

Gas exchanges and growth of sesame (*Sesamum indicum*, L.) cultivated under saline waters and nitrogen-potassium fertilizers

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Abstract

The synergistic action between K and N (NO_3^-) favors the absorption of both ions by plants. A suitable combination of these macronutrients may be an alternative capable of alleviating the nutritional imbalance due to the excessive absorption of chloride and sodium by the plant and inhibitory competition between nitrate and potassium. In addition, it can favor the control of the turgidity of the cells, through the osmoregulation, elevate the synthesis of organic solutes, promoting the ionic homeostase, and consequently decrease the effect saline stress on the plants. In this context, this study aimed to evaluate the gas exchanges and growth of sesame cv. BRS G4 as a function of irrigation with water of increasing electrical conductivity (ECw) and fertilization with different combinations of nitrogen (N) and potassium (K). This experiment was conducted under greenhouse conditions. Treatments were distributed in randomized blocks and analyzed in 5 x 4 factorial scheme, consisting of five levels of ECw (0.6; 1.2; 1.8; 2.4 and 3.0 dS m^{-1}) and four combinations of N and K: N/K₂O (70/50; 100/75; 130/100 and 160/125% of the recommendation for pot experiments), with three replicates, totaling 60 experimental units. Except for the ECw level of 0.6 dS m^{-1} , the other salt concentrations evaluated in this study compromised the gas exchanges and consequently growth of sesame cv. BRS G4, at 50 days after sowing. Fertilization with N and K in the combination of 70/50% of the recommendation of N/K₂O led to the greatest growth of sesame cv. BRS G4. The N/K₂O combinations of 130/100, 160/125 and 160/125% reduced the negative effects of saline irrigation of 1.2, 1.8 and 2.4 dS m^{-1} , respectively, on the stomatal conductance of sesame plants. The combined supply of N and K₂O in combinations of 100/75, 130/100 and 130/100% promoted higher CO₂ assimilation rate in sesame plants using water of 1.2 and 1.8 and 2.4 dS m^{-1} , respectively.

Keywords: *Sesamum indicum*, L., salinity, physiology, mineral fertilization.

Abbreviations: DAS_ days after sowing; ECw_water electrical conductivity; PH_plant height; SD_stem diameter, NL_number of leaves LA_leaf area; *g*_s_stomatal conductance; *A*_assimilation rate, *E*_transpiration; *C*_i_internal CO₂ concentration.

Introduction

Sesame (*Sesamum indicum*, L.) stands out as one of the most produced oilseed crops in the world, occupying the ninth position, whose mean production is 481.40 kg ha^{-1} . In Brazil, its mean yield is of the order of 600.0 kg ha^{-1} , being mainly used either fresh or constituting products of food and baking industry (Queiroga and Silva, 2008). According to Araújo et al. (2012), this crop has stability of production with respect to the factor water, i.e., low water requirement compared with other cultivated species. It has good adaptability to hot climate, which are important adaptive characteristics for semi-arid regions. In addition, its cultivation generates

income, mainly for small producers, which makes it an excellent alternative for the socioeconomic development of this region.

However, the occurrence of high temperatures, low rainfall, irregular distribution of rains and intense evaporation, which are characteristics of the semi-arid region of Northeast Brazil, makes a successful sesame production dependent on the use of irrigation, although this crop has traits that are favorable for its planting and production in this region. It should be mentioned that the semi-arid region of Northeast Brazil faces serious problems regarding its water resources,

because a major part of the water sources available for irrigation in this region has high concentration of dissolved salts (Souza et al., 2017).

Water with high concentrations of salts causes reduction in the soil osmotic potential, which leads to water deficit in the crops, resulting in stomatal closure, limitation on CO₂ assimilation and transpiration, with consequent reduction in the photosynthetic rate (Silva et al., 2010). Besides, it causes toxicity and nutritional imbalance in plants (due to the accumulation of sodium and chloride in the tissues), culminating with the inhibition of growth and production of the crops (Pedrotti et al., 2015). Consequently, the feasibility of using these resources is associated with the development of efficient alternatives that allow these waters to be utilized (Alves et al., 2011) in the agricultural expansion of this region.

