

## GROWTH AND GAS EXCHANGES OF COTTON UNDER WATER SALINITY AND NITROGEN-POTASSIUM COMBINATION<sup>1</sup>

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**ABSTRACT** - The objective of this study was to evaluate the growth and gas exchanges of white-fiber cotton cv. BRS 368 RF irrigated using waters with increasing levels of salinity under nitrogen-potassium combination in a greenhouse in the municipality of Campina Grande-PB, Brazil. The treatments were distributed in randomized blocks, corresponding to five levels of electrical conductivity of water - ECw (0.7; 2.2; 3.7; 5.2 and 6.7 dS m<sup>-1</sup>) and four combinations of fertilization nitrogen-potassium - N/K<sub>2</sub>O (70/50, 100/75, 130/100, 160/125% of the recommended dose for pot experiments), with three replicates. Irrigation with water of electrical conductivity from 0.7 dS m<sup>-1</sup> compromised the growth and gas exchanges of cotton cv. BRS 368 RF, and dry biomass of leaf, stem and root were the most sensitive variables to salt stress. The nitrogen-potassium combination of 100/75% of the recommendation for pot experiments resulted in higher growth in plant height, but the highest stomatal conductance of cotton cv. BRS 368 RF was recorded in plants cultivated with 75/50% N/K<sub>2</sub>O, at 77 days after sowing. The variation in the N/K<sub>2</sub>O combination did not interfere with biomass accumulation in cotton plants. The treatment with water of lowest salinity (0.7 dS m<sup>-1</sup>) and the combination of 160/125% of the recommendation promoted greater leaf expansion, at 107 days after sowing.

**Keywords:** *Gossypium hirsutum* L.. Salinized waters. Photosynthesis. Phytomass.

## CRESCIMENTO E TROCAS GASOSAS DO ALGODOEIRO SOB SALINIDADE DA ÁGUA E COMBINAÇÃO NITROGÊNIO-POTÁSSIO

**RESUMO** - Objetivou-se com este trabalho avaliar o crescimento e as trocas gasosas do algodoeiro de fibra branca cv BRS 368 RF irrigado com águas de salinidade crescente sob combinação de nitrogênio-potássio em casa de vegetação no município de Campina Grande-PB. Os tratamentos foram distribuídos em blocos casualizados, correspondentes a cinco níveis de condutividade elétrica da água - CEa (0,7; 2,2; 3,7; 5,2 e 6,7 dS m<sup>-1</sup>) e quatro combinações de nitrogênio-potássio - N/K<sub>2</sub>O (70/50, 100/75, 130/100, 160/125% da dose recomendada para ensaios em vasos), com três repetições. A irrigação com água de condutividade elétrica a partir de 0,7 dS m<sup>-1</sup> comprometeu o crescimento e as trocas gasosas do algodoeiro cv. BRS 368 RF, sendo a fitomassa seca de folha, caule e raiz as variáveis mais sensíveis ao estresse salino. A combinação nitrogênio-potássio de 100/75%, da recomendação para estudo de vasos resultou em maior crescimento em altura das plantas, mas a maior condutância estomática do algodoeiro cv. BRS 368 RF foi registrada nas plantas cultivadas com 75/50% de N/K<sub>2</sub>O, aos 77 dias após a semeadura. A variação da combinação N/K<sub>2</sub>O não interferiu no acúmulo de fitomassas das plantas de algodoeiro. O tratamento com água de menor salinidade (0,7 dS m<sup>-1</sup>) e a combinação de 160/125% da recomendação promoveu maior expansão foliar das plantas, aos 107 dias após a semeadura.

**Palavras-chave:** *Gossypium hirsutum* L., Águas salinizadas, Fotossíntese, Fitomassa.

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<sup>1</sup>Received for publication in 10/11/2019; accepted in 03/02/2020.

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## INTRODUCTION

Cotton (*Gossypium* spp.) is the oilseed crop that has the most important fiber in the world, cultivated in more than 80 countries, with a global production of 123 million bales (480 pounds per bale) (SAEED et al., 2014). Brazil is the world's fifth largest producer of cotton and the third largest exporter of this fiber (COUTINHO et al., 2015).

The maintenance of agricultural activity in Northeastern Brazil relies on the use of irrigation due to the irregularity/scarcity of rainfall throughout the cycle of crops. In this region, most of the water used in irrigation has high contents of dissolved salts, so it becomes necessary to use these waters to meet the demand of crops (SILVA et al., 2014). However, the use of water with excess salts can inhibit plant growth and production, due to the reduction in the osmotic potential of the soil solution, and may also cause toxicity, nutritional imbalance or both simultaneously (SYVERTSEN; GARCIA-SANCHEZ, 2014).

Thus, cotton cultivation emerges as an option for generating jobs and income in this region as well as for the use of these waters, considering that it tolerates high salt concentrations in the root zone (LIMA et al., 2016). According to Ayers and Westcot (1999), this is a salt-tolerant crop with threshold salinity of 7.7 dS m<sup>-1</sup> in the saturation extract and of 5.1 dS m<sup>-1</sup> in the water. However, the effect of salinity may vary according to environmental conditions, genetic material used, development stages, as well as irrigation and fertilization management practices (LEMES et al., 2018).

In this context, fertilization management stands out as an important strategy, capable of favoring the acquisition of nutrients by plants under saline conditions (SILVA et al., 2011), particularly the combined supply of N/K, as these are two of the macronutrients most required by the majority of crops. Nitrogen has structural function and is part of several organic compounds that are vital to plants, such as amino acids, proteins and proline, among others, increasing the osmotic adjustment capacity of plants to salinity and increasing crop tolerance to salt stress (MASOULEH et al., 2019).

