Journal of Agricultural Science; Vol. 9, No. 11; 2017
ISSN 1916-9752 E-ISSN 1916-9760
Published by Canadian Center of Science and Education

Biomass and Rootstock Quality of Guava (*Psidium guajava* L.) Saline Water Irrigated under Nitrogen Fertilization

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Received: August 25, 2017 Accepted: September 21, 2017 Online Published: October 15, 2017

doi:10.5539/jas.v9n11p162 URL: https://doi.org/10.5539/jas.v9n11p162

Abstract

The increase in N concentration in the root zone of plants under saline conditions can inhibit the absorption of chloride and reduce the osmotic, toxic and nutritional effects caused by this ion. Thus, this study aimed to evaluate biomass and rootstock quality of guava. cv. Paluma irrigated with saline water under four N rates. The experiment was carried out in a greenhouse at the Center of Science and Agri-food Technology of the Federal University of Campina Grande (UFCG) at Pombal-PB, Brazil. The experimental design was randomized block with a 5 × 4 factorial arrangement. The treatments were levels of electrical conductivity of water - ECw (0.3. 1.1. 1.9. 2.7 and 3.5 dS m^{-1}), with 70, 100, 130 and 160% of the N rate recommended for guava seedlings (541, 773, 1004.9 and 1236.8 mg N dm^{-3} of soil) and four replicates totaling 80 plots, each one with five plants. Salt stress caused by electrical conductivity of irrigation water of 1.4 dS m^{-1} onwards affected negatively the formation of phytomass of guava cv. Paluma rootstock, and this effect was mitigated on root dry matter and Dickson quality index by the increase in nitrogen rate up to 819.38 mg of N dm^{-3} of soil.

Keywords: *Psidium guajava* L., saline stress, seedling production

1. Introduction

The common guava (*Psidium guajava* L.) is a species over all the subtropical and tropical regions worldwide, due to its easy adaptation to different edaphoclimatic conditions. In the Brazilian Northeast region, it is among the fruit crops of high economic value cultivated under irrigation (Gurgel et al., 2007), since its fruit has great acceptance in domestic and foreign markets, because of its pleasant taste, strong aroma and protein-mineral quality (Cavalcante et al., 2005).

In the semiarid region of the Northeast, a large portion of the water used for irrigation comes from small and medium-sized reservoirs (superficial water) and wells (ground water), which have high salt content with an ECw between 1.97 and 2.98 dS m^{-1} (Medeiros et al., 2003). The use of this water for irrigating crops, including guava, may compromise formation of seedlings and productive capacity (Cavalcante et al., 2007).

The excess of salts in the irrigation water may increase soil pH, electrical conductivity in the saturation extract of soil and compromise Na/Ca, Na/Mg and Na/Ca+Mg ratios in plants, causing nutritional imbalance, toxic and osmotic effects (Pereira et al., 2006). According to Nobre et al. (2010), Na^+ and Cl^- are the most frequently accumulated ions in salinized soils and the main ones to harm plant metabolism. Additionally, under saline conditions high Cl^- concentration may interfere with the absorption of NO_3^- through ionic competition, causing N deficiency (White & Broadley, 2001). In studies using saline water in irrigation, salinity negatively affected the production of guava seedlings of cultivars ‘Paluma’, ‘Ogawa’, ‘Rica’, ‘Pentecoste’, ‘Surubim’ and ‘IPA-B38’, reducing growth and phytomass accumulation in plant parts (Ferreira et al., 2001; Cavalcante et al., 2005; Gurgel et al., 2007; Cavalcante et al., 2010).

For the expansion of guava plantations in regions with low quality irrigation water, such as saline water, besides evaluating genotypes tolerant to salinity (Cavalcante et al., 2005; Gurgel et al., 2007), technologies for reducing effects of salts on plants during seedling formation should be evaluated. In this regard, Del Amor et al. (2000) report evidence of competition in absorption of nitrogen (nitrate) and chloride, so that an increase in N concentration in the root zone under saline conditions can inhibit absorption of chloride and minimize osmotic toxic effects caused by chloride.

Therefore, N fertilization is an alternative because it enhances plant growth and reduces the effect of salinity on plants (Flores et al., 2001). The explanation may be related to the structural function of N, because it participates in organic compounds for plants, such as amino acids, proteins, proline, which increases the capacity of plants for osmotic adjustment, increasing the resistance to the water stress caused by salinity (Parida & Das, 2005).