Thus, mineral fertilization, especially with nitrogen and potassium emerges as an alternative to reduce the negative effects of salinity in crops. Furthermore, K is vital for photosynthesis and formation and translocation of carbohydrates (Araújo et al., 2012). It acts as an enzyme activator, improves water balance, increases water use efficiency, competes with Na⁺, and promotes the accumulation of N compounds in plants by increasing N use efficiency (Heidari and Jamshid, 2010). N is part of several organic molecules, such as amino acids, enzymes, proteins and proline, which promote the osmotic adjustment capacity of plants (Oliveira et al., 2014), mitigating the negative effects caused by salt stress on the crops.

Although the benefits of nitrogen and potassium fertilizations are recognized, little is known about their interaction as an alternative to mitigate salt stress on the sesame crop. In this context, this study aimed to evaluate the gas exchanges and growth of sesame cv. BRS G4, as a function of irrigation with water of different salinity levels and fertilization with combinations of nitrogen and potassium.

Results and discussion

Effect of saline stress and nitrogen and potassium fertilization on the gas exchange of sesame

According to the mean comparison test for stomatal conductance (Fig 1A), when sesame plants cv. BRS G4 were irrigated with highly saline water (3.0 dS m⁻¹), no statistical differences were observed among the combinations of fertilization with N/K₂O at 50 DAS. In general, when sesame plants were irrigated with 1.2 dS m⁻¹ saline water, the increment in N/K₂O ratio was increase in stomatal conductance. Plants irrigated with 2.4 and 1.8 dS m⁻¹ saline water showed the highest *g_s* values (0.143 and 0.170 mol H₂O m⁻² s⁻¹, respectively) under fertilization with the N/K₂O combination of 160/125% (Fig 1A). Plants irrigated with 1.2 dS m⁻¹ saline water, showed the highest *g_s* values under fertilization of 130/100% of the N and K (of recommended dose), respectively. However, for the other N/K₂O combinations no significant difference was observed. In sesame plants subjected to irrigation with the lowest salinity level (0.6 dS m⁻¹), no significant difference was found among plants which received the N/K₂O combinations of 70/50 and 100/75%. However, they showed higher *g_s* values compared with those under 160/125% of nitrogen/potassium. The

highest stomatal conductance was obtained under fertilization with 70/50% (Fig 1A).

Based on the interaction among the evaluated factors, the highest stomatal conductance was found in plants irrigated with 1.2 dS m⁻¹ salinity and under N/K₂O combination of 130/100%. In plants subjected to ECw of 1.8 and 2.4 and dS m⁻¹, the best *g_s* values were observed under the combination of 160/100% of the recommendation for both levels of salinity (Fig 1A), indicating that increase in the combined supply of N and K positively acts by reducing the deleterious effects of irrigation water salinity on sesame *g_s*. Feijão et al. (2011) explained that improvements in photosynthetic parameters are also due to the N status in the plant, as this nutrient is used for the synthesis of components of the photosynthetic elements. Potassium supply reduces the harmful effects of high-saline waters by acting on improvement of water balance in the plant (Gurgel et al., 2010). This favors the increase of *g_s*, consequently improving gas exchanges.

For CO₂ assimilation rate at 50 DAS (Fig 1B), it can be noted that plants subjected to irrigation with increased salinity levels loss *A* and, according to the means comparison test (Fig 1B), plants irrigated with lowest salinity level (0.6 dS m⁻¹) showed no significant difference among the different combinations of N/K₂O. Sesame plants grown under ECw of 1.2 dS m⁻¹ and fertilized with 100/75% (Fig 1B) were superior to those which received 130/100 and 160/125% of the recommendation. Thus, it can be inferred that the response of sesame plants to the combined application of N/K₂O was more pronounced, when the waters of higher levels of salinity were used.