Potassium is vital for several biological processes in plant cells, such as enzymatic activation, respiration, photosynthesis and improvement in water balance (PRAZERES et al., 2015; HASANUZZAMAN et al., 2018). Understanding the interactions between salinity and fertilizers is of significant importance to increase crop yield and fertilizer use efficiency (ZENG; XU; WU, 2014).

In this context, the objective of this study was to evaluate the growth by biometric variables and by biomass formation and gas exchanges of white-fiber cotton cv. BRS 368 RF under irrigation water

salinity and nitrogen-potassium combinations.

## MATERIAL AND METHODS

The study was carried out under greenhouse conditions (ambient protected from rain, wind and insects), at the Center of Technology and Natural Resources of the Federal University of Campina Grande (CTRN/UFCG), located in the municipality of Campina Grande, PB, Brazil, at the local geographic coordinates 7° 15' 18" S latitude, 35° 52' 28" W longitude and altitude of 550 m.

The treatments were arranged in randomized blocks, using the 5 x 4 factorial scheme, with three replicates, relative to five levels of irrigation water electrical conductivity - EC<sub>w</sub> (0.7; 2.2; 3.7; 5.2 and 6.7 dS m<sup>-1</sup>) and four combinations of nitrogen-potassium fertilization - N/K<sub>2</sub>O (70/50, 100/75, 130/100, 160/125% of the recommendation), and the dose of 100% recommendation corresponded to 100 mg N and 150 mg K<sub>2</sub>O kg<sup>-1</sup> of soil of the for pot experiments (NOVAIS; NEVES; BARROS, 1991). The combined fertilization with nitrogen and potassium was split and applied at 10, 30 and 60 days after sowing (DAS), in order to increase the efficiency and utilization of these nutrients by plants. Urea was used as a nitrogen source and potassium chloride as source of potassium.

The saline waters were prepared by dissolving NaCl, CaCl<sub>2</sub>.2H<sub>2</sub>O and MgCl<sub>2</sub>.6H<sub>2</sub>O, in the proportion of 7:2:1, respectively, in public-supply water (EC<sub>w</sub> = 0.6 dS m<sup>-1</sup>) of the municipality of Campina Grande, PB, and the quantities were determined based on the relationship between EC<sub>w</sub> and the concentration of salts (mmol<sub>c</sub> L<sup>-1</sup> = 10\*EC<sub>w</sub> dS m<sup>-1</sup>) according to Richards (1954). After preparation, the waters were stored in 200-L plastic containers, one for each EC level studied, properly protected to avoid evaporation, entry of rainwater and contamination with materials that may compromise its quality.

The plants were grown in plastic containers adapted as drainage lysimeters with capacity for 20 L. A drain, consisting of tube, was installed at the bottom of each lysimeter to drain the leachate into a collector in order to evaluate the drained water and determine water consumption by plants. The tip of the drain inside the pot was envolved with a non-woven geotextile (Bidim OP 30) to avoid clogging by soil material.

The lysimeters were filled with a 0.5-kg-layer of crushed stone type zero, followed by 25 kg of a Neossolo Regolítico (Entisol) with sandy loam texture, collected from 0-20 cm depth in Lagoa Seca, PB, Brazil, with physical and chemical attributes obtained according to the methodology proposed by EMBRAPA (2017): Ca<sup>2+</sup>, Mg<sup>2+</sup>, Na<sup>+</sup>, K<sup>+</sup>, Al<sup>3+</sup> + H<sup>+</sup> = 2.60; 3.66; 0.16; 0.22 and 1.93 cmol<sub>c</sub> kg<sup>-1</sup>, respectively; pH (water 1:2.5) = 5.9; EC<sub>se</sub> =

1.0 dS m<sup>-1</sup>; organic matter = 1.36 dag kg<sup>-1</sup>; sand, silt and clay = 73.29, 14.21, and 12.50 dag kg<sup>-1</sup>, respectively; bulk density (kg dm<sup>-3</sup>) = 1.39; moisture content at 33.42 and 1519.5 kPa = 11.98 and 4.32 dag kg<sup>-1</sup>, respectively.

The cotton cultivar was BRS 368 RF, a transgenic material resistant to glyphosate herbicide, with fiber yield of 40% and production potential under irrigated conditions between 4,200 and 4,500 kg ha<sup>-1</sup>. It has short size and semi-early to early cycle, with opening of the first flower at 55 days after sowing with total cycle of 165 to 170 days (EMBRAPA, 2011).

Prior to sowing, the soil moisture content was raised to field capacity level with the respective waters of each treatment. After sowing, irrigation was performed daily by applying in each lysimeter a volume of water sufficient to keep soil moisture close to field capacity. The applied volume was determined according to the water requirement of the plants, estimated by water balance: volume applied minus volume drained in the previous irrigation, plus a leaching fraction of 0.10 to prevent excessive accumulation of salts in the soil (AYERS; WESTCOT, 1999).