Guava is highly demanding in N, and this nutrient is the second most required during the initial growth stage. Nitrogen accumulations were 552 and 585 mg plant^{-1} 120 days after transplantation in cultivars 'Paluma' and 'Século XXI', respectively (Franco et al., 2007). This result was confirmed by Dias et al. (2012) who observed positive effects of 773 mg N dm^{-3} of substrate on growth, phytomass production and quality of guava seedlings cv. 'Paluma' 120 days after transplantation.

There are studies about the importance of N fertilization on the initial growth of guava crop, but only a few of them evaluated the interaction between salinity and N fertilization, especially with the cv. 'Paluma', one of the most used cultivars in the Brazilian northeast region. Thus, this study aimed to evaluate the effect of saline water on phytomass accumulation and rootstock quality of guava cv. 'Paluma', under different nitrogen rates.

2. Materials and Methods

2.1 Experiment Localization and Treatments

The experiment was carried out in a greenhouse from March to October 2014, at the Center of Sciences and Agri-food Technology (CCTA) of the Federal University of Campina Grande (UFCG), in Pombal-PB, Brazil (6º47′20″ S; 37º48′01″ W; 184 m).

A randomized block design was used with a 5 × 4 factorial arrangement of treatments: 1) salinity of irrigation water, with five levels of electrical conductivity (ECw): 0.3, 1.1, 1.9, 2.7 and 3.5 dS m^{-1}; 2) four N rates: 70, 100, 130 and 160% of the N rates recommended for production of guava seedlings (541, 773, 1004.9 and 1236.8 mg N dm^{-3}), in four blocks (80 plots total), each one with five plants.

Saline waters were prepared using supply water, from the Piancó River, Pombal-PB. with ECw of 0.3 dS m^{-1}, mixed with requisite amounts of salts of NaCl, $CaCl_2 \cdot 2H_2O$ and $MgCl_2.6H_2O$ in equivalent proportion of 7:2:1, which is the predominant ratio in the main water sources available for irrigation in the Brazilian northeast region. The calculations were based on the following relation between ECw and salt concentration: mmol$_c$ L^{-1} = EC × 10 (Rhoades et al., 1992). Nitrogen rates were determined based on the mean rate of 773 mg dm^{-3}, recommended by Dias et al. (2012) for seedling production of guava. cv. 'Paluma' propagated by herbaceous cutting method, which corresponded to a 100% N rate.

2.2 Plant Material and Management of the Experiment

The guava cultivar 'Paluma' was sown on March 18, 2014, at a depth of 1.0 cm, using two seeds per polytube (capacity = 288 cm^3), supported on metal benches, at a height of 0.8 m from the ground.

The polytubes were filled with a substrate composed of Fulvic Neosol + sand + weathered cattle manure, in 82%, 15% and 3% (volume basis) proportion, respectively. The substrate was analyzed for physical and chemical characteristics (Table 1) in the laboratory, according to the methodologies of Claessen (1997).

For phosphate fertilization, the rate of 100 mg of P dm^{-3} was applied in the form of single superphosphate, which was ground and mixed with the substrate at planting (Corrêa et al., 2003). For potassium fertilization (potassium chloride), the rate of 726 mg of K dm^{-3} of substrate, recommended by Franco et al. (2007), was divided into four equal applications at 60, 90, 120 and 150 days after emergence (DAE), via fertigation with water of EC of 0.3 dS m^{-1} for all treatments.

Thinning was performed when plants showed two pairs of fully expanded true leaves (15 DAE), leaving only the most vigorous seedling per tube.

Saline water application started at 25 DAE. Irrigations were applied according to the treatments, based on plant water demand, corresponding to the difference between the applied volume and the volume drained in the previous irrigation. A leaching fraction of 0.15 was applied in intervals of fifteen days, based on the volume applied during this period.

