

REGULAR ARTICLE

## Evaluation of the salinity tolerance of cotton trees under saline stress on two substrates

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### Regular Section

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### Statements and Declarations

#### Data availability

All data will be shared if requested.

#### Institutional Review Board Statement

Not applicable.

#### Conflicts of interest

The authors declare no conflict of interest.

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#### Autor contribution

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### Abstract

The aim of this study was to evaluate the salinity tolerance of cotton plants grown under saline stress in two substrates. The experiment was carried out in an experimental area at the Centec Cariri School of Technology (FATEC Cariri), located in the city of Juazeiro do Norte, Ceará state. A completely randomized design was applied, using a 4 x 2 factorial scheme consisting of four irrigation water salinity levels (0.5; 3.0; 5.0, and 10 dS m<sup>-1</sup>) and two substrates (0 - 20 cm soil layers and earthworm humus) with four replications. To assess the tolerance of cotton plants to salinity, the methodology proposed by Fageria (1985) was used. The percentage reductions in plant height, absolute growth rate, stem diameter, number of leaves, and leaf area of cotton plants submitted to different levels of irrigation with saline water in the two substrates were quantified, comparing them to the control (plants cultivated with water whose electrical conductivity is equal to 0.5 dS m<sup>-1</sup>). Based on the found results, it was found that the used type of substrate changed the classification of cotton salinity tolerance, especially at the highest salinity levels tested (5.0 and 10 dS m<sup>-1</sup>). When considering the data of stem diameter and leaf area, cotton plants grown in soil substrate showed greater tolerance to salinity. However, from the data regarding absolute growth rate and number of leaves, cotton plants grown in earthworm humus substrate were more tolerant to salinity.

### Keywords

*Gossypium hirsutum* L., growth. saline water. earthworm humus.



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### Introduction

Soil salinity is one of the biggest challenges for agricultural production, especially in arid and semi-arid regions of the planet (El Hasini et al., 2019). According to the Food and Agriculture Organization of the United Nations, the global area of soils affected by salts is about 424 million hectares of topsoil (0–30 cm) and 833 million hectares of subsoil (30–100 cm) (based on 73% of land mapped so far) (FAO, 2022). By 2050, it is expected that 50% of the world's arable land will be affected by salinity (Jamil et al., 2011).

Salt stress can severely affect crop growth and development throughout the life cycle, including seed germination, seedling establishment and production (Zhu, 2016). These effects are associated with low osmotic potential of the soil solution, nutritional imbalance, ionic toxicity, hormonal imbalance, and induction of oxidative stress, or a combination of these factors (Abdelhamid et al., 2013; Schoebitz et al., 2013; Nadeem et al., 2013; Mansour and Ali, 2017).

Cotton (*Gossypium hirsutum* L.) is among the most suitable species for cultivation in saline conditions because it is tolerant to saline stress and consumes a relatively low daily amount of water (Oliveira et al., 2013). Thus, its cultivation appears as an option for generating employment and income in arid and semi-arid regions, such as the Brazilian Northeast (Lima et al., 2016). However, although the crop is considered tolerant to salinity (Dong, 2012; Mahajan and Tuteja, 2005), numerous studies have shown that its growth and production are markedly reduced when irrigated with waters having higher saline concentrations (Capitulo et al. 2016; Min et al. 2016; Santos et al. 2016; Dias et al. 2020; Ribeiro et al. 2020; Guo et al. 2020; Pereira et al. 2020; Hamani et al. 2021).

In recent years, one of the challenges faced by researchers has been to find alternatives to minimize the deleterious effects of salts on cultures. In this context, some studies carried out by Ribeiro et al. (2013) on yellow passion fruit, Almeida et al. (2019) in eggplant plants, Dias et al. (2019) on arugula, and Dias et al. (2022) on radish, showed that the choice of substrate to be used can mitigate the deleterious effects of salts in these cultures.

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In this context, the aim of the present study was to evaluate the salinity tolerance of cotton plants grown under saline stress in two substrates.

## Materials and methods

The experiment was conducted from April to June 2022 in an experimental area at the Centec Cariri School of Technology (FATEC Cariri), belonging to the Institute for Technological Education (CENTEC), located in the city of Juazeiro do Norte, Ceará state, with the geographic coordinates 07°12'47"S, 39°18'55"W. The city, located at 377 meters of altitude, has a climate between Tropical Semiarid to Tropical Semiarid Mild, with an average temperature of 24 to 26 °C, and rainy season from January to May. The average rainfall is 925 mm (Lima et al., 2012). Within the Köppen climate types (Köppen and Geiger, 1928), one can identify as predominant in Juazeiro do Norte the climate class BSW'h',

that is, Semi-arid Climate, with a short rainy season starting in the summer and reaching its peak in the transition between summer and autumn.