The growth of cotton cv. BRS 368 RF was evaluated along the vegetative and fruiting stages, at 65 and 107 days after sowing (DAS), respectively, based on the measurements of plant height (PH), stem diameter (SD), number of leaves (NL) and leaf area (LA) and, at the end of the experiment (120 DAS), through the dry biomass of leaves (LDB), stem (SDB) and roots (RDB). PH was obtained by using as reference the distance from the collar to the insertion of the apical meristem. SD was measured at 2 cm from the collar, and NL was determined by counting the leaves of each plant. LA was obtained by measuring the midrib length of all leaves, following the methodology described by Grimes and Carter (1969), according to Equation 1:

$$y = 0.26622 x^{2.3002} \quad (1)$$

Where: y is leaf area per leaf and x is the midrib length of the cotton leaf. Leaf area per plant (cm<sup>2</sup>) was determined by the sum of the area of all leaves.

At 77 DAS, at the flowering stage, leaf gas exchanges of cotton were measured by determining the stomatal conductance ( $g_s$  - mol H<sub>2</sub>O m<sup>-2</sup> s<sup>-1</sup>), CO<sub>2</sub>

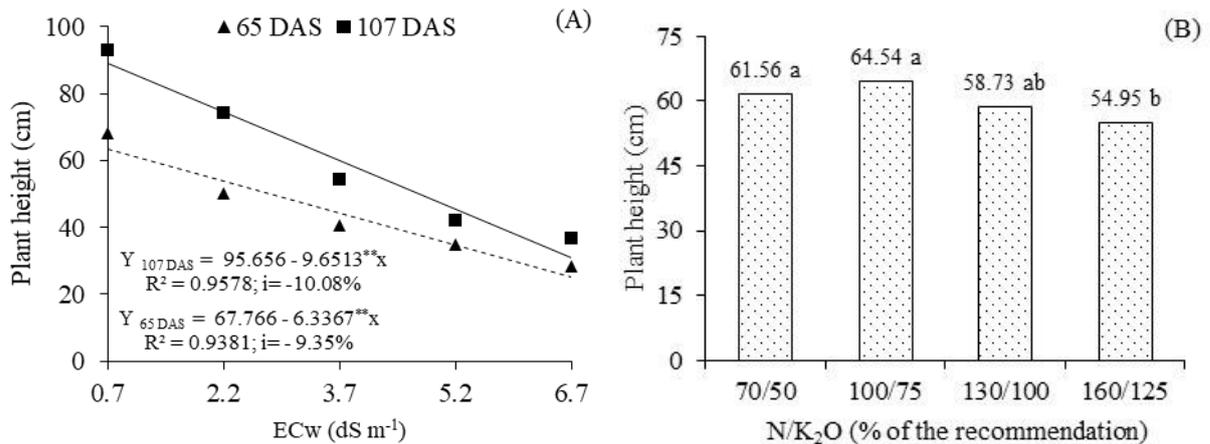
assimilation rate ( $A$  - μmol m<sup>-2</sup> s<sup>-1</sup>), transpiration ( $E$  - mmol of H<sub>2</sub>O m<sup>-2</sup> s<sup>-1</sup>) and intercellular CO<sub>2</sub> concentration ( $C_i$  - μmol mol<sup>-1</sup>), with a portable infrared gas analyzer (IRGA) (LCPro+ - Portable Photosynthesis System®). These data were used to calculate instantaneous water use efficiency - WUE<sub>i</sub> ( $A/E$ ) [(μmol m<sup>-2</sup> s<sup>-1</sup>)(mmol H<sub>2</sub>O m<sup>-2</sup> s<sup>-1</sup>)<sup>-1</sup>] and instantaneous carboxylation efficiency - CE<sub>i</sub> ( $A/C_i$ ) [(μmol m<sup>-2</sup> s<sup>-1</sup>)(μmol mol<sup>-1</sup>)<sup>-1</sup>].

At the end of the reproductive cycle, the plants were removed from the pots and the leaves, stem and roots were separated to quantify the dry mass of each organ. After being placed in paper bags, these materials were dried in an air circulation oven at 65 °C until constant weight. After drying, dry biomass leaf, stem and root were determined.

The collected data were subjected to analysis of variance by F test and by linear and quadratic regression for water salinity levels and means comparison test (Tukey at 0.05 probability level) for the combinations of nitrogen-potassium fertilization, using the statistical program SISVAR (FERREIRA, 2011).

## RESULTS AND DISCUSSION

Plant height was linearly inhibited by the increase in irrigation water salinity in both evaluations (Figure 1A). The increase in water salinity caused reductions of 9.35 and 10.08% in height with per unit increase in electrical conductivity, which corresponded to decreases of 60.03 and 65.13% among plants irrigated with waters of 0.7 and 6.7 dS m<sup>-1</sup> at 65 and 107 DAS, respectively (Figure 1A). Thus, it is evident that the longer exposure of plants to salt stress has contributed to the intensification of deleterious effects on the growth in plant height, as verified at 107 DAS. According to Acosto-Motos et al. (2017), in the long term, salt stress affects the photosynthetic process, due to the accumulation of salts in the leaves. Bezerra et al. (2018) state that the decrease is a response to the reduction in the osmotic potential of the soil due to the increase in irrigation water salinity and accumulation of Cl<sup>-</sup> and Na<sup>+</sup> ions. Under these conditions both simultaneously hamper photosynthesis and other biochemical processes that are essential to plant growth.



