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Physiological indices of okra under organomineral fertilization and irrigated with salt water[1]

Índices fisiológicos de quiabeiro irrigado com água salina sob adubação organomineral

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HIGHLIGHTS:
Salt water reduces gas exchange in the okra crop, but organomineral fertilization mitigates these effects.
The use of organic and mineral fertilization is viable in okra crops under salt stress.
Organic fertilizer (100% bovine biofertilizer) decreases internal CO_2 concentration.

ABSTRACT: Proper management of brackish water can increase plant production in the Brazilian semiarid region. Organomineral fertilization contributes to minimizing the harmful effects of salinity. As such, the present study aimed to assess the physiological indices of okra crops grown under organomineral fertilization and irrigated with salt water. A completely randomized design was used, in a 6 × 2 factorial scheme with six repetitions, corresponding to six types of fertilization: T1 - mineral fertilization with NPK; T2 - bovine biofertilizer; T3 - fertilization with plant ash; T4 - mineral fertilizer (50%) + bovine biofertilizer (50%); T5 - mineral fertilizer (50%) + plant ash (50%); T6 - control; and two electrical conductivities of the irrigation water (0.5 and 5.0 dS m^{-1}). The physiological indices of okra plants were analyzed 30 and 60 days after transplanting (DAT). Although salt stress negatively affects the physiological indices of okra, organomineral fertilization partially mitigates these effects. Thus, organic and mineral fertilizers are recommended in okra crops under saline conditions.

Key words: *Abelmoschus esculentus*, physiology, plant nutrition

RESUMO: O manejo adequado de águas salobras pode aumentar a produção vegetal no Semiárido brasileiro. A fertilização organomineral contribui para a minimização dos efeitos deletérios da salinidade. Nessa perspectiva, objetivou-se avaliar os índices fisiológicos do quiabeiro cultivado sob adubação organomineral e irrigado com água salina. O experimento foi no delineamento experimental inteiramente casualizado no esquema fatorial 6 x 2, com seis repetições, referente a seis formas de adubação: T1 - Adubação mineral com NPK; T2 - Adubação com biofertilizante bovino; T3 - Adubação com cinza vegetal; T4 - Adubo mineral (50%) + biofertilizante bovino (50%); T5 - Adubo mineral (50%) + cinza vegetal (50%); T6 - Controle; e a duas condutividades elétricas da água de irrigação (0,5 e 5,0 dS m^{-1}). Aos 30 e 60 dias após o transplantio, foram analisados os índices fisiológicos do quiabeiro. O estresse salino afeta de forma negativa os indicies fisiológicos da cultura do quiabo, no entanto, a fertilização organomineral atenua parcialmente os efeitos dos sais sobre a fisiologia do quiabeiro. Desta forma, recomenda-se o uso da adubação orgânica e mineral no cultivo do quiabeiro em condições de salinidade.

Palavras-chave: *Abelmoschus esculentus*, fisiologia, nutrição vegetal

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Introduction

Okra [*Abelmoschus esculentus* (L.) Moench], a flowering plant in the family Malvaceae, with African origins, is primarily used for human consumption and has recently attracted interest from the industrial sector for fiber production. It is grown in tropical and subtropical areas due to its hardiness and low production costs (Marin et al., 2017; Torres et al., 2014; Sales et al., 2019).

In regions with a hot dry climate, such as the Brazilian semiarid, which is subject to drought at certain times of the year, irrigation is essential in order to safeguard production; however, limitations such as low quality water persist (Ribeiro et al., 2016).

The accumulation of ions such as Na^+ and/or Cl^- in chloroplasts causes several disturbances in plant biomolecular processes, resulting in osmotic effects, limiting water transport and restricting stomatal opening as well as the photochemical processes involved (Silveira et al., 2016).

There are several alternatives that can be used to mitigate the effects of salt stress on plants, including the use of organic liquid biofertilizers (Souza et al., 2019), nitrogen or potassium fertilization, or a combination of these in the form of organomineral fertilizers (Santos et al., 2019).

