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Physiology and growth of maize under salinity of water and application of hydrogen peroxide[1]

Fisiologia e crescimento do milho sob salinidade da água e aplicação de peróxido de hidrogênio

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HIGHLIGHTS:

- *Electrical conductivity of water (ECw) of 2.0 dS m^{-1} reduces growth and chlorophyll content in maize.*
- *Hydrogen peroxide up to a concentration of 171.66 μmol L^{-1} increases leaf area and total biomass in maize.*
- *Salt stress combined with high H_2O_2 concentrations inhibits stomatal conductance, transpiration, and CO_2 assimilation rate.*

ABSTRACT: The salinity of irrigation water or soil is the abiotic factor that most negatively impacts the yield of crops, including green maize, so it is necessary to find alternatives to ensure production. In this context, this study aimed to evaluate gas exchange, photosynthetic pigments, and growth of maize under salinity of irrigation water and application of hydrogen peroxide. The experiment was conducted at the Center of Sciences and Agrifood Technology, Pombal, PB, belonging to the Federal University of Campina Grande, in the period from January to February 2015, using the maize hybrid 'AG 1051'. The treatments consisted of two salinity levels of irrigation water (0.3 and 2.0 dS m^{-1}) and five concentrations of hydrogen peroxide (0, 40, 80, 160, and 320 μmol L^{-1}) applied via irrigation water. The experimental design was completely randomized, in a 2 × 5 factorial scheme, with four replicates. ECw of 2.0 dS m^{-1} reduces transpiration, stomatal conductance, total chlorophyll, carotenoids, and initial growth, but does not affect the dry mass accumulation of maize plants. Application of H_2O_2 via soil varying from 0 to 320 μmol L^{-1} causes reductions in the CO_2 assimilation rate and transpiration, as well as at concentrations from 0 to 160 μmol L^{-1} for stomatal conductance of plants irrigated with ECw of 2.0 dS m^{-1}. Application of H_2O_2 via soil up to a concentration of 320 μmol L^{-1} increases the plant height, but reduces culm diameter of maize.

Key words: *Zea mays* L., saline water, mitigator

RESUMO: A salinidade da água de irrigação ou do solo são fatores abióticos que mais impacta negativamente na produtividade das culturas, entre elas, o milho verde, sendo necessário encontrar alternativas para garantir a produção. Nesse contexto, objetivou-se avaliar as trocas gasosas, os pigmentos fotossintéticos e o crescimento do milho sob salinidade da água de irrigação e aplicação de peróxido de hidrogênio. O experimento foi conduzido no Centro de Ciências e Tecnologia Agroalimentar, Pombal, PB, pertencente à Universidade Federal de Campina Grande, no período de janeiro a fevereiro de 2015, utilizando-se do híbrido de milho 'AG 1051'. Os tratamentos foram constituídos por dois níveis de salinidade da água de irrigação (0,3 e 2,0 dS m^{-1}) e cinco concentrações de peróxido de hidrogênio (0, 40, 80, 160 e 320 μmol L^{-1}) aplicadas via água de irrigação. O delineamento experimental foi o inteiramente casualizado, no esquema fatorial 2 × 5, com quatro repetições. A CEa de 2,0 dS m^{-1} reduz a transpiração, condutância estomática, clorofila total, carotenóides e crescimento inicial, mas não afeta o acúmulo de massa seca das plantas de milho. A aplicação de H_2O_2 via solo variando de 0 a 320 μmol L^{-1} promoveu redução na taxa de assimilação de CO_2 e transpiração, bem como em concentrações de 0 a 160 μmol L^{-1} para condutância estomática de plantas irrigadas com CEa de 2,0 dS m^{-1}. Aplicação de H_2O_2 via solo até a concentração de 320 μmol L^{-1} aumentou a altura de plantas, porém reduziu o diâmetro do colmo de milho.

Palavras-chave: *Zea mays* L., águas salinas, atenuante

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Introduction

In the current scenario of climate change, the low and irregular distribution of rainfall, together with high temperatures and high evapotranspiration, has limited the availability of good quality water in semi-arid regions. As an alternative to meet the water needs of crops, it is necessary to use irrigation; however, when managed improperly, it can lead to soil salinization, reducing the availability of water for plants, in addition to causing problems such as nutritional imbalance and toxicity of specific ions (Kaya et al., 2015b).

In physiology, the first response of the plant to salt stress is stomatal closure, as a way to prevent water loss, resulting in changes in stomatal conductance, internal carbon concentration, transpiration, and CO_2 assimilation rate (Arif et al., 2020). In addition to limiting gas exchange, stomatal closure induces the production of reactive oxygen species (ROS) due to the imbalance between the photochemical phase and the Calvin cycle, with consequences for plant metabolism (Taiz et al., 2017; Foyer, 2018).

However, it is possible to consider that the production of maize in regions with high levels of salts in the soil and/or water can be optimized using strategies to mitigate the deleterious effects of salinity, since this crop is moderately sensitive, with water salinity threshold of 1.1 dS m^{-1} and soil salinity (saturation extract) threshold of 1.7 dS m^{-1}. In this regard, the application of hydrogen peroxide (H_2O_2) can increase the plant's ability to survive adverse conditions (Silva et al., 2019b), following the example of studies conducted by Gondim et al. (2013) and Silva et al. (2016), who evaluated maize plants from seeds pre-treated with H_2O_2 and subjected to salinity and observed a reduction in the sensitivity of plants to salinity.