A completely randomized design was applied to this experiment, using a 4 x 2 factorial scheme consisting of four irrigation water salinity levels (0.5; 3.0; 5.0, and 10 dS m<sup>-1</sup>) and two substrates (0 - 20 cm soil layer and earthworm humus), with four replications. Each experimental unit was represented by a 550 mL plastic cup containing one plant, totalling 32 experimental units.

The soil was collected in an experimental area of Fatec Cariri at a depth of 0 to 20 cm, being sieved and after that, used to fill the plastic cups. The earthworm humus was purchased in agricultural stores in the city of Juazeiro do Norte. The chemical characterization of the soil and earthworm humus can be found in Table 01.

**Table 1:** Characterization of the soil from the 0-20 cm soil layer and earthworm humus used in the experiment.

0 - 20 cm Soil Layer									
ECse	pH	SB	V	T	Ca <sup>2+</sup>	Mg <sup>2+</sup>	Na <sup>+</sup>	K <sup>+</sup>	H <sup>+</sup> +Al <sup>3+</sup>
dS m <sup>-1</sup>	-	cmol <sub>c</sub> /dm <sup>3</sup>	%	cmol <sub>c</sub> /dm <sup>3</sup>	-----		cmol <sub>c</sub> /dm <sup>3</sup>	-----	
0.15	6.0	2.83	58	4.91	1.90	0.70	0.03	0.20	2.08
Earthworm Humus									
ECse	pH	Humidity	C	OM	Ca <sup>2+</sup>	Mg <sup>2+</sup>	P	K <sup>+</sup>	N
dS m <sup>-1</sup>	-	%	g/kg	cmol <sub>c</sub> /dm <sup>3</sup>	-----		g/kg	-----	
3.43	7.50	64.90	216.95	374.01	14.70	4.95	7.31	13.50	13.34

pH – hydrogen ion potential; ECse – electrical conductivity of the saturation extract; SB – sum of exchangeable bases; T – cation exchange capacity; V – base saturation; C – carbon; OM – organic matter.

The cotton cultivar BRS 433FL B2RF was chosen, having been evaluated for 37 days (April 25, 2022 to June 2, 2022). The seeds were acquired at the Experimental Field of Embrapa Algodão, located in the city of Barbalha, Ceará state. Sowing was carried out by placing one seed per cup, 2 cm deep. The application of saline treatments started eight days after sowing (DAS).

The different salinity levels of irrigation water (3.0; 5.0, and 10.0 dS m<sup>-1</sup>) were obtained by the addition of sodium chloride (NaCl) to the local water supply (ECa = 0.5 dS m<sup>-1</sup>) following the methodology proposed by Richards (1954): C = ECs x 640, where C = salt concentration, mg L<sup>-1</sup>; ECs = electrical conductivity of the solution, dS m<sup>-1</sup>. After preparing the saline solutions, they were stored in plastic buckets of 5 L capacity, properly closed to avoid evaporation. Irrigation was performed manually on alternate days, being performed slowly until the water drained into the cup, thus reaching the field capacity in all cups.

At 37 days after sowing (DAS) measurements of plant height, stem diameter, number of leaves, and leaf area were performed. The measurement of plant height (PH) was determined using a graduated ruler expressed in centimetres, while the stem diameter was measured with the aid of a digital ruler. To obtain the number of leaves (NF), only the leaves that were photosynthetically active were considered. The leaf area (LA) was obtained through the equation below proposed by Tollenaar (1992):

$$LA = L \times W \times 0,75 \quad (1)$$

Where: LA = leaf area, in cm<sup>2</sup>; L = length of the main vein of the cotton sheet, in cm; W = width perpendicular to the main vein of the cotton sheet, in cm.

The absolute growth rate (AGR) was calculated from the height growth data (cm day<sup>-1</sup>) using the data that were collected and applied in Equation 2:

$$AGR = \frac{PH_2 - PH_1}{T_2 - T_1} \quad (2)$$

Where, AGR - Absolute growth rate in relation to height; PH<sub>2</sub> and PH<sub>1</sub> - Variation of plant growth (in height) between two consecutive samples taken at times T1 and T2 (days); T1 and T2 - Time interval between assessments, in days, without considering pre-existing values, prior to this variation.