Several sources have been studied and applied to soil under salt water irrigation, as reported by Sousa et al. (2020), who used organic bovine biofertilizer in okra plants, Guedes Filho et al. (2015), who fertilized sunflowers with urea, and Prazeres et al. (2015), who applied potassium chloride to cowpea.

In light of the above, the present study aimed to assess the physiological indices of okra plants under organomineral fertilization irrigated with salt water.

Material and Methods

The experiment was conducted from September to December 2018, in the Professor Luiz Antônio da Silva Teaching Garden of the University of International Integration of the Afro-Brazilian Lusophony (UNILAB), in Redenção, Ceará state (CE), Brazil. Climate in the region is classified as tropical wet, with rain predominantly occurring in summer and fall (Köppen, 1923).

The substrate used was obtained by mixing so-called arisco sand, sand and bovine manure at a ratio of 4:2:1, respectively. The chemical analysis results are presented in Table 1.

A completely randomized design was used, in a 6 x 2 factorial scheme with six repetitions, in which the first factor corresponds to different types of fertilization: T1 - mineral fertilization with NPK (100% of the recommended dose); T2 - bovine biofertilizer (100%); T3 - fertilization with plant ash (100%); T4 - mineral fertilizer (50%) + bovine biofertilizer (50%); T5 - mineral fertilizer (50%) + plant ash (50%); T6 - control (no fertilization); and the second to two electrical conductivities of the irrigation water - ECw (0.5 and 5.0 dS m^{-1}).

Okra seeds (‘Santa Cruz 47’ cultivar) were sown in seed trays and transplanted to plastic pots containing 23 kg of substrate 15 days after seedling establishment, under full sun conditions.

Percolation lysimeters were prepared by drilling a hole into the bottom of each pot and attaching a hose to drain the water. To prevent leaks, the hoses were connected to 1 L PET bottles sealed with glue, which received the water drained each day.

Irrigation began 22 days after transplanting (DAT) and was performed daily using the percolation lysimeter method, supplying enough water every 24 hours to maintain the substrate at field capacity.

The salt water applied in irrigation was obtained in accordance with the methodology of Rhoades et al. (2000), whereby the amount of NaCl, $CaCl_2.2H_2O$ and $MgCl_2.6H_2O$ used to prepare the irrigation water was determined to obtain the desired ECw at a ratio of 7:2:1, from water used to supply the experimental area, which represents the control treatment (0.5 dS m^{-1}).

The mineral fertilization recommendations of Trani (2013) for okra, consisting of 80 kg ha^{-1} of N, 100 kg ha^{-1} of P_2O_5 and 60 kg ha^{-1} of K_2O, were adopted.

The biofertilizer was prepared using fresh bovine manure via aerobic fermentation, with the addition of nonsaline water (0.5 dS m^{-1}) at a ratio of 1:1, for 30 days. Plant ash (from sugarcane burning) was added in treatments T3 (1.0 kg) and T5 (500 g).

Table 2 shows the mineral concentrations of the bovine biofertilizer and plant ash.

At 30 and 60 DAT, the following physiological indices were analyzed: photosynthesis (A), transpiration (E), stomatal conductance (g_s), internal CO_2 concentration (C_i) and leaf temperature (TL), using an infrared gas analyzer (LI 6400 XT, LICOR) in an open system, with an air flow rate of 300 mL min^{-1}; measurements were taken between 10 a.m. and 12 p.m. on fully expanded leaves. Instantaneous water use efficiency (WUEi) was calculated as the ratio between the net photosynthetic rate (An) and transpiration rate (E).

The data were submitted to analysis of variance and Tukey’s test, using Assistat 7.7 Beta software (Silva & Azevedo, 2016).