Thus, it is necessary to assess the effectiveness of H_2O_2 in activating the plant's defense system and its effect on initial growth and gas exchange. This study aimed to evaluate gas exchange, photosynthetic pigments, and growth of maize under salinity of irrigation water and application of hydrogen peroxide.

Material and Methods

The study was conducted during January and February 2015, under open sky conditions, at the Center for Sciences and Agrifood Technology of the Federal University of Campina Grande (CCTA/UFCG), located in Pombal, PB, Brazil, whose altitude is 144 m, and coordinates 6° 48' 16" S and 37° 49' 15" W. The climatic characteristics recorded during the experiment are shown in Figure 1.

The treatments consisted of a combination of two levels of irrigation water salinity (0.3 and 2.0 dS m^{-1}) and five concentrations of hydrogen peroxide (0, 40, 80, 160, and 320 μmol L^{-1}), forming a 2 × 5 factorial, totaling 10 treatments and 40 experimental units, distributed in a completely randomized design, with four repetitions. The experimental unit consisted of two plants per pot. The salinity levels were based on the threshold water salinity of the maize crop (1.1 dS m^{-1}), selecting a low salinity level and another level above the threshold. Hydrogen peroxide concentrations were based on the study conducted by Godim et al. (2013) in maize plants using seeds pre-treated with hydrogen peroxide, but we used lower concentrations.

Considering that in the literature there are already studies with peroxide application via leaves and they obtained promising results, we chose to test the application via irrigation water, to check if it has the same effectiveness.

Three applications of hydrogen peroxide were made, one per day, during the first three days after sowing (DAS) via irrigation water, using an A.R. source. The H_2O_2 solutions were prepared from a 1 mmol L^{-1} solution of H_2O_2, based on molecular weight, until reaching the desired concentrations. In each event, 0.3 L of the hydrogen peroxide solution was applied. As the application of the product was carried out via irrigation water, it was not necessary to isolate the treatments, since there was no possibility of drift.

The cultivation was performed in pots with capacity of eight cubic decimeters (8 dm^3), filled with soil classified as Vertisol, with clayey texture and cation exchange capacity (CEC): 49.15 $cmol_c\ dm^{-3}$; exchangeable sodium percentage (ESP): 2%; pH (in soil:water 1:2.5): 7.14; electrical conductivity of the

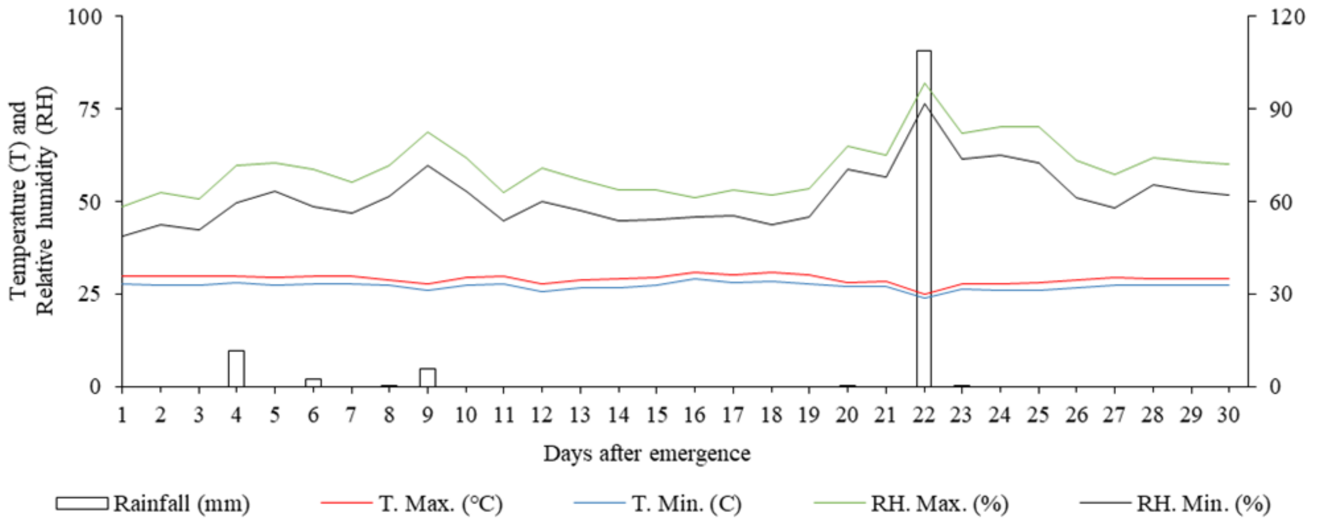


Figure 1. Maximum (T max.) and minimum (T min.) temperature (°C), maximum and minimum relative humidity of air (RH - %), and rainfall (mm) observed during the experimental period

saturation extract (EC): 0.64 dS m^{-1}; P: 98.7 mg dm^{-3}; K: 399.5 mg dm^{-3}, Na: 214.3 mg dm^{-3}; Ca: 36.60 $cmol_c$ dm^{-3}; Mg: 10.60 $cmol_c$ dm^{-3}; H + Al: 0 $cmol_c$ dm^{-3}.

The maize seeds used were the double-crossed hybrid 'AG 1051', as it is a genetic material indicated for the production of green corn and silage. Five seeds were sown per pot at a depth of 2.0 cm. Emergence in more than 50% of the plants was observed four days after sowing, and thinning was performed seven days after sowing, leaving two plants per pot.

