlenmeyer® flasks were subjected to 90 °C for 120 minutes in a drying oven, and then the final electrical conductivity (EC2) was measured. Electrolyte leakage was expressed as the percentage of conductivity relative to the total conductivity after treatment for 120 min at 90 °C, according to a methodology adapted from Scotti-Campos et al. (2013), considering Equation 4:

$$\mathrm{EL}\ (\%) = \frac{\mathrm{EC1}}{\mathrm{EC2}} \times 100 \quad (4)$$

where: EL - electrolyte leakage (%); EC1 - initial electrical conductivity (dS m^{-1}); and EC2 - final electrical conductivity (dS m^{-1}).

Contents of chlorophyll *a*, chlorophyll *b*, total chlorophyll and carotenoids were determined according to Arnon (1949), using plant extracts obtained from the sample of leaf discs with an area of 12 mm, collected from the third leaf fully expanded from the apical bud. Each sample received 5 mL of Dimethyl sulfoxide and was stored at ambient temperature; after 48 hours, a spectrophotometer was used to take readings of the photosynthetic pigments at the absorbance wavelengths of 470, 647 and 663 nm, respectively, according to Equations 5, 6, 7 and 8.

$$\mathrm{Chl}\ a = (12.25 \times \mathrm{ABS663}) - (2.79 \times \mathrm{ABS647}) \quad (5)$$

$$\mathrm{Chl}\ b = (21.5 \times \mathrm{ABS647}) - (5.10 \times \mathrm{ABS647}) \quad (6)$$

$$\mathrm{Chl}\ total = (7.15 \times \mathrm{ABS663}) + (18.71 \times \mathrm{ABS647}) \quad (7)$$

$$\mathrm{Car} = (1000 \times \mathrm{ABS470} - 1.82 \times \mathrm{Chl\ a} - 85.02 \times \mathrm{Chl\ b})/198 \quad (8)$$

Where: Chl *a* - chlorophyll *a*; Chl *b* - chlorophyll *b*; Chl *total* - total chlorophyll; Car – carotenoids. The values obtained for the contents of chlorophyll *a*, chlorophyll *b*, total chlorophyll and carotenoids (Car) in the leaves were expressed in µg mL^{-1}.

Chlorophyll *a* fluorescence was determined by means of an OS5p pulse-modulated fluorometer from Opti Science, using the Fv/Fm protocol to determine the fluorescence induction variables: Initial fluorescence (F_0), Maximum fluorescence (Fm), Variable fluorescence (Fv) and the quantum efficiency of photosystem II (Fv/Fm). The protocol was performed at 7:00 a.m., after the leaves were adapted to the dark for 30 minutes using a clip of the equipment, to ensure that all primary acceptors were fully oxidized, i.e., that the reaction centers were open.

Relative growth rates in plant height and stem diameter were determined at 58 and 85 DAS, according to the methodology of Benincasa (2003), using Equation 9 and Equation 10.

$$\mathrm{RGR}_{PH} = \frac{\mathrm{Ln}(\mathrm{PH}_2) - \mathrm{Ln}(\mathrm{PH}_2)}{\mathrm{T}_2 - \mathrm{T}_1} \quad (9)$$

$$\mathrm{RGR}_{SD} = \frac{\mathrm{Ln}(\mathrm{SD}_2) - \mathrm{Ln}(\mathrm{SD}_2)}{\mathrm{T}_2 - \mathrm{T}_1} \quad (10)$$

Where PH_1 – Plant height at 58 DAS; PH_2 – Plant height at 85 DAS; SD_1 – Stem diameter at 58 DAS; SD_2 – Stem diameter at 85 DAS; T_1 – Period in days of the evaluation at 58 DAS and T_2 – Period in days of the evaluation at 85 DAS.

The collected data were subjected to the homogeneity test (Levene test) and, when normal distribution was observed, analysis of variance was performed by the F test at 0.05 probability level. When significant, polynomial regression analysis was performed for the levels of electrical conductivity of irrigation water. All statistical analyses were performed using the statistical software R-Studio (R CORE TEAM, 2024).

RESULTS AND DISCUSSION

There was a significant effect ($p \leq 0.01$) for the interaction between the factors (ECw × CHT) on the relative water content (RWC) and electrolyte leakage (EL) in the leaf blade of eggplant plants cv. Preta Comprida (Table 2).