By evaluating the joint effect of water salinity and N/K₂O combinations, it can be noted that the increase in N/K₂O combinations associated with the highest salinity level (3.0 dS m⁻¹) caused reduction in the CO₂ assimilation of sesame plants (Fig 1B). The sources of N and K₂O used in the present study had salt indices of 75 and 116, respectively (Rarder et al., 1943). The highest ECw level, associated with the highest values of the N/K₂O combination, intensified the effect of salt stress, due to the osmotic effect caused by the increase of salts dissolved in the irrigation water associated with the use of fertilizers (Prazeres et al., 2015).

According to the mean comparison test (Fig 1A), sesame plants subjected to irrigation using water with ECw of 1.2, 1.8 and 2.4 dS m⁻¹ obtained the highest CO₂ assimilation rates when they were grown under combinations of 100/75%, 130/100% and 130/100% of the recommendation of N/K₂O, respectively (Fig 1B). This indicates that the supply of these nutrients acts in combination by reducing stress caused by the ECw, possibly due to the synergism between N and K, improving the efficiency of use of these nutrients by the plant.

It is known that N participates in several biomolecules, such as proteins, nucleic acids, amino acids and proline (McAllister et al., 2012). Also, K is vital for photosynthesis, carbohydrate formation (Araújo et al., 2012) and improvement of water balance (Heidari and Jamshid, 2010), which justifies the increments in *g_s* and *A* caused by the increase in the combined supply of N and K₂O, represented by fertilization with greater combinations of N/K₂O. Therefore, fertilization with an adequate combination of N/K₂O can result in higher photosynthetic rate and greater production of photoassimilates, as well as increased

accumulation of organic compounds, indicating that the combined supply of N and K contributes to better development of sesame under salt stress conditions.

The increase in irrigation water salinity adversely affects leaf transpiration in sesame plants cv. BRS G4. According to the regression equation (Fig 2A), the obtained data fitted best to a linear model. Transpiration decreased by 13.64% per unit increase in EC_w, which corresponded to a percentage decline of 35.65% in the *E* of plants irrigated with of 3.0 dS m⁻¹ water in comparison to those subjected to EC_w of 0.6 dS m⁻¹. This reduction in transpiration is caused by the increase in EC_w due to stomatal limitation. This stands out as a defense strategy of the plant to minimize the excessive dehydration or a consequence of water imbalance in leaf epidermis (Ribeiro et al., 2009), because *E* is closely related to stomatal opening. When stomatal conductance decreases due to the stress caused by salinity, as observed in this study (Fig 1A), there is an increase in the resistance to water diffusion from inside the leaf to the atmosphere.

The internal CO₂ concentration of sesame plants cv. BRS G4 increased linearly as the levels of irrigation water salinity increased. According to the regression equation (Fig 2B), plants irrigated with highest level of electrical conductivity (3.0 dS m⁻¹) obtained *C_i* of 273.16 μmol mol⁻¹, whereas plants subjected to the lowest salinity level (0.6 dSm⁻¹) showed *C_i* of 197.73 μmol mol⁻¹. In other words, there was an increment of 38.15% in the internal CO₂ concentration of sesame plants irrigated with EC_w of 3.0 dS m⁻¹ compared with those subjected to 0.6 dS m⁻¹. Corroborating this result, Tatagiba et al. (2014) claimed that plants subjected to salt stress exhibit reduction in stomatal conductance, leaf transpiration and photosynthesis, but the internal CO₂ concentration increases. Thus, the increment in internal CO₂ concentration can lead to loss of mesophyll capacity to absorb carbon, i.e., the CO₂ entering the mesophyll cells is not being fixed in the carboxylation stage, possibly because of damages to their structure.

According to Freire et al. (2014), the increase of *C_i* within the leaves indicates that CO₂ is not being used for synthesis of sugars in the photosynthetic process. This shows that some non-stomatal factor is interfering with this process, which leads to increased resistance to CO₂ diffusion to the substomatal chamber.