Table 2. Nutritional composition of the bovine biofertilizer and plant ash

Organic fertilizer	N	P	K^+	Ca^{2+}	Mg^{2+}	Fe	Cu	Zn	Mn
	(g L^{-1})					(mg L^{-1})			
Biofertilizer	0.82	1.40	1.0	2.5	0.75	141.60	1.92	68.2	14.72
Plant ash	0.40	1.13	54.4	28.7	13.9	7819.1	10.5	37.8	240.8

Table 1. Chemical characteristics of the substrate used in okra cultivation

OM	N	Ca^{2+}	K^+	Mg^{2+}	Na^+	$H^+ + Al^{3+}$	ECse	SB	P	CEC	V
(g kg^{-1})		cmol$_c$ kg^{-1}					(dS m^{-1})		(mg kg^{-1})	(%)	
9.15	0.54	2.0	3.31	2.0	1.31	0.83	0.5	9.43	59	6.7	91

OM - Organic matter; ECse - Electrical conductivity of the saturated extract of the substrate; SB - Sum of bases (Ca^{2+} + Mg^{2+} + Na^+ + K^+); CEC - Cation exchange capacity - [Ca^{2+} + Mg^{2+} + Na^+ + K^+ + (H^+ + Al^{3+})]; V - Base saturation - (Ca^{2+} + Mg^{2+} + Na^+ + K^+/ CTC) x 100

Results and Discussion

Analysis of variance demonstrated significant interaction between fertilization and salinity for the variable transpiration (E) at 30 and 60 DAT (Table 3).

Additionally, this interaction significantly influenced leaf temperature (LT) and instantaneous water use efficiency (WUEi) at 30 DAT. Salinity was significant for photosynthesis (A) at 30 and 60 DAT, and for stomatal conductance (gs) and WUEi at 60 DAT. Finally, at 30 DAT, internal CO_2 concentration (Ci) showed significant results for fertilization and salinity.

Figure 1 shows that the photosynthetic rate declined at 30 and 60 DAT (Figures 1A and B, respectively), when plants were irrigated with highly saline water (5.0 dS m^{-1}).

The lower photosynthetic rates due to salt stress can be attributed to the decline in cell expansion that precedes photosynthesis inhibition, causing partial stomatal closing and thereby reducing the CO2 available to leaves (Gomes et al., 2015). This result can be justified by decreased stomatal conductance in the present study (Figure 2). Nascimento et al. (2012) found that salt stress causes an imbalance between the production and removal of reactive oxygen species (ROS). This increases ROS levels and triggers oxidative stress, with a series of negative implications for metabolic mechanisms associated with photosynthesis.

Similar results were reported by Sousa et al. (2020) for okra plants irrigated at an ECw of 2.0; 3.0; 4.0 and 5.0 dS m^{-1} when compared to controls (1.0 dS m^{-1}), and by Pereira Filho et al. (2019) in fava bean.

At 30 DAT, salt stress reduced the transpiration rate of okra (Table 4) by 31.7; 29.1 and 19.4% in treatments T1, T4 and T6, respectively, whereas the rise in ECw from 0.5 to 5.0 dS m^{-1} resulted in declines of 34.2, 49.0, 37.1 and 28.5% in T1, T2, T3 and T5, respectively (Table 4).

The effect of T1 for both assessments may have been due to the positive effect of photosynthesis in treatments involving irrigation with low saline water (0.5 dS m^{-1}). However, highly saline water (5.0 dS m^{-1}) decreased the transpiration rate

Table 4. Mean transpiration values (E) in okra plants grown under organomineral fertilization and irrigated with salt water, at 30 and 60 days after transplanting (DAT)