Table 1. Physical and chemical characteristics of the substrate used in the experiment

Textural Class	Apparent (Bulk) density	Total porosity	Organic matter	P	Exchangeable cation			
					Ca^{2+}	Mg^{2+}	Na^+	K^+
	----------- g cm^{-3} ----------	-------- % -------	----- g kg^{-1} -----	-- mg dm^{-3} --	------------ $cmol_c$ dm^{-3} -----------			
Sandy loam	1.38	47.00	32	17	5.4	4.1	2.21	0.28

Saturation Extract										
pH_{PS}	ECse	Ca^{2+}	Mg^{2+}	K^+	Na^+	Cl^-	SO_4^{2-}	CO_3^{2-}	HCO_3^-	Saturation
	-- dS m^{-1} --	--- $mmol_c$ dm^{-3} ---								--------- % ----------
7.41	1.20	2.50	3.75	4.74	3.02	7.50	3.10	0	5.63	27.00

Note. Ca^{2+} and Mg^{2+} extracted with KCl 1 M at pH 7.0; Na^+ and K^+ extracted with NH_4OAc 1 M at pH 7.0; Organic matter: determined by wet digestion Walkley-Black method; pH_{PS}: pH of saturated paste of the substrate; ECse: Electrical conductivity of the saturation extract of the substrate at 25 °C.

Fertilization began at 25 DAE and was divided into 14 equal applications, performed every 10 days, using urea (45% of N) as the N source, and application was similar to that used for K fertilization.

2.3 Variables Measured

Fresh and dry phytomass accumulation and rootstock quality of guava were evaluated at 190 DAE. Variables were: stem fresh matter (StFM), leaf fresh matter (LFM), shoot fresh matter (ShFM), as well as stem dry matter (StDM), leaf dry matter (LDM), shoot dry matter (ShDM), root dry matter (RDM) and total dry matter (TDM).

The stalk of each plant was cut close to the soil, separated into stem and leaves and immediately weighed on a precision scale (0.001 g), for evaluating StFM and LFM; the sum of these two variables accounted for the ShFM. For DM evaluation, roots were removed from the substrate with a 3-mm-mesh sieve, and leaves, stems and roots were separately placed in identified paper bags and dried in a forced-air oven at 65 ºC until constant mass, for determination of LDM, StDM and RDM; ShDM was determined as the sum of StDM and LDM, and TDM as the sum of ShDM and RDM.

Rootstock quality was determined through the Dickson quality index, according to Equation 1 (Dickson et al. 1960).

$$DQI = [TDM/(PH/SD) + (ShDM/RDM)] \tag{1}$$

Where, DQI = Dickson quality index; TDM = total dry matter of the plant (g); PH = plant height (cm); SD = stem diameter (mm); ShDM = shoot dry matter (g) and RDM = root dry matter (g).

2.4 Statistical Analysis

The data were evaluated through analysis of variance by F test ($p \le 0.05$). If significant, linear and quadratic polynomial regressions were performed using SISVAR/UFLA. The selection of the regression was based on the best fit, considering R^2 and a plausible biological explanation.

3. Results and Discussion

Irrigation water salinity and N rates had a significant effect ($p \le 0.05$) on StFM, StDM, LFM, LDM, ShFM, ShDM and TDM (Table 2). In addition, according to the ANOVA, there was significant interactive effect ($p \le 0.05$) between the factors irrigation water salinity and N fertilization on RDM and IQD.

There was quadratic response of StFM and StDM with the increase in salinity of the irrigation water at 190 DAE and the highest values (2.98 and 1.25 g), were obtained at the ECw levels of 1.4 dS m^{-1} and 1.3 dS m^{-1}, respectively (Figure 1A). This behavior may be related to the adaptation of the rootstock to the saline stress because, according to Flowers (2004), plants adapt or acquire tolerance to salinity when they are able to adjust the osmotic potential of the cells with that in the soil, or even maintaining high contents of K, Ca and NO_3 and low contents of Na and Cl inside the tissues, especially in the leaves (Dias & Blanco, 2010). Cavalcante et al. (2005) and Cavalcante et al. (2007) also observed adaptation of guava seedlings, cv. 'Surubim', to irrigation water salinity at 180 DAE, with greater accumulation of StDM in plants irrigated with water of 1.5 dS m^{-1}.

Table 2. Summary of the analysis of variance for stem fresh matter (StFM), stem dry matter (StDM), leaf fresh matter (LFM), leaf dry matter (LDM), shoot fresh matter (ShFM), shoot dry matter (ShDM), root dry matter (RDM), total dry matter (TDM) and Dickson quality index (IQD) of rootstocks of guava. cv. 'Paluma', irrigated with saline water under different nitrogen fertilization rates at 190 DAE

