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Tolerance of peanut (*Arachis hypogea*) genotypes to salt stress in the initial phase

Francisco V. da S. Sá¹, Manoel G. dos Santos¹, Aurélio P. Barros Júnior¹, José R. T. de Albuquerque¹, Almir R. E. de Souza² & Rayanne M. P. Ribeiro¹

¹ Universidade Federal Rural do Semi-Árido/Centro de Ciências Agrárias. Mossoró, RN, Brasil. E-mail: vanies_agronomia@hotmail.com (Corresponding author) - ORCID: 0000-0001-6585-8161; manoel.galdino5@gmail.com - ORCID: 0000-0003-4972-5849; aurelio.barros@ufersa.edu.br - ORCID: 0000-0002-6983-8245; ricardoalbuquerqueagro@gmail.com - ORCID: 0000-0003-4113-1501; rayanne_tab@hotmail.com - ORCID: 0000-0001-8826-6142 ² Instituto Federal de Alagoas. Piranhas, AL, Brasil. E-mail: almir.souza@ifal.edu.br - ORCID: 0000-0002-9266-5063

ABSTRACT: This study aimed to evaluate the emergence, growth, biomass accumulation and tolerance of peanut genotypes under salt stress. The experiment was conducted in a protected environment (greenhouse), evaluating six peanut genotypes (Tatuí, L7151, Caiapó, IAC8112, IAC881 and Havana), which were subjected to two levels of irrigation water salinity (0.5 [control] and 3.5 dS m⁻¹), arranged in a 6 x 2 factorial scheme, in a randomized block design, with five repetitions, with two plants per plot. Plants were cultivated for 30 days after sowing in lysimeters with capacity for 0.5 dm³, filled with a mixture of non-saline, non-sodic soil and commercial substrate in 1:1 proportion on volume basis. During this period, plants were evaluated for emergence, growth, biomass accumulation, tolerance to salinity and dissimilarity. The genotypes Tatuí and L7151 are the most sensitive to salt stress in the emergence stage. Irrigation with high-salinity water reduced the growth and biomass accumulation of the peanut genotypes, and Caiapó and IAC8112 were the least affected. The classification of salinity tolerance had the following sequence: Caiapó > IAC8112 > Havana > Tatuí > IAC881 > L7151.

Key words: irrigation, salinity, genetic improvement

Tolerância de genótipos de amendoim (*Arachis hypogea*) ao estresse salino na fase inicial

RESUMO: Objetivou-se avaliar a emergência, o crescimento, o acúmulo de biomassa e a tolerância de genótipos de amendoim sob estresse salino. O experimento foi desenvolvido em ambiente protegido (casa de vegetação), avaliando seis genótipos de amendoim (Tatuí, L7151, Caiapó, IAC8112, IAC881 e Havana) submetidos a dois níveis de salinidade da água de irrigação (0,5 (controle) e 3,5 dS m⁻¹), arranjados em esquema fatorial 6 x 2, em delineamento experimental de blocos casualizado, com cinco repetições, com duas plantas por parcela. As plantas foram cultivadas durante 30 dias após a semeadura, em lisímetros com capacidade para 0,5 dm³, preenchidos com uma mistura de solo não salino e não sódico, com substrato comercial na proporção de 1:1 base em volume. Durante esse período as plantas foram avaliadas quanto à emergência, crescimento, acúmulo de biomassa e tolerância à salinidade e dissimilaridade. Os genótipos Tatuí e L7151 são os mais sensíveis ao estresse salino na fase de emergência. A irrigação com água de alta salinidade reduziu o crescimento, o acúmulo de biomassa dos genótipos de amendoim, sendo os genótipos Caiapó e IAC8112 os menos afetados. A classificação de tolerância à salinidade teve a seguinte sequência: Caiapó > IAC8112 > Havana > Tatuí > IAC881 > L7151.