Effect of saline stress and nitrogen and potassium fertilization on sesame growth

Based on the data presented in Fig 2, it can be observed that the increase in irrigation water salinity led to decrease in the growth of sesame cv. BRS G4 plants, reflecting the negative effects of salt stress on photosynthetic rate and increase in internal CO₂ concentration in the substomatal chamber (Fig 1B and 2B). According to the regression equation (Fig 2C), the increase in EC_w caused a 14.11% reduction in plant height per unit, corresponding to a decline of 37.01% in plants irrigated using 3.0 dS m⁻¹ water, compared to those cultivated under the lowest salinity level (0.6 dS m⁻¹). The reduction in SD caused by salt stress was lower than that observed for PH, being equal to 31.91%, i.e., decrease of 2.82 mm in the SD of sesame cv. BRS G4 plants subjected to irrigation with the highest salinity level (3.0 dS m⁻¹), compared to the lowest EC_w level (Fig 2D).

As observed for plant height and stem diameter (Fig 2C and 2D), the number of leaves of sesame cv. BRS G4 was also negatively influenced by irrigation with saline water. According to the regression equation (Fig 2E), it decreased by 9.42% per unit increase of EC_w, which corresponded to a reduction of 24.01% (4.44 leaves) in the number of leaves of plants irrigated with 3.0 dS m⁻¹, compared with those subjected to the lowest EC_w level (0.6 dS m⁻¹). For leaf area, there was greater intensity of the negative effects of water salinity. According to the regression equation (Fig 2F), the reduction in LA was 16.75% per unit increase in EC_w, i.e., when sesame plants were irrigated with 3.0 dS m⁻¹ water. Their leaf area was on average 44.69 cm² inferior to the value of plants subjected to 0.6 dS m⁻¹. The deleterious effects of EC_w was observed on the PH, SD, NL and LA of the sesame crop irrigated with saline water equal to or greater than 1.2 dS m⁻¹, compared to plants irrigated with 0.6 dS m⁻¹ (Fig 2). Therefore, it can be inferred that irrigation using water with EC_w higher than 0.6 dS m⁻¹ compromises the growth of sesame cv. BRS G4. Moreover, the fact that salt stress is more pronounced on sesame leaf area (Fig 2F) indicates that the stressed plants reduced not only their number of leaves but also the expansion of their leaf blade, certainly as an adaptive mechanism aiming to reduce water loss by transpiration.

The reduction in PH, SD, NL and LA of sesame cv. BRS G4 is due to the increase in the concentration of salts in the soil solution from the irrigation water, which reduces the osmotic potential of the soil, limiting water absorption by plants and consequently compromising their photosynthetic and metabolic processes (Dias et al., 2017). This causes them to reduce their energy expenditure, and as a result, there is a decrease in plant growth (Liu and Jiang, 2015). Silva et al. (2017) explained that the decline in the number of leaves and leaf area are common morphological alterations in plants under salt stress, standing out as an important adaptive mechanism of plants grown under excessively saline conditions, aiming at reduction of the transport of Na⁺ and Cl⁻ in the xylem and concomitant maintenance of water in plant tissues.

Based on the mean comparison test (Fig 3A), the N/K₂O combination of 160:125%, which corresponded to the greater combined supply of N and K, caused losses in SD, NL and LA, leading to the lowest values (6.74 mm, 14.06 leaves and 54.82 cm², respectively) for these variables (Fig 3A, 3B and 3C). In addition, the N/K₂O combinations of 70/50, 100/75 and 130/100% caused the highest values of SD, statistically differing only from the treatment with 160/125%. Lower values of growth variables obtained from the N/K₂O combination of 160/125% are probably due to the intensification of the osmotic effect caused by the accumulation of salts in the soil, enhanced by inadequate management of fertilizers with high salt index. As already mentioned the photosynthetic rate decreased in plants irrigated with high-salinity water.

For the number of leaves (Fig 3B), sesame plants fertilized with N/K₂O combination of 70/50% differed statistically from plants subjected to the other combinations. However, when 100/75% is compared with 130/100%, there was no significant difference, as observed for 130/100% and 160/125%. In contrast, for leaf area (Fig 3C), plants which

Table 1. Chemical and physical characteristics of the Eutrophic Regolithic Neosol used in the experiment*.

