Original Research Article

PRODUCTION OF TAMARIND SEEDLINGS UNDER IRRIGATION WITH SALT WATER AND NITROGEN DOSES

. **ABSTRACT**

Aims: The objective of this work was to evaluate the effect of nitrogen fertilization on the production of *Tamarindus indica* L. seedlings irrigated with saline water.

Study design: A randomied complete block design was used in a 4 x 4 factorial scheme, whose factors were the electrical conductivity levels of the irrigation water.

Place and Duration of Study: The work was conducted under protected environment conditions (greenhouse) at the Federal University of Campina Grande, Center for Agro-Food Sciences and Technology, Campus of Pombal-PB, Brazil, in the period from January and March 2017.

Methodology: Doses of 0, 100, 200 and 300 mg of N dm⁻³ of nitrogen and electrical conductivity levels of the irrigation - ECw $(0.3, 2.1, 3.7, 3.7)$ and 5.2 dS m^{-1}) on growth of *Tamarindus indica* L.

Results: The growth of the seedlings was reduced by the increase of the electric conductivity of the irrigation water, however, irrigation with ECw water 2.15 dS $\text{m}^{\text{-1}}$, promotes acceptable reductions of 10% in the morphology of the tamarind tree.

Conclusion: The use of nitrogen fertilization with urea did not attenuate the deleterious effects of irrigation water salinity or promoted improvements in the quality of tamarind seedlings.

Keywords: Tamarindus indica L., nitrogen fertilization, saline stress.

1. INTRODUCTION

The tamarind tree (*Tamarindus indica* L.) belongs to the Fabaceae family, the Caesalpinoideae subfamily. It is a species of ample adaptive capacity in the tropical regions of Brazil. Due to the presence of a deep root system and resistance to prolonged droughts, its cultivation is indicated in semi-arid regions [1]. This may be due to its region of origin, Southern Africa, which has similar climatic characteristics to those of semi-arid regions of Brazil [2].

, ice creams, pastes, sweets, liqueurs, jellies and also as an ingredient in condiments and sauces, a fact that makes it one of the main exotic tropical fruits [3]. Suralkar et al. [4] still emphasize the importance of this culture in medicine, with potent anti-inflammatory and analgesic applications, which can boost the expansion of cultivated areas in Brazil.

Another factor that exalts this culture as an alternative to the northeastern region of the country is its moderate tolerance to salinity in the seedling stage, becoming a potentially viable species to evaluate the influence of the management in reducing the negative effects of salinity during plant growth [1]. On this account, seedling formation is one of the crucial stages of the production process and can enable farmers to obtain better performance plants to withstand adverse field conditions [5].

Salinity is considered an adversity in the production of the crops, whose effect compromises the germination viability of the seeds, due to the reduction of the osmotic potential, increase in ionic toxicity and imbalance in the absorption of water and nutrients, thus limiting the productive expression of most crops [6].

In the production process, nitrogen fertilization appears as a technique capable of reducing the deleterious effects of saline stress, since nitrogen acts by increasing the tolerance of the plants through a greater accumulation of nitrogenous organic compounds, such as proline, free amino acids, glycinetatin, which play a significant role in the cellular osmotic balance, besides stabilizing subcellular structures (membranes and proteins) under saline stress [7]. Such behavior has contributed to attenuate the effects of salinity on seedling production, as presented by Souza et al. [8] in guava tree (*Psidium guajava* L.), Bezerra et al. [9] passion fruit tree (*Passiflora edulis* L.) and Pinheiro et al. [10] in acerola cherry tree (*Malphigia emarginata* D.C).

Therefore, the objective of this work was to evaluate the effect of nitrogen fertilization on the production of *Tamarindus indica* L. seedlings irrigated with saline water.

2. MATERIAL AND METHODS

2.1 *Location of the experiment*

The work was conducted between January and March 2017, under protected environment conditions (greenhouse), at the Federal University of Campina Grande, Center of Science and Technology Agri-Food of the Campus of Pombal - PB, whose geographic coordinates are 06° 46' 13"S of latitude and 37° 48' 06" W of longitude and 178 meters of altitude, located in the micro region of the Backwoods of Paraíba [11].