The irrigations were performed manually twice a day (at 8:00 a.m. and 5:00 p.m.), and the volume to be applied was determined by drainage lysimetry. The amount of water applied per pot varied from 0.5 to 2.0 L per day during the experiment. The irrigation water with 2.0 dS m^{-1} was prepared by adding NaCl to the local municipal water, which had an electrical conductivity of 0.3 dS m^{-1}, based on the relationship between ECw and the concentration of salts ($mmol_c$ L^{-1} = 10 × ECw) (Richards, 1954). Irrigation with water of different salinities began at 10 DAS, when the emergence of plants had stabilized.

For nutritional management, macro and micronutrients were applied in the amounts of 730, 495, and 748 mg dm^{-3} of the soil of N, P, and K, respectively, in the form of urea, ammonium sulfate, monopotassium phosphate, and potassium chloride, divided into 4 applications, the first as basal and the others at 8, 18, and 28 days after emergence (DAE), via fertigation. For fertilization with micronutrients, boric acid and zinc sulfate were used as sources. Cultural practices and phytosanitary control were carried out whenever necessary in accordance with the needs of the crop (Cruz & Pereira Filho, 2002).

The evaluations were performed at 30 DAE. At this time, gas exchange was determined based on CO_2 assimilation rate (A - μmol CO_2 m^{-2} s^{-1}), stomatal conductance (gs - mol H_2O m^{-2} s^{-1}), transpiration (E - mmol H_2O m^{-2} s^{-1}), and intercellular CO_2 concentration (Ci - μmol CO_2 mol^{-1}), measured with an infrared gas analyzer (IRGA) LCpro (Analytical Development, Kings Lynn, UK), using a constant light source of 1200 μmol photons m^{-2} s^{-1} and CO_2 concentration of the environment. For the IRGA readings, the intermediate leaves were chosen, the 4th or 5th leaf counted from the apex of the plant, so as not to underestimate the values using old or very young leaves, evaluating one plant per plot. Readings were taken between 7 and 9 a.m.

To determine the contents of chlorophyll and carotenoids, leaf samples were collected at 30 DAE and sent to the Plant Physiology Laboratory of CCTA/UFCG to extract the pigments in 80% acetone. After being macerated and filtered in 0.45-micron filter paper, they were quantified by spectrophotometry, as described in Lichtenthaler (1987).

For the evaluation of growth and accumulation of dry mass, at 30 DAE the plants were collected by cutting them close to the ground. In these plants, plant height, culm diameter, number of leaves per plant, leaf area, leaf and culm dry mass, and total dry mass of the shoot were evaluated. Plant height was measured with a tape measure, from the collar, that is, approximately close to the ground, to the apex of the plant. Culm diameter was measured with a digital Vernier caliper. The number of leaves was obtained by counting only the fully formed leaves (visible ligule).

The leaf area (cm^2 per plant) was determined from the collection of eight leaf discs of known area (1.4 cm^2), summing the area of all discs, multiplying the value by the leaf dry mass, and dividing the result by the dry mass of the discs. The discs were dried in an oven with forced-air circulation at 70 °C for 72 hours.

The total dry biomass was determined by adding the dry mass of the leaves and the culm, also obtained after drying in an oven with forced air circulation at 65 °C for 72 hours.

For statistical analyses, the data were preliminarily subjected to the normality test (Kolmogorov-Smirnoff) using Sisvar software (Ferreira, 2019), followed by an analysis of variance at 0.05 probability level and, in cases of significance, linear and quadratic regression analysis was performed, using the statistical program SAEG, Version 9.1.

RESULTS AND DISCUSSION

There were significant effects of isolated factors as well as that of interaction between irrigation water salinity and H_2O_2 levels for CO_2 assimilation rate (A), transpiration (E), stomatal conductance (gs), and intercellular CO_2 concentration (Ci) ($p \leq 0.01$).

It was observed that the plants irrigated with water of 0.3 dS m^{-1} obtained a higher CO_2 assimilation rate (29.5 μmol CO_2 m^{-2} s^{-1}) at the estimated H_2O_2 concentration of 71 μmol L^{-1}. For plants irrigated with 2.0 dS m^{-1} water, the data did not obtain adequate fits ($y = 23.928 - 0.0011^{ns}x$, $R^2 = 0.0194$), with an average CO_2 assimilation rate of 23.8 μmol CO_2 m^{-2} s^{-1}, a value considered normal for plants with C4 metabolism (Figure 2A). According to Silva et al. (2021a), when H_2O_2 is applied at an appropriate concentration, it stimulates some physiological processes, including CO_2 assimilation rate and stomatal conductance.