		Chemical characteristics							
pH (H ₂ O) (1:2.5)	OM dag kg ⁻¹	P (mg kg ⁻¹)	K ⁺	Na ⁺	Ca ²⁺	Mg ²⁺	Al ³⁺ + H ⁺	ESP (%)	ECse (dS m ⁻¹)
6.24	1.079	48.00	0.28	1.82	7.41	5.23	3.07	10.22	2.50
Physical characteristics									
Size fraction (g kg ⁻¹)			Textural class	Water content (kPa)		AW	Total porosity m ³ m ⁻³	AD (kg dm ⁻³)	PD
Sand	Silt	Clay		33.42	1519.5				
656.6	175.0	168.4	SL	28.84	10.42	18.42	0.53	1.27	2.74

*Determined according to methodologies proposed by Donagema et al. (2011). OM – Organic matter: Walkley-Black wet digestion; Ca²⁺ and Mg²⁺ extracted with 1 M KCl at pH 7.0; Na⁺ and K⁺ extracted with 1 M NH₄OAc at pH 7.0; Al³⁺ and H⁺ extracted with 0.5 M CaOAc at pH 7.0; ESP – Exchangeable sodium percentage; ECse – Electrical conductivity of the saturation extract; SL – Sandy loam; AW – Available water; AD – Apparent density; PD – Particle density.

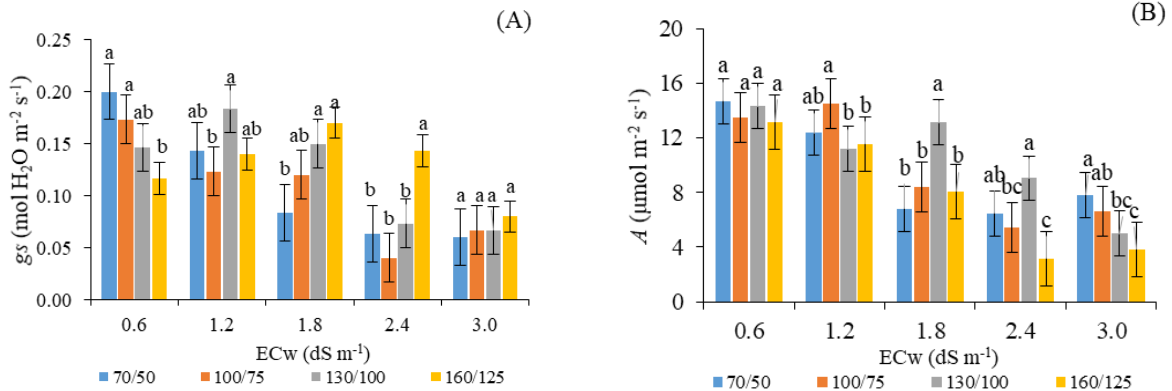


Fig 1. Stomatal conductance – g_s (A) and CO₂ assimilation rate – A (B) of sesame cv. BRS G4, as a function of the interaction between the combination of fertilization with nitrogen and potassium – N/K₂O and irrigation water electrical conductivity – EC_w, at 50 days after sowing. Means with different letters indicate that the treatments differ by Tukey test, $p < 0.05$; Bars represent mean standard error ($n = 3$).

