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GAS EXCHANGES IN *Handroanthus impetiginosus* SEEDLINGS UNDER ARTIFICIAL SHADING AND IRRIGATION WITH BRACKISH WATER**TROCAS GASOSAS EM MUDAS DE *Handroanthus impetiginosus* SOB SOMBREAMENTO ARTIFICIAL E IRRIGAÇÃO COM ÁGUAS SALOBRAS****Luciana Luzia Pinho¹, Claudivan Feitosa de Lacerda², João Alencar de Sousa³, Antônio Marcos Esmeraldo Bezerra⁴, Jonnathan Richeds da Silva Sales⁵, Juvenaldo Florentino Canjá⁶**

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ABSTRACT: The Caatinga biome has peculiar climate and soil conditions. Although native vegetation has mechanisms of adaptation to conditions imposed by the environment, little is known about the development of forest species under the influence of abiotic factors. Thus, the objective of the present work was to evaluate the effects of irrigation with brackish water on the leaf gas exchange of *Handroanthus impetiginosus* seedlings grown in different types of artificial shading. The experiment was conducted at the Center of Education and Research in Urban Agriculture of the Federal University of Ceará, under a randomized blocks design with four repetitions. The plots were composed of four levels of shading, formed by black shading screen of the sombrite type: 0 (full sun), 30, 50 and 70% and the subplots were formed by five increasing levels of electrical conductivity of the irrigation water: 0.4, 1.6, 2.8, 4.0, and 5.2 dS m⁻¹. At 60 days after sowing (DAS) net photosynthesis rates, transpiration, stomatal conductance, internal CO₂ concentration, and water use efficiency were determined. The higher electrical conductivities of the water (4.0 and 5.2 dS m⁻¹) caused damage to the photosynthetic apparatus of *Handroanthus impetiginosus* seedlings, however, the use of artificial shading attenuated the harmful effects of salinity, and the black shading screen with 50% shade was the one that most favored leaf gas exchange, allowing tolerance of up to 2.8 dS m⁻¹.

Keywords: Ipê-roxo, salinity, shading screens.

RESUMO: O bioma Caatinga possui condições singulares de clima e solo. Embora a vegetação nativa possua mecanismos de adaptação às condições impostas pelo ambiente, pouco se sabe sobre o desenvolvimento de espécies florestais sob influência de fatores abióticos. Assim, o objetivo do presente trabalho foi avaliar os efeitos do sombreamento artificial e da irrigação com águas salobras sobre as trocas gasosas foliares de mudas de *Handroanthus impetiginosus*. O experimento foi conduzido no Núcleo de Ensino e Pesquisa em Agricultura Urbana da Universidade Federal do Ceará, sob o delineamento de blocos casualizados em esquema de parcelas subdivididas com quatro repetições. As parcelas foram formadas por quatro níveis de sombreamento: 0 (pleno sol), 30, 50 e 70% e as subparcelas foram formadas por cinco níveis crescentes de condutividade elétrica da água de irrigação: 0,4; 1,6; 2,8; 4,0 e 5,2 dS.m⁻¹. Aos 60 dias após a semeadura foram determinadas as taxas de fotossíntese líquida, transpiração, condutância estomática, concentração interna de CO₂ e eficiência momentânea do uso da água. O aumento da concentração de sais ocasionou danos ao aparato fotossintético das mudas de *H. impetiginosus*, no entanto, o uso do sombreamento artificial atenuou os efeitos nocivos da salinidade.

Palavras-chave: Ipê-roxo, salinidade, telas de sombreamento.

INTRODUCTION

Caatinga is one of the main Brazilian biomes, located in the Northeast region, is restricted to Brazil and covers an area of approximately 910,000 km² (SILVA et al., 2017). This vegetation is characterized by being constantly exposed to severe environmental conditions, such as water, salt, and light stress, in addition to frequent anthropic interference (SÁ et al., 2021).