2.2 *Description of treatments*

The experimental design was a randomized complete block (RCBD), in a 4×4 factorial scheme, whose factors consisted of the electrical conductivity levels of the irrigation water - ECw (0.3, 2.1, 3.7 and 5, 2 dS m⁻¹) and nitrogen doses (0, 100, 200 and 300 mg N dm⁻³), with four replicates and two plants per plot.

Salinity waters were obtained from the water supply (ECa of 0.3 dS m^{-1}) by the addition of sodium chloride (NaCl), obeying the relationship between ECa and the concentration of salts (mmolc L^{-1} = CE x 10) [12].

Nitrogen doses were based on the results found by Morales-Payan [13] with the estimated dose of nitrogen of 200 mg dm⁻³ presenting the best results.

2.2 *Production of seedlings*

The experiment was conducted in polyethylene bags with 1150 mL capacity, with lateral perforations to allow free drainage of water. The bags were placed on wooden benches at a height of 0.6 m from the ground to facilitate cultural treatment and application of treatments.

The substrate used was composed of 82% Fluvent soil, 15% fine sand and 3% bovine manure tanned. The physical and chemical characteristics (Table 1) were obtained according to the methodology proposed by Claessen [14] in the Soil and Plant Laboratory of the CCTA / UFCG.

pH – hydrogen potential, Ca2+ and Mg2+ extracted with 1 M L-1 KCl at pH 7.0; Na⁺ and K⁺ extracted using 1 M L-1 NH4OAc at pH 7.0; Al3++H⁺ extracted using 0.5 M L-1 CaOAc pH 7.0; ECse – electrical conductivity of the saturation extract; AD - apparent density; DP –particle density.

For the sowing, two seeds of wild tamarind were used per polyethylene bag, which were previously submitted to the method of overcoming dormancy, where an incision was made on the opposite side to the protrusion of the primary root. During the germination and emergence period of the seedlings, the soil was maintained with moisture near field capacity, with daily irrigation with local water supply (0.3 dS m^{-1}) .

At 15 days after the emergence (DAE) the thinning was performed, leaving the seedling more vigorous, and irrigation was initiated with saline water, which were performed based on the water requirement of the plant, by the process of drainage lysimeter, by providing it on a daily basis, the amount of evapotranspiration so as to lift the soil at the level of field capacity, which was determined by the applied water volume difference and retained in the drain previously distributed in 15 plants of the experiment [15]. Every 15 days, a 10% leaching fraction was applied based on the volume applied in this period, in order to reduce the salinity of the substrate saturation extract.

Nitrogen fertilization was initiated at 20 DAE, divided into 8 applications in equal parts, performed weekly using urea as nitrogen source (45% N), with applications made via fertigation through water with electrical conductivity of 0.3 dS $m⁻¹$ for all treatments.

The crop managements were carried out during the whole period of conduction of the experiment, as manual control of invasive plants and superficial scarification of the substrate, for removal of compacted layers.

2.3 *Analyzed variables*

At 70 DAE, the growth of tamarind seedlings was evaluated by shoot height (SH), with a millimeter ruler and stem diameter (SD), with a digital caliper. In the period from 20 to 70 DAE, the physiological variables related to the absolute and relative growth rate of shoot height (AGRSH and RGRSH) and stem diameter (AGRSD and RGRSH) were evaluated, using the method proposed by Benicasa [16], as described in equations 1 and 2, respectively:

$$
AGR = \frac{SH2-SH1}{\pi z - t1} \tag{1}
$$

wherein:

AGR= absolute growth rate; $SH2$ = plant growth at time t2; $SH1$ = plant growth at time t1; t2 -11 = time difference between samples.

The relative growth rate (RGR) was obtained by equation 2, where growth was measured as a function of the pre-existing matter, which was adapted for height and diameter.

$$
RGR = \frac{ln SH2 - ln SH1}{t2 - t1}
$$

wherein:

RGR = relative growth rate; $SH2$ = plant growth at time t2; $SH1$ = plant growth at time t1; t 2 $-$ t 1 = time difference between samples; ln = natural logarithm.

