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Growth and physiology of maize under water salinity and nitrogen fertilization in two soils

Régis dos S. Braz¹, Claudivan F. de Lacerda¹, Raimundo N. de Assis Júnior¹, Jorge F. da S. Ferreira², Adriana C. de Oliveira¹ & Aureliano de A. Ribeiro¹

- ¹ Universidade Federal do Ceará. Fortaleza, CE, Brasil. E-mail: regismta@bol.com.br (Corresponding author) ORCID: 0000-0002-2992-0623; cfeitosa@ufc.br ORCID: 0000-0002-5324-8195; assisjr@ufc.br ORCID: 0000-0002-5892-0454; drica_fj@hotmail.com.br ORCID: 0000-0003-3931-7913; alburibeiro@hotmail.com ORCID: 0000-0001-5823-7615
- ² United States Department of Agriculture/US Salinity Laboratory. Riverside, CA, USA. E-mail: jorge.ferreira@ars.usda.gov ORCID: 0000-0003-4550-6761

ABSTRACT: The nitrogen fertilization, in general, minimizes the effects of salinity on plants, but the mitigating action depends on plant species, cultivar, soil texture and plant age, among other factors. The objective of this study was to evaluate the effects of irrigation water salinity and nitrogen fertilization on maize physiology and growth (*Zea mays*) in soils with different textures. The experiment was carried out in a greenhouse in Fortaleza City, CE, Brazil, in PVC soil columns (100 x 20 cm). The design was completely randomized using the factorial 2 x 2 x 4, referring to two soils, an Ultisol and a Quartzipsamment, nitrogen doses of 105 and 210 kg ha⁻¹, under irrigation with saline water of 0.5, 2.0, 4.0 and 6.0 dS m⁻¹, with four repetitions. With the exception of stem diameter, salinity of water impaired plant growth and leaf gas exchanges. The Ultisol provided better growth and physiological responses compared to the Quartzipsamment. The nitrogen doses did not mitigate growth variables except root biomass. The gas exchanges increased with the highest nitrogen dose, without interaction with the salinity.

Key words: Zea mays, leaf gas exchange, soil texture

Crescimento e fisiologia do milho sob salinidade da água e adubação nitrogenada em dois solos

RESUMO: A adubação nitrogenada, em geral, minimiza os efeitos da salinidade às plantas, mas a ação mitigadora depende da espécie vegetal, cultivar, textura do solo e idade das plantas, entre outros fatores. Assim, objetivou-se avaliar os efeitos da salinidade da água de irrigação e adubação nitrogenada na fisiologia e crescimento do milho (*Zea mays*) em solos com texturas diferentes. O experimento foi realizado em casa de vegetação em Fortaleza, CE, utilizando-se colunas de solo de policloreto de vinila - PVC (100 x 20 cm). O delineamento foi inteiramente casualizado usando o fatorial 2 x 2 x 4, referente a dois solos, um Argissolo Vermelho-Amarelo e um Neossolo Quartzarênico, doses de nitrogênio de 105 e 210 kg ha⁻¹, sob irrigação com águas de salinidade 0,5, 2,0, 4,0 e 6,0 dS m⁻¹, com quatro repetições. Com exceção do diâmetro do caule, a salinidade da água prejudicou o crescimento e as trocas gasosas foliares das plantas. O Argissolo proporcionou melhores respostas no crescimento e fisiologia em comparação ao Neossolo. As doses de nitrogênio não mitigaram as variáveis de crescimento, exceto a biomassa radicular. As trocas gasosas aumentaram com a maior dose de nitrogênio, sem interação com a salinidade.

Palavras-chave: Zea mays, trocas gasosas foliares, textura do solo



Introduction

Plant growth and development are results of genetic factors, environmental factors and their interactions, and their quantification makes it possible to improve agricultural production (Oliveira et al., 2010). Among the environmental factors which inhibit the development of agricultural crops, the excess of salts in soil or in the irrigation water stands out (Guimarães et al., 2013; Azevedo et al., 2017).