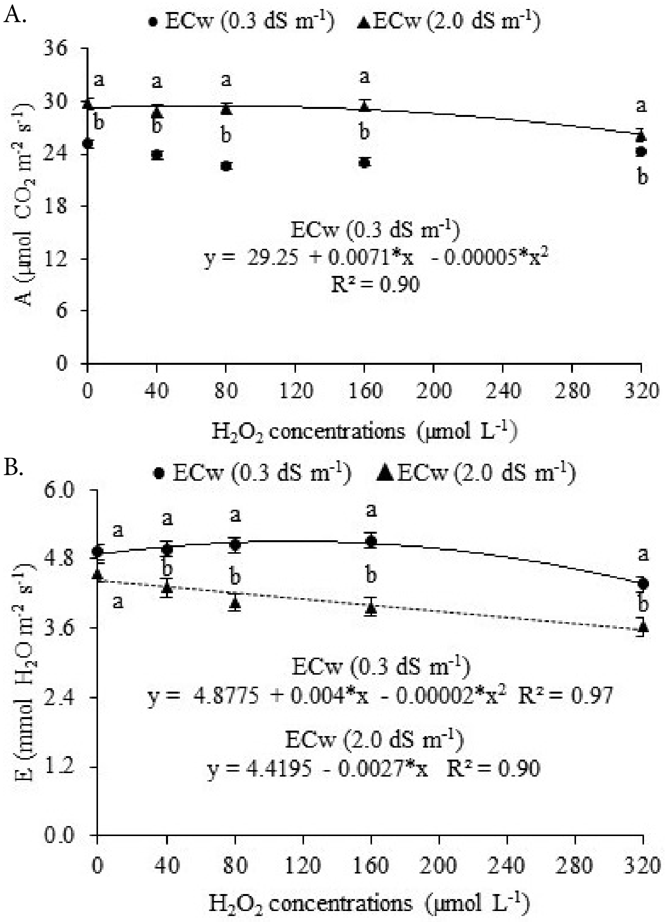
When analyzing the interaction between the electrical conductivity of irrigation water and hydrogen peroxide concentrations (Figure 2A), it was observed that the CO_2 assimilation rate of plants subjected to ECw of 0.3 dS m^{-1} differs significantly from that of plants irrigated with water of 2.0 dS m^{-1}. Plants grown under ECw of 0.3 dS m^{-1} stood out with the highest CO_2 assimilation rate, regardless of the concentration of H_2O_2 applied.

The low CO_2 assimilation rate, recorded in plants subjected to ECw of 2.0 dS m^{-1}, occurred due to the osmotic adjustment that the plant performs to continue the metabolic processes (Sousa et al., 2021); no benefit was observed from the application of H_2O_2 in attenuating the deleterious effects

Table 1. Summary of F test for CO_2 assimilation rate (A), transpiration (E), stomatal conductance (gs), and intercellular CO_2 concentration (Ci) in maize plants subjected to different levels of irrigation water salinity and concentrations of hydrogen peroxide (H_2O_2) 30 days after emergence

Source of variation	F Test			
	A	E	gs	Ci
Salinity (ECw)	**	**	**	**
Hydrogen peroxide (H_2O_2)	**	**	**	*
Interaction (ECw × H_2O_2)	**	**	**	**
CV (%)	3.60	9.07	7.98	15.99

CV (%) - Coefficient of variation; ** - significant at 0.01 probability level, by the F test



* - Significant at $p \leq 0.05$ by F test. Means of ECw compared by F teste at $p \leq 0.05$. Means followed by the same letter for determined concentration of H_2O_2 indicate no significant difference between them. Vertical bars indicate standard error of mean (n = 4)

Figure 2. CO_2 assimilation rate (A) and transpiration (B) of maize plants subjected to different salinity levels (ECw) of irrigation water and concentrations of hydrogen peroxide (H_2O_2), 30 days after emergence

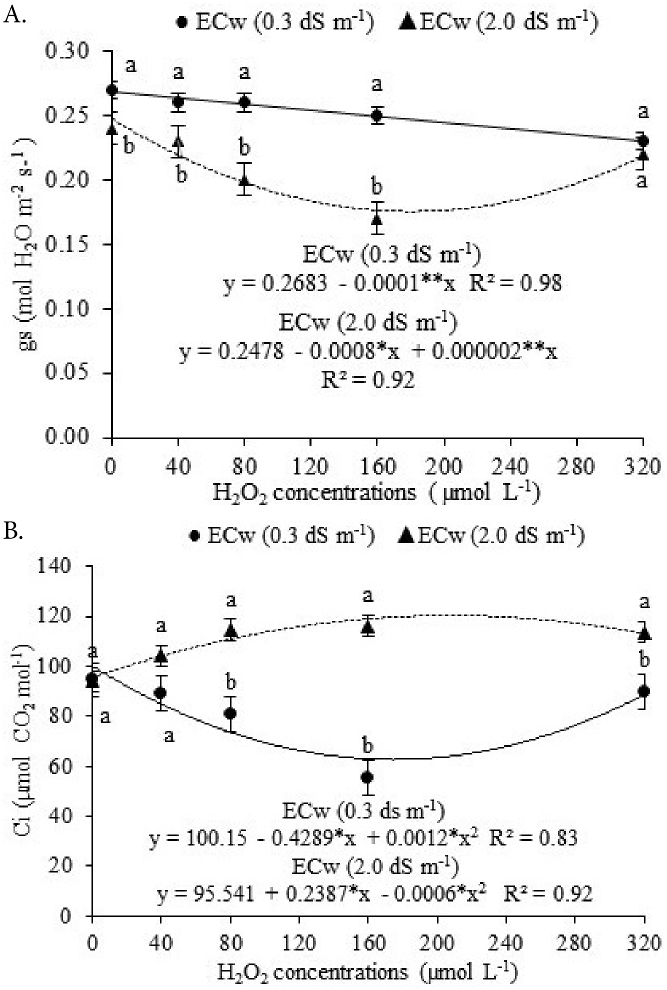
of salinity on this physiological parameter. Such results are in agreement with those found by Sousa et al. (2021) who evaluated the influence of nitrogen doses on growth and gas exchange of the 'AG 1051' maize crop irrigated with saline water and observed that the ECw of 3.0 dS m^{-1} reduced the parameters of gas exchange, in particular the CO_2 assimilation rate, at 30 DAS.