When speaking of the Northeast region, it is immediately associated with the phenomenon of drought, which is one of the main abiotic factors that limit agricultural activity in arid and semi-arid regions, making irrigation indispensable to develop agricultural production (QIU et al., 2021). However, given the hydrogeological conditions of the region, we are faced with the frequent occurrence of water sources with inferior quality, which when used in irrigation can cause damage to both soil and plants. However, due to the water scarcity in the region, the use of brackish water in irrigation has become a viable alternative, provided that adequate management techniques are adopted (SABINO et al., 2021; PIRASTEH-ANOSHEH et al., 2022).

Artificial shading is another interesting strategy for the semi-arid region, since it has gained prominence due to its ability to mitigate the effects of water deficit, being able to modify the temperature inside, creating favorable conditions for plant development (ROSA et al., 2021), and can even help reduce the harmful effects of salinity. The Ipê-roxo (*Handroanthus impetiginosus*) is a deciduous species native to the Caatinga with high economic value. Because of its exuberant beauty, it is widely

used in urban afforestation and in recovery programs of degraded areas. Its wood is widely used for timber purposes, besides having high medicinal value, being also used in the pharmaceutical industry due to its analgesic and anti-inflammatory properties (RIBEIRO; COELHO, 2021). However, although this species has the ability to adapt to semi-arid conditions, the low water availability of the region can compromise its existence (SILVA et al., 2020).

The native species of the Caatinga, due to their physiological mechanisms, have a high potential for tolerance to high temperatures, water deficit, and salt stress, conditions that are predominant in the Northeast region (SABINO et al., 2021). However, information on the production of purple trumpet ipê seedlings under such conditions is still quite scarce, which makes this study extremely relevant for techniques in seedling production. Thus, the objective of the present study was to evaluate the effects of artificial shading and irrigation with brackish water on the leaf gas exchange of *Handroanthus impetiginosus* seedlings.

MATERIAL AND METHODS

The experiment was conducted in the period from October 2019 to January 2020, at the Center of Education and Research in Urban Agriculture (NEPAU) belonging to the Plant Science Department of the Federal University of Ceará (UFC), Pici Campus, Fortaleza, Ceará, Brazil, located at the geographic coordinates 03°44'17" S, 38°34'22" W and altitude of 19 m. The meteorological data obtained during the experimental period are shown in Table 1.

Table 1. Average values of temperature (°C) and relative humidity (%) of the environments (full sun, 30, 50, and 70% shade) during the experimental period

Environments	Temperature (°C)		Relative humidity (%)	
	Mín	Máx	Mín	Máx
Full Sun	34,8	29,1	63,9	53,7
Screened with 30% shading	35,3	28,7	64,1	52,4
Screened with 50% shading	34,6	28,2	64,7	51,1
Screened with 70% shading	33,5	27,9	65,6	50,3

The experimental design used was a randomized blocks in a subdivided plot scheme, with four repetitions. The plots were composed of four levels of shading, formed by a black shading screen (full sun, 30, 50, and 70%) and the subplots were composed of five increasing levels of electrical conductivity of the irrigation water (0.4, 1.6, 2.8, 4.0, and 5.2 dS m⁻¹), totaling 80 experimental units. The seedlings were produced in the greenhouse, where the seeds were sown in styrofoam trays of 128 cells filled with substrate composed of arisco + vermicompost in the proportion 2:1 and after their establishment, at 10 days after sowing, the seedlings were repotted into polyethylene plastic pots with 7 L capacity, containing a 0.5 L layer of gravel n. 0 at the bottom and filled with arisco + vermicompost + soil (classified as Red-Yellow Argissolo) in the proportion 7:1:2.