To assess the tolerance of cotton plants to salinity, the methodology proposed by Fageria (1985) was applied. The percentage reductions in plant height (PH), absolute growth rate (AGR), stem diameter (SD), number of leaves (NF), and leaf area (LA) of cotton plants submitted to different irrigation levels were quantified with saline water in the two substrates, comparing them to the control (plants cultivated with water whose electrical conductivity is equal to 0.5 dS m<sup>-1</sup>), according to Equation 3, proposed by Fageria (1985):

$$RP (\%) = \frac{(PSTS - PCTS)}{PSTS} \times 100 \quad (3)$$

Where: RP - Reduction of production; PSTS – value obtained in the irrigated treatment with non-saline water; PCTS – value obtained in treatments irrigated with saline water.

The results were interpreted by observing the percentage reduction intervals, with this reduction of 0-20% being classified as tolerant (T); of 20.1 – 40%, moderately tolerant (MT) of 40.1 – 60% moderately sensitive (MS) and > 60 classified as sensitive (S) (Fageria, 1985).

**Table 2:** Percentage reduction in plant height (PH), absolute growth rate (AGR), stem diameter (SD), number of leaves (NF), leaf area (LA), and classification regarding salinity tolerance of plants of cotton plants subjected to different levels of irrigation water salinity in two substrates.

Variables	Soil Layer: 0 - 20 cm			
	0.5	3.0	5.0	10.0
-----dS m <sup>-1</sup> -----				
AP	0.0 <sup>T</sup>	5.57 <sup>T</sup>	7.27 <sup>T</sup>	18.40 <sup>T</sup>
TCA	0.0 <sup>T</sup>	18.20 <sup>T</sup>	28.57 <sup>MT</sup>	44.50 <sup>MS</sup>
DC	0.0 <sup>T</sup>	2.67 <sup>T</sup>	5.35 <sup>T</sup>	10.71 <sup>T</sup>
NF	0.0 <sup>T</sup>	16.66 <sup>T</sup>	20.83 <sup>MT</sup>	25 <sup>MT</sup>
AF	0.0 <sup>T</sup>	11.43 <sup>T</sup>	16.30 <sup>T</sup>	33.46 <sup>MT</sup>
Earthworm Humus				
AP	0.0 <sup>T</sup>	5.99 <sup>T</sup>	13.34 <sup>T</sup>	15.61 <sup>T</sup>
TCA	0.0 <sup>T</sup>	20.73 <sup>MT</sup>	29.06 <sup>MT</sup>	30.08 <sup>MT</sup>
DC	0.0 <sup>T</sup>	7.90 <sup>T</sup>	12.34 <sup>T</sup>	23.45 <sup>MT</sup>
NF	0.0 <sup>T</sup>	8.37 <sup>T</sup>	12.5 <sup>T</sup>	21.87 <sup>MT</sup>
AF	0.0 <sup>T</sup>	22.22 <sup>MT</sup>	27.56 <sup>MT</sup>	50.30 <sup>MS</sup>

<sup>T</sup>: Tolerant; <sup>MT</sup>: Moderately tolerant; <sup>MS</sup>: Moderately sensitive; <sup>S</sup>: sensitive.

Considering data on plant height (PH), the cotton plant proved to be tolerant in relation to all irrigation water salinity levels in both evaluated substrates (Table 2). Daniel et al. (2021) observed that the height of cotton plants did not differ statistically when subjected to different saline concentrations. Likewise, Oliveira et al. (2013), evaluating the tolerance of cotton cultivars (BRS Verde and BRS Topázio) in different salinity conditions, observed that there was no significant reduction in plant height with the increase in irrigation water salinity. A plant's ability to tolerate the effects of salinity depends on its genetic makeup or variations in its physiological processes that allow plants to cope with salinity stress, which include the degree of ion exclusion and tolerance to osmotic stress (Naeem et al. 2020; Ashraf et al., 2008).

For absolute growth rate (AGR) data, cotton plants grown in soil substrate were tolerant when irrigated with water whose salinity levels were up to 3.0 dS m<sup>-1</sup>. At irrigation water salinity levels of 5.0 and 10.0 dS m<sup>-1</sup>, plants were classified as moderately tolerant and moderately sensitive, with percentage reductions of 28.57 and 44.50%, respectively. Plants grown in earthworm humus substrate, apart from the control treatment, were considered moderately tolerant to salinity, with reductions of 20.73; 29.06, and 30.08 % at salinity levels of 3.0, 5.0, and 10.0 dS m<sup>-1</sup>, respectively (Table 2).

Plant growth rates can be limited by different abiotic factors, acting alone or simultaneously (Lacerda et al., 2016; Silva et al., 2016), such as saline stress. In cotton plants,

## Results and discussion

Table 2, below, shows the percentage reductions in plant height, absolute growth rate, stem diameter, number of leaves, leaf area, and classification according to salinity tolerance of cotton plants subjected to different levels of irrigation water salinity in two substrates.