Regarding transpiration, when the plants were irrigated with water of 0.3 dS m^{-1}, there was a quadratic behavior with the increase in the concentration of H_2O_2, with maximum transpiration of 5.07 mmol H_2O m^{-2} s^{-1} at the estimated H_2O_2 concentration of 100 μmol L^{-1} (Figure 2B). When the plants were irrigated with water of 2.0 dS m^{-1}, there was a reduction in the transpiration rate of 2.44% for each 40 μmol L^{-1} increase of H_2O_2 (Figure 2B). Thus, under stress conditions, the increase in the concentration of this compound may have helped the plants to identify the stress and reduce the transpiration rate, especially because there was no significant effect on CO_2 assimilation rate, inducing acclimation of plants to salt stress.

When analyzing the effect of ECw at each concentration of H_2O_2 (Figure 2B), it was observed that transpiration was negatively influenced by ECw, with lower values obtained at ECw of 2.0 dS m^{-1}, with greater intensity at concentrations of 160 and 320 μmol L^{-1}. This can be explained by the fact that, when plants are under stress, there is an increase in the production of ROS, including H_2O_2, which in excess can induce toxicity and plant death. Thus, the application of high concentrations of H_2O_2 probably may have contributed to the reduction of transpiration of the plants. Significant differences were observed in transpiration rate at all doses of peroxide, except for the control treatment.

For stomatal conductance (Figure 3A), it was found that the increase in the concentration of H_2O_2 caused a reduction in the values of 'gs' at both levels of salinity (ECw = 0.3 and 2.0 dS m^{-1}), an interesting fact because when the plants were irrigated with low-salinity water, the decrease in CO_2 assimilation rate and transpiration only occurred when they were under the concentration of 320 μmol L^{-1}, which denotes an adaptation. When plants were irrigated with water of higher salinity, the behavior of gs was similar to that of transpiration, emphasizing that the CO_2 assimilation rate was not affected, which may be related to greater efficiency in the use of CO_2.

When analyzing the irrigation water salinity levels at each H_2O_2 concentration (Figure 3A), there was a reduction in gs with the ECw 2.0 dS m^{-1} at the concentrations of 0, 40, 80, and 160 μmol L^{-1}, compared to the ECw of 3.0 dS m^{-1}.



**, * significant at $p \leq 0.01$, 0.05, respectively by F test. Means of ECw compared by F test at $p \leq 0.05$. Means followed by the same letter for determined concentration of H_2O_2 indicate no significant difference between them. Vertical bars indicate standard error of mean (n = 4)

Figure 3. Stomatal conductance (A) and intercellular CO_2 concentration (B) of maize plants subjected to different salinity levels (ECw) of irrigation water and concentrations of hydrogen peroxide (H_2O_2), 30 days after emergence

For intercellular CO_2 concentration, there was a significant difference between salinity levels for H_2O_2 concentrations 80, 160, and 320 μmol L^{-1}. When studying the intercellular concentration of CO_2, one notices an opposite behavior to that of CO_2 assimilation rate. In this context, when detailing the result observed in plants irrigated with water of 0.3 dS m^{-1} (Figure 3B), one notices that the lowest values were observed with the application of 178.70 μmol L^{-1} of H_2O_2; in this case, the reduction in Ci and the increase in CO_2 assimilation rate mean an increase in the efficiency of carboxylation, a situation raised when studying the 'gs', which was reduced with the increase in the concentration of H_2O_2, that is, the increase of this compound may have enabled better adaptation of the plants to the environment that, by itself, can cause other abiotic stresses by temperature and relative humidity of the air (Taiz et al., 2017).

For plants irrigated with water of 2.0 dS m^{-1}, the highest intercellular CO_2 concentration was 119.28 μmol CO_2 mol^{-1} at a concentration of 198.91 μmol L^{-1} H_2O_2, promoting a 26.08% increase compared to the concentration of 0 μmol L^{-1} of H_2O_2 (Figure 3B); in this case, as there was no reduction in CO_2 assimilation rate, but there was a decrease in the values of 'gs' and 'E', it can be said that the accumulated CO_2 was strategic for the acclimatization of plants, ensuring its absorption and assimilation.

The physiological parameters serve to check for alterations in plants subjected to stress conditions. Therefore, the reduction observed in these variables when the ECw was increased from 0.3 to 2.0 dS m^{-1} is common; however, it was expected that the increase in the concentration of H_2O_2 could be more significant in favoring these variables, alleviating the salt stress in pre-treated maize plants, as verified by Gondim et al. (2013), who reported that the pre-treatment of seeds with H_2O_2 was able to increase the values of photosynthetic rate and stomatal conductance, compared to plants that did not have pre-treated seeds.

The application of H_2O_2 at low concentrations, as a pretreatment before subjecting plants to abiotic stresses, activates physiological responses that allow them to tolerate salinity (Gondim et al., 2012; Silva et al., 2016). Silva et al. (2021a) observed that the use of hydrogen peroxide, via soaking of seeds of passion fruit plants at concentrations between 10 and 30 μM, mitigated the deleterious effects of salinity on stomatal conductance, transpiration, CO_2 assimilation rate, and instantaneous carboxylation efficiency.

Silva et al. (2019a), when studying the foliar application of hydrogen peroxide in yellow passion fruit under salinity, observed that the 25 μM concentration favored transpiration, CO_2 assimilation rate, and internal carbon concentration of the plants.

In this study, when plants were irrigated with water of electrical conductivity 2.0 dS m^{-1}, a reduction in some physiological parameters was observed in plants from the H_2O_2 treatment, but the CO_2 assimilation rate was maintained. This effect is related to the association of salt stress together with the high concentration of H_2O_2 above 70 μmol L^{-1}, because H_2O_2 is a ROS which in turn is already produced naturally in the plant; however, in excess ROS can induce toxicity and plant death (Petrov & van Breusegem, 2012; Smirnoff & Arnaud, 2019).