Salinity inhibits plant growth due to osmotic effects, toxic effects and damage to the absorption of essential nutrients. It also cause negative effects on gas exchanges as a result of the reduction in available water caused by decrease in the osmotic potential of the soil solution (Andrade et al., 2018).

The response of plants to salinity is complex, and physiological, biochemical and molecular adjustment mechanisms are necessary for their survival in saline environments (Willadino et al., 2017). The efficiency of these mechanisms depends on the species, genotype or cultivar, concentration and composition of salts in the irrigation water or soil solution.

Research studies point to the increase in nitrogen (N) supply as a management strategy capable of minimizing the effects of salinity on plants (Oliveira et al., 2014). This action results in greater accumulation of organic compounds containing N such as proline (Perveen & Nazir, 2018), which can increase the capacity for cellular osmotic adjustment, increasing the tolerance to water and salt stresses. However, salinity may interfere in the acquisition and utilization of N (Feijão et al., 2013) and soil physical properties may affect its availability to plants (Stevenson, 1982; Luizão et al., 2004).

Considering that water and/or soil salinity compromises the morphophysiological attributes of plants and that N attenuates salinity-induced damage, the objective was to evaluate the effects of irrigation using water with increasing salinity levels on the growth and physiology of the double-hybrid maize 'BRS 2020' in soils of different textures.

MATERIAL AND METHODS

The study was carried out in a greenhouse at the Federal University of Ceará (UFC), in Fortaleza, CE, Brazil (03° 45' S; 38° 33' W), at 19 m of altitude, in the period from April 5 to June 11, 2016. Data of temperature, air relative humidity and light intensity inside the greenhouse during the experimental period were obtained from a data logger (HOBO* U12-012 Temp/RH/Light/Ext model) programmed to record data every 15 min. The average daily temperature ranged from

25.8 to 31.6 °C; air relative humidity from 64.1 to 91.8% and light intensity (between 6 and 18 h) from 5,231 to 18,535 Lux.

Materials of the soils Ultisol and Quartzipsament were collected in the layers of 0-20, 21-60, 61-100 (Ultisol) and 0-20 and 21-100 cm (Quartzipsament), which correspond to their pedogenetic horizons, were used as substrate, following the same sequence found in the field to fill the columns. Both soils were collected and separated according to their horizons, pounded to break up clods, sieved through 5-mm mesh and characterized with respect to their physical and chemical attributes (Table 1).

Particle-size analysis was performed by the pipette method, soil bulk density by volumetric ring method, particle density by the volumetric flask method, electrical conductivity was measured in saturated paste extract and pH in water

The experimental design was completely randomized in a $2 \times 2 \times 4$ factorial arrangement, corresponding to two soils (Ultisol and Quartzipsamment), two N doses (210 and 105 kg ha⁻¹) and four levels of irrigation water salinity (0.5; 2.0; 4.0 and 6.0 dS m⁻¹), with four repetitions.

The polyvinyl chloride (PVC) columns with dimensions of 100 x 20 cm, used to cultivate the plants, received an internal layer of glue and sand to improve the adhesion between the soil and the PVC, decreasing the preferential water flow. The base was closed with a PVC cap, which was perforated and attached to a tube to collect drained water. In the assembly, the sequence of soil horizons as they were in the field was followed, compacting them up to a density of 1.53 kg dm⁻³. Sand was added to the column up to 10 cm height and the remainder was filled with the soils to be evaluated. For the Ultisol, the first layer (20 cm) corresponded to the A horizon, the second (25 cm) to the E horizon and the third (40 cm) to the textural B horizon. For the Quartzipsamment, the first layer (20 cm) was formed by the A horizon and the second (65 cm) by the C horizon.