Palavras-chave: irrigação, salinidade, melhoramento genético



Introduction

Peanut (*Arachis hypogea* L.) is an oilseed species of socioeconomic and food importance in Brazil and worldwide. Estimates from the United States Department of Agriculture (USDA) highlight China, India and the United States as the world's largest peanut producers, with productions of 17.5, 6.5 and 3.3 billion tons, respectively, in the 2017/18 season. In this survey, Brazil is pointed as the twelfth country in the world ranking, producing an average of 460,000 tons in a planted area of 130 thousand hectares (USDA, 2018).

In Northeast Brazil, the area planted with peanuts is 3,075 thousand hectares and the production is 3,111 thousand tons, with yield of 1,012 kg ha⁻¹, with the states of Sergipe, Bahia, Ceará and Maranhão as the main producers. However, peanut cultivation in the states of Ceará, Paraíba, Pernambuco and Rio Grande do Norte tends to increase, especially in the areas of family farming (IBGE, 2018). In these states, agricultural crops are mainly cultivated under the edaphoclimatic conditions of the semiarid region and are subjected to conditions of quantitative and qualitative water deficit, due to either low water availability or high concentrations of salts dissolved in the water available for irrigation (Medeiros et al., 2003; Rodrigues et al., 2016; Santos et al., 2012; Sousa et al., 2014).

Peanut cultivation in the semiarid region is conditioned by its tolerance to water and salt stresses, but there are few studies evaluating peanut tolerance to salinity (Costa et al., 2014; Santos et al., 2012; Sousa et al., 2012; 2014). Sousa et al. (2014), evaluating the growth and biomass accumulation of the peanut cultivar BRS-1 under saline water irrigation, found that water salinity above 1.5 dS m⁻¹ caused drastic reductions in growth and biomass accumulation. The tolerance of crops to salinity varies between species and between cultivars, genotypes and accessions of the same species (Fageria et al., 2010; Sá et al., 2016a,b). Thus, identifying and using salt-tolerant peanut varieties will promote better performance in areas prone to salinity problems. Thus, this study aimed to evaluate the emergence, growth, biomass accumulation and tolerance of peanut genotypes under salt stress.

MATERIAL AND METHODS

The experiment was carried out in a protected environment (greenhouse) at the Centro de Ciências Agrárias of the Universidade Federal Rural do Semi-Árido, Mossoró, RN,

Brazil (5° 11' S, 37° 20' W, 18 m of altitude). Six peanut genotypes (Tatuí, L7151, Caiapó, IAC8112, IAC881 and Havana) from the collection of UFERSA were subjected to two levels of irrigation water salinity (0.5 [control] and 3.5 dS m⁻¹), arranged in a 6 x 2 factorial scheme, in a randomized block design, with five repetitions and two plants per plot. These levels of salinity were chosen because they commonly occur in the waters used for irrigation in the Brazilian northeast region (Medeiros et al., 2003).

Peanut plants were grown in lysimeters with capacity for 0.5 dm³ of substrate, until 30 days after sowing (DAS). The substrate for seedling production was composed of soil collected from the A horizon of an Ultisol (Lima et al., 2017) and commercial substrate (TOP Plant*) in 1:1 proportion on volume basis, respectively, which were characterized according to the methodology of Donagema et al. (2011) (Table 1). For sowing, four seeds were distributed in each lysimeter, totaling eight seeds per treatment.

For the control treatment, supply water available at the site (EC $_{\rm w}$ = 0.5 dS m $^{-1}$) was used. Irrigation waters of desired electrical conductivity were prepared by adding NaCl salts to the local supply water, considering the equivalent relationship between EC $_{\rm w}$ and the concentration of salts (10 * mmol $_{\rm c}$ L $^{-1}$ = 1 dS m $^{-1}$ of EC $_{\rm w}$) according to Rhoades et al. (1992), valid for EC $_{\rm w}$ from 0.1 to 5.0 dS m $^{-1}$, which encompasses the levels tested. To prepare the solutions with the respective levels of electrical conductivity (EC), the salts were weighed according to each treatment and dissolved in water. The conductivity values were checked using a portable conductivity meter adjusted at a temperature of 25 °C. After preparation, the saline solutions were stored in plastic containers (100 L), one for each EC level and protected from evaporation, entry of rainwater and contamination with materials that could compromise their quality.