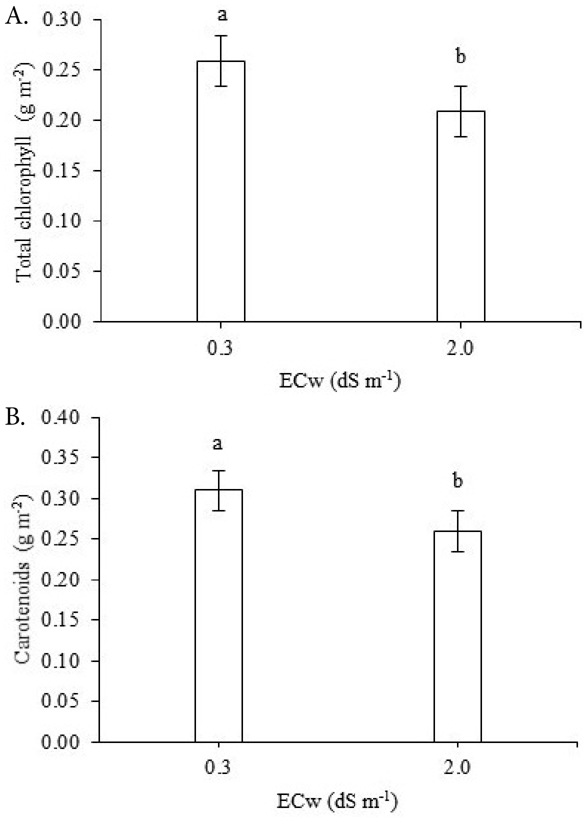
There was no significant interaction between irrigation water salinity and H_2O_2 concentration for chlorophyll a, chlorophyll b, total chlorophyll, and carotenoid contents in maize plants. However, there were significant effects of irrigation water salinity levels on total chlorophyll and carotenoids. For hydrogen peroxide concentrations, no significant effect was observed (Table 2).

When studying the total chlorophyll and carotenoid contents as a function of salinity, it was noted that the increase in the concentration of salts in the irrigation water caused a reduction in the contents of these variables, that is, there was a degradation of total chlorophyll and carotenoids with increasing salinity, a fact that explains the lower gas exchange observed in plants under salt stress, regardless of the concentration of H_2O_2 applied (Figure 4). The decrease in pigment synthesis in plants

Table 2. Summary of F test for chlorophyll a (Chl a), chlorophyll b (Chl b), total chlorophyll (Chl t) and carotenoids (Car) in maize plants subjected to different levels of irrigation water salinity and concentrations of hydrogen peroxide (H_2O_2) 30 days after emergence

Source of variation	F Test			
	Chl a	Chl b	Chl t	Car
Salinity (ECw)	ns	ns	**	*
Hydrogen peroxide (H_2O_2)	ns	ns	ns	ns
Interaction (ECw × H_2O_2)	ns	ns	ns	ns
CV (%)	17.98	18.11	20.10	20.27

CV (%) - Coefficient of variation; * and **- significant at 0.05 and 0.01 probability level, respectively, by the F test



Means compared by F test at $p \leq 0.05$. Means followed by same letter indicate no significant difference between them. Vertical bars indicate standard error of mean (n = 4)

Figure 4. Total chlorophyll (A) and carotenoids (B) contents of maize plants subjected to different levels of irrigation water salinity and concentrations of hydrogen peroxide (H_2O_2), 30 days after emergence

under high salinity conditions possibly stimulated the activity of the enzyme chlorophyllase, which acts on the degradation of photosynthetic pigments and destroys the structure of chloroplasts, also causing imbalance and loss of activity of pigment proteins (Silva et al., 2021b; Lima et al., 2021).

High levels of carotenoids can contribute to the maintenance of chlorophyll levels in plants under salt stress (Kim et al., 2012; Kachout et al., 2013). Thus, it can be said that the reduced contents of carotenoids may have contributed to the non-significance in the contents of chlorophyll a and b, especially in plants irrigated with water of 2.0 dS m^{-1}, since the concentration of total chlorophyll in maize plants decreases with increase in water salinity (Kaya et al., 2015a).

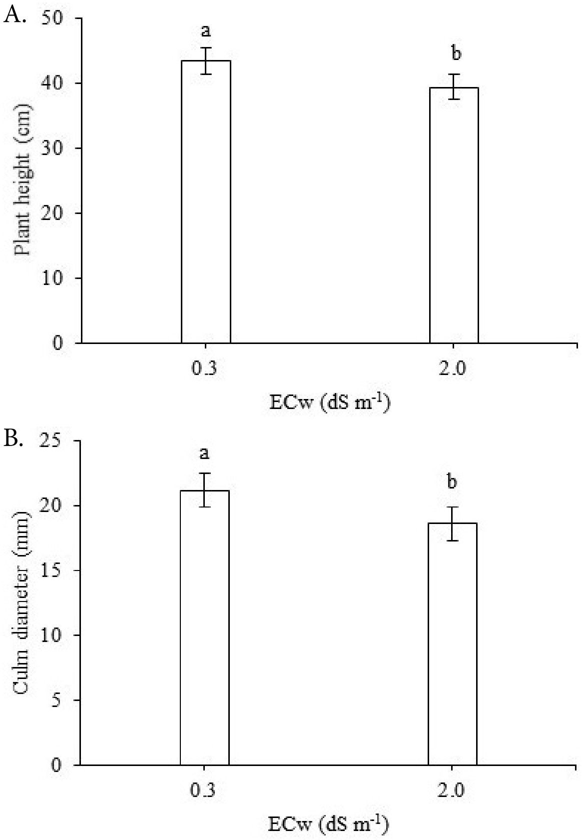
There was no significant interaction between the salinity of irrigation water and levels of H_2O_2 for plant height, culm diameter, number of leaves, leaf area, leaf dry biomass, leaf dry biomass, and total dry biomass. However, the salinity and H_2O_2 factors were significant for plant height and culm diameter, and for the number of leaves, leaf area, and total dry biomass of plant there was a significant effect of the H_2O_2 factor (Table 3).