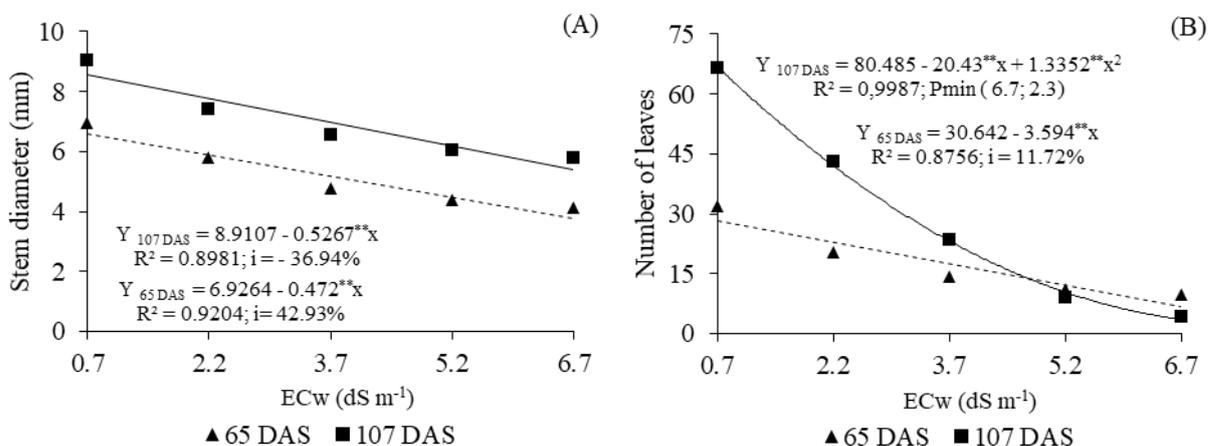
Means followed by the same letter do not differ by Tukey test ( $p < 0.05$ ).

**Figure 1.** Plant height of cotton plants cv. BRS 368 RF, as a function of irrigation water salinity - ECw (A), at 65 and 107 days after sowing (DAS), and as a function of combined fertilization with nitrogen-potassium - N/K (B), at 107 DAS.

Based on the comparison of means (Figure 1B), the height of cotton cv. BRS 368 RF responded to the effects of the nitrogen-potassium combinations, with the highest values of 61.56, 64.54 and 58.73 cm obtained in plants fertilized with combinations of 70/50, 100/75 and 130/100% of N/K<sub>2</sub>O, while the lowest PH (54.95 cm) was obtained in plants receiving 160/125% of N/K<sub>2</sub>O, but with no significant difference from those fertilized with 130/100% of N/K<sub>2</sub>O. It must be pointed out that there were no significant differences ( $p > 0.05$ ) between the combinations 75/50, 100/75 and 130/100% of N/K<sub>2</sub>O. The increase in K<sup>+</sup> concentration in the soil solution more than the nutritional needs of the crops negatively interferes with the absorption of Ca<sup>2+</sup> and Mg<sup>2+</sup> ions, as it competes for the same absorption sites in root tissues, inducing calcium and magnesium deficiency in plants (SILVA; TREVIZAM, 2015). This situation is consistent with the inhibition in the growth of cotton plants since Ca acts in the stabilization of cell membrane and its deficiency causes destructuring of cell walls with subsequent collapse of tissues (TAIZ et al., 2017), while Mg<sup>2+</sup>

plays an important role in metabolic processes, being a cofactor of enzymatic activity with ATP and stabilizer of the ribosomal structure, besides being a central atom in the formation of the chlorophyll structure (TANOI; KOBAYASHI, 2015). Therefore, the reduction of this element in leaf tissues compromises photosynthesis, as well as protein synthesis, negatively affecting plant growth.

The exposure of cotton plants to irrigation with salinized waters also led to reductions in stem diameter and number of leaves, at 65 and 107 DAS (Figure 2A and 2B). On the same evaluation dates, the increase in water salinity reduced stem diameter by 42.93 and 36.94%, respectively, at 65 and 107 DAS, in cotton plants subjected to ECw = 6.7 dS m<sup>-1</sup>, compared to those irrigated with 0.7 dS m<sup>-1</sup> water. The reduction in SD may be related to the osmotic effects that inhibit root growth and the absorption of water and nutrients by the plant (ACOSTO-MOTOS et al., 2017), leading to reduction in cell expansion and division (BEZERRA et al., 2018), resulting in a reduction in the size of vascular cells and tissues of the stem of plants exposed to salt stress (FARHANA et al., 2014).



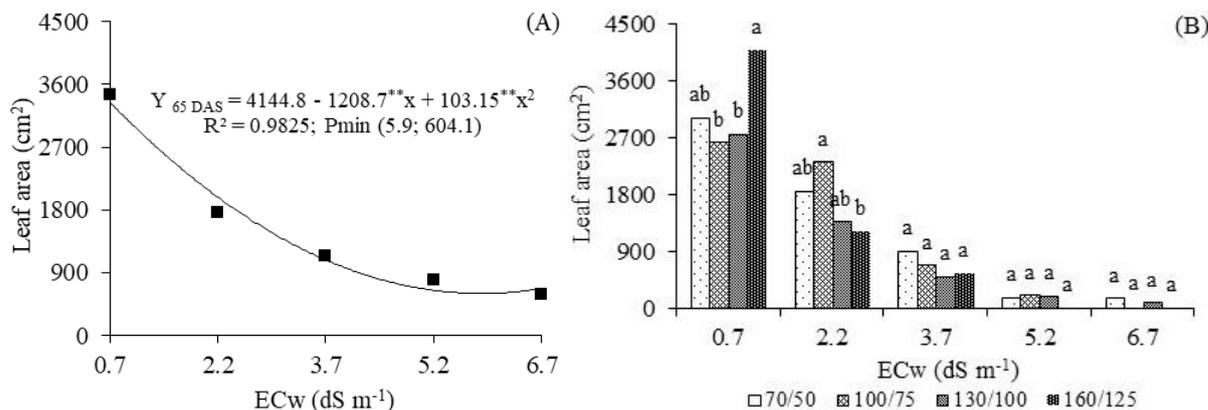
**Figure 2.** Stem diameter (A) and number of leaves (B) of cotton cv. BRS 368 RF, as a function of the irrigation water salinity - ECw, at 65 and 107 days after sowing (DAS).