The irrigations were performed daily, in order to leave the soil with moisture close to the maximum holding capacity, based on the drainage lysimetry method, applying a leaching fraction (LF) of 0.20 in addition to the water requirement. The volume applied (V_a) per container was obtained by the difference between the previous volume applied ($V_{\rm prev}$) minus the average drainage (D), divided by the number of containers (n), as indicated in Eq. 1:

$$Va = \frac{Vprev - \left(\frac{D}{n}\right)}{(1 - LF)} \tag{1}$$

Table 1. Physical and chemical characteristics of an Ultisol collected in the 0-30 cm layer and chemical characteristics of the commercial substrate used in the experiment

Soil													
Clay		Sand		Silt	BD		PD	Porosity		Textural class			
(g kg ⁻¹)				(kg dm ⁻³)				(%)			lexiulai ciass		
100.0		890.0		10.0 1.		2	2.51		37.45		Loamy Sand		
EC 1:2.5	рH	P	K ⁺	Ca+	² Mg ⁺²	Na+	Al ³⁺	H+ + Al3	+ SB	T	MO	ESP	
(dS m ⁻¹)	H ₂ O	(mg dm ⁻³)		(cmol _c dm ⁻³)						(g kg ⁻³)	(%)		
0.16	6.72	1.2	0 0.20	1.40	0.50	0.05	0.00	0.70	2.15	2.85	13.23	1.75	
Commercial Substrate													
EC (1:2.5		рН	P	SB	T	0.M.	K ⁺	Ca ⁺²	Mg ⁺²	Na+	Al³+	H++AI3+	
(dS m ⁻¹)		H₂O	(mg dm ⁻³)	cmol _c dm ⁻³)		(g kg ⁻³)		(cmol₀ dm					
1.65	į	5.75	86.00	59.61	71.49	570.0	1.67	11.60	28.50	17.84	0.00	11.88	

 P, K^*, Na^* - Mehlich-1 extractor; $Al^{3*}, Ca^{2*}, Mg^{2*}$ - 1.0 mol L^+ KCl extractor; BD - Bulk density; PD - Particle density; PD - PA - PA

During the experiment, the emergence of peanut seedlings was monitored by daily counts, considering as emerged those with cotyledon above soil level. The seedlings counted daily were not discarded, so a cumulative value was obtained. After stabilization of emergence at 20 days after sowing (DAS), emergence percentage (EP) (%) was determined based on the relationship between the number of plants emerged and the number of seeds sown.

Along the experiment, morphological aspects were monitored based on the growth analysis of the plants at 30 DAS, by the determination of plant height (PH) (cm), measured using a graduated ruler (mm) as the distance between the collar and the apex of the plant, stem diameter (SD), measured with a digital caliper one centimeter above soil surface, and number of leaves (NL), determined by counting fully expanded leaves.

After growth analysis, the plants were collected, separated into shoots and roots, dried in an air circulation oven at 65 °C and, after reaching constant weight, weighed on an analytical scale (0.0001 g) to determine shoot dry mass (SDM) (g) and root dry mass (RDM) (g). These data were used to determine the total dry mass (TDM) by summing SDM and RDM.

Total dry mass production data were used to calculate the percentages partitioned among the vegetative organs and the salinity tolerance index. For this, the data of saline treatments were compared to those of the control (EC $_{\rm w}=0.5$ dS m $^{-1}$), according to the methodology of Fageria et al. (2010), based on four levels of classification: T (tolerant; 0-20%), MT (moderately tolerant; 21-40%), MS (moderately sensitive; 41-60%) and S (sensitive; > 60%), as presented in Eq. 2.

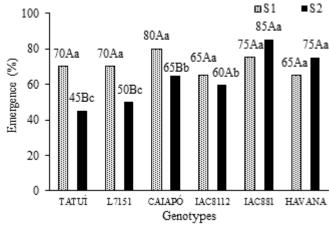
$$STI(\%) = \frac{TDM \text{ production in saline treatment}}{TDM \text{ production in control treatment}} \times 100 \quad (2)$$

These indices were calculated using the total dry mass production of each genotype as the main parameter to determine the tolerance of the materials to salt stress.