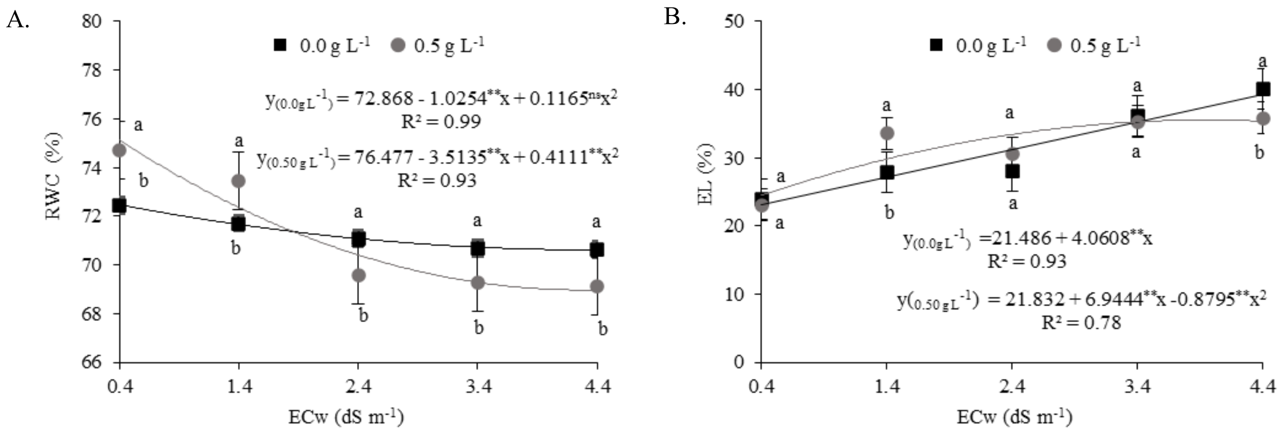
The increase in electrical conductivity in the irrigation water negatively affected the relative water content (RWC) in the leaf blade of eggplant cv. Preta Comprida (Figure 2A). There was a significant difference between chitosan concentrations at all salinity levels. Plants that received the chitosan concentration of 0.50 g L^{-1} showed the maximum estimated value for RWC (75.14%) when subjected to ECw of 0.4 dS m^{-1} and when compared with plants that were not subjected to chitosan applications (0.0 g L^{-1}) irrigated with the same salinity level, there were increases of 3.54%. From the ECw of 1.9 dS m^{-1}, chitosan intensified the deleterious effects of salt stress, and the lowest RWC (68.97%) was observed under water salinity of 4.4 dS m^{-1}.

In the decomposition of chitosan concentrations considering each ECw level, significant differences were observed in the RWC of plants irrigated with the different water salinity levels; plants under chitosan concentration of 0.50 g L^{-1} were statistically superior to those that received 0.0 g L^{-1}, even those irrigated with ECw of 1.9 dS m^{-1}. At the other ECw levels, foliar application of 0.50 g L^{-1} resulted in lower values for relative water content compared to plants that received 0.0 g L^{-1}. The reduction in water content in plants is related to the osmotic effect caused by the increase in osmotic pressure in the soil due to the accumulation of salts, causing a restriction in the absorption of water and nutrients, thus reducing the amount of water in the leaf blade (SILVA et al., 2021).

Table 2. Summary of the analysis of variance for relative water content (RWC) and electrolyte leakage (EL) in the leaf blade of eggplant cv. Preta Comprida irrigated with brackish water and under foliar application of chitosan, at 85 days after sowing (DAS).

Source of variation	DF	Mean squares	
		RWC	EL
Electrical conductivity (ECw)	4	23.15**	257.93**
Linear regression	1	72.71**	19.04**
Quadratic regression	1	75.85**	23.5**
Chitosan (CHT)	1	0.05^{ns}	1.95^{ns}
Interaction (ECw × CHT)	4	7.31**	7.14**
Blocks	3	0.9^{ns}	6.3^{ns}
CV (%)	-	1.16	5.92

DF - Degrees of freedom; CV - Coefficient of variation; *,**, ^{ns} - Significant at $p \leq 0.05$, $p \leq 0.01$ and not significant, respectively.



Means with equal letters indicate no significant difference between chitosan concentrations by the F test ($p \leq 0.05$).
*,**, ^{ns} - Significant at $p \leq 0.05$, $p \leq 0.01$ and not significant, respectively

Figure 2. Relative water content - RWC (A) and electrolyte leakage - EL (B) in the leaf blade of eggplant cv. Preta Comprida, as a function of the interaction between the levels of electrical conductivity of irrigation water - ECw and chitosan concentrations, at 85 days after sowing.

Increasing irrigation water salinity increased electrolyte leakage in the leaf blade of eggplant (Figure 2B) under the chitosan concentration of 0.0 g L^{-1}, causing an increase of 70.29% when comparing plants grown under ECw of 0.4 dS m^{-1} with those under 4.4 dS m^{-1}. On the other hand, it was verified that the chitosan concentration of 0.5 g L^{-1} attenuated the deleterious effects of salinity only on plants cultivated under ECw of 4.4 dS m^{-1}. Regarding the effects of chitosan concentrations at each water salinity level on EL, there was a significant difference between chitosan applications (0.0 and 0.50 g L^{-1}) for plants grown under ECw of 1.4, 2.4 and 4.4 dS m^{-1}. Possibly, this result is associated with the increase in the ionic concentration of Na^+ and Cl^- in the root environment, which contributes to membrane rupture and instability, leading to increased formation of reactive oxygen species (ROS), resulting in peroxidation and oxidation of the cell membrane (XAVIER et al., 2022). Salt stress can induce the production of ROS, due to its toxic effect, causing photo-oxidative damage to the photosynthetic system, peroxidation of cell membrane, and increased cellular protoplasmic damage (YUDINA et al., 2020).