SV	FD	Mean Square								
		StFM	StDM	LFM	LDM	ShFM	ShDM	RDM	TDM	DQI
Salinity (S)	4	2.23*	0.44*	7.33*	0.79*	20.89*	2.34*	0.13*	3.49*	0.017*
Nitrogen rates (NR)	3	9.12*	2.02*	30.50*	3.17*	66.97*	10.20*	0.18**	13.01*	0.032*
Interaction (S*NR)	12	0.24^{ns}	0.05^{ns}	0.88^{ns}	0.10^{ns}	1.70^{ns}	0.26^{ns}	0.040*	0.46^{ns}	0.004*
Blocks	3	0.38^{ns}	0.06^{ns}	0.51^{ns}	0.05^{ns}	2.05^{ns}	0.09^{ns}	0.039^{ns}	0.16^{ns}	0.0009^{ns}
Residue	57	0.22	0.04	1.19	0.10	1.77	0.23	0.02	0.29	0.0014
CV (%)		17.65	18.07	25.21	23.31	19.22	19.49	20.61	17.17	17.58

Note. ns, non-significant; *, ** significant at $p \leq 0.05$ and $p \leq 0.01$; SV = Sources of variation; FD = Freedom degree; CV = coefficient of variation.

StFM and StDM were negatively affected by the increase in N rates at 190 DAE, and the data fitted to a linear regression equation (Figure 1B) with reductions of 11.01 and 11.82%, respectively, for each 30% increase in recommended N rate. It is believed that, with the increment in N fertilization rates, the seedlings may have absorbed larger amount of N in the ammoniacal form promoting a toxic effect of ammonium on the plant, reflecting in the reduction of StFM and StDM accumulation, as observed by Souza et al. (2016) in Crioula guava rootstock.

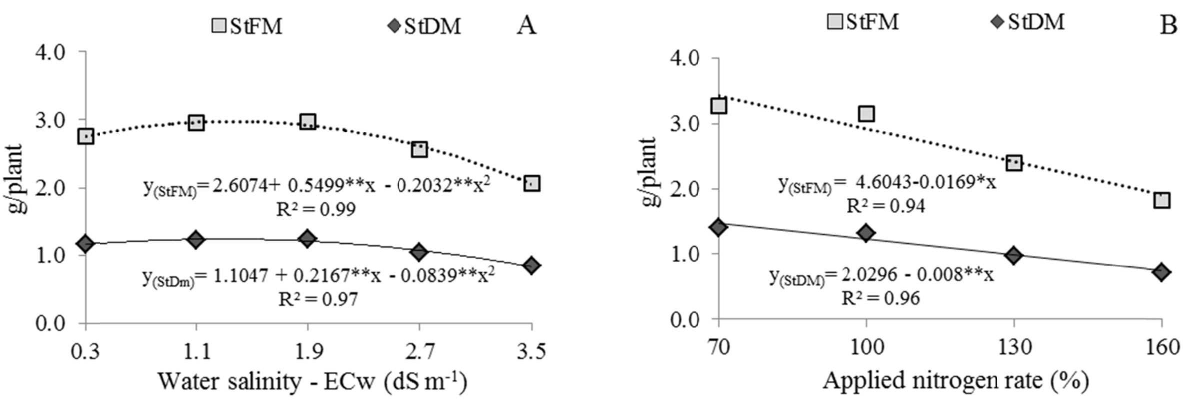

Figure 1. Stem fresh matter (StFM) and stem dry matter (StDM) of guava rootstocks as a function of irrigation water salinity (A) and the percentage of recommended nitrogen rate (B) at 190 DAE

Note. ** and * = significant at 0.01 and 0.05 probability levels ($p \leq 0.01$ and $p \leq 0.05$).

Regarding LFM and LDM (Figure 2A), there was a quadratic response to the increment in salinity of irrigation water and their respective maximum values of 4.94 and 1.53 g were obtained at ECw levels of 1.6 and 1.4 dS m^{-1}. Under these conditions, plants may have acquired adaptation to salinity in order to avoid greater water loss through transpiration creating mechanisms such as accumulation of wax on leaf surface and increase in the number of palisade and spongy cells in the leaf mesophyll (Parida & Das, 2005), thus incrementing leaf phytomass. Cavalcante et al. (2005) and Cavalcante et al. (2007) also studied the adaptation of seedlings of 'Surubim' guava to irrigation water salinity in the period of 180 DAE and observed the highest value of leaf dry matter at the ECw level of 1.5 dS m^{-1}.