At 70 DAE, after a destructive evaluation of the experiment, the plants were collected, then washed (roots), fractionated in leaf, stem and root, then conditioned in previously identified paper bags and put to dry in a forced circulation oven, which was maintained at a temperature of 65 ° C until obtaining a constant mass for determination of the dry biomass of the leaf (DBL), stem (DBS) and root (DBR), with these values the dry shoot biomass of the aerial part was determined - DSBA (DBL + DBS) and total dry biomass - TDB (DSBA + DBR) in a precision balance of 0.01 g.

In this study, the quality of the seedlings was obtained through the Dickson quality index (DQI), using the Dickson et al. [17], describe in equation 3.

$$
DQI = \frac{TDB}{\left(\frac{SH}{SD}\right) + \left(\frac{DSBA}{DBR}\right)}
$$

 (3)

wherein:

 $DQI = Dickson$ quality index; $SH =$ shoot height (cm); $SD =$ stem diameter (mm); $TDB =$ total dry biomass (g); DSBA = aerial part dry shoot biomass (g); DBR = root dry biomass (g).

2.3 *Statistical analysis*

The data were evaluated through analysis of variance by the F test (1 and 5% probability) and in the case of a significant effect, for both factors studied, linear and quadratic polynomial regression analysis was performed using the statistical software SISVAR [18].

3. RESULTS AND DISCUSSION

According to the analysis of variance (Table 2), there is an isolated effect of salinity levels and nitrogen rates on shoot height, stem diameter and absolute growth rate of shoot height; for the absolute and relative growth rate of the diameter of the stem, one can only notice the effect of the nitrogen doses. Interactive effect among the factors was observed only in the variable relative growth rate of shoot height.

Table 2. Summary of analysis of variance of plant height (SH) and stem diameter (SD) at 70 DAE, and absolute growth rate (AGR_{SH}) and relative plant height (RGR_{SH}) and absolute growth rate (AGR_{SD}) and relative stem diameter (RGR_{SD}) of tamarind **seedlings irrigated with saline waters and nitrogen doses in the period from 20 to 70 DAE.**

Source of variation	DF	Mean Squares							
		SH	SD	AGR _{SH}	RGR_{SH}	AGR_{SD}	RGR_{SD}		
Saline levels (SL)	3	489	0.24	0.24	0.00014	0.00009^{ns}	$0.00001^{\overline{ns}}$		
Linear regression		1062 [^]	0.17^{ns}	0.50	0.00025	0.000002 ^{ns}	0.000007		
Quadratic regressior		46 ^{ns}	0.53	0.02 ^{ns}	0.00001 ^{ns}	0.000054^{ns}	0.000000^{ns}		
Nitrogen doses (ND)	3	473**	0.77 ^{**}	0.23	0.00013	0.00034	0.00002		
Linear regression		1225 **	1.89 ^{**}	0.59^{\degree}	0.00032	0.000902	0.00007 **		
Quadratic regressior		43 ^{ns}	0.28	0.04 ^{ns}	0.00006	0.000085	0.000006^{ns}		
Interaction (SL*ND)	9	46 ^{ns}	0.07 ^{ns}	0.02 ^{ns}	0.00001	0.00001 ^{ns}	0.00000^{ns}		
Blocks	3	31 ^{ns}	0.11^{ns}	001 ^{ns}	0.00000^{ns}	0.00001 ^{ns}	0.00000^{ns}		
CV(%)		11.82	7.57	15.21	8.30	13.62	10.66		
** $P > 0.01 \cdot * P > 0.05 \cdot {}^{ns}P > 0.05$									

*** P<0.01; * P<0.05; nsP>0.05*

The increase in saline levels caused linear decreases in the height of the tamarind trees (Figure 1A), presenting reductions of 4.42% per unit increase of ECw, leading to a loss of 10.94 cm (22.55%) in the level salinity of 5.4 dS $m⁻¹$ when compared to plants irrigated with water supply (0.3 dS m^{-1}) . Such behavior may be related to the closure of the stomata as a way to reduce transpiration, reducing the photosynthetic rate, and consequently decreasing plant growth under these conditions [19].

*** P<0.01; * P<0.05*

Nitrogen levels also promoted a linear and decreasing behavior in the shoot height variable (Figure 1B), causing a reduction of 8.10% for each increase of 100 mg dm⁻³ in N doses, a fact that in the dose of 300 mg dm⁻³ provided a decrease of 11.73 cm (22.55%). It is denoted to be this reduction due to the release of hydrogen in the soil solution by the nitrification process of the ammonia present in the urea, thus providing a nutritional imbalance [20].