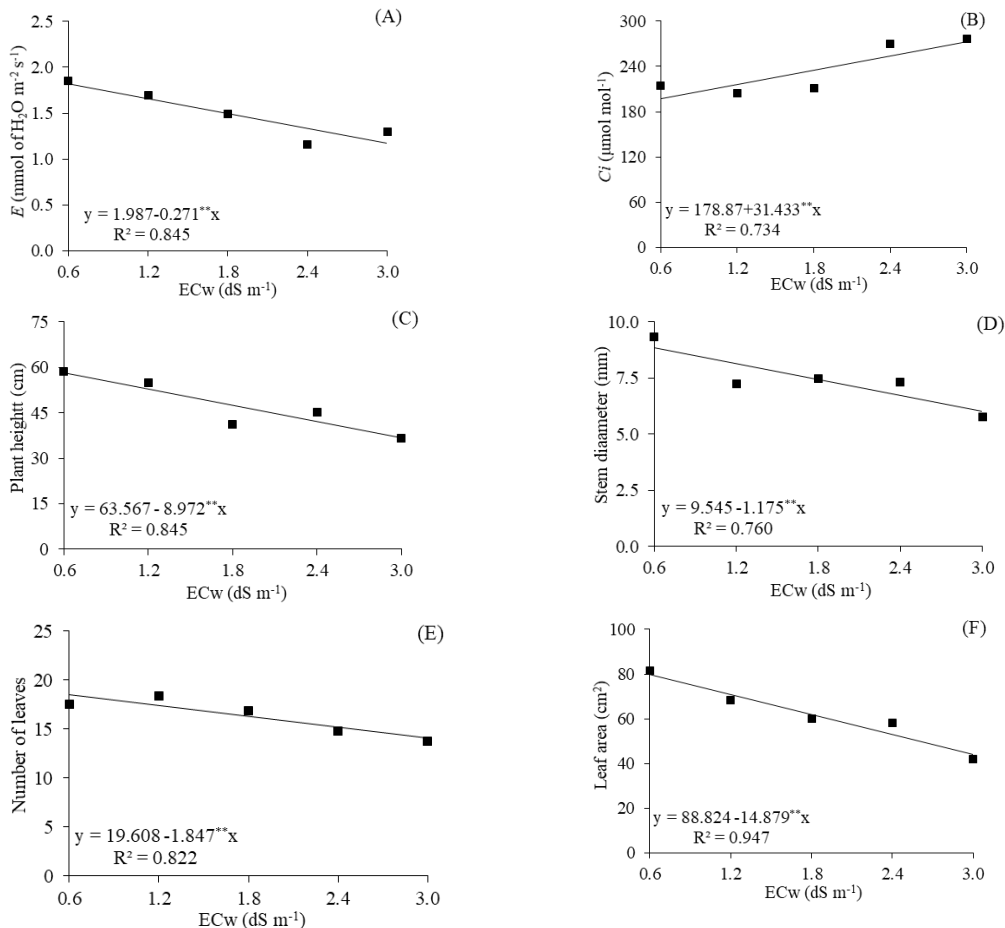


Fig 2. Transpiration – E (A) and internal CO₂ concentration – C_i (B), plant height (C), stem diameter (D), number of leaves (E) and leaf area (F) of sesame cv. BRS G4, as a function of electrical conductivity of irrigation water – EC_w, at 50 days after sowing.