On both dates, the increase in water salinity hampered leaf production by cotton plants (Figure 2B). The regression equations showed that the increase in irrigation water salinity from 0.7 to 6.7 dS m<sup>-1</sup> resulted in reductions of 21.5 and 63.3 leaves plant<sup>-1</sup>, respectively at 65 and 107 DAS. The reduction in NL can be considered a mechanism of acclimatization of plants to salt stress, aiming to reduce the transpiration rate and consequently the absorption of water and toxic ions, in an attempt to inhibit excessive accumulation in plant organs (SILVA et al., 2011), resulting in damage to essential biochemical processes.

The leaf area of cotton plants was also reduced by the increase in irrigation water salinity (Figure 3A). Cotton plants had the lowest LA (604.12 cm<sup>2</sup>) when subjected to irrigation with water of 5.9 dS m<sup>-1</sup>, corresponding to a reduction of 2,745.13 cm<sup>2</sup> (81.96%) compared to those irrigated

using water with lowest salinity (0.7 dS m<sup>-1</sup>). Naturally, the reduction in the number of leaves (Figure 2B) contributed to the decrease in LA, but the large reduction in LA also reflects the inability of cotton plant to continue its leaf expansion process under the imposed salinity level.

As leaf area - LA constitutes a variable that indicates the efficiency in light capture, photosynthesis and formation of photoassimilates, it contributes to plant growth (BRITO et al., 2014) and reflects on plant tolerance to salinity. However, the capacity for leaf expansion and for fixing carbon under salt stress depends on the intensity of the stress imposed on plants (CHAMEKH et al., 2014). Under these conditions, the properties of cell walls change and the rates of turgor and photosynthesis of the leaves decrease (ACOSTO-MOTOS et al., 2017).



Means followed by the same letter do not differ by Tukey test ( $p < 0.05$ ) at the same ECw level.

**Figure 3.** Leaf area of cotton cv. BRS 368 RF, as a function of irrigation water salinity - ECw at 65 days after sowing (DAS) (A) and as a function of the interaction between ECw levels and N/K<sub>2</sub>O combinations (B) at 107 DAS.

At 107 DAS the interaction between the factors studied (NS x N/K<sub>2</sub>O) had significant effects on cotton leaf area (Figure 3B). Further analysis of the interaction showed that under ECw of 0.7 dS m<sup>-1</sup>, plants that received 160/125% differed statistically from those that received 100/75 and 130/100% of N/K<sub>2</sub>O, while for plants irrigated using water with ECw of 2.2 dS m<sup>-1</sup> there was statistical difference between the N/K<sub>2</sub>O combinations of 100/75 and 160/125%. When evaluating the LA of cotton irrigated with waters of 3.7, 5.2 and 6.7 dS m<sup>-1</sup>, it was verified that there was no significance between the different combinations of fertilization. This fact indicates that, regardless of the N/K<sub>2</sub>O combination, the salt stress caused by irrigation with high-salinity water (ECw > 3.7 dS m<sup>-1</sup>) inhibits the growth of cotton plants cv. BRS 368 RF, at 107 DAS (Figure 3B).

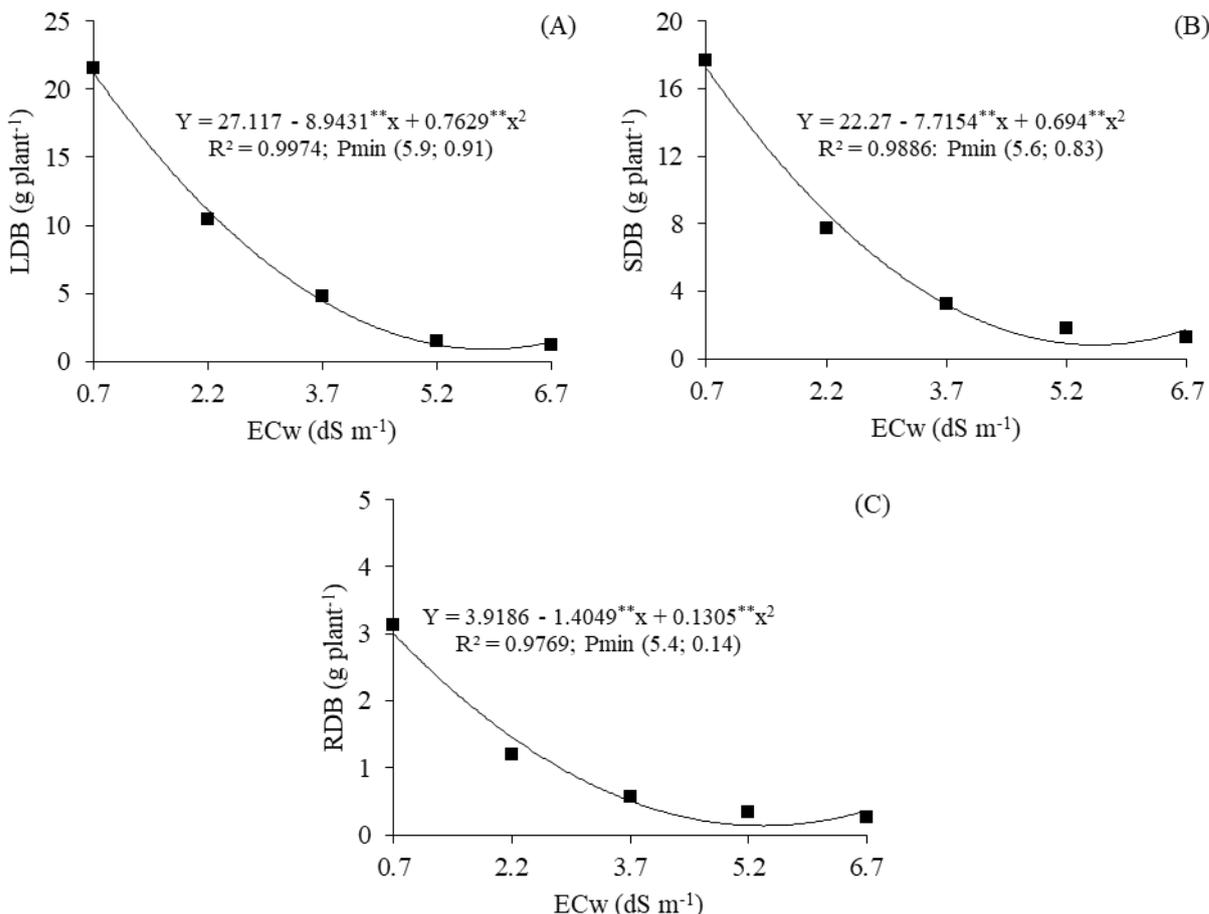
By analyzing the LA data (Figure 3B), it can be noted that plants irrigated with 0.7 dS m<sup>-1</sup> water and fertilized with N/K<sub>2</sub>O in the combination of 160/125% had an increase in this variable, whereas