The diameter of the stem was negatively affected by the increased salinity of the irrigation water, where according to regression studies (Figure 2A), there was a decrease of 1.14% per unit increase in the electrical conductivity of the water, reduction of 5.81% when compared to the plants irrigated with ECw 0.3 dS m^{-1} . This fact is possibly promoted by the concentration of salts in the soil solution, which makes it difficult for water to be absorbed by plants due to the greater energy expenditure to maintain its turgidity [21].

Figure 2. Effect of conductivity of irrigation water (A) and nitrogen doses (B) on stem diameter of tamarind seedlings at 70 DAS

*** P<0.01; * P<0.05*

In the treatments with nitrogen fertilization, the increase of N doses affected the stem expansion of the tamarind seedlings (Figure 2B). A linear decreasing effect of 4.22% was observed at each increase of 100 mg N dm^{-3} , depreciation of 12.66% in the highest nitrogen dose tested. The ionic competition between cations and ammonium, formed after the urea hydrolysis process, can provide a accented reduction in plant growth, as it induces potassium, calcium and magnesium deficiency soon after the weekly application of the fertilizer [22].

The absolute growth rate for shoot height from 20 to 70 DAE decreased with increasing salinity of irrigation water from 0.82 to 0.58 cm day⁻¹ (Figure 3A), resulting in a loss of 29, 27% between plants irrigated with water of higher and lower electrical conductivity (5.4 and $0.3 \, \text{m}^{-1}$). A result of the increase in the concentration of the Na and Cl ions in the soil solution, which tend to reduce the amount of nutrients absorbed by the plant due to ionic competition and decrease of solutes input in the plant [23].

Figure 3. Isolated effect of irrigation water conductivity (A) and nitrogen doses (B) on the absolute growth rate of shoot height in the period from 20 to 70 DAE

*** P<0.01; * P<0.05*

Similar reductions were observed with increasing doses of nitrogen in AGRSH at 20 to 70 DAE (Figure 3B), in which a total decrease of 28.9% was observed when compared to plants fertilized with 300 mg dm⁻³ (0.59 cm day⁻¹) and to those that did not receive nitrogen fertilization (0.83 cm day⁻¹). Being possibly explained by the increase of saline stress near the root system, caused by high urea salinity (75%), thus compromising soil solution absorption and cell expansion [24;25].

For the variable relative growth rate of shoot height (Figure 4), there is a decreasing linear behavior with the increase in the electrical conductivity of the irrigation water at all the nitrogen doses tested (100, 200 and 300 mg dm⁻³), with reductions of 3.8; 4.5 and 4.83% per unit increment in ECw, reaching a salt level of 5.4 ds m⁻¹ an inhibition of 19.38; 22.95 and 24.63%, when purchased the plants irrigated with ECw of 0.3 dS $m⁻¹$, respectively. The reductions occurring in this period are reflections of energy expenditure for the synthesis of osmotically active organic compounds that are essential in the compartmentalization of salts and regulation of ion transport [26].

Figure 4. Relative growth rate of shoot height as a function of salinity of irrigation water and nitrogen doses in tamarind seedlings in the period from 20 to 70 DAE

*** P<0.01; * P<0.05*

The absolute (Figure 5A) and relative (Figure 5B) growth rates of stem diameter were affected by increasing nitrogen doses, with linear reductions of 10 and 6.47% at each addition of 100 mg of N dm⁻³, which provided a total reduction of 30% in AGRSD and 19.41% in RGRSD in the highest nitrogen dose tested (300 mg of N dm⁻³) when compared to control plants (0 mg dm^{-3}) . Silva et al. [27] attributes this behavior to the rapid transformation of urea into ammonium in the soil, thereby exponentially increasing the concentration of this element available to the plant, leading to toxicity.

Figure 5. Effect of nitrogen rates on the absolute (A) and relative (B) growth rates of stem diameter of tamarind seedlings in the period from 20 to 70 DAE

*** P<0.01; * P<0.05*

According to the results of the analysis of variance (Table 3), we can observe interaction of the factors saline levels and nitrogen doses for the dry stem, leaf and shoot biomass, in addition to Dickson quality index. However, for the dried root biomass only the isolated effect of the two factors can be observed.