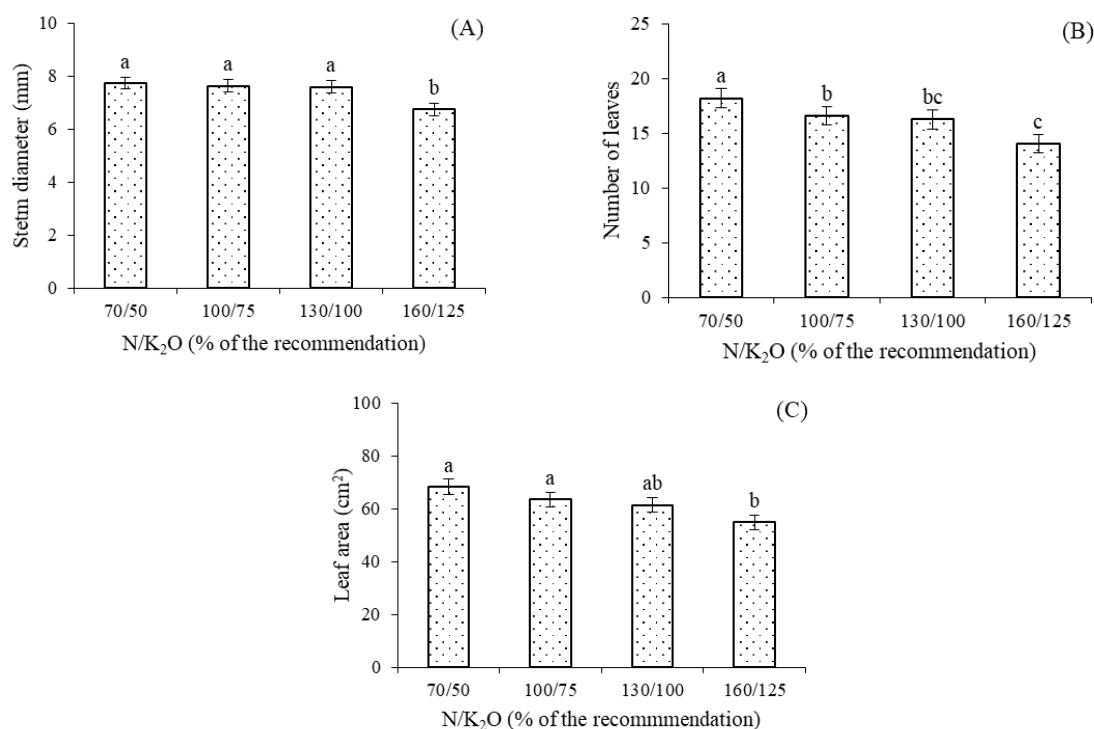


Fig 3. Stem diameter (A), number of leaves (B) and leaf area (C) of sesame cv. BRS G4, under different combination of nitrogen and potassium – N/K₂O fertilization and irrigated with saline waters, at 50 days after sowing; Means with different letters indicate that the treatments differ by Tukey test, $p < 0.05$; Bars represent mean standard error ($n = 3$).

received the 70/50% combination did not differ significantly only from those fertilized with 160/125% of N/K₂O. For both number of leaves and leaf area, the highest values were found using the N/K₂O combination of 70/50%. Thus, it can be inferred that the combination of 70/50% presents itself as the best N/K fertilization management for the crop, given the increased growth of the plants, possibly as a consequence of the better levels of stomatal conductance and CO₂ assimilation rate obtained in sesame plants subjected to this combination and irrigated with 0.6 dS m⁻¹ water (Fig 1). Additionally, the N/K₂O combination of 70/50% has economic importance, because it not only promotes the growth of sesame but also also represents reduction of costs related to lower requirement of inputs.

In general, plants tend to respond differently to fertilizer management, especially N and K. It is known that mineral fertilizers have distinct salt indices and, in excess, can be detrimental to plant metabolism. Therefore, fertilization is of great importance in a balanced combination. Thus, the supply of N having urea as source contributes to the acidification of soils (Lopes, 1989) for releasing H⁺ ions during the nitrification process, which, even at low concentration, directly affects plant growth by affecting nutrient availability. In addition, high K doses reduce calcium and magnesium absorption in plants due to competitive inhibition (Silva and Trevizam, 2015).

A study conducted by Lacerda et al. (2003) evaluated the growth of sorghum subjected to iso-osmotic solutions of NaCl/KCl salts at concentrations of 0.0/2.0, 71.5/0.5, 71.0/1.0, 70.0/2.0, 68.0/4.0 and 64.0/8.0 mM applied gradually. The authors found reductions in shoot dry matter and leaf area. These authors concluded that the increase in

the concentration of K⁺ salts, particularly KCl can cause greater reductions in growth than NaCl concentrations.