Three seeds of the double-hybrid maize 'BRS 2020' were sown in each column, with thinning performed 10 days after sowing and leaving one plant per column. After thinning, saline waters began to be applied through daily irrigation, maintaining a leaching fraction of 0.15. Low-salinity water was obtained by diluting well water (EC of 1.0 dS m $^{-1}$) in distilled water until reaching EC $_{\rm w}$ of 0.5 dS m $^{-1}$, which was measured using a portable conductivity meter. The other saline levels of irrigation water were obtained by adding NaCl and CaCl $_{\rm h}$ 20 at equivalent proportion of 7:3, respectively.

Fertilization with 2.62 and 1.31 g of N per soil column in the form of urea and 1 g of K per column was divided into

Table 1. Physical and chemical characterization of the soils used in the experiment, according to the profile depth

Soil layer	Sand	Silte	Clay	Soil texture	ρς	EC _{es}	рН	P	Ca	Mg	Na	K	C	N
(cm)		(g kg ⁻¹)		Sour texture	(kg dm ⁻³)	(dS m ⁻¹)	μп	(mg kg ⁻¹)	(cmol _c kg ⁻¹)			(g kg ⁻¹)		
						Ultisol								
0-20	825	107	68	Sandy loam soil	1.47	0.12	6.3	33	1.90	0.30	0.16	0.43	12.3	1.24
21-60	845	85	70	Sandy loam soil	1.46	0.10	6.2	8	0.70	0.50	0.17	0.24	7.32	0.75
61-100	527	157	316	Clay loam soil	1.35	0.11	5.7	5	1.10	0.60	0.18	0.90	3.36	0.35
Quartzipsamment														
0-20	923	53	24	Sandy soil	1.54	0.36	5.3	2	0.90	0.60	0.21	0.59	7.56	0.77
21-100	929	42	29	Sandy soil	1.55	0.46	5.4	2	0.50	0.40	0.28	0.67	2.16	0.22

 $[\]rho_s$ – Soil bulk density; EC_{es} – Electrical conductivity of the substrate

four applications: 15% at 10 days after sowing (DAS), 25% at 25 DAS, 25% at 40 DAS and 35% applied at 55 DAS. At sowing, each column received 1.12 g of P and 1 g of the FTE BR 12 mix, containing Ca (7.1%), S (5.7%), B (1.8%), Cu (0.8%), Mn (2.0%), Mo (0.1%) and Zn (9.0%). Magnesium was supplied as magnesium sulfate through irrigation water.

At 67 days after sowing, the following variables were evaluated: growth in height, stem diameter at the base of the plant, leaf area using a leaf area integrator, dry biomass of roots, dry biomass of stem + sheaths, dry biomass of leaf blades and total dry biomass. Measurements of stomatal conductance (gs), photosynthesis (A), transpiration (E) and internal CO₂ concentration (Ci) were performed at 15, 30, 45 and 60 days after the treatments began, in the first fully expanded leaf, from 8:00 to 10:30 a.m., using a portable infrared gas analyzer (LI6400XT model, Licor, USA) under saturating light of 1,800 µmol m⁻² s⁻¹ and ambient conditions of temperature and CO₂ concentration.

The data were subjected to the Kolmogorov-Smirnov normality test, as a prerequisite for analysis of variance by F test. Means referring to soils and N doses were compared by F test, which is conclusive for factors with two levels, whereas those referring to water salinity were compared by regression using the program Assistat version 7.7 (Silva & Azevedo, 2016).

RESULTS AND DISCUSSION

Except for root dry biomass, the interaction between water salinity, N and soil type did not interfere significantly in any of the variables evaluated and the interaction between soil and water salinity caused significant effects only on root dry biomass (RDB) and total dry biomass (TDB) (Table 2).

The Ultisol resulted in higher growth and biomass production of the plants than the Quartzipsament, with superiority of 22.78, 12.58, 32.38, 45.94, 36.47, 38.91 and 41.03%, respectively for the growth in height, stem diameter, leaf area, dry biomass of roots, stem + sheaths, leaves and total dry biomass (Table 2). This superiority of the Ultisol over the Quartzipsament can be a response to the lower sand proportion and higher availability of nutrients (Table 1), especially potassium, which performs osmoregulatory

function in plants under salt stress, providing better conditions for root and shoot growth of plants in general, including maize.