Source of variation	DF	Mean Squares						
		DSB	DLB	DRB	DSB	TDB	DQI	
Saline levels (SL)	3	1,08	$2,02^{\degree}$	$0,35^{n}$	6.98°	9.19	0,0120	
Linear regression		$2,82$ ^{**}	$4,58^{\degree}$	$0,68^\circ$	14,63	21,68	0,0246	
Quadratic regression		0.02 ^{ns}	$0,10^{ns}$	0.01 ^{ns}	$0,029^{ns}$	0.08 ^{ns}	0,0091	
Nitrogen doses (ND)	3.	0,56	0,96	0,07	$2,90^{\degree}$	3,78	0,0057	
Linear regression		1,43	2,86	$0,12^{-}$	8,33	53,14	0,0106	
Quadratic regression		0,14	$0,002^{ns}$	$0,01^{\text{ns}}$	$0,11^{\text{ns}}$	$0,20^{ns}$	0,0059	
Interaction (SL*ND)		0.08	0.10^{-7}	0.02 ^{ns}	0.26	0.37	0,0027	
Blocks	3	0,067 ^{ns}	0.05 ^{ns}	0,01 ^{ns}	$0,12^{ns}$	0,30 ^{ns}	$0,0004^{\text{ns}}$	
CV(%)		13,59	13,94 $\pm 5.331 \pm 5.355$ ± 0.55	21,09	12,30	12,23	13,83	

Table 3. Summary of variance analysis for dry stem biomass (DSB), leaf (DLB), root (DRB), shoot (DSB), total (TDB) and Dickson quality index (DQI) of tamarind seedlings under different levels of irrigation water salinity and nitrogen doses at 70 DAE.

*** P<0.01; * P<0.05; nsP>0.05*

For the dry stem biomass (Figure 6A), the nitrogen doses of 0, 100 and 200 mg dm⁻³ provided a linear and decreasing effect, with unit reductions of 7.04; 8.52 and 7.21%, which led to a depreciation of 0.66 at the level of 5.4 dS m⁻¹; 0.87 and 0.59 g when compared to plants irrigated with water of electrical conductivity of 0.3 dS $m⁻¹$, respectively. However, the dose of 300 mg N dm $³$ showed a quadratic effect, with the point of maximum gain occurring</sup> in the ECw of 2.2 dS m⁻¹, giving a 16.10% increase in DSB in relation to the plants submitted to ECw of 0.3 dS m^{-1} . This consequence of the limitation in height and diameter of the stem caused by the reduction of the cellular expansion, probably due to the accumulation of NaCl in the soil solution, which causes osmotic (physiological dry), toxic (accumulation of ions) and nutritional effects absorption) characteristic of saline stress environments [28].

Figure 6. Dry stem (A) and leaf biomass (B) as a function of salinity of irrigation water and nitrogen doses in tamarind seedlings at 70 DAE *** P<0.01; * P<0.05*

For the dry biomass of the leaf, the doses of 0; 200 and 300 mg of N dm⁻³ presented similar behavior to DSB (Figure 6B), with losses of 0.97; 0.38 and 0.57 g when compared to higher and lower salinity of the irrigation water, that is, reductions 41,22; 23.25 and 35.46% when using the highest salt level, respectively. The dose of 200 mg of N dm-3, even showing the smallest reductions with the increase in salinity, presented values lower than the plants that did not receive the nitrogen fertilization (0 mg of N dm⁻³), being more relevant when irrigated with water of supply of 0.3 dS $m⁻¹$ with a reduction of 29.56% in the DLB. Rebouças et al. [29] attributed this behavior to the water deficiency conditions induced by the osmotic (physiological dry) effect that leads to morphological and anatomical changes in the plants, among them, the reduction of leaf size and number.

In relation to the effects of salinity of irrigation water on root dry biomass (Figure 7A), the regression equation shows a linear and decreasing effect, with a decrease of 6.02% per unit increase in ECw, equivalent to a reduction of 30.7% (0.28 g) in the plants submitted to irrigation water salinity of 5.4 dS m^{-1} , when compared to the irrigated plants with the lowest saline level (0.3 dS m^{-1}) . This result can be a consequence of the high sodium content in relation to the other cations, being adsorbed by the exchange complex and, in this case, dispersing the clay particles, causing the soil to lose its structure, becoming impermeable, fact that hinders root growth [30;31].