in those subjected to 2.2 dS m<sup>-1</sup>, the increase in nitrogen/potassium supply reduced leaf expansion, suggesting that for this salinity level (2.2 dS m<sup>-1</sup>) the highest mineral fertilization intensified the negative effects of salinity, since potassium chloride was the source of K<sub>2</sub>O used in this study, a fertilizer that has a high salt index (116). In addition, when used in excess, K<sup>+</sup> promotes competition with Ca<sup>2+</sup> and Mg<sup>2+</sup> as previously discussed for the data of plant height (Figure 1B), reinforcing the hypothesis that determining an adequate N/K<sub>2</sub>O combination for the maintenance of osmotic and ionic homeostasis may be an alternative capable of alleviating the negative effects of salt stress.

The degenerative effects of water salinity on biometric growth were also observed on biomass formation. The data relative to leaf, stem and root dry biomass were described by the quadratic mode (Figure 4), whose lowest estimated accumulations of LDB, SDB and RDB were obtained under ECw of 5.9, 5.6 and 5.6 dS m<sup>-1</sup>, respectively. On the other

hand, those cultivated with ECw of 0.7 dS m<sup>-1</sup> had the highest values. By comparing the data of biomass for the different organs of cotton plants, it was

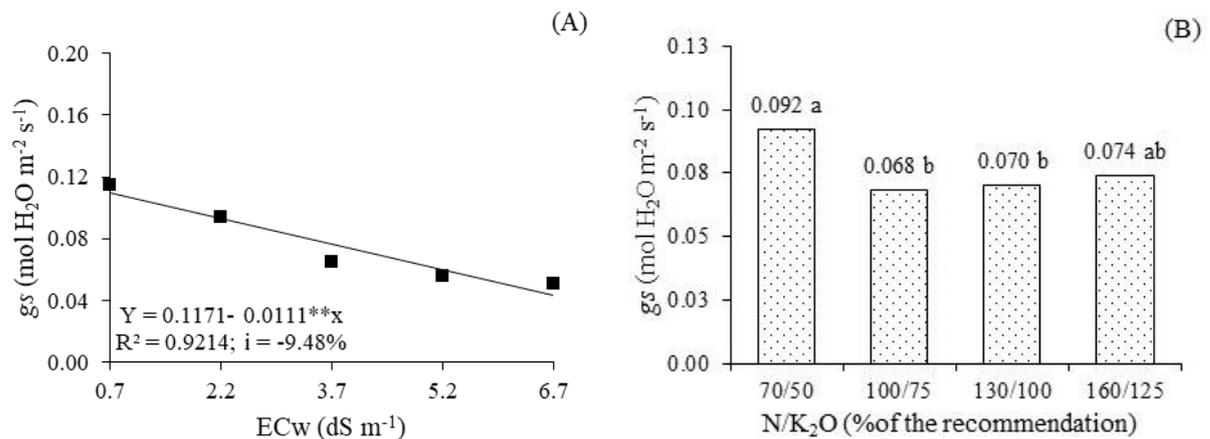
possible to note reductions of 20.32, 16.38 and 2.86 g plant<sup>-1</sup>, representing percentage reductions of 95.71, 95.18 and 95.33% in dry biomass of leaves, stem and roots, respectively.



**Figure 4.** Dry biomass of leaf - LDB (A), stem - SDB (B) and root - RDB (C) of cotton cv. BRS 368 RF, as a function of irrigation water salinity - ECw, at 120 days after sowing (DAS).