Additionally, the higher clay proportion of the Ultisol possibily promote higher aggregation and contribute to aggregate stability, resulting in greater soil porosity for water storage, water dynamics and nutrients for plant growth (Wang et al., 2017). Similar responses have been obtained by Santos et al. (2017) for stem height and diameter in soybean, Fraga Júnior et al. (2012) for leaf area in citrus and by Braccini et al. (1999) for dry biomass in maize, all with higher results in clayey soils compared to soils with lower clay content.

In relation to N fertilization, significance was only observed on RDB, in which the dose of 210 kg ha⁻¹ resulted in higher root biomass. The increase of N supply, in general, stimulates phytomass production, indicating that N can favor the processes of plant growth and development, due to higher carbon fixation (Martuscello et al., 2006). In addition, roots have faster osmotic adjustment and slower loss of turgor than the shoots (Oliveira et al., 2016). Similar responses in maize also showed that N fertilization promoted increase in RDB (Morais et al., 2015).

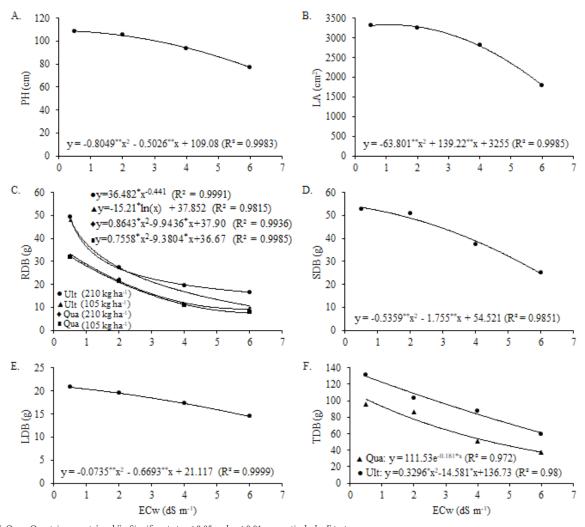
Compared to the control treatment (0.5 dS m⁻¹), PH decreased by 28.65% as a function of the increase in irrigation water salinity up to the level of 6.0 dS m⁻¹ (Figure 1A). Soil salinity limits plant growth due to excess of salts in the root zone, causing a greater osmotic effect outside the roots and restricting the water flow from the soil to the plants (Lima et al., 2014). With the increase in the osmotic pressure of the medium, there is a reduction in the readily available water, affecting cell division and elongation (Taiz et al., 2017). The decrease of PH in maize has also been documented by Gomes et al. (2011) and Souza et al. (2014) using irrigation water with EC levels of 5.0 and 4.5 dS m⁻¹, respectively.

Compared to the control, plants irrigated with EC $_{\rm w}=6.0$ dS m $^{-1}$ had a 46.3% decrease in LA (Figure 1B). Plants under water and salt stresses have smaller LA, thus reducing the transpiration surface and water loss (Taiz et al., 2017). The reduction in LA also contributes to maintaining the balance between absorption and transpiration, which may have increased water use efficiency, reducing the accumulation of salts in the plant (Nobre et al., 2011). However, the reduction

Table 2. Summary of analysis of variance, by mean squares, for plant height (PH), stem diameter (SD), leaf area (LA), root dry biomass (RDB), dry biomass of stem + sheaths (SDB), dry biomass of leaf blades (LDB) and total dry biomass (TDB) of maize plants under water salinity and nitrogen fertilization in soils with different textures