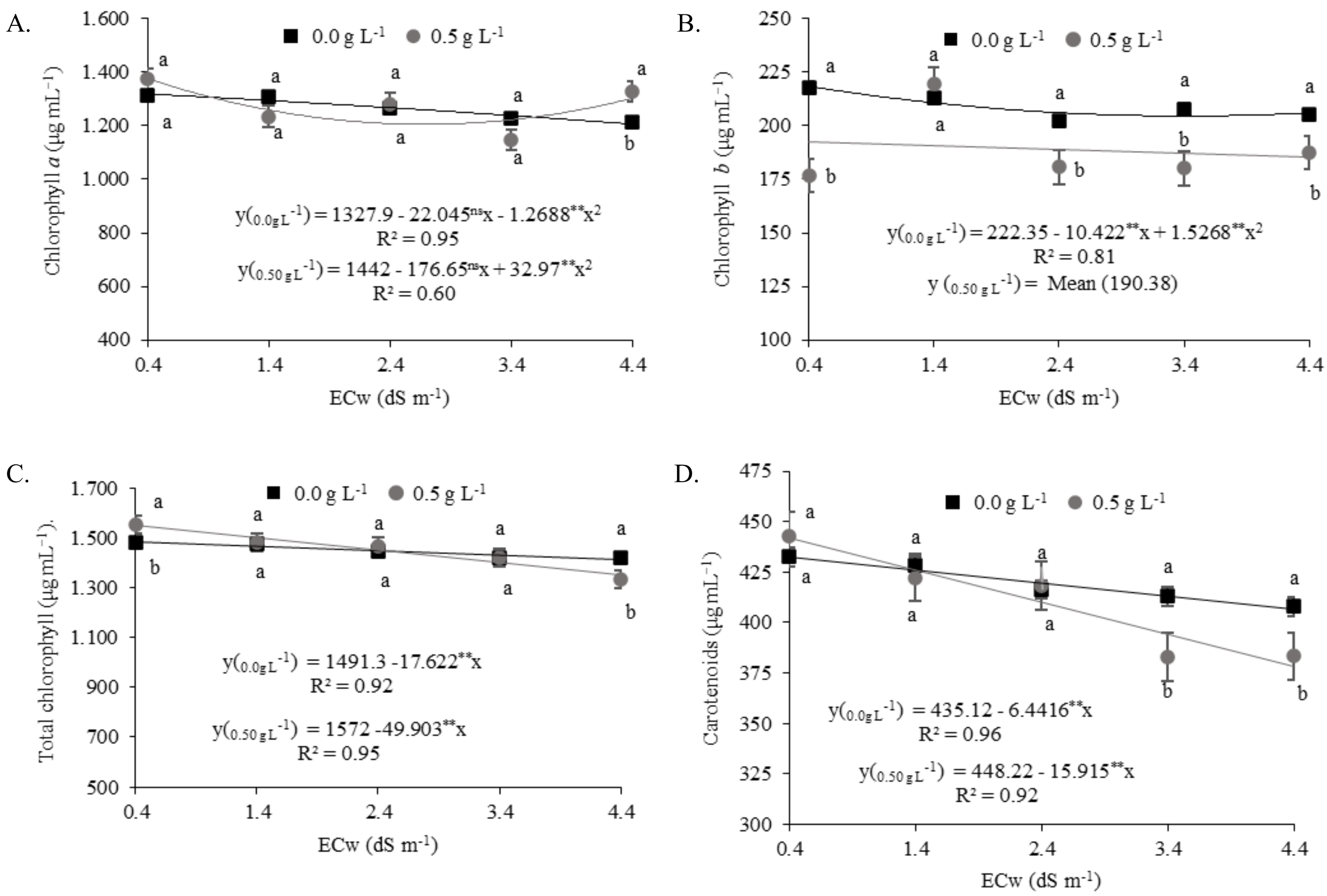
The interaction between the factors levels of electrical conductivity of irrigation water and concentrations of chitosan (ECw × CHT) significantly affected the contents of chlorophyll *a* (Chl *a*), chlorophyll *b* (Chl *b*), total chlorophyll (Chl *t*) and carotenoids (Car) of eggplant cv. Preta Comprida, at 85 DAS (Table 3).