Although the literature indicates threshold salinity of 5.1 dS m<sup>-1</sup> in irrigation water for white-fiber cotton, according to Ayers and Westcot (1999), the crop has the same qualitative standards observed in other plant species cultivated under salt stress, a fact observed through the inhibition of growth (PH, SD, NL, LA) and biomass accumulation, as a consequence of the use of its resources (energy and metabolic precursors) for the activation of defense mechanisms to the detriment of the biomass accumulation (KUMAR et al., 2017). In addition, high salt concentrations cause disturbances in mineral nutrition, alteration of membrane permeability and cellular osmotic balance, generation of oxidative stress (increased levels of reactive oxygen species), or inhibition of different enzymatic activities (SHANKAR; KUMAR; AGRAWAL, 2016).

Stomatal conductance decreased linearly with the increase in irrigation water salinity (Figure 5A), with losses of 9.48% per unit increase in ECw and of 60.91% between plants irrigated with waters of 6.7 dS m<sup>-1</sup> and 0.7 dS m<sup>-1</sup>. The limitation in water availability in response to osmotic stress resulting from the increase in ECw restricts stomatal conductance, constituting an important strategy of plants to avoid severe dehydration and maintain high cell water potential (LIRA et al., 2018). The results (Figures 1, 2, 3 and 4) are in agreement with Marchiori et al. (2017), who concluded that the changes observed in crop growth reflect a possible state of water deficit in plants and that, in this situation, it can restrict the dilution of ions such as Na<sup>+</sup> and Cl<sup>-</sup> in plant tissues, causing greater accumulation of these elements to the detriment of biomass, with consequent toxicity.



Means followed by the same letter do not differ by Tukey test ( $p < 0.05$ ).

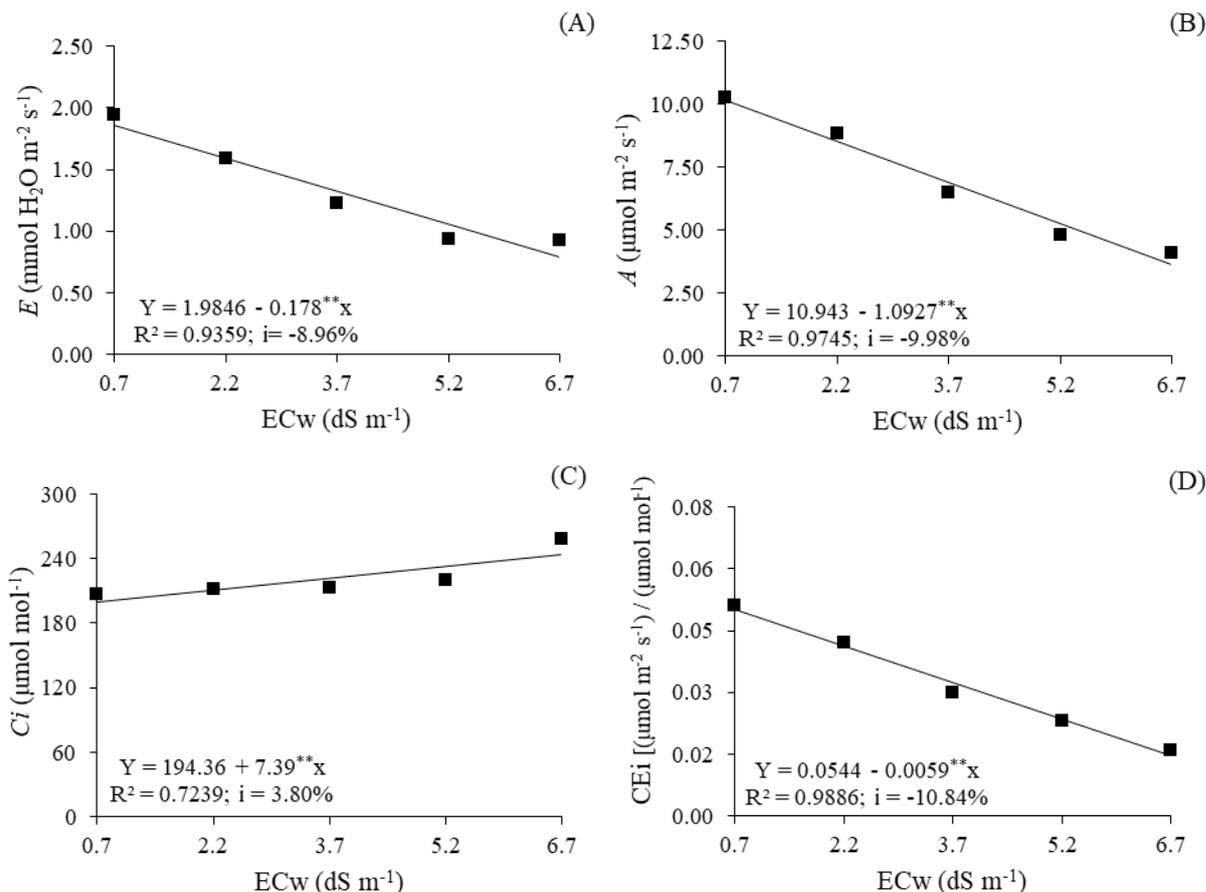
**Figure 5.** Stomatal conductance -  $g_s$  of white-fiber cotton cv. BRS 368 RF as a function of irrigation water salinity - ECw (A) and as a function of fertilization with combinations of nitrogen-potassium - N/K<sub>2</sub>O (B), at 77 days after sowing (DAS).