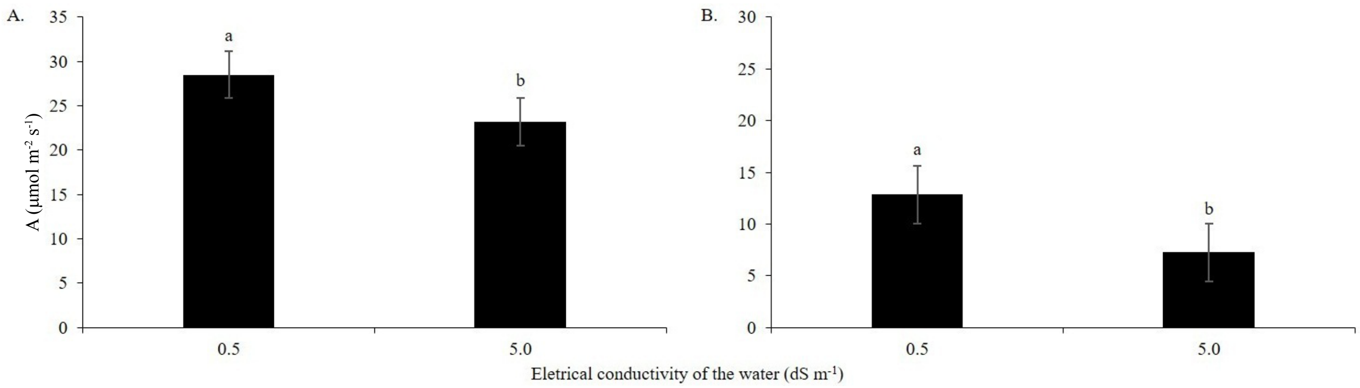
Fertilizers	E (mmol m^2 s^{-1})			
	30 DAT		60 DAT	
	Electrical conductivity (dS m^{-1})			
	0.5	5.0	0.5	5.0
T1	8.52 abA	5.82 bB	3.88 aA	2.55 bB
T2	6.43 bA	5.84 bA	3.98 aA	2.03 bB
T3	7.75 bA	6.72 abA	3.80 aA	2.39 bB
T4	10.43 aA	7.39 abB	3.77 aA	4.26 aA
T5	7.73 bA	8.42 aA	4.24 aA	3.03 bB
T6	9.18 abA	7.40 abB	3.69 aA	3.95 abA

Means followed by the same lowercase letter in the column and uppercase letter in the row do not differ according to Tukey's test at p ≤ 0.05; T1 - 100% mineral fertilization; T2 - 100% bovine biofertilizer; T3 - 100% plant ash; T4 - 50% mineral + 50% bovine; T5 - 50% mineral + 50% ash and T6 - Control

Table 3. Analysis of variance summary for the variables photosynthesis (A), transpiration (E), stomatal conductance (gs), internal CO_2 concentration (Ci), leaf temperature (LT) and instantaneous water use efficiency (WUEi) in okra plants grown under organomineral fertilization with high and low salinity irrigation at 30 and 60 days after transplanting (DAT)

SV	DF	Mean squares					
		A	E	gs	Ci	LT	WUEi
				30 DAT			
Fertilization (F)	5	21.45^{ns}	7.82**	8.91×10^{-3ns}	713.09*	7.12**	0.77**
Salinity (S)	1	412.75**	23.83**	0.19^{ns}	24110.41**	38.45**	0.36*
F x S	5	18.80^{ns}	3.85**	8.81×10^{-3ns}	524.40^{ns}	5.88**	0.82**
Treatments	11	57.92**	7.47**	0.02**	2754.35**	9.40**	0.76**
Residue	36	8.55	0.81	1.96×10^{-3}	249.02	0.12	0.05
CV (%)		15.43	11.81	14.68	7.46	1.07	6.71
				60 DAT			
Fertilization (F)	5	10.19^{ns}	1.39^{ns}	0.01^{ns}	1877.85^{ns}	2.28^{ns}	0.86^{ns}
Salinity (S)	1	374.30**	8.82**	0.14**	90.97^{ns}	19.13^{ns}	10.21**
F x S	5	14.13^{ns}	1.96*	0.01^{ns}	1351.37^{ns}	2.21^{ns}	1.65^{ns}
Treatments	11	45.08**	2.32**	0.02**	1476.09^{ns}	3.78*	2.07^{ns}
Residue	36	6.29	0.58	7.52×10^{-3}	757.81	0.25	1.05
CV (%)		24.93	21.97	22.88	10.41	1.46	34.74

SV - Sources of variation; DF - Degrees of freedom; *; ** and ^{ns} - Significant according to F test at p ≤ 0.05, p ≤ 0.01 and not significant, respectively; CV - Coefficient of variation



Means followed by the same letter do not differ according to Tukey's test at p ≤ 0.05

Figure 1. Mean photosynthesis values of okra plants irrigated under high and low salinity, at 30 (A) and 60 (B) days after transplanting (DAT)

(partial stomatal closure), reducing CO_2 absorption (Prazeres et al., 2015).

In order to adapt to excess salt, plants lower leaf water potential (by reducing the transpiration rate) in order to maintain a gradient that favors water absorption and flow in the soil-root-stem system, which typically increases the expenditure of metabolic energy (Taiz et al., 2017).