Considering the values of plant height and culm diameter at the salinity levels of irrigation water, it can be observed that plants irrigated with 0.3 dS m^{-1} water have higher mean values than plants irrigated with 2.0 dS m^{-1} water (Figures 5A and B). The reduction in growth under salt stress can be attributed to the reduction of the osmotic potential caused by the increased concentration of soluble salts in the soil solution, which directly affects water absorption by plants. In addition, the absorption of nutrients by plants is affected, as specific ions such as Na$^+$ may compete with other essential nutrients (Byrt et al., 2018).

Plant height increased linearly with increasing H_2O_2 concentration, obtaining 22.29% when compared to the concentration of 320 μmol L^{-1} with 0 μmol L^{-1} (Figure 6A), which indicates that regardless of whether the water is saline or not, the increase in H_2O_2 concentration favored the primary growth of the plants. According to Farouk & Amira, (2018), this is because H_2O_2, when used in adequate concentrations, has a beneficial effect on plants, favoring greater absorption of water and nutrients, including essential ions for plant growth and development, such as N, P, and K.

For culm diameter, the opposite situation occurred, with a linear reduction of 5.36% when comparing the concentration of 0 μmol L^{-1} with that of 320 μmol L^{-1} of H_2O_2 (Figure 6B). It can be observed that high concentrations of H_2O_2, combined with salt stress, caused greater elongation of the plants, but smaller diameter.

The number of leaves per plant increased with increasing H_2O_2 concentration up to 151.66 μmol L^{-1}, promoting an increment of 5.8% compared to the concentration of 0 μmol L^{-1} of H_2O_2 (Figure 6C). The leaf area also increased with the increment in the concentration of H_2O_2, with a value of 2325.55 cm^2 at the estimated concentration of 172.69 μmol L^{-1}, an increment of the order of 15.67% in leaf area compared to the 0 μmol L^{-1} concentration of H_2O_2, regardless of the salinity of the irrigation water (Figure 6D).



Means compared by F test at p ≤ 0.05. Means followed by same letter indicate no significant difference between them. Vertical bars indicate standard error of mean (n = 4)

Figure 5. Plant height (A) and culm diameter (B) of maize plants subjected to different levels of irrigation water salinity and concentrations of hydrogen peroxide (H_2O_2) 30 days after emergence

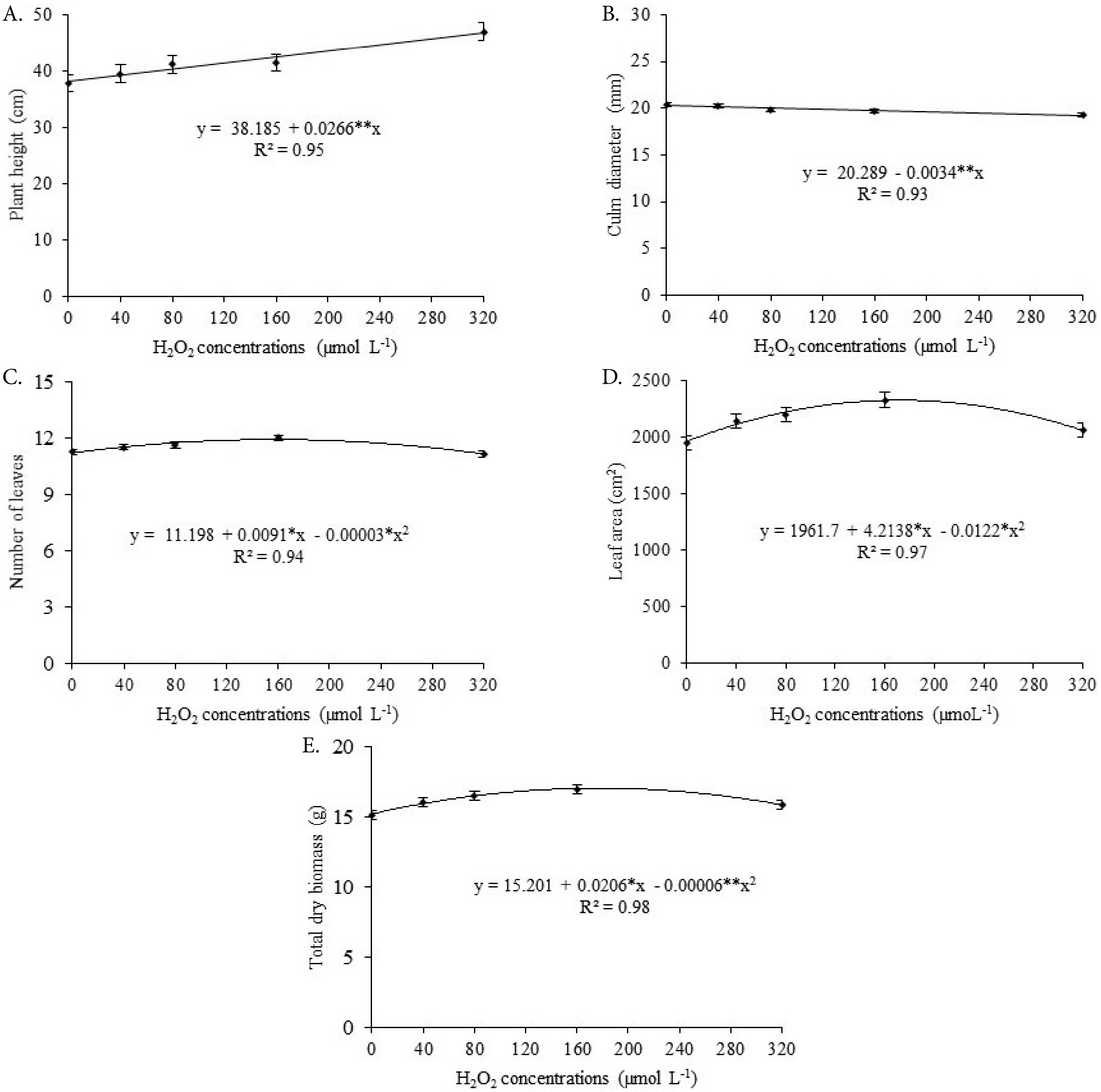
In this study, even though there was no significant interaction, the highest total dry biomass was 16.96 g at the estimated concentration of 171.66 μmol L^{-1}, promoting an increase of 12.16% in the total dry mass of the shoot compared to the concentration of 0 μmol L^{-1} (Figure 6E).