Regarding the chlorophyll *a* contents (Figure 3A), foliar application of chitosan at a concentration of 0.50 g L^{-1} resulted in a maximum value of 1376.62 μg mL^{-1} in plants cultivated under irrigation with ECw of 0.4 dS m^{-1}. On the other hand, the absence of foliar application of chitosan (0.0 g L^{-1}) resulted in the lowest contents (1206.3 μg mL^{-1}) of Chl *a* in plants subjected to ECw of 4.4 dS m^{-1}. Foliar applications of chitosan at concentrations of 0.0 and 0.50 g L^{-1} differed significantly only in plants irrigated with ECw of 4.4 dS m^{-1}. At the other ECw levels, there were no significant differences between the concentrations of chitosan. The beneficial effect of chitosan may be associated with its action in the activation of plant signaling mechanisms, thus resulting in the differential expression of stress-tolerant genes (GOHARI et al., 2023). In addition to inducing antioxidant systems to resist stress, chitosan can also activate enzymes and defense genes, increasing plant resistance against stress, intensifying photosynthetic activity (ELSHARKAWY et al., 2022).

Table 3. Summary of the analysis of variance for chlorophyll *a* (Chl *a*), chlorophyll *b* (Chl *b*), total chlorophyll (Chl t) and carotenoids (Car) of eggplant cv. Preta Comprida irrigated with brackish water and under foliar application of chitosan, at 85 days after sowing (DAS).

Source of variation	DF	Mean square			
		Chl *a*	Chl *b*	Chl t	Car
Electrical conductivity (ECw)	4	24878.67**	783.74**	22829.56**	2597.30**
Linear regression	1	25.05**	2.45**	15.91**	4.02**
Quadratic regression	1	62.06**	8.78**	20.15*	5.37**
Chitosan (CHT)	1	775.06 ns	4084.83**	155.75ns	928.33**
Interaction (ECw × CHT)	4	14398.85*	614.21**	6597.48**	599.73**
Blocks	3	15148.10*	415.45ns	512.41ns	30.14ns
CV (%)	-	5.59	1.81	2.46	2.00

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



Means with equal letters indicate no significant difference between chitosan concentrations by the F test ($p \leq 0.05$). *,**, ns - Significant at $p \leq 0.05$, $p \leq 0.01$ and not significant, respectively. Vertical error bars represent the standard error of the mean (n = 4).

Figure 3. Contents of chlorophyll *a* - Chl *a* (A), chlorophyll *b* - Chl *b* (B), total chlorophyll - Chl t (C) and carotenoids - Car (D) of eggplant plants cv. Preta Comprida, as a function of the interaction between the levels of electrical conductivity of irrigation water - ECw and chitosan concentrations, at 85 days after sowing (DAS).

The chlorophyll *b* contents (Figure 3B) of eggplant plants cv. Preta Comprida grown under chitosan concentration of 0.50 g L^{-1} were not satisfactorily described by any regression model, obtaining an average of 190.38 µg mL^{-1}. On the other hand, in the case of plants cultivated in the absence of foliar application of chitosan (0.0 g L^{-1}), the maximum estimated value (218.42 µg mL^{-1}) was obtained in plants irrigated with ECw of 0.4 dS m^{-1}. When comparing the Chl *b* contents of plants grown under ECw of 4.4 dS m^{-1} to that of plants irrigated with water of 0.4 dS m^{-1}, a decrease of 5.66% (12.40 μg mL^{-1}) was observed. Regarding the decomposition of chitosan concentrations considering the different levels of water salinity, significant differences were observed, with higher Chl *b* contents in plants grown in the absence of foliar application of chitosan (0.0 g L^{-1}) compared to those that received 0.50 g L^{-1} at all water salinity levels, except for the ECw of 1.4 dS m^{-1}. The reduction in chlorophyll contents in plants subjected to salt stress may be associated with increased chlorophyllase enzyme activity, lipid peroxidation and the generation of reactive oxygen species in the cellular system (SILVA et al., 2017).

It is verified that, regardless of the concentration of chitosan applied in eggplant plants cv. Preta Comprida, there were decreases in total chlorophyll contents (Figure 3C) of 4.75% and 12.86% for the concentrations of 0.0 and 0.5 g L^{-1}, respectively, when comparing plants irrigated with ECw of 0.4 dS m^{-1} with those under 4.4 dS m^{-1}. On the other hand, when observing the effects of chitosan concentrations at each level of water salinity (Figure 3C), it can be seen that the application of this compound differed statistically under ECw levels of 0.4 and 4.4 dS m^{-1}. The beneficial effect of chitosan on plants grown under low water salinity may be associated with the induction of defense mechanisms, contributing to the synthesis of antioxidant metabolites and preventing the accumulation of reactive oxygen species (GOHARI et al., 2023).