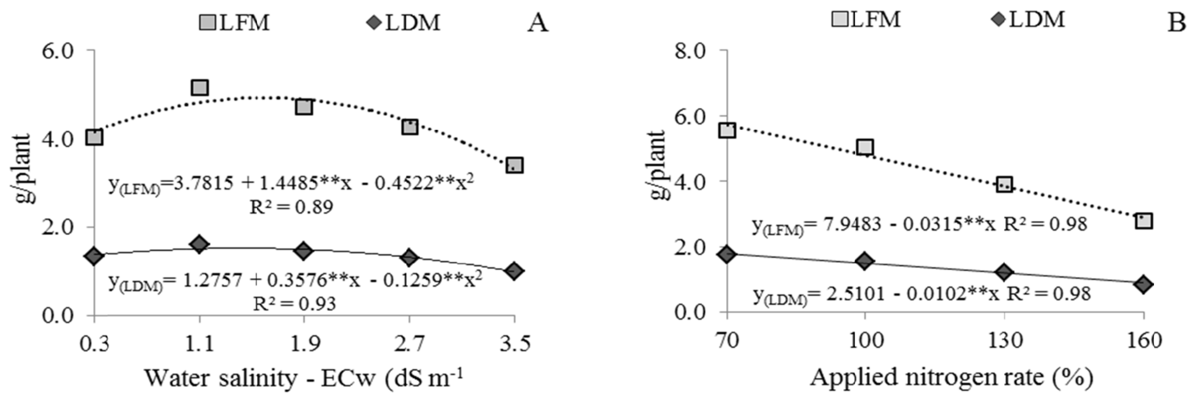

Figure 2. Leaf fresh matter (LFM) and leaf dry matter (LDM) of guava seedlings as a function of irrigation water salinity (A) and the percentage of recommended nitrogen rate (B) at 190 DAE

Note. ** = significant at 0.01 probability levels ($p \leq 0.01$).

LFM and LDM reached the highest values at the N rate of 70% (541.1 mg of N dm^{-3}) and with the increment in N fertilization there were linear reductions (Figure 2B) in these variables equivalent to 11.89 and 12.19%, respectively, for each 30% increment in N. Souza et al. (2016) also observed that increasing N rates above 541.1 mg of N dm^{-3} promoted decreases on LFM and LDM in guava seedlings of the Crioula genotype. This result can be explained by the same causes that affected StFM and StDM when subjected to the increase in N rates.

The data of ShFM and ShDM best fitted to a quadratic equation (Figure 3A) and the highest values of 7.96 and 2.77 g, corresponding to the respective phytomass values, were obtained in plants irrigated with ECw of 1.5 and 1.4 dS m^{-1}. It is inferred that at 190 DAE 'Paluma' guava plants are tolerant to salinity, thus reaching higher values of shoot phytomass until the salinity level of 1.5 dS m^{-1}.

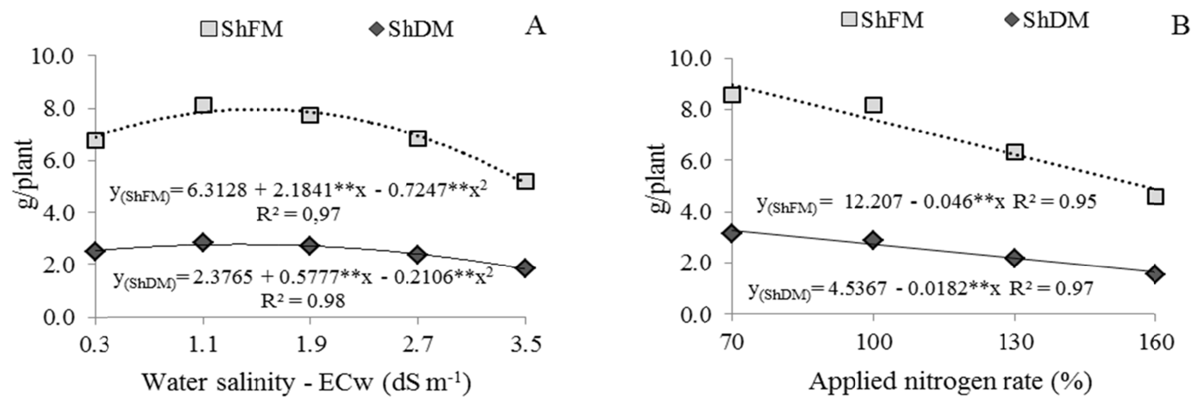

Figure 3. Shoot fresh matter (ShFM) and shoot dry matter (ShDM) of guava seedlings as a function of irrigation water salinity (A) and the percentage of recommended nitrogen rate (B) at 190 DAE

Note. ** = significant at 0.01 probability levels ($p \leq 0.01$).