The addition of H_2O_2 to leaf tissues exogenously or its endogenous induction acts as an induction signal for the expression of genes referring to catalase, ascorbate peroxidase, guaiacol peroxidase, and glutathione reductase (Petrov & van Breusegem, 2012), whereby the accumulation of H_2O_2 in appropriate amounts benefits plants by mediating acclimation and tolerance to biotic and abiotic stresses. In this context, the application of H_2O_2 at the time of sowing can induce

Table 3. Summary of F test for plant height (PH), culm diameter (CD), number of leaves (NL), leaf area (LA), culm dry biomass (CDB), leaf dry biomass (LDB), and total dry biomass (TDB) of maize plants subjected to different levels of irrigation water salinity and concentrations of hydrogen peroxide (H_2O_2) 30 days after emergence

Source of variation	F Test						
	PH	CD	NL	LA	CDB	LDB	TDB
Salinity (ECw)	**	**	ns	ns	ns	ns	ns
Hydrogen peroxide (H_2O_2)	**	**	*	*	ns	ns	*
Interaction (ECw × H_2O_2)	ns	ns	ns	ns	ns	ns	ns
CV (%)	6.42	2.87	5.14	16.83	17.79	16.59	16.55

CV (%) - Coefficient of variation; * and ** significant at p ≤ 0.05 and 0.01, respectively by F test



* and ** significant at $p \leq 0.05$ and 0.01, respectively by F test. Vertical bars indicate standard error of mean (n = 4)

Figure 6. Plant height (A), culm diameter (B), number of leaves (C), leaf area (D), and total dry biomass (E) of maize plants under different levels of irrigation water salinity and concentrations of hydrogen peroxide (H_2O_2), 30 days after emergence

the emergence of more resistant seedlings by accelerating enzymatic activity and metabolic processes, before the plants are exposed to salt stress.

In some studies, H_2O_2 has been used to combat salinity. For example, Souza et al. (2019), when studying the exogenous application of hydrogen peroxide at 20 µM, found beneficial effects on the accumulation of phytomass and quality of cashew rootstocks. Silva et al. (2019c) found that foliar application of 15 µM promoted better leaf area in the melon plant under saline conditions. Gondim et al. (2011), when spraying maize seedlings with H_2O_2 at a concentration of 10 mM, under salinity of 0 and 80 mM, found that H_2O_2 induced the acclimation of maize plants to salt stress, partially reversing the deleterious effects of salinity on growth.

Working with maize plants from seeds pretreated with H_2O_2 and subjected to salinity, Godim et al. (2013) observed that pretreatment conferred salinity tolerance to plants. Silva et al. (2019d), studying the application of hydrogen peroxide in sunflower plants under salt stress, found that pretreatment with hydrogen peroxide via seed and the combination via seed + leaves were able to reduce the deleterious effects of salinity, promoting higher production of biomass. In yellow passion fruit, Silva et al. (2021a) found that the use of hydrogen peroxide, via seed soaking at concentrations between 10 and 30 µM induces the acclimatization of plants to salt stress, mitigating the deleterious effects of salinity on the relative growth rate in culm diameter and leaf area.

CONCLUSIONS

1. Irrigation water salinity of 2.0 dS m^{-1} reduces transpiration, stomatal conductance, total chlorophyll, carotenoids, and

initial growth, but does not affect the dry mass accumulation of maize plants.

2. Application of H_2O_2 via soil varying from 0 to 320 μmol L^{-1} causes reductions in the CO_2 assimilation rate and transpiration, as well as at concentrations from 0 to 160 μmol L^{-1} for stomatal conductance of plants irrigated with ECw of 2.0 dS m^{-1}

3. Application of H_2O_2 via soil up to a concentration of 320 μmol L^{-1} increases the plant height, but reduces culm diameter of maize.

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