The seedlings were transplanted out in the late afternoon, in order to minimize factors that could cause stress, such as light and high temperatures. The seedlings remained in their respective shaded environments and were irrigated with water with an electrical conductivity of 0.4 dS m⁻¹ for 15 days to adapt and for their establishment not to be compromised. After this period, the salinity treatments were applied. Before the application of the saline treatments, fertilization was performed according to the recommendations of Souza et al. (2006), which consisted in applying the following doses in mg kg⁻¹ of substrate: 150 of N, 200 of P, and 150 of K. Irrigation was done manually, with a 2-day irrigation shift and the amount of water applied was determined based on the water consumption of the plants (Equation 1), through the difference between the volume of water applied and the drained volume (BERNARDO et al., 2019). A leaching fraction of 15% was adopted in order to avoid excessive salt accumulation in the root zone of the seedlings (AYERS; WESTCOT, 1999).

$$TIR = ((VA - VD) / ((1 - LF))) \quad (01)$$

Where: TIR: total irrigation required (mL); VA: water volume applied (mL); VD: water volume drained (mL); FL: leaching fraction.

The salts used to prepare the brackish solutions used in the irrigations were NaCl, CaCl₂.2H₂O and MgCl₂.6H₂O added to well water of initial electrical conductivity of 0.4 dS m⁻¹, in the proportion 7:2:1, which is a representative approximation of most water sources available for irrigation in the Northeast Region of Brazil (MEDEIROS, 1992).

At 60 days after sowing (DAS), measurements of net photosynthesis rates (A, μmol m⁻² s⁻¹), transpiration (E, mmol m⁻² s⁻¹), stomatal conductance (gs, mol m⁻² s⁻¹), and internal CO₂ concentration (C_i, ppm) were performed using an infrared gas analyzer - IRGA (LI-6400XT, Li-Cor, USA). With the photosynthesis and transpiration data, the water use efficiency (EUA) was calculated.

The data obtained was submitted to the Kolmogorov-Smirnov test to verify its normality of distribution. When the normality condition was not satisfied, the data were transformed into x0.5. Subsequently, the analysis of variance (ANOVA) was performed and when significant by the F test (p ≤ 0.05), the means were submitted to regression analysis, adopting the mathematical model based on the significance and the determination coefficient. Statistical analyses were performed with the help of the statistical software ASSISTAT version 7.7 (SILVA; AZEVEDO, 2016).

RESULTS AND DISCUSSION

Analyzing the results of variance analysis (Table 2) for gas exchange data, there was a significant interaction between shading and salinity for the variables internal CO₂ concentration (C_i), transpiration (E) and water use efficiency (EUA). Stomatal conductance (gs) and net photosynthesis (A) were statistically significant at the 1% level for the factors alone.

Table 2. Summary of the analysis of variance concerning the data of stomatal conductance (g_s), internal CO_2 concentration (C_i), net photosynthesis (A), transpiration (E) and water use efficiency (EUA) of *Handroanthus impetiginosus* seedlings under artificial shading and irrigation with brackish water.

SV	DF	Mean Square				
		g_s	C_i	A	E	EUA
Blocks	3	0,022*	1051,9	0,377	0,359	0,485
Shading (A)	3	0,149**	4542,5**	74,92**	0,895**	4,813**
Residue (a)	9	0,005	488,74	3,398	0,327	0,292
Salinity (B)	4	0,062**	7840,7**	47,76**	0,414**	1,705**
Interaction (A x B)	12	0,001 ^{ns}	1077,5**	2,301 ^{ns}	0,053**	0,924*
Residue (b)	48	0,003	353,3	1,538	0,016	0,371
CV (%) A		33,07	6,87	28,96	11,84	21,05
CV (%) B		26,50	5,84	19,48	8,19	23,71

SV: sources of variation; DF: degrees of freedom; CV: coefficient of variation; *: significant at 5% probability level; **: significant at 1% probability level; NS: not significant

It is observed that the mean g_s fitted a quadratic polynomial model with increasing shading levels (Figure 1A). The highest

stomatal conductance ($0.28 \text{ mol m}^{-2} \text{ s}^{-1}$) was obtained at 57% shading.