Treatments	PH	SD	LA	RDB	SDB	LDB	TDB
Soil (A)	37.92**	14.32**	27.96**	291.15**	40.43**	52.65**	101.64**
N dose (B)	0.08 ^{ns}	0.08 ^{ns}	3.90 ^{ns}	11.70**	0.06 ^{ns}	2.11 ^{ns}	2.19 ^{ns}
Salinity (C)	26.26**	2.64 ^{ns}	23.12**	703.98**	40.70**	11.73**	106.46**
AxB	0.003 ^{ns}	2.05 ^{ns}	2.46 ^{ns}	1.99 ^{ns}	2.25 ^{ns}	1.65 ^{ns}	2.13 ^{ns}
AxC	0.57 ^{ns}	1.24 ^{ns}	1.92 ^{ns}	31.53**	1.60 ^{ns}	1.50 ^{ns}	2.97*
BxC	0.32 ^{ns}	0.70 ^{ns}	0.57 ^{ns}	5.69**	0.67 ^{ns}	0.49 ^{ns}	1.19 ^{ns}
AxBxC	0.98 ^{ns}	1.03 ^{ns}	1.63 ^{ns}	3.12*	0.60 ^{ns}	1.45 ^{ns}	0.54 ^{ns}
CV (%)	13.29	12.28	21.08	8.76	19.40	17.95	13.51
	cm	mm	cm ²	g	g	g	g
Ultisol	204.8 a	17.0 a	3181.9 a	26.97 a	47.90 a	20.92 a	95.79 a
Quartzipsamment	166.8 b	15.1 b	2403.6 b	18.48 b	35.10 b	15.06 b	67.92 b
210 kg ha ⁻¹ N	184.9 a	16.0 a	2938.2 a	23.57 a	41.74 a	18.57 a	83.90 a
105 kg ha ⁻¹ N	186.7 a	16.1 a	2647.4 a	21.87 b	41.25 a	18.57 a	79.81 a

ns - Not significant; 'and " - Significant by F test, at $p \le 0.05$ and $p \le 0.01$, respectively; Means followed by the same letter in the column do not differ statistically by F test at $p \le 0.05$



Ult - Ultisol; Qua - Quartzipsament; 'and " - Significant at $p \le 0.05$ and $p \le 0.01$, respectively, by F test **Figure 1.** Plant height-PH (A), leaf area-LA (B), root dry biomass-RDB (C), dry biomass of stem + sheaths-SDB (D), dry biomass of leaf blades-LDB (E) and total dry biomass-TDB (F) of maize plants under salt stress in two soils

in the area for photosynthesis can cause yield losses. Similar results were observed in sweet corn and popcorn with LA reductions of 28.2 and 28.7%, respectively, when irrigated using water with EC of 4.5 dS m⁻¹, compared to 0.5 dS m⁻¹ (Souza et al., 2014).

The root dry biomass (RDB) decreased in the Ultisol and Quartzipsament as the EC_w increased, regardless of the applied N dose (Figure 1C). However, it is observed that in both soils this decrease was lower at the highest N dose, especially at the highest levels of salinity, indicating that N can reduce the effects of salinity on the reduction of root biomass.

The difference found in the root dry biomass content between the Ultisol and the Quartzipsament can be attributed to the fact that clay soils have a higher nitrogen retention capacity than sandy soils. In this case, a higher amount of N in the Ultisol may provide increased biomass production. Since N performs structural function, participating in organic compounds that are vital to plants, it plays the role of promoting growth and can inhibit the effects of salinity on plants. Melo et al. (2015) also observed lower losses in RDB due to N fertilization.

The increase in EC_w caused losses of 52.14 and 30.45% in SDB (Figure 1D) and LDB (Figure 1E), respectively. The

threshold water salinity for the maize crop is $1.1~dS~m^{-1}$ (Ayers & Westcot, 1999). Salinity levels above the threshold of the species or cultivar reduces growth due to increased metabolic energy spent on the absorption of water and nutrients and on osmotic adjustment, besides reducing crop growth and production (Rhoades, 2000). The ionic effect caused by the accumulation of ions in plant tissues also reduces plant growth (Munns & Tester, 2008), affecting biomass production. Similar responses were found in popcorn, which showed a reduction in SDB and LDB as salinity increased (Oliveira et al., 2016).