Data on ShFM and ShDM in plants fertilized with increasing N rates (Figure 3B) best fitted to linear regression equations and there were reductions of 11.30 and 12.03% in ShFM and ShDM for each 30% increment in N rate. In this context, it is emphasized that increasing nitrogen rates from 70% N (541.1 mg N dm^{-3}) does not mitigate the effect of salinity on the shoot, but contributes to the increase of saline stress, justified by salinization of the substrate due to the increasing rates of urea that has relatively high saline index (index of 75%) and the toxic effect of the ammonium ion (NH_4^+), excessively absorbed by the plants with increased nitrogen fertilization (Souza et al., 2016; Silva et al., 2017).

TDM data best fitted to a quadratic regression equation as irrigation water salinity increased (Figure 4A), with maximum value of 3.54 g obtained in plants irrigated with water of 1.3 dS m^{-1}. The explanation is based on the adaptation of plants to salinity in the period of 190 DAE, as observed for fresh and dry matter of leaves, stems and shoots (Figures 1A, 2A and 3A). These results differ from those reported by Ferreira et al. (2001), Gurgel et al. (2007), Cavalcante et al. (2010) and Souza et al. (2016), who observed reductions in TDM accumulation with the increment in salinity of irrigation water in guava seedlings from the cultivars 'Rica', 'Ogawa' and 'Paluma' between 50 and 80 DAE. In this case, it is suggested that the tolerance or adaptation of guava rootstocks to salinity with respect to TDM is higher in the period of 190 DAE, as reported by Tester and Davenport (2003), who claim that plant adaptation to salinity may vary among genotypes of the same species and with the crop phenological stage.

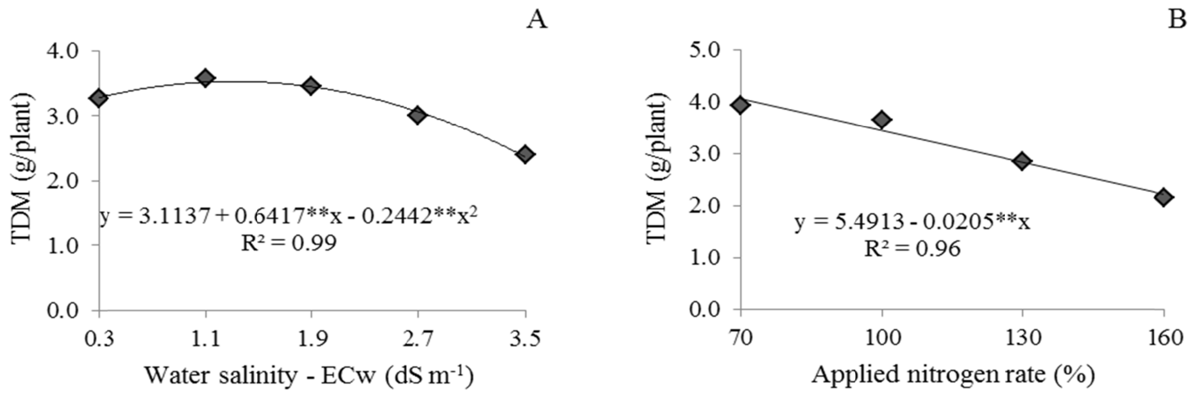

Figure 4. Total dry matter (TDM) of guava seedlings as a function of irrigation water salinity (A) and the percentage of recommended nitrogen rate (B) at 190 DAE

Note. ** = significant at 0.01 probability levels ($p \leq 0.01$).

According to Figure 4B, the highest accumulation of TDM in guava rootstock. cv. 'Paluma', was obtained at the lower N rate of 70% (541.1 mg of N dm^{-3}), and the increase in N fertilization linearly inhibited the TDM, causing a decrease of 13.66% for each 30% increase in N rate. Similarly, Souza et al. (2016) observed in Crioula guava rootstock irrigated with saline waters, that N rates 541.1 mg of N dm^{-3} onwards reduced accumulation of TDM, due to an increase in saline stress because of increase in the rate of urea (75% salinity index).