Similar data were reported by Sousa et al. (2020) in okra, whereby the use of a bovine biofertilizer as low saline levels (1.0 and 2.0 dS m^{-1}) positively affected the transpiration rate. Additionally, Prazeres et al. (2015) found that mineral fertilization increased transpiration in bean plants only when low salinity water was used (0.7 dS m^{-1}).

As shown in Figure 2, the salinity of irrigation water exhibited an isolated effect on stomatal conductance at 60 DAT.

Stomatal closure declines in plants under salt stress, reducing CO_2 assimilation in the leaf mesophyll and directly affecting internal CO_2 concentration as well as photosynthesis (Taiz et al., 2017).

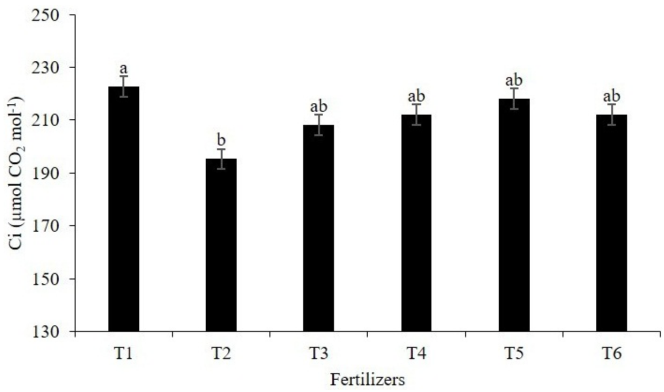
Since sodium in the soil occurs primarily as cations (Na^+), it competes for absorption sites in the roots with other minerals such as potassium, (K^+), resulting in possible potassium deficiency.

Melo et al. (2014) found that K accumulation in plants favors an osmotic gradient that facilitates water movement, regulating stomatal opening and playing a vital role in cell turgidity. However, these functions are significantly compromised in saline environments due to increased stomatal resistance and, consequently, lower gs.

Similar results were obtained by Sousa et al. (2020), who observed decreased stomatal conductance in okra plants irrigated with salt water. Souza et al. (2019) observed that an increase in salinity from 0.5 to 4.5 dS m^{-1} lowered stomatal conductance in fava beans.

In the present study, analysis of the effect of fertilization (Figure 3) on internal CO_2 concentration in okra at 30 DAT indicated that mineral fertilizer (T1) produced the greatest internal accumulation of this compound (222.71 µmol CO_2 mol^{-1}).

The high internal carbon concentration observed may be related to greater stomatal opening, evident in stomatal conductance at 30 DAT (Figure 2). In general, greater stomatal opening favors the entry of CO_2 from atmosphere into the leaf mesophyll, increasing its concentration in the substomatal cavity (Prazeres et al., 2015).



Means followed by the same letter do not differ according to Tukey's test at $p \leq 0.05$; T1 - 100% mineral fertilization; T2 - 100% bovine biofertilizer; T3 - 100% plant ash; T4 - 50% mineral + 50% bovine; T5 - 50% mineral + 50% ash and T6 - Control

Figure 3. Mean internal CO_2 concentration in okra plants under organomineral fertilization

According to Figure 4, at 30 DAT the increase in salinity reduced internal CO_2 concentration (189.04 µmol mol^{-1}) when compared to plants irrigated with water at 0.5 dS m^{-1} (233.86 µmol mol^{-1}).

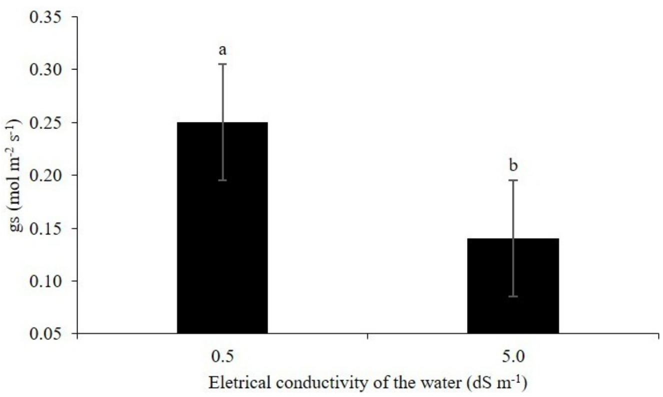
This response can be attributed to the fact that higher salt concentrations trigger soil osmotic pressure, restricting gs and CO_2 assimilation (Ribeiro et al., 2016). The proportional reductions in A (Figure 1) and gs (Figure 2) caused by salt stress indicate that the decline in Ci may be related to osmotic effects, which limit the entry of CO_2 for assimilation.