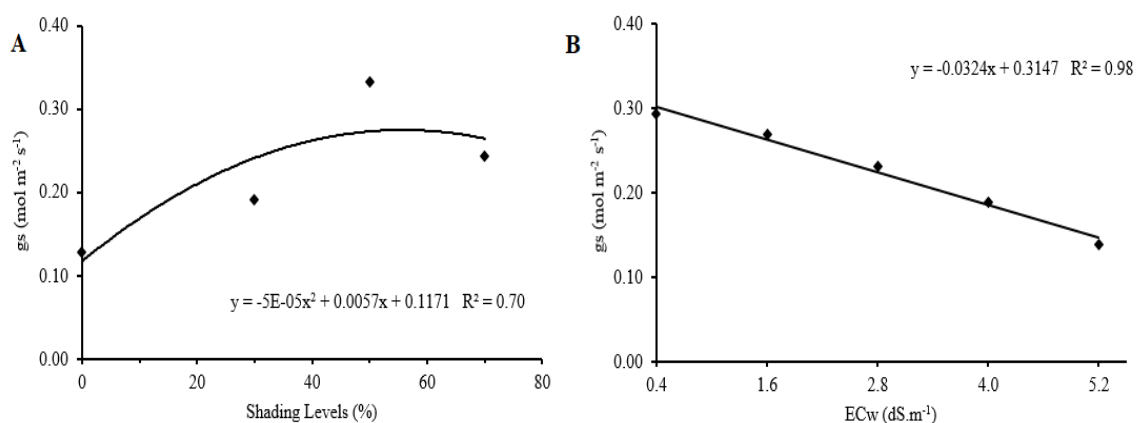


Figure 1. Stomatal conductance (g_s) in *Handroanthus impetiginosus* seedlings under artificial shading (A) and electrical conductivity of irrigation water (EC_w) (B)

The higher vapor pressure deficit (VPD) in the full sun and 30% shaded environments may have been responsible for the lower stomatal conductance values in these environments, resulting from the response mechanisms to the imposed abiotic stresses (TAIZ et al., 2017). With respect to salinity (Figure 1B), a linear decreasing behavior of g_s

was observed with increasing water electrical conductivity, with a reduction of $0.03 \text{ mol m}^{-2} \text{ s}^{-1}$ for each unitary increment of EC_w . Stomatal conductance is related to the water dynamics of the plant, thus, the lower the availability of water, either by the higher incidence of solar radiation or by the osmotic effect resulting from salt stress, the lower the g_s

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values will be, in order to avoid the loss of tissue turgescence (ROSA et al., 2017; LIMA et al., 2020).

The internal CO₂ concentration was significantly ($p < 0.01$) influenced by the

interaction between the shading and salinity factors. There was an increase in the average C_i of the seedlings of all environments with the increase of the electrical conductivity of the water (Figure 2).

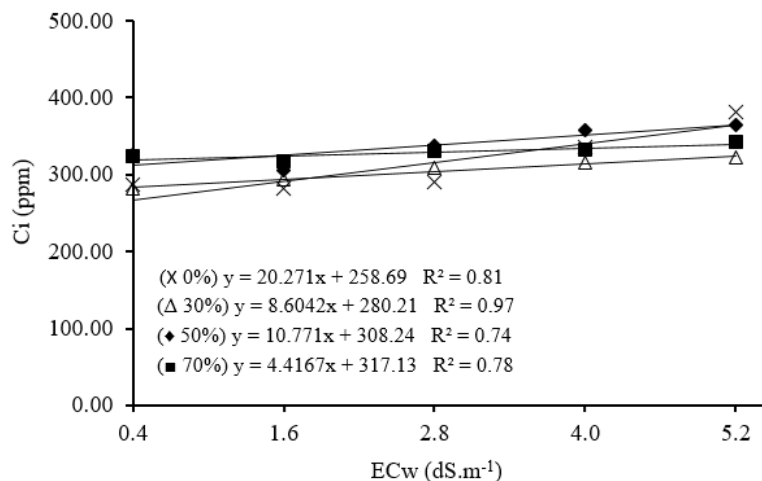


Figure 2. Internal CO₂ concentration (C_i) in *Handroanthus impetiginosus* seedlings under artificial shading and irrigation water electrical conductivity (ECw)