Figure 7. Effect of conductivity of irrigation water (A) and nitrogen doses (B) on the dry biomass of the root of tamarind seedlings at 70 DAS *** P<0.01; * P<0.05*

The increasing doses of nitrogen, according to the regression equation (Figure 7B), affected the root dry biomass of the tamarind seedlings, with a linear and decreasing reduction of 3.8% in each increase of 100 mg of N dm⁻³, which led to a depreciation of 11.4% in the N dm⁻³ 300 mg dose in relation to plants without nitrogen fertilization. It is denoted that this a consequence of the reduction of potassium uptake by competition with ammonium, which provides a reduction in nitrate reductase enzyme activation, reducing the nitrate uptake even though it is present in the rhizosphere, thus stagnating root growth [32].

According to the regression equations presented in Figure 8A, dry shoot biomass showed a linear decreasing response with increasing salinity of irrigation water in the plants without (0 mg of N dm⁻³) and with (100; 200 and 300 mg of N dm⁻³) nitrogen fertilization. This situation caused in each unit increase of the EC a depreciation of 7.66; 8.47; 6.04 and 4.82%, resulting in irrigation water salinity of 5.4 dS m^{-1} decrease of 1.64; 1.80; 0.99 and 0.69 g when compared to the irrigated plants with 0.3 dS m^{-1} at doses of 0, 100, 200 and 300 mg of N dm⁻³, respectively. For Bosco et al. [33], the stomatal closure and consequent reduction of the normal $CO₂$ flow towards the carboxylation site is one of the main factors responsible for the reduction of shoot biomass accumulation due to the limitation of photosynthesis.

Figure 8. Dry shoot (A) and total (B) biomass as a function of salinity of irrigation water and nitrogen doses in tamarind seedlings at 70 DAE *** P<0.01; * P<0.05*

The total dry biomass was affected by the interaction between irrigation water salinity and nitrogen doses, and according to regression equation (Figure 8B), the nitrogen doses of 0, 100, 200 and 300 mg dm⁻³ were affected linear and decreasing effect of 7.16; 8.15; 5.79 and 5.64% in each unit increase of the electrical conductivity of the irrigation water, respectively. In this way reductions of 1.90 are observed; 2.17; 1.18 and 1.05 g in the plants irrigated with ECw of 5.4 dS m⁻¹ when compared to that of 0.3 dS m⁻¹. Maia et al. [34] attributed the inhibition of growth in plants under salt stress to the damages occurred in the cell membranes, due to the displacement of the structural Ca^{+2} of the cell surface by the excess of external Na+, leading to the destabilization of the plasma membrane and consequent leakage of cytoplasmic components. The lack of response of the nitrogen doses may be a reflection of the availability of the element through the substrate.

Nitrogen doses, with increased salt levels, showed a linear and decreasing behavior in the Dickson quality index variable (Figure 9), with a decrease of 6.82; 5.92; 2.58 and 2.14% in each unit increase of the irrigation ECw, corresponding to doses of 0, 100, 200 and 300 mg of N dm⁻³. Taking into account the minimum value of DQI of 0.2 for the seedlings to present satisfactory development in the field [35], we can establish the maximum electrical conductivity of 4.07; 5.04; 5.96 and 1.22 dS m^{-1} at the respective nitrogen doses - 0, 100, 200 and 300 mg dm 3 . This fact shows that even when the Ureia toxin was reduced, nitrogen-fertilized plants with the exception of the 300 mg dose of N dm⁻³ showed a higher balance in plant growth at high salinity levels when compared to plants without nitrogen fertilization.

doses in tamarind seedlings at 70 DAE *** P<0.01; * P<0.05*

4. CONCLUSION

The growth and quality of tamarind seedlings is reduced by the increase in the electrical conductivity of the irrigation water, however, irrigation with ECw water of 2.15 dS m^{-1} , promotes acceptable reductions of 10% in tamarind seed morphology.

The use of nitrogen fertilization with urea did not attenuate the deleterious effects of irrigation water salinity nor did they promote improvements in quality of tamarind seedlings.

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