Silva et al. (2019) reported contrasting results, whereby a one unit increase in the electrical conductivity of irrigation water (0.5; 2.5; 5.0; 7.5 and 10.0 dS m^{-1}) raised the internal CO_2 concentration of sorghum.

With respect to the leaf temperature of okra (Table 5), a significant interaction was observed at 30 DAT. Temperature increased as the electrical conductivity of irrigation water rose. The highest LT values were recorded in plants fertilized with plant ash (35.58 °C), followed by 50% mineral fertilizer + 50% bovine biofertilizer (35.47 °C).

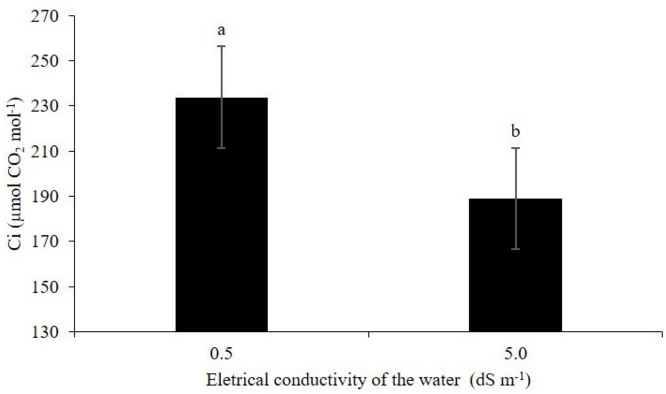
Leaf temperature is inversely proportional to transpiration rate, that is, as transpiration decreased with increased salinity (Table 5), leaf water content declined and leaf temperature rose.

Lacerda et al. (2011) reported contrasting results, whereby salt water irrigation increased the solar radiation intercepted by



Means followed by the same letter do not differ according to Tukey's test at $p \leq 0.05$

Figure 2. Mean stomatal conductance (gs) values of okra plants irrigated under high and low salinity, at 60 days after transplanting (DAT)



Means followed by the same letter do not differ according to Tukey's test at $p \leq 0.05$

Figure 4. Mean internal CO_2 concentration (Ci) in okra plants irrigated under high and low salinity, at 30 days after transplanting (DAT)

Table 5. Mean leaf temperature values (LT) in okra plants grown under organomineral fertilization and irrigated with salt water, at 30 days after transplantig (DAT)

Fertilizers	LT (°C)	
	Electrical conductivity (dS m^{-1})	
	0.5	5.0
T1	30.94 dB	33.24 dA
T2	31.57 dB	35.31 abA
T3	32.43 cB	35.58 aA
T4	34.37 aA	35.47 aA
T5	32.80 bcA	34.27 cA
T6	33.53 bB	34.72 bcA

Means followed by the same lowercase letter in the column and uppercase letter in the row do not differ according to Tukey's test at $p \leq 0.05$; T1 - 100% mineral fertilization; T2 - 100% bovine biofertilizer; T3 - 100% plant ash; T4 - 50% mineral + 50% bovine; T5 - 50% mineral + 50% ash and T6 - Control

the basal leaves of cowpea plants, improving the photosynthesis rate and leaf temperature.

In regard to WUEi, the addition of salt to irrigation water did not expose plants to stress at 30 DAT, demonstrating that okra is resistant to salt water up to this developmental stage (Table 6).