There was a reduction in TDB as the EC $_{\rm w}$ increased, with losses of 54.45 and 60.72% in the Ultisol and Quartzipsament, respectively (Figure 1F). The smaller loss of TDB in the Ultisol suggests that, even under salinity conditions, the more clayey texture of the Ultisol provides greater capacity of water and nutrient retention, increasing the availability of both to the plant. The high concentration of salts in the irrigation water can lead to break in water potential homeostasis, ionic imbalance at the soil-plant interface and cause toxicity in the plant, altering its growth and dry matter production (Asch et al., 2000). Sousa et al. (2012) also reported that maize TDB decreased with the use of irrigation water with EC $_{\rm w}$ = 3.4 dS m $^{-1}$.

The soil type was significant for gs, A, E and Ci and the N doses were significant for gs, A and E (Table 3). The salinity

Table 3. Summary of ANOVA and means for stomatal conductance (gs), transpiration (E), photosynthesis (A), and internal CO_2 concentration (Ci) in maize plants under salt stress and nitrogen doses in two soils

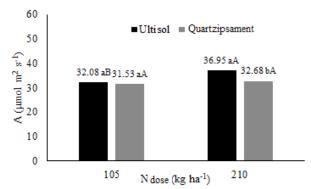
SV	gs	E	A	Ci	
Soil (A)	52.95**	5.39*	9.80**	4.95*	
N dose (B)	9.65**	4.67*	15.33**	1.59 ^{ns}	
Salinity (C)	113.33**	18.39**	34.60**	20.09**	
AxB	0.49 ^{ns}	3.12 ^{ns}	5.87*	1.57 ^{ns}	
AxC	8.01**	0.62 ^{ns}	0.08^{ns}	2.10 ^{ns}	
BxC	1.16 ^{ns}	1.44 ^{ns}	1.22 ^{ns}	2.21 ^{ns}	
AxBxC	2.59 ^{ns}	1.24 ^{ns}	2.30^{ns}	1.14 ^{ns}	
CV (%)	7.75	13.29	9.23	27.44	
	mol m ² s ⁻¹	mmol m ² s ⁻¹	μmol	m ² s ⁻¹	
Ultisol	0.22 a	6.19 a	34.52 a	64.57 a	
Quartzipsament	0.19 b	5.73 b	32.11 b	55.41 b	
210 kg ha-1 N	0.21 a	6.18 a	34.82 a	57.39 a	
105 kg ha ⁻¹ N	0.20 b	5.75 b	31.81 b	62.59 a	

SV - Source of variation; "s - Not significant, 'and " - Significant by F test, at $p \leq 0.05$ and $p \leq 0.01$, respectively; Means followed by the same letter for the same soil or N dose in the column do not differ statistically by F test at $p \leq 0.05$

levels significantly influenced all variables. There were also significant interactions of 'soils x doses' for A and 'soil x salinity' for gs.

Plants cultivated in the Ultisol, compared to the Quartzipsament, showed higher values of gs, A, E and Ci with successive increments of 15.79, 8.03, 7.51 and 16.53% (Table 3), which could possibly be attributed to the fact that the Ultisol has more nutrients, promoting better development of both shoots and root system, whereas the highest N dose led to increments in gs, A and E. However, there was no effect of N doses on Ci. Since N is part of the main components of the photosynthetic system, the increase in its availability generally results in positive effects on the carbon assimilation rate (Correia et al., 2005).

The interaction between soils and N doses affected photosynthesis (Figure 2). For the dose of 210 kg ha⁻¹, the highest photosynthetic rates were found in the Ultisol, showing

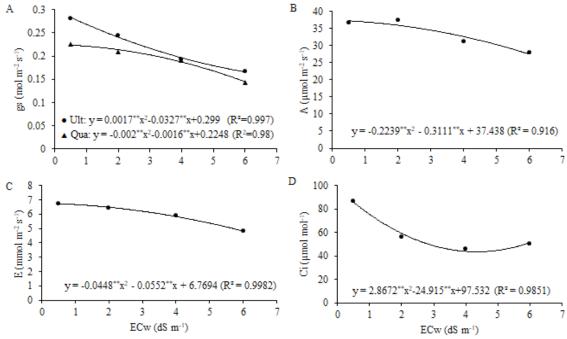


Means followed by the same letter do not differ statistically by F test at $p \leq 0.05;$ The lowercase letters compare the soils in each N dose and the uppercase letter the N doses in each soil