According to the follow-up analysis for the salinity levels at each N rate (Figure 5A), the increase in water salinity did not cause significant difference in RDM of plants fertilized with the N rate of 70% (541.1mg of N dm^{-3}), obtaining average weight of 0.76 g/plant; however, there was significant influence on RDM under the other rates. Data best fitted to a quadratic regression equation for the N rates of 100 (773) and 130% (1004.9 mg of N dm^{-3}), and the highest values, corresponding to 0.86 and 0.76 g/plant, were obtained at the ECw levels of 1.6 and 1.3 dS m^{-1}, respectively (Figure 5A). Differently, Cavalcante et al. (2010) observed that water salinity linearly reduced RDM in 'Paluma' guava seedlings. However, it is observed (Figure 5A) that this effect was mitigated in the plants that received N fertilization at the rate of 100% N (773 mg of N dm^{-3}), providing a greater accumulation of RDM up to electrical conductivity of 1.6 dS m^{-1}. Blanco et al. (2008) affirm that the adequate supply of nitrogen under saline conditions can reduce the Cl/N ratio in the plant as a result of the higher absorption of NO_3^- in relation to Cl^-, reestablishing the nutritional balance, which can mitigate the osmotic effects, toxic and nutritional caused by saline stress. In addition, high values of NO_3^- in relation to Cl^- under salinity, can increase the synthesis of photoassimilates in the plants, and it can be accumulated as a reserve in the root, resulting in higher RDM.

The follow-up analysis for N rates at each level of irrigation water salinity (Figure 5B) shows that there was no significant difference in RDM accumulation with the increment in N rates in plants under ECw of 0.3 and 3.5 dS m^{-1}, showing averages of 0.76 and 0.55 g/plant. The N rate of 70% (541.1 mg of N dm^{-3}) (Figure 5B) promoted the highest RDM values (0.86 and 0.93 g/plant) in plants under salinity levels of 1.1 and 1.9 dS m^{-1}, becoming close to the value of 552 mg of N/plant accumulated in 'Paluma' guava seedlings under hydroponic cultivation (Franco et al., 2007). In plants irrigated with water salinity of 2.7 dS m^{-1}, the data best fitted to a quadratic regression equation and RDM reached its highest value (0.87 g/plant) at the N rate of 106% (819.38 mg of N dm^{-3}). In general, it is verified that in the plants irrigated with ECw of 1.1, 1.9 and 2.7 dS m^{-1}, N fertilization mitigated the effect of saline stress on RDM, thus, making it essential to determine optimal rates for fertilization of salinity sensitive crops, which may promote an increase in plant tolerance to this type of stress (Kaya et al., 2007).

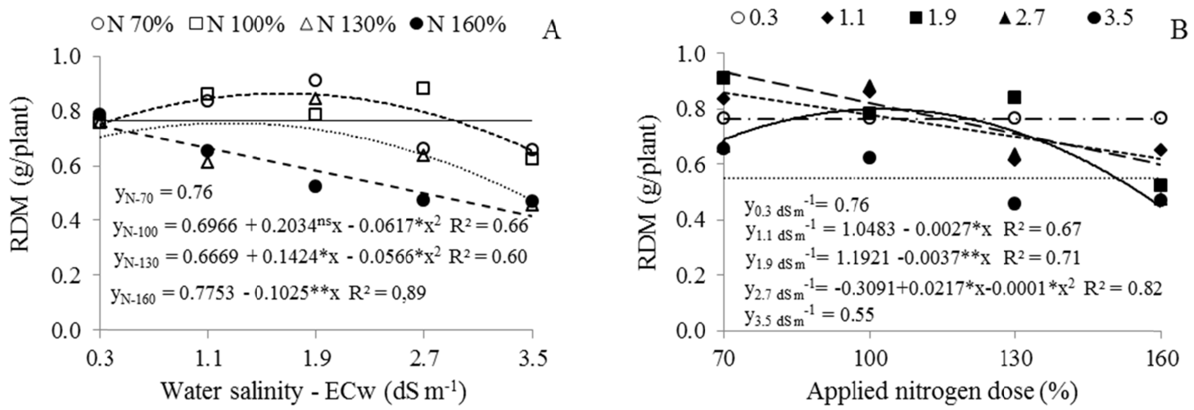

Figure 5. Follow-up analysis of the factor irrigation water salinity at each percentage of recommended nitrogen rate (A) and the factor percentage of recommended nitrogen rate at each salinity level (B) for root dry matter (RDM) of guava seedlings at 190 DAE

Note. ns,** and * = non-significant, significant at 0.01 and 0.05 probability levels ($p \leq 0.01$ and $p \leq 0.05$).