Irrigation with saline water negatively influenced the synthesis of carotenoids in eggplant cv. Preta Comprida (Figure 3D), regardless of the concentration of chitosan applied, whose decreases were 5.94% (25.76 μg mL^{-1}) and 14.41% (63.66 μg mL^{-1}) for plants grown under chitosan concentrations of 0.0 and 0.50 g L^{-1}, respectively, when comparing the ECw levels of 0.4 and 4.4 dS m^{-1}. Regarding the effects of chitosan concentrations at each water salinity level (Figure 3D), it can be seen that foliar application of 0.50 g L^{-1} of chitosan resulted in a significant effect under ECw of 4.4 dS m^{-1}, i.e., an effect similar to that observed for Chl *a* (Figure 3A). The reduction in the synthesis of photosynthetic pigments in eggplant plants may be associated with the stress caused by the excess of dissolved salts in irrigation water, which may have stimulated the activity of the enzyme chlorophyllase, which acts in the degradation of photosynthesizing pigment molecules, or with the photooxidation caused by oxidative stress (DIAS et al., 2019).

There was a significant effect of the interaction between the factors (ECw × CHT) on all the analyzed variables of chlorophyll *a* fluorescence (Table 4) of eggplant cv. Preta Comprida, at 85 days after sowing.

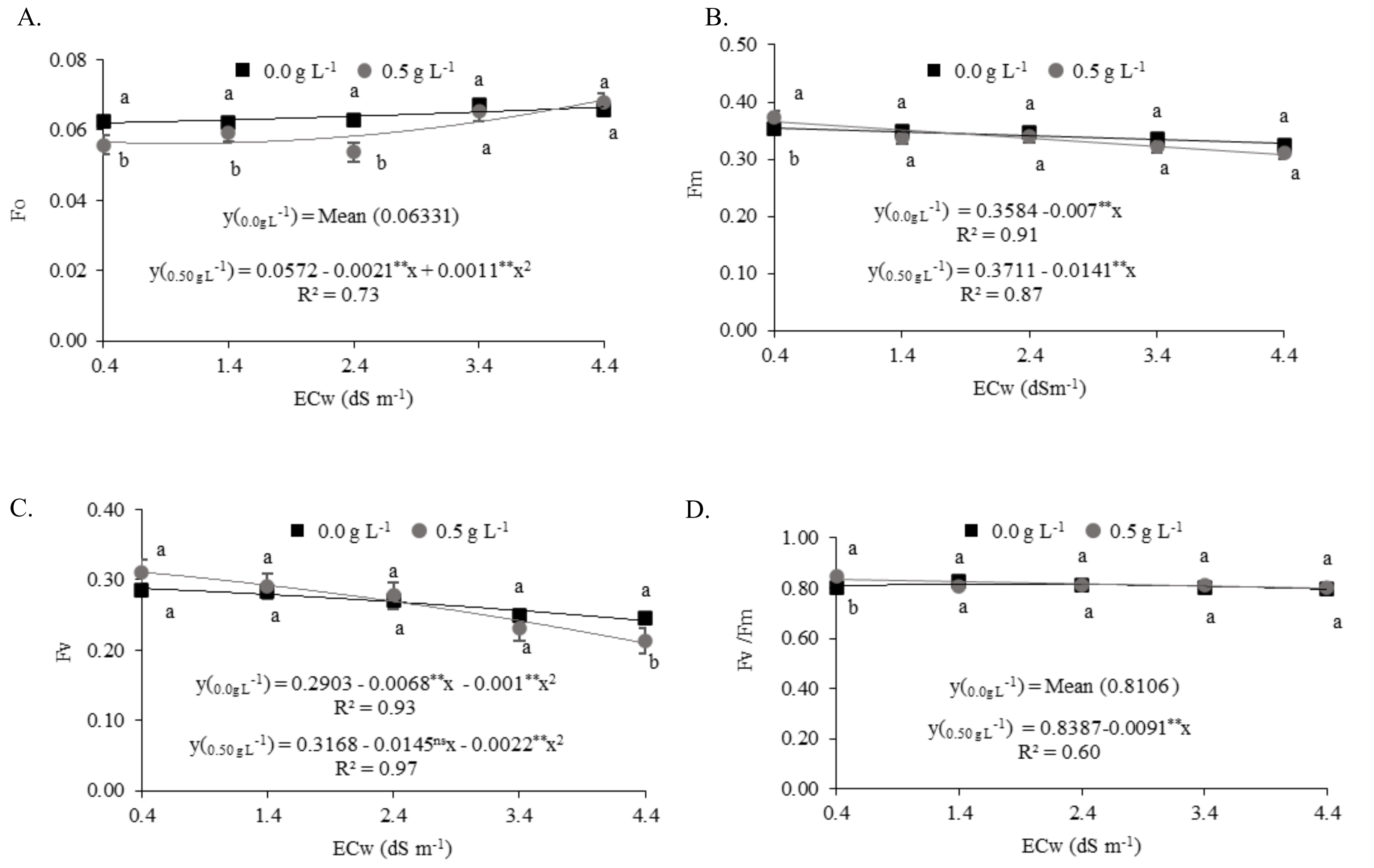
Table 4. Summary of the analysis of variance for initial fluorescence (F_0), maximum fluorescence (Fm), variable fluorescence (Fv) and quantum efficiency of photosystem II (Fv/Fm) of eggplant cv. Preta Comprida irrigated with brackish water and under foliar application of chitosan, at 85 days after sowing.