The obtained data were subjected to analysis of variance (F test) and, in cases of significance, the Student's t-test was applied for the levels of irrigation water salinity while Scott-Knott means grouping test was applied for the genotypes, both at p \leq 0.05, using the statistical program SISVAR* 5.6 (Ferreira, 2011). The data were standardized with mean zero and variance one. Subsequently, cluster analysis was performed by the hierarchical method, Ward's minimum variance, using the Euclidean distance as a measure of dissimilarity (Hair et al., 2009).

RESULTS AND DISCUSSION

The emergence of peanut plants was significantly influenced (p < 0.05) by the interaction between irrigation water salinity levels and genotypes, and it was verified that the genotypes IAC8112, IAC881 and Havana were not influenced by salt stress in the stage of emergence. However, there were reductions of 35.71, 28.57 and 18.75% in the emergence percentage of the genotypes Tatuí, L7151 and Caiapó, respectively (Figure 1). Such behavior is possibly related to the reduction of seed imbibition capacity as the water salinity level increases, because



Uppercase letters compare salinity levels by Student's t-test, $p \leq 0.05$. Lowercase letters compare genotypes by the Scott-Knott means grouping test, $p \leq 0.05$

Figure 1. Emergence percentage of peanut genotypes subjected to irrigation with saline water (S1 = 0.5 and S2 = 3.0 dS m⁻¹) at 20 days after sowing

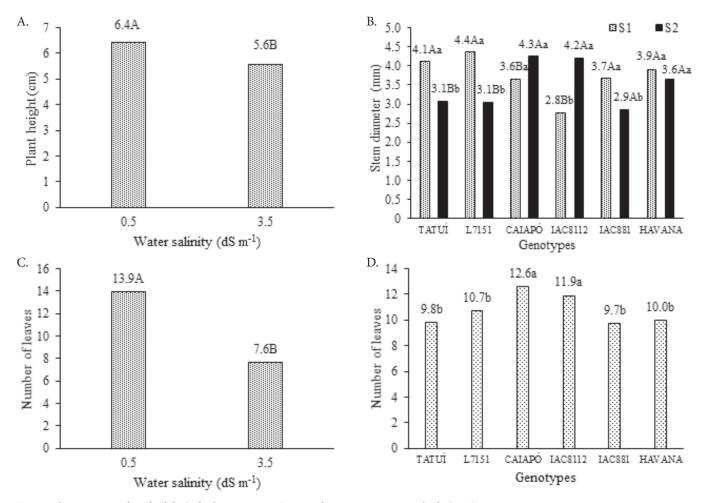
the osmotic potential of the substrate tends to decrease, which slows down or reduces the germination process with negative effects on seedling emergence (Albuquerque et al., 2016).

Irrigation with high-salinity water (3.5 dS m⁻¹) reduced (p \leq 0.05) the growth in height and production of leaves, regardless of the studied peanut genotype, causing reductions of 12.5 and 45%, respectively (Figures 2A and C). Sousa et al. (2012), evaluating the growth of the peanut cultivar PI-165317 irrigated with saline water, observed that the increase in water salinity drastically reduced peanut growth. Difference (p \leq 0.05) was observed between the peanut genotypes for number of leaves, with Caiapó and IAC8112 showing the highest number of leaves, regardless of irrigation water salinity (Figure 2D). This indicates that there is a difference in the growth potential of the peanut genotypes.

For stem diameter, significant interaction (p \leq 0.05) was observed between salinity levels and peanut genotypes, verifying reductions of 24.3, 29.5, 21.6 and 7.7% in the genotypes Tatuí, L7151, IAC881 and Havana, respectively (Figure 2B). It is also observed that the stem diameters of the genotypes Caiapó and IAC8112, under conditions of high salinity (3.5 dS m⁻¹), was 19.4 and 50.0% larger than those of plants cultivated under low salinity (0.5 dS m⁻¹), respectively (Figure 2B).