According to the follow-up analysis of salinity levels at each N rate (Figure 6A) and based on regression equations, the DQI of guava rootstocks showed a quadratic response with the increment in irrigation water salinity for the N rates of 70, 100 and 130%, with the highest values of 0.28. 0.27 and 0.23 obtained at the ECw levels of 1.6, 1.7 and 1.2 dS m^{-1}, respectively. These DQI values show that the seedlings had good quality for this N rate, because they remained above the minimum standard value of 0.20 established by Hunt (1990). Besides favoring crop growth, adequate N rates can reduce the effect of salinity on plants, because they stimulate the production of organic solutes, which increases the capacity for osmotic adjustment, and plant resistance to water and saline stress (Silva et al., 2008).

As to the follow-up analysis of N rates at each water salinity level (Figure 6B), it is verified that there was no significant effect on plants irrigated with the ECw of 0.3 dS m^{-1}, whose average DQI was 0.23. However, it is observed that the N rate of 70% (541.1 mg of N dm^{-3}) favored higher DQI values (0.28, 0.30, and 0.22) in plants irrigated with salinity levels of 1.1, 1.9 and 3.5 dS m^{-1}, and the increase in N fertilization caused linear decreases of 8.43, 11.10, and 12.67% for each 30% increment in N rate. However, in plants under water salinity of 2.7 dS m^{-1}, DQI data best fitted to a quadratic regression equation, with the highest value (0.261) obtained at the N rate of 105% (811.65 mg of N dm^{-3}). These data are consistent with those of Dias et al. (2012), who observed higher DQI values in 'Paluma' guava seedlings at the N rate of 800 mg dm^{-3} in plants irrigated with non-saline water. Additionally, under salinity conditions (Figure 6B), N fertilization close to this rate also reduces the negative effect of saline stress on rootstock quality.

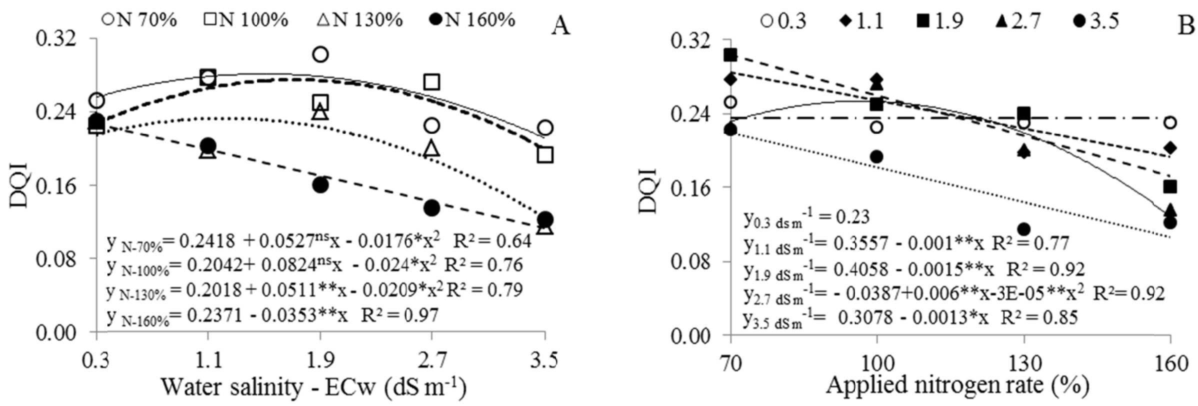

Figure 6. Follow-up analysis of the factor irrigation water salinity at each percentage of recommended nitrogen rate (A) and the factor percentage of recommended nitrogen rate at each salinity level (B) for the DQI of guava seedlings at 190 DAE

Note: ns,** and * = non-significant, significant at 0.01 and 0.05 probability levels ($p \leq 0.01$ and $p \leq 0.05$).

4. Conclusions

Salt stress provoked by electrical conductivity of irrigation water of beyond 1.4 dS m^{-1} affects negatively the formation of phytomass and quality of guava cv. Paluma rootstock seedlings, and this effect is mitigated on root dry matter and Dickson quality index by the increase in nitrogen rate up to application of 106% (819.38 mg of N dm^{-3} of soil) of the recommended rate for production of seedlings of the crop.

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