Figure 2. Photosynthesis (A) in maize plants under salt stress and nitrogen fertilization in two soils

greater utilization of N in this soil, especially due to its better structuring, water retention and nutrient adsorption. However, there was no interaction between the dose of 105 kg ha⁻¹ and the two soils, or between both N doses and Quartzipsament.

In comparison to the control, the losses of gs in plants subjected to EC_w of 6.0 dS m⁻¹ were 40.78% in the Ultisol and 36.43% in the Quartzipsament (Figure 3A). However, it is worth pointing out that, although plants cultivated in the Ultisol had the highest losses of gs, this soil led to higher values of gs compared to the Quartzipsament. Stomatal closure is one of the first responses of the plant to stress in order to avoid excessive water loss (Gomes et al., 2011) and occurs due to the decrease of water flow in the plant as a result of the high concentration of salts in the root zone. The reduction in the osmotic potential of the soil solution decreases water availability to plants and causes a water stress that consequently leads to stomatal closure. Sousa et al. (2016) also verified, in the maize crop, that irrigation with saline waters causes reduction in gs.



Ult - Ultisol; Qua - Quartzipsament; ** - Significant at $p \le 0.01$ by F test

Figure 3. Stomatal conductance-gs (A), net photosynthesis-A (B), transpiration-E (C) and internal CO₂ concentration-Ci (D) of maize plants as a function of irrigation water electrical conductivity (EC_w)

Photosynthesis was reduced by 23.53% in plants irrigated with EC_w = 6.0 dS m⁻¹, compared to the control plants. Photosynthesis is one of the main physiological processes affected by salinity and can be inhibited by the accumulation of Na⁺ and/or Cl⁻ ions in chloroplasts, which affect the biochemical and photochemical processes involved in photosynthesis (Taiz et al., 2017). Additionally, the water deficit induced by salinity tends to cause stomatal closure, which results in limitation in Ci and may also affect the concentration of the pigments involved in photosynthesis. With the reduction in gs, the availability of CO₂ to the leaves decreases (Gomes et al., 2011), causing a reduction in the net photosynthesis due to the decrease in the partial pressure of CO₂ in the intercellular spaces or in the substomatal chamber (Amorim et al., 2010). Results similar to the present study were obtained by Silva et al. (2013), who found that the increase in irrigation water salinity reduced photosynthesis in cowpea.

The increase in salinity also reduced E, causing losses of 28.67% when plants were subjected to the maximum EC_w (Figure 3C). Plants under salt stress generally show a reduction in the water flow to the shoots, due to the decrease in gs, reducing the flow of water vapor to the atmosphere (Gonçalves et al., 2010). Sousa et al. (2016) also reported that E was reduced by salinity in maize.

The CO_2 concentration (Ci) decreased as the irrigation water salinity increased to 4.0 dS m⁻¹, with a reduction of 46.95% compared to the control (Figure 3D). Subsequently, Ci increased with the EC_{w} of 6.0 dS m⁻¹. The decrease in Ci is related to lower gs, because the increase in the concentration of salts in soil reduces the osmotic potential of its solution, causing water stress and leading to stomatal closure, which results in a limitation of Ci. Thus, stomatal closure usually reduces gs, Ci and CO_2 assimilation rate (Musyimi et al., 2007). However, the subsequent increase in Ci with the highest salinity level points to a non-stomatal effect on photosynthesis, indicating a more severe stress condition for maize plants.

Conclusions

- 1. The nitrogen doses did not attenuate the effects of salinity on maize plants.
- 2. Although root biomass increased with the additional nitrogen fertilization, there were different effects depending on the class of soil.

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