Ghanem et al. (2011), evaluating tomato plants under salt stress, reported that the increment in salinity increases the synthesis and accumulation of cytokinins in the root of tomatoes. These authors mentioned that the increase in root cytokinin synthesis modified both the hormonal and ionic state of the shoots and improved the reductions of growth and yield induced by salinity. It should be added that cytokinins are phytohormones responsible for regulating cell synthesis and expansion (Taiz et al., 2015). Salt stress probably stimulated the synthesis and/or accumulation of phytoregulators in the peanut genotypes, thus stimulating their growth.

Irrigation with saline water increases the concentration of salts in the substrate, inhibiting plant growth, due to the reduction in the osmotic potential of the soil solution, restricting the availability of water (Esteves & Suzuki, 2008;



Uppercase letters compare salinity levels by Student's t-test, $p \le 0.05$. Lowercase letters compare genotypes by the Scott-Knott means grouping test, $p \le 0.05$. Figure 2. Plant height (A), stem diameter (B) and number of leaves (C and D) of peanut genotypes subjected to irrigation with saline water (S1 = 0.5 and S2 = 3.0 dS m⁻¹) at 30 days after sowing

Sousa et al., 2010, 2014). However, the higher the level of severity of the salt stress, the greater the dependence of plant development on genetic factors such as plant species, cultivar and phenological stage (Correia et al., 2009; Sousa et al., 2014; Sá et al., 2017).

The increase in water salinity from 0.5 to 3.5 dS m⁻¹ reduced the root dry mass accumulation of the peanut plants regardless of the genotype studied, causing a percentage reduction of 56.3% (Figure 3A). For shoot dry mass, there was significant interaction ($p \le 0.05$) between peanut genotypes and salinity levels, and only Caiapó was not influenced by the salinity levels. For the other genotypes, Tatuí, L7151, IAC8112, IAC881 and Havana, there were reductions of 54.5, 66.7, 45.4, 66.7 and 72.7% in shoot dry mass accumulation, when they were irrigated with high-salinity water (3.5 dS m⁻¹), respectively (Figure 3B).

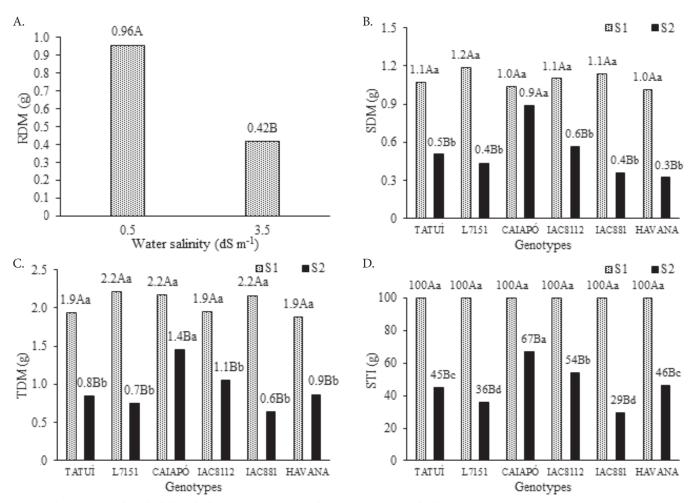
Total dry mass accumulation followed the same tendency of shoot dry mass accumulation. It was observed that the genotype Caiapó was significantly influenced by the salinity levels, and that the dry mass accumulation of the genotypes Tatuí, L7151, IAC8112, IAC881 and Havana were drastically reduced due to irrigation with high-salinity water (3.5 dS m⁻¹) (Figure 3C).

The accumulation of salts in the substrate due to the successive irrigations with saline water causes osmotic and

ionic effects on plants. The osmotic effect hampers and/or restricts water absorption by the root system, whereas the ionic effect caused by the excessive absorption of ions causes toxicity by specific ions, especially those of sodium and chlorine, leading to physiological, nutritional and hormonal interactions that affect the synthesis and accumulation of photoassimilates and, consequently, plant development (Munns & Tester, 2008; Graciano et al., 2011; Syvertsen & Garcia-Sanchez, 2014; Taiz et al., 2015).