Based on the means comparison test for  $g_s$  (Figure 5B), the N/K<sub>2</sub>O combination of 70/50% was statistically different from those of 100/75 and 130/100%. However, plants grown with 100/75, 130/100 and 160/125% of N/K<sub>2</sub>O did not differ from each other. A study conducted by Prazeres et al. (2015) also found that the increase in potassium supply when KCl was used as a source of K<sub>2</sub>O negatively affected the stomatal conductance of common bean. This situation indicates that there is a negative interaction between excessive potassium supply and salinity, thus promoting an intensification of the osmotic effect, which limits CO<sub>2</sub> diffusion through the stomata.

Transpiration and the rate of CO<sub>2</sub> assimilation by plants, as observed for stomatal conductance, were also inhibited by the increase in the salt concentration of the waters (Figures 6A and 6B). The increase in ECw resulted in reductions of 8.96 and 9.98% per unit increase in ECw for  $E$  and  $A$ , respectively. When comparing plants irrigated with waters of 6.7 and 0.7  $\text{dS m}^{-1}$ , there were reductions of 57.41% in transpiration (Figure 6A) and 64.41% in CO<sub>2</sub> assimilation rate (Figure 6B). These reductions, in general, refer to the lower stomatal opening due to the action of salts, causing decreases in the rates of transpiration and CO<sub>2</sub> assimilation. In this situation, salinity limits  $g_s$  and, as an effect, the entry of CO<sub>2</sub> into the leaf mesophyll, reducing photosynthesis due to the decrease in the partial pressure of this gas in the intercellular spaces, besides reducing the loss of water in the form of vapor through the stomata, raising leaf temperature (LIMA et al., 2017). In the transpiration process, energy in the form of latent heat is transferred from the leaf to the air, and its decrease may constitute a mechanism of acclimatization to salt stress (SOUSA et al., 2016).

The increment in irrigation water salinity increased the intercellular CO<sub>2</sub> concentration by 3.80% per unit increment in ECw, i.e. the  $C_i$  of plants irrigated with the highest salinity level (6.7  $\text{dS m}^{-1}$ ) was 22.22% higher compared to those irrigated with 0.7  $\text{dS m}^{-1}$  water (Figure 6C). This increase in  $C_i$ , even with the reduction in  $g_s$  (Figure 5A), suggests that the accumulation of CO<sub>2</sub> in the leaf mesophyll, observed in this study, occurred due to possible restrictions in carbon metabolism, which can be reaffirmed after analyzing the CE<sub>i</sub> response of cotton in this study (Figure 6D). The CE<sub>i</sub> measures the capacity of the RuBisCO enzyme to capture carbon dioxide from the atmosphere and fix it in an organic form, starting the Calvin cycle. According to the regression equation (Figure 6D), the irrigation of plants using water with ECw of 6.7  $\text{dS m}^{-1}$  resulted in a 70.41% reduction in the carboxylation capacity, compared to plants subjected to 0.7  $\text{dS m}^{-1}$ .

The decrease in  $C_i$  under saline conditions occurs when there is a reduction in  $g_s$ , reflecting stomatal limitation, and in this case the plant uses CO<sub>2</sub> from respiration to maintain a minimum level of photosynthetic rate when the  $C_i$  values are very low (CAMPOS et al., 2014). In addition, under salt stress conditions, there is a decrease in the transport of cytokinins from the root to the aerial part that is involved in the restoration of RuDP (RuBisCO), in the photosynthetic process (BARROS et al., 2010). In accordance with the results obtained, the reduction of photosynthesis, transpiration and stomatal conductance, as well as increments in the intercellular CO<sub>2</sub> concentration under salt stress, have also been observed in studies conducted by Stepien and Klobus (2006) and Soares et al. (2018).



**Figure 6.** Transpiration rate -  $E$  (A),  $\text{CO}_2$  assimilation rate -  $A$  (B), intercellular  $\text{CO}_2$  concentration -  $C_i$  (C) and instantaneous carboxylation efficiency - CEi (D) of white-fiber cotton cv. BRS 368 RF as a function of irrigation water salinity - ECw at 77 days after sowing (DAS).

The results obtained in this study demonstrate that the adequate supply of combined mineral fertilizers (N and K) constitutes an important technique to mitigate the deleterious effects caused by irrigation with high-salinity water on crops, especially in the Northeast region, where this problem is aggravated by the edaphoclimatic conditions, so that crops of socioeconomic importance for this region, such as cotton, which is notably tolerant to salt stress, have their potential yield drastically reduced due to the intensity and time of exposure to the stress, as well as to the incipient technical-scientific information for cultivation conditions.

## CONCLUSIONS

Irrigation using water with electrical conductivity from  $0.7 \text{ dS m}^{-1}$  compromises the growth and gas exchanges of cotton cv. BRS 368 RF, and dry biomass of leaf, stem and root are the most sensitive variables to salt stress.

The N/ $\text{K}_2\text{O}$  combination of 100/75% of the recommendation results in higher growth in plant

height, but the highest stomatal conductance of cotton cv. BRS 368 RF is obtained in plants grown with 75/50% N/ $\text{K}_2\text{O}$ , at 77 days after sowing.

The variation in the N/ $\text{K}_2\text{O}$  combination does not interfere with the biomass accumulation of cotton plants.

Water salinity of  $0.7 \text{ dS m}^{-1}$  and fertilization with 160/125% N/ $\text{K}_2\text{O}$  promotes greater leaf expansion in cotton at 107 days after sowing.

## ACKNOWLEDGMENTS

To the National Postdoctoral Program (PNPD/CAPES/UFCEG) for granting the scholarship to the second author and to the National Institute of Science and Technology in Salinity - INCTSal, for funding the project.

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