It is possible to observe that the highest internal CO₂ concentrations were observed in the 50 and 70% shaded environments from the application of the control treatment until the concentration of 4.0 dS m⁻¹. This behavior is directly related to the higher stomatal conductance observed in these environments, since it favors the entry of carbon and consequently increases its concentration inside the stomatal cells (TAIZ et al., 2017).

However, the full sun environment showed higher C_i values in the treatment with higher salinity (5.2 dS m⁻¹), possibly due to damage to the photosynthetic apparatus of the seedlings, caused by the higher concentration of salts applied, causing this available CO₂ not to be used in the photosynthetic process, as can be seen in Figure 3, resulting in increased C_i within the stomatal cells. In addition, the higher temperatures observed in the full sun environment may also have contributed to the inhibition of enzymes involved during the CO₂ fixation process.

Silva et al. (2017) point out that when plants are subjected to the influence of two or more simultaneous abiotic factors, one can potentiate the negative effects of the other, compromising the establishment of species that

are not adapted to their environmental conditions.

High C_i values combined with low g_s values, evidenced mainly in the full sun environment, are an indication that the CO₂ fixation process during the carboxylation stage was compromised, and may be related to damage in the photosynthetic apparatus (OLIVEIRA et al., 2020). Ribeiro et al. (2021) also observed reductions in photosynthesis and stomatal conductance of *Handroanthus impetiginosus* seedlings even with the increase in internal CO₂ concentration, where the authors attributed this behavior to the lower concentration of ATP and NADPH, due to a reduction in the photochemical phase of photosynthesis. Photosynthesis showed statistical significance at the 1% probability level for the shading and salinity factors alone (Figure 3A). With respect to shading levels, a quadratic polynomial fit of the averages of A was observed with a maximum value obtained with 45% shading (7.74 $\mu\text{mol m}^{-2} \text{s}^{-1}$). For salinity, there was a linear decreasing behavior of the photosynthesis averages with increasing ECw, with a reduction of 0.94 $\mu\text{mol m}^{-2} \text{s}^{-1}$ for each unit increase in the electrical conductivity of the irrigation water.

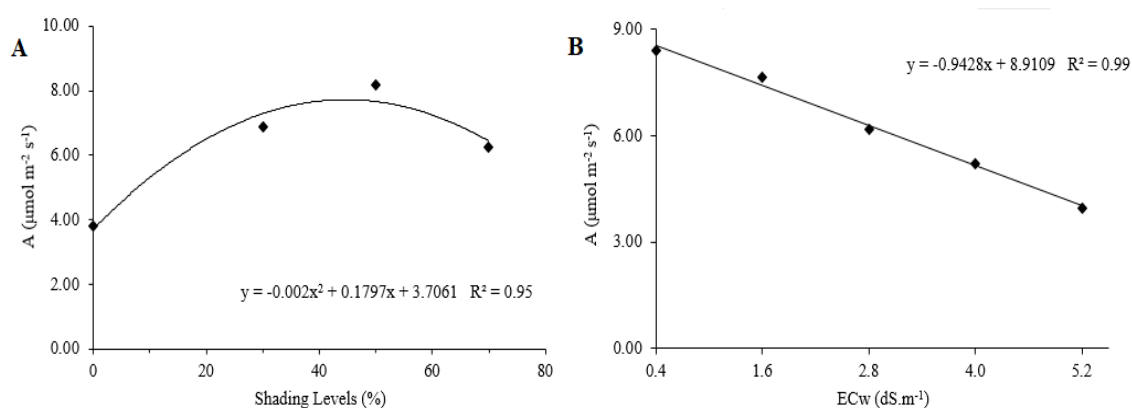


Figure 3. Photosynthetic rate (A) in *Handroanthus impetiginosus* seedlings under artificial shading (A) and electrical conductivity of irrigation water (CEw) (B)