Reduction in peanut biomass accumulation has also been observed by Santos et al. (2012) and Costa et al. (2014), evaluating irrigation with saline water in the cultivation of peanut BR-1. Sá et al. (2016a, b; 2017) also verified drastic reductions in the dry mass accumulation of cowpea and castor bean genotypes subjected to salt stress in the initial development stage. The researchers found that ionic and osmotic effects are critical in the initial development stage, exerting major pressure of selection on cultivated plants.

Salinity tolerance index (STI) was affected (p \leq 0.05) by the interaction between genotypes and salinity levels, with the genotypes formed four groups, when subjected to irrigation with high-salinity water (3.5 dS m⁻¹). The first group consisted of the genotype Caiapó with STI of 67%, the second group was formed by IAC8112 with STI of 57%, the third group comprise the genotypes Tatuí and Havana with STI of 45 and 46%,



Uppercase letters compare salinity levels by Student's t-test, $p \le 0.05$. Lowercase letters compare genotypes by the Scott-Knott means grouping test, $p \le 0.05$. Figure 3. Root dry mass - RDM (A), shoot dry mass - SDM (B), total dry mass - TDM (C) and salinity tolerance index - STI (D) of peanut genotypes subjected to irrigation with saline water (S1 = 0.5 and S2 = 3.0 dS m⁻¹) at 30 days after sowing

respectively, and the fourth group contained the genotypes L7151 and IAC881 with STI of 36 and 29%, respectively (Figure 3D).

Thus, based on the classification of Fageria et al. (2010), which contains four levels of classification: T (tolerant; 0-20%), MT (moderately tolerant; 21-40%), MS (moderately sensitive; 41-60%) and S (sensitive; > 60%), it is possible to classify Caiapó as moderately tolerant, IAC8112, Tatuí and Havana as moderately sensitive, and L7151 and IAC881 as sensitive.

In the study of the dissimilarity among the peanut genotypes, based on the Euclidean distance, the formation of three distinct groups was observed between the combinations: salinity levels (S) and genotypes (G) (Figure 4). The first group (I) is characterized by the low salinity level, where all genotypes can express their maximum vigor. The second group (II) can be characterized by genotypes with higher tolerance to salt stress, with high performance in germination and allocation of shoot and root phytomass. The genotypes Caiapó, IAC8112, IAC881 and Havana stand out in this group because, even under high salinity of 3.5 dS m⁻¹, they are present in the group of high performance, denoting their tolerance to salinity. The third group (III) is characterized by high salinity and low performance for germination and dry mass accumulation of shoots and roots, and the genotypes Tatuí and L7151 stood out as more sensitive to salt stress in the initial development stage.

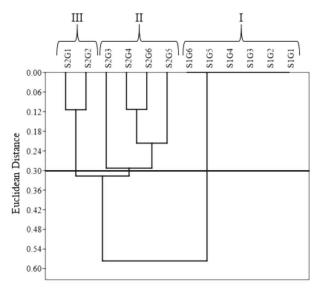


Figure 4. Dissimilarity of peanut genotypes (G1 - Tatuí, G2 - L7151, G3 - Caiapó, G4 - IAC8112, G5 - IAC881 and G6 - Havana) subjected to irrigation with saline water (S1 = 0.5 and S2 = 3.0 dS m^{-1}) at 30 days after sowing

Thus, based on the emergence percentage (Figure 1), salinity tolerance index (Figure 3D) and Euclidean distance (Figure 4), it is possible to characterize the hierarchy of salinity tolerance as Caiapó > IAC8112 > Havana > Tatuí > IAC881

> L7151. Although the genotype IAC881 showed optimum performance in terms of emergence percentage, this material did not obtain good dry mass yield when subjected to salt stress. The genotype L7151 is the most sensitive to salinity, with low performance in the initial development stage.

Conclusions

- 1. The genotypes Tatuí and L7151 are the most sensitive to salt stress in the emergence stage.
- 2. Irrigation with high-salinity water reduced the growth and biomass accumulation of peanut genotypes, and Caiapó and IAC8112 are the least affected.
- 3. The classification of salinity tolerance of genotypes had the following sequence: Caiapó > IAC8112 > Havana > Tatuí > IAC881 > L7151.

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