Santos et al. (2016) observed that the absolute growth rate decreased from 2.63 to 1.11 cm day<sup>-1</sup> between plants irrigated with water with lower (0.7 dS m<sup>-1</sup>) and higher saline levels (8.7 dS m<sup>-1</sup>), causing a reduction of 57.63%.

When considering the stem diameter (SD) data, the cotton plants grown in the soil substrate showed to be tolerant in all irrigation water salinity levels. In earthworm humus, this happened up to a salinity of 5.0 dS m<sup>-1</sup>. At the highest salinity level of irrigation water (10.0 dS m<sup>-1</sup>), the plants in earthworm humus were classified as moderately tolerant, with a percentage reduction of 23.45% (Table 2).

In two cotton cultivars (BRS Topázio and BRS Verde), Oliveira et al. (2013) found that the stem diameter was not significantly affected by the salinity of the irrigation water, with average values of 5.26 and 5.45 mm for the cultivar Topázio and Verde, respectively. On the other hand, Capitolo et al. (2016) observed that increasing levels of water salinity inhibited the growth of cotton stem diameter. The authors found a linear decrease, with a reduction of 5.76% per unit increment of the irrigation water electrical conductivity. When the plants were irrigated with water with salinity of 6.0 dS m<sup>-1</sup>, there was a reduction in CO of 28.48% (0.021 mm) in relation to plants cultivated with water with low salinity levels (1.5 dS m<sup>-1</sup>).

For leaf number (LN) data, cotton plants grown in soil layer 0 – 20 cm were classified as tolerant up to a salinity of

3.0 dS m<sup>-1</sup>. In the other irrigation water salinity levels (5.0 and 10.0 dS m<sup>-1</sup>), the plants were classified as moderately tolerant, with percentage reductions of 20.83 and 25%, respectively. For plants grown in earthworm humus, this only happened at the highest salinity level (10.0 dS m<sup>-1</sup>), with a reduction of 21.87% (Table 2). Nascimento et al. (2019), when analysing the number of cotton leaves at 80 and 100 days after sowing (DAS), found percentage reductions of 5.31% and 6.97%, respectively, per unit increment of irrigation water salinity, which corresponds to a reduction of approximately 13 and 25 leaves, respectively, in plants irrigated with water with the highest salinity level (7.5 dS m<sup>-1</sup>) in relation to those irrigated with the lowest level (1.5 dS m<sup>-1</sup>).

The greatest differences for the classification of cotton salinity tolerance among the tested substrates were observed when leaf area (LA) data were used. While in the soil substrate, the cotton plants were classified as tolerant when irrigated with water whose salinity was up to 5.0 dS m<sup>-1</sup>. For plants grown in the earthworm humus substrate, this was only observed for the control treatment (0.5 dS m<sup>-1</sup>). At irrigation water salinity levels of 3.0 and 5.0 dS m<sup>-1</sup>, plants were classified as moderately tolerant, with percentage reductions of 22.22 and 27.56%, respectively. At the highest saline level (10.0 dS m<sup>-1</sup>), the plants were classified as moderately sensitive, with a reduction of 50.30% (Table 2).

Leaves are sensitive organs and reduce in size and number in the presence of high concentrations of salts (Dias et al., 2017). This reduction is a mechanism to avoid water loss through transpiration, which can favour the retention of toxic ions in the roots, limiting their accumulation in the shoots (Acosta-Motos et al., 2017).

Similarly, Lima et al. (2017) found that the growth of cotton (BRS Rubi cultivar) suffered a decline in the evaluated variables with an increase in salinity in the irrigation water, with leaf area being the most sensitive to this increase in saline concentrations. For the same cultivar, Dias et al. (2020) found that cotton plants had the smallest leaf area (604.12 cm<sup>2</sup>) when subjected to irrigation with water whose salinity equalled 5.9 dS m<sup>-1</sup>, corresponding to a reduction of 2745.13 cm<sup>2</sup> (81.96%) compared to those irrigated with water of lower salinity (0.7 dS m<sup>-1</sup>).

## Conclusions

The used type of substrate changed the classification of the tolerance of cotton plants, especially at the highest salinity levels that have been tested (5.0 and 10 dS m<sup>-1</sup>). When considering the data of stem diameter and leaf area, cotton plants grown in soil substrate showed greater tolerance to salinity. However, using absolute growth rate and number of leaves data, cotton plants grown in earthworm humus substrate were more tolerant to salinity. Conducting studies addressing this issue can provide more accurate information on the management of cotton when irrigated with water with high saline concentrations.

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