Regarding the environments, it is observed that even in those that had higher levels of incident radiation there was availability of CO₂ (Figure 2) for the photosynthetic process, however, the lowest values of A were observed in the plants that were in these environments. Possibly, the higher temperatures may have inhibited CO₂ assimilation in the seedlings, since temperatures above 30° inhibit CO₂ assimilation in C3 plants, such as purple ipê, reducing the activation of RuBisCo and consequently the photosynthetic process (OLIVEIRA et al., 2020). Similar results were observed by Bessa et al., (2017) who observed that salinity caused reductions in net photosynthesis of *Handroanthus impetiginosus* seedlings, with average values of 6.83 $\mu\text{mol m}^{-2} \text{s}^{-1}$. With respect to salinity, it is observed that there was a slight increase in C_i with the increase in the electrical conductivity of the water. From this result it is possible to infer that the main responsible for the reduction in photosynthetic rate (Figure 3B) were not stomatal limitations, since CO₂ was not a limiting factor, allowing us to attribute this

reduction to the inhibition of photochemical and enzymatic processes related to carbon assimilation (LACERDA et al., 2020). If the internal CO₂ concentration is increasing, then the carbon that is reaching the mesophyll cells is not being fixed in the carboxylative phase, possibly because of damage to its structure due to excess salts, consequently causing decreases in photosynthetic rate (PEREIRA et al., 2020). Lima et al. (2017) point out that in addition to the osmotic effect, another factor that may be responsible for reduced photosynthesis in plants exposed to salt stress is the accumulation of Na⁺ and Cl⁻ ions, which are primarily responsible for damage to enzyme and membrane structures.

Transpiration was influenced by the interaction between the shading and salinity factors ($p < 0.05$). It can be seen that regardless of the level of shading, all environments showed linear reductions with the increase in the electrical conductivity of the water. With increasing EC_w, decreases of 0.39, 0.23, 0.13, and 0.20 $\text{mmol m}^{-2} \text{s}^{-1}$ were observed for the full sun, 30, 50, and 70% shade environments, respectively (Figure 4).

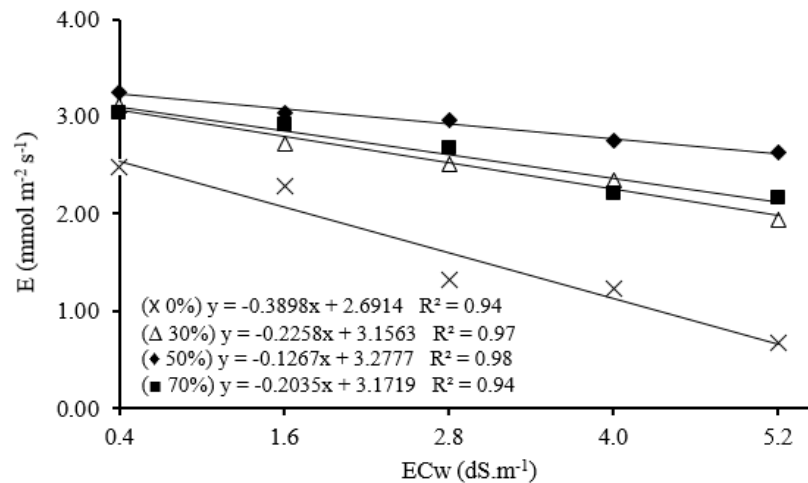


Figure 4. Transpiration (E) in *Handroanthus impetiginosus* seedlings under artificial shading and irrigation water electrical conductivity (CEw)

The increase in transpiration in the 50% shade environment indicates that there was possibly a greater stomatal opening of the plants in this environment, while in the full sun environment, the soil absorbs a greater amount of thermal energy, increasing water evaporation and creating a water deficit condition. The reduction of transpiration rate is a strategy of plants in order to tolerate stress, from osmotic adjustment (CAMPELO et al., 2015). The aforementioned authors also observed reductions in transpiration of yellow ipê seedlings, due to stomatal closure as a function of the water availability condition.