Source of variation	DF	Mean square			
		F_0	Fm	Fv	Fv/Fm
Electrical conductivity (ECw)	4	0.000^{**}	0.012^{**}	0.007^{**}	0.001^{**}
Linear regression	1	0.001^{**}	0.004^{**}	0.005^{**}	0.008^{**}
Quadratic regression	1	0.002^{**}	0.008^{**}	0.01^{**}	0.008^{**}
Chitosan (CHT)	1	0.000^{**}	0.000^{ns}	0.000^{ns}	0.001^{ns}
Interaction (ECw × CHT)	4	0.000^{**}	0.0004^{*}	0.001^{*}	0.001^{**}
Blocks	3	1.3×10^{3ns}	6.8×10^{3ns}	6.4×10^{3ns}	6.3×10^{3ns}
CV (%)	-	2.87	3.21	7.2	1.63

DF - Degrees of freedom; CV - Coefficient of variation; *,**, ^{ns} - Significant at $p \leq 0.05$, $p \leq 0.01$ and not significant, respectively.

Regarding the initial fluorescence (Figure 4A), plants cultivated under ECw of 0.4 dS m^{-1} and foliar application of chitosan at a concentration of 0.50 g L^{-1} obtained the lowest value (0.056) and, when compared with the control treatment irrigated with the same salinity level, there was a decrease of 9.58% (0.0053). Chitosan application at a concentration of 0.50 g L^{-1} promoted an increase of 22.50% (0.012) in the initial fluorescence, with the highest value (0.069) being reached in plants irrigated with ECw of 4.4 dS m^{-1}. When analyzing the effects of chitosan concentrations at the different ECw levels, it was observed that the concentration of 0.0 g L^{-1} was superior to 0.50 g L^{-1} in plants cultivated under water salinity of 0.4, 1.4 and 2.4 dS m^{-1}. The negative effect of salinity may be associated with the reduction in energy capture in the reaction centers, probably because the excessive accumulation of specific ions causes an imbalance in the metabolic activity of the plant, leading to the formation of reactive oxygen species, which limits the energetic activity of the photosynthetic pigments (YOUSEFZADEH et al., 2023).

Regarding the maximum fluorescence (Figure 4B), it is observed that the increase in ECw levels promoted linear decreases of 1.95 and 3.79% per unit increment in ECw in plants cultivated under foliar application of 0 and 0.50 g L^{-1} of chitosan. When comparing the Fm of plants grown under concentrations of 0.0 and 0.50 g L^{-1} between ECw levels of 4.4 and 0.4 dS m^{-1}, reductions of 8.55% (0.028) and 15.43% (0.0564) were observed. In the decomposition of the interaction of chitosan concentrations at each ECw level, significant differences were observed only in plants grown under ECw of 0.4 dS m^{-1}. The decrease in maximum fluorescence may be associated with the effects caused by salt stress, causing a slowdown in photosynthetic activities, aimed at mitigating the toxic effects caused by salinity (MONTEIRO et al., 2018).



Means with equal letters indicate no significant difference between chitosan concentrations by the F test ($p \leq 0.05$). *,**, ^{ns} - Significant at $p \leq 0.05$, $p \leq 0.01$ and not significant, respectively. Vertical error bars represent the standard error of the mean ($n = 4$).

Figure 4. Initial fluorescence – F_0 (A), maximum fluorescence – Fm (B), variable fluorescence – Fm (C) and quantum efficiency of photosystem II – Fv/Fm (D) in eggplant plants cv. Preta Comprida, as a function of the interaction between the levels of electrical conductivity of irrigation water – ECw and chitosan concentrations, at 85 days after sowing (DAS).

When evaluating chlorophyll *a* fluorescence in soursop plants under salt stress (0.8 to 4.0 dS m^{-1}), Silva et al. (2022) observed a 3.31% reduction in maximum fluorescence as a function of the unit increase in irrigation water salinity. According to the authors, this is associated with low efficiency in the photoreduction of quinones and in the flow of electrons between photosystems, resulting in a lower activity of photosystem II in the thylakoid membrane and directly influencing the flow of electrons between photosystems.

For variable fluorescence (Figure 4C), it was verified that foliar application of 0.0 and 0.5 g L^{-1} of chitosan promoted the maximum estimated values of 0.28 and 0.31, respectively, under water salinity of 0.4 dS m^{-1}. When comparing the Fv of plants that received 0.0 and 0.50 g L^{-1} of chitosan, reductions of 16.14 and 32.27% were observed between the ECw levels of 4.4 and 0.4 dS m^{-1}. Regarding the decomposition of chitosan concentrations considering each ECw level (Figure 4C), there were significant differences in the Fv of plants grown under ECw of 0.4 and 4.4 dS m^{-1}. Fv represents the plant's ability to transfer the energy of excited electrons in pigment molecules to form NADPH, ATP, and reduce ferredoxin, which increases CO_2 assimilation in the biochemical phase of photosynthesis. Thus, the reduction in Fv indicates that the photosynthetic apparatus was damaged by salt stress, affecting photosystem II and negatively compromising the photosynthetic process (SILVA et al., 2022).