By contrast, Nobre et al. (2014) studied the growth, water consumption and use efficiency of castor bean plants under salt and nitrogen stress and found a decline in efficiency when the EC of water increased. However, in the present study, analysis of the effect of water at 0.5 dS m^{-1} demonstrated that the combined action of 50% mineral fertilizer + 50% bovine biofertilizer exposed okra plants to stress (2.72 μmol m^{-2} s^{-1} (mol H_2O m^{-2} s^{-1})$^{-1}$). The highest WUEi was obtained under 100% bovine fertilization (4.35 μmol m^{-2} s^{-1} (mol H_2O m^{-2} s^{-1})$^{-1}$). The superior performance of this input in terms of WUEi may be associated with the gradual availability of N, P and K, with adequate amounts favoring better plant-water relations.

Plants responded positively to treatments involving irrigation with saline water at 5.0 dS m^{-1} and fertilization, followed by the control. Additionally, the greatest efficiency was obtained with mineral fertilization, with 4.20 (μmol m^{-2} s^{-1} (mol H_2O m^{-2} s^{-1})$^{-1}$). The chemical attributes of this fertilizer may have promoted osmotic adjustment in the roots, ensuring better water absorption.

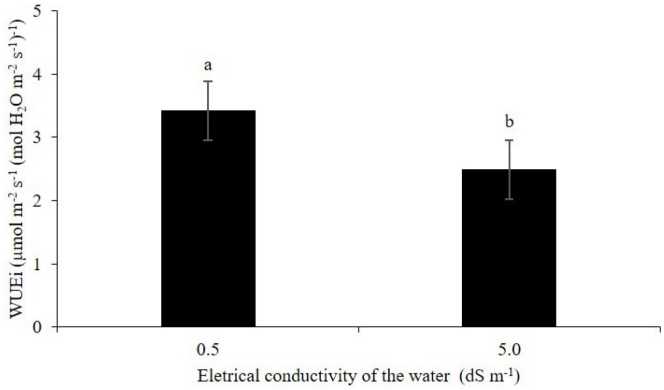
The WUEi of okra plants at 60 DAT was negatively affected by increased electrical conductivity. As shown in Figure 5, plants irrigated with 0.5 dS m^{-1} water exhibited higher WUEi (μmol m^{-2} s^{-1} (mol H_2O m^{-2} s^{-1})$^{-1}$) when compared to 5.0 dS m^{-1} (μmol m^{-2} s^{-1} (mol H_2O m^{-2} s^{-1})$^{-1}$).

Dias et al. (2016) reported that a higher salt concentration in irrigation water lowers soil osmotic and water potential, preventing the crop from absorbing the available water. Similar results were found by Lima et al. (2018), who observed reduced instantaneous water use efficiency at high saline levels in cotton, with a linear decrease of 8.17% per unit increase in ECw.

Table 6. Mean instantaneous water use efficiency values (WUEi) in okra plants grown under organomineral fertilization and irrigated with salt water, at 30 days after transplantig (DAT)

Fertilizers	WUEi (μmol m^{-2} s^{-1} (mol H_2O m^{-2} s^{-1})$^{-1}$)	
	Electrical conductivity (dS m^{-1})	
	0.5	5.0
T1	3.48 bcB	4.20 aA
T2	4.35 aA	3.74 abB
T3	3.84 bA	3.44 bB
T4	2.72 dB	3.76 abA
T5	3.50 bcA	3.47 bA
T6	3.15 cdA	3.48 bA

Means followed by the same lowercase letter in the column and uppercase letter in the row do not differ according to Tukey's test at $p \leq 0.05$; T1 - 100% mineral fertilization; T2 - 100% bovine biofertilizer; T3 - 100% plant ash; T4 - 50% mineral + 50% bovine; T5 - 50% mineral + 50% ash and T6 - Control



Means followed by the same letter do not differ according to Tukey's test at $p \leq 0.05$

Figure 5. Mean instantaneous water use efficiency (WUEi) in okra plants irrigated under high and low salinity, at 60 days after transplantig (DAT)

Conclusions

1. Although salt stress negatively affects the physiological indices of okra, organomineral fertilization partially mitigates these effects.
2. Organic and mineral fertilizers are recommended in okra crops under saline conditions.
3. Irrigation with water at 5.0 dS m^{-1} increases the leaf temperature of okra plants.

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