Thus, the higher VPD in environments with higher incidence of radiation promotes a condition of low water availability, inducing stomatal closure with consequent reduction in transpiration rate.

The momentary water use efficiency (EUA) was significant at the 5% probability level for the interaction between shading and salinity (Figure 5).

It can be seen that the mean EUA of the seedlings that were in full sun showed a quadratic fit while the seedlings that were under the other shading levels showed a linear decreasing behavior with increasing ECw.

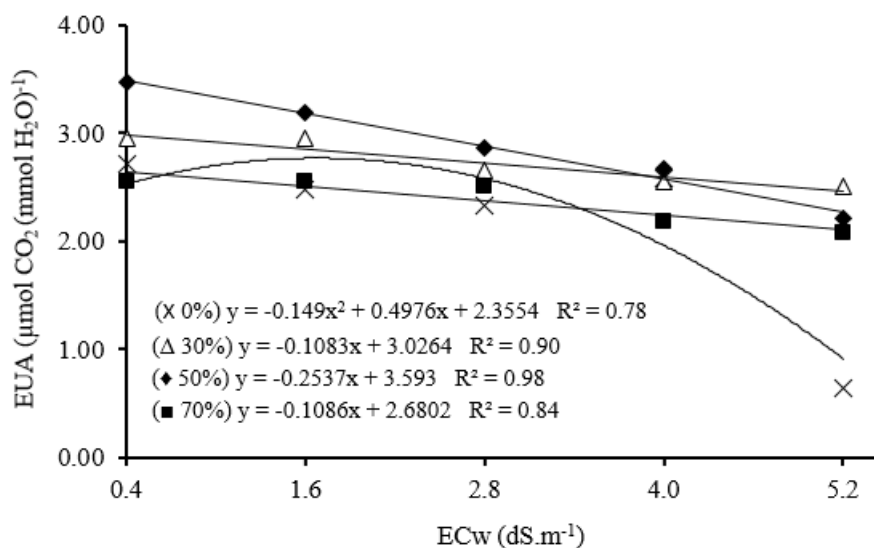


Figure 5. Water use efficiency (EUA) in *Handroanthus impetiginosus* seedlings under artificial shading and irrigation water electrical conductivity (CEw)

It was observed that the seedlings that received the higher salinity treatments showed clear symptoms of toxicity (Figure 5), causing a restriction of the photosynthetic rate even when CO₂ was available. Thus, reduced photoassimilate production occurred, negatively affecting their development by reducing biomass production per unit of transpired water and consequently restricting *EUA* (HATFIELD; DOLD, 2019). However, it is observed that even with the increase in salinity, the seedlings in the 50% shaded environment presented a greater *EUA* when compared to those in full sun, showing that artificial shading is able to attenuate the harmful effects of salts in the irrigation water.

CONCLUSIONS

The highest levels of electrical conductivity of the irrigation water (4.0 and 5.2 dS m⁻¹) caused the greatest reductions in leaf gas exchange, showing damage to the photosynthetic apparatus of *Handroanthus impetiginosus* seedlings, compromising photosynthesis and consequently their development.

Artificial shading proved to be advantageous for the development of *Handroanthus impetiginosus* seedlings under salinity conditions, with the black screen with 50% shading being the one that most favored leaf gas exchange, allowing it to tolerate an electrical conductivity of up to 2.8 dS m⁻¹.

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