The data on the quantum efficiency of photosystem II of eggplant cv. Preta Comprida (Figure 4D) under foliar application of 0.0 g L^{-1} were not described by the regression models tested, obtaining a mean value of 0.8106. On the other hand, the application of 0.50 g L^{-1} resulted in linear decreases of 1.08% per unit increase in ECw, i.e., a reduction of 4.36% between plants cultivated under ECw of 4.4 and 0.4 dS m^{-1}. The decrease in Fv/Fm may be associated with changes in photosystem II caused by salt stress and the inhibition of the breakdown of the water molecule, which is essential for obtaining the necessary electrons in the photochemical phase of photosynthesis (OLIVEIRA et al., 2018).

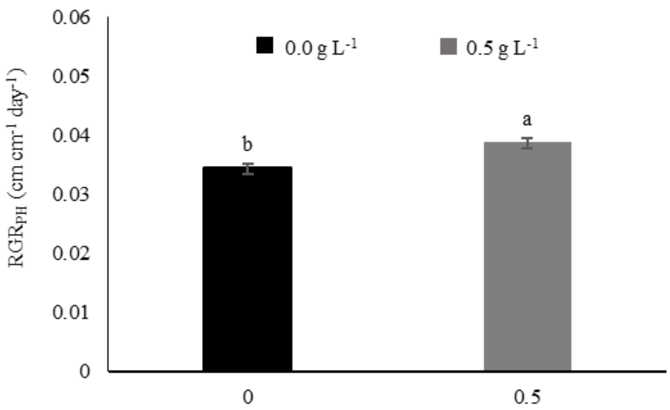
The levels of electrical conductivity of the water and the interaction between the factors (ECw × CHT) did not significantly influence the relative growth rate in stem diameter of eggplant cv. Preta Comprida. There was an individual effect of the concentrations of chitosan (CHT) on the relative growth rate in plant height (RGR_{PH}) of eggplant plants cv. Preta Comprida (Table 5). Salinity in irrigation water and chitosan concentrations did not significantly influence any of the variables measured.

Table 5. Summary of the analysis of variance for relative growth rate in plant height (RGR_{PH}) and stem diameter (RGR_{SD}) of eggplant cv. Preta Comprida irrigated with brackish water and under foliar application of chitosan, at 85 days after sowing.

Source of variation	DF	Mean squares	
		RGR_{PH}	RGR_{SD}
Electrical conductivity (ECw)	4	0.000001^{ns}	0.0000012^{ns}
Linear regression	1	0.000001^{ns}	0.0000031^{ns}
Quadratic regression	1	0.000010^{ns}	0.000011^{ns}
Chitosan (CHT)	1	0.00004^{**}	0.000021^{ns}
Interaction (ECw × CHT)	4	0.000031^{ns}	0.000001^{ns}
Blocks	3	0.000^{ns}	0.000010^{ns}
CV (%)	-	9.11	20.11

DF - Degree of freedom; CV - Coefficient of variation; *,**, ^{ns} - Significant at $p \leq 0.05$, $p \leq 0.01$ and not significant, respectively.

Foliar application of chitosan at a concentration of 0.5 g L^{-1} resulted in a statistically superior relative growth rate (Figure 5) in plant height compared to the control treatment (0.0 g L^{-1}). An increase of 12.62% in RGR_{PH} was observed between plants subjected to foliar application of 0.5 and 0.0 g L^{-1} of chitosan. The increased growth of eggplant plants may be associated with the beneficial effect on water and nutrient absorption, possibly due to the better mitotic division and cell enlargement required for growth, similar to the effects of hormones (GOHARI et al., 2023).



Means followed by equal letters did not differ from each other for the chitosan concentrations, using the F test ($p \leq 0.05$). Vertical error bars represent the standard error of the mean (n = 4).

Figure 5. Relative growth rate in plant height (RGR_{PH}) in eggplant cv. Preta Comprida, as a function of chitosan concentrations, at 85 days after sowing (DAS).

The changes in the morphophysiological variables for each chitosan concentration can be observed in Pearson's correlation matrix (Figure 6). Both chitosan concentrations (0 g L^{-1} and 0.50 g L^{-1}) promoted a high correlation between the relative water content and variable fluorescence (0.60), as well as the photosynthetic pigment variables (Car and Chl t), which were positively correlated with maximum fluorescence and variable fluorescence, with correlation values higher than 0.70.