Materials and methods

Localization, experimental procedure, treatments

The study was carried out under greenhouse conditions from May to July 2017, at the Center of Technology and Natural Resources of the Federal University of Campina Grande (CTRN/UFCEG), located in the municipality of Campina Grande, PB, Brazil, at the geographic coordinates 7° 15' 18" S, 35° 52' 28" W and altitude of 550 m. The experimental design was randomized blocks with three replicates in a 5 x 4 factorial scheme, and treatments consisted of five levels of irrigation water electrical conductivity - EC_w (0.6; 1.2; 1.8; 2.4 and 3.0 dS m⁻¹) and four combinations of nitrogen and potassium: N/K₂O (70/50; 100/75; 130/100 and 160/125% of the recommendation for pot experiments). The doses relative to 100% (recommended dose) corresponded to 100, 300 and 150 mg kg⁻¹ of N, P₂O₅ and K₂O, respectively, according to Novais et al. (1991). Nitrogen (N) and potassium (K) fertilizations were carried out at 15, 25 and 35 days after sowing (DAS), using urea as source of N and potassium chloride as source of K while P was applied at sowing as super phosphate simples.

Plant material, establishment and management of the experiment

The plant material used in this study was the sesame cultivar BRS G4. Plants were grown in pots adapted as drainage lysimeters, connected at the bottom to a 4-mm-diameter

hose to drain the leachate into a container in order to evaluate the drained water and determine water consumption by plants. The tip of the drain inside the pot was involved with a nonwoven geotextile (Bidim OP 30) to prevent clogging by soil material.

The lysimeters were filled with a 0.5-kg layer of crushed stone (No. 0), followed by 25 kg of a Eutrophic Regolithic Neosol with sandy loam texture collected in the 0-20 cm layer, in the rural area of the municipality of Lagoa Seca, Paraíba. The soil material was properly pounded to break up clods and sieved, and its physical-hydraulic and chemical characteristics are presented in Table 1.

The irrigation waters with their respective values of electrical conductivity were prepared by dissolving the salts NaCl, CaCl₂.2H₂O and MgCl₂.6H₂O, at the equivalent proportion of 7:2:1, respectively, in water from the public supply system of Campina Grande, PB. Prior to sowing, the soil was brought to field capacity using the respective waters of each treatment. After sowing, irrigation was carried out daily by applying in each lysimeter a volume of water to maintain the soil moisture close to field capacity. The volume applied was determined based on 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.

Traits measured

The effects of the different salinity levels and combinations of N and K on the gas exchanges of sesame cv. BRS G4 were evaluated based on stomatal conductance (*g_s*), transpiration (*E*), CO₂ assimilation rate (*A*) and internal CO₂ concentration (*C_i*) using a portable Infrared Gas Analyzer (IRGA), 'LCPro +' model of ADC BioScientific Ltda. Changes caused in plant growth were also measured by collecting data of plant height (PH), stem diameter (SD), number of leaves (NL) and leaf area (LA) at 50 DAS. PH was measured as the distance from the collar to the apical meristem and SD was measured at 2 cm from the collar. NL was determined by counting the leaves, and LA was determined by following the methodology described by Silva et al. (2002), through Equation 1:

$$LA = 0.3552 \times L^2 \quad (1)$$

Where; L corresponds to the midrib length of the sesame leaf.

Statistical analysis

After collection, the data were subjected to analysis of variance by F test and, when significant, polynomial regression analysis was carried out for the factor water salinity levels and means comparison test (Tukey test at 0.05 probability level) was applied for the combination of fertilization with N and K, using the statistical software SISVAR 4.2 (Ferreira, 2011).

Conclusions

Water electrical conductivity above 0.6 dS m⁻¹ compromises the gas exchanges and consequently growth of sesame cv. BRS G4, at 50 days after sowing. Fertilization with nitrogen and potassium in the combination of 70/50% of the recommended dose of N/K₂O leads to greater growth of sesame cv. BRS G4. Combinations of 130/100, 160/125 and 160/125% of N/K₂O reduce the negative effects of irrigation with water of 1.2 and 1.8 and 2.4 dS m⁻¹, respectively, on the stomatal conductance of sesame plants. The combined supply of N and K₂O in combination with 100/75, 130/100 and 130/100% promoted higher CO₂ assimilation rate in sesame plants irrigated using water of 1.2 and 1.8 and 2.4 dS m⁻¹, respectively.

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