Regarding the chitosan concentration of 0.0 g L^{-1} (Figure 6), there was a negative correlation between electrolyte leakage and the variables chlorophyll b (-0.63), carotenoids (-0.72) and total chlorophyll (-0.62), except for the initial fluorescence, which showed a positive correlation of 0.66. This result is possibly a consequence of the negative effect of salt stress, altering photosynthetic processes, which reduces the amount of photoassimilates. Under stress conditions, plants undergo physiological disturbances that lead to enzymatic changes and can affect the protein stability of photosynthetic enzymes, such as ribulose-1,5-bisphosphate carboxylase/oxygenase (RuBisCO) (SHUMILINA et al., 2019).

On the other hand, with foliar application of chitosan at a concentration of 0.5 g L^{-1}, there was a significant positive correlation between the relative water content and the maximum fluorescence (0.67), the variable fluorescence (0.66) and the quantum efficiency of photosystem II (0.60). Regarding carotenoids and total chlorophyll, a significant positive correlation of 0.05 was observed with the variables of chlorophyll *a* fluorescence (0.5), except for initial fluorescence, which showed negative correlations of -0.78 and -0.77 with these variables, respectively. Possibly, these effects are associated with the role of chitosan in preventing Na^+ absorption, thereby increasing chlorophyll biosynthesis, due to the critical increase in cytokinin production (BAKHOUM; SADAK; BADR, 2020).

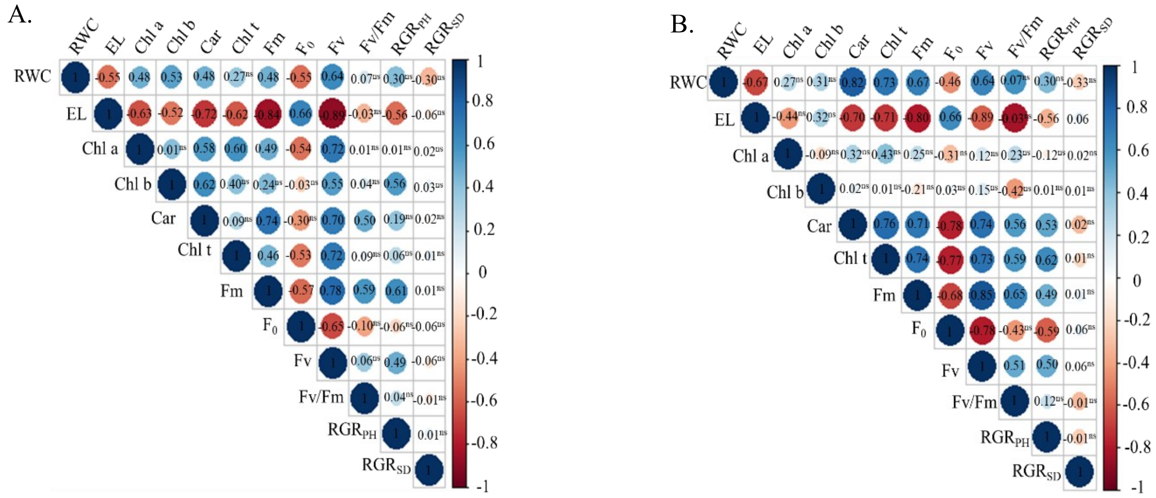


Figure 6. Pearson's correlation matrix for the variables analyzed within the levels of electrical conductivity and the chitosan doses of 0.0 g L^{-1} (A) and 0.5 g L^{-1} (B), for the variables RWC, EL, Chl *a* (μg mL^{-1}), Chl *b* (μg mL^{-1}), Chl *t* (μg mL^{-1}), Car (μg mL^{-1}), F_0, Fm, Fv, Fv/Fm, RGR_{PH} (cm cm^{-1} day^{-1}) and RGR_{SD} (cm cm^{-1} day^{-1}).

When evaluating the effects of salinity on spearmint plants under foliar application of chitosan, Gohari et al. (2023) observed that the spraying of chitosan increased the contents of photosynthetic pigments under salt stress (50 mM NaCl), leading to the highest values for Chl *a* (19.79 mg g^{-1}), Chl *b* (8.66 mg g^{-1}) and carotenoids (11.53 mg g^{-1}), corresponding to increases of 12.96, 66.86 and 41.82%, respectively.

CONCLUSIONS

Application of chitosan at a concentration of 0.50 g L^{-1} attenuates the effects of water salinity up to 1.9 dS m^{-1} on the relative water content in the leaf blade of eggplant cv. Preta Comprida. At this same concentration, there are beneficial effects on the synthesis of chlorophyll *a*, total chlorophyll and carotenoids under water salinity of 0.4 dS m^{-1}. Irrigation water salinity above 0.8 dS m^{-1} increases electrolyte leakage and inhibits the synthesis of photosynthetic pigments and chlorophyll *a* fluorescence of eggplant cv. Preta Comprida, regardless of the foliar application of chitosan. Application of chitosan promotes an increase in the relative growth rate in plant height for eggplant cv. Preta Comprida in the period of 58-85 days after sowing.

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