Physiological and vegetative behavior of banana cultivars under irrigation water salinity

Edvaldo B. Santana Júnior[1], Eugênio F. Coelho[2], Karoline S. Gonçalves[2] & Jailson L. Cruz[2]

[1] Instituto Federal do Piauí. Paulistana, PI, Brasil. E-mail: edvaldobispo@gmail.com - ORCID: 0000-0001-6133-4096
[2] Embrapa Mandioca e Fruticultura. Cruz das Almas, BA, Brasil. E-mail: ecoelho@cnpmf.embrapa.br - ORCID: 0000-0001-7079-6858; karolinesg@yahoo.com.br
  (Corresponding author) - ORCID: 0000-0002-1124-9309; jailson.cruz@cnpmf.embrapa.br - ORCID: 0000-0002-2536-6143

ABSTRACT: The purpose of the study was to evaluate the effect of salinity levels of irrigation water on physiology and growth of banana cultivars during the vegetative stage. The experiment was carried out following a completely randomized design in split plots, with four levels of salinity in irrigation water (0.5, 2.0, 3.0 and 4.0 dS m^{-1}) and four banana cultivars (Pacovan, Prata Anã, BRS Platina and Princesa) with three repetitions. Stomatal conductance, transpiration, relative water content, leaf water potential and proline concentration were evaluated at 150 days after transplanting (DAT), besides plant height, stem diameter and leaf area. All variables showed sensitivity to the increase of salinity level of irrigation water. The results of soil water extraction, relative water content in leaf (RWC), leaf water potential and proline content differed (p ≤ 0.05) among the cultivars. Prata Anã and BRS Platina cultivars were the ones with lowest sensitivity, while Pacovan and BRS Princesa cultivars showed larger variation of soil water extraction and RWC with the increase in irrigation water salinity (electrical conductivity), being considered the ones of highest sensitivity to salinity in this study. Banana crop growth as a consequence of physiological effects was inhibited by the increase in water salinity, mainly in "BRS Princesa" cultivar.

Key words: *Musa* spp., relative transpiration, electrical conductivity, drip irrigation

Comportamento fisiológico e vegetativo de cultivares de bananeira sob salinidade da água de irrigação

RESUMO: Objetivou-se neste trabalho avaliar o efeito de níveis de salinidade da água de irrigação na fisiologia e crescimento durante a fase vegetativa em cultivares de bananeira. O experimento foi conduzido utilizando delineamento inteiramente casualizado em esquema de parcelas subdivididas, com quatro níveis de salinidade da água de irrigação (0,5; 2,0; 3,0 e 4,0 dS m^{-1}) e quatro cultivares de bananeira (Prata Anã, BRS Platina, Princesa e Pacovan) com três repetições. Foram realizadas as seguintes avaliações: condutância estomática, transpiração, conteúdo relativo de água na folha, pressão de turgescência das folhas e teor de prolina, aos 150 dias após o transplantio, e altura de planta, diâmetro de caule e área foliar. Todas as variáveis avaliadas apresentaram sensibilidade ao aumento do nível de salinidade da água. Os resultados de extração de água do solo, conteúdo relativo de água, pressão de turgescência e o teor de prolina na folha diferiram (p ≤ 0.05) entre as cultivares. As cultivares Prata Anã e BRS Platina apresentaram menor sensibilidade, enquanto a Pacovan e BRS Princesa maior variação na extração de água do solo e no conteúdo relativo de água na folha com o aumento da salinidade da água de irrigação (condutividade elétrica) sendo consideradas as de maior sensibilidade à salinidade nesse estudo. O crescimento da bananeira como consequência dos efeitos na fisiologia foi inibido pelo incremento da salinidade, principalmente na cultivar BRS Princesa.

Palavras-chave: *Musa* spp., transpiração relativa, condutividade elétrica, gotejamento

INTRODUCTION

Banana (*Musa* spp) is considered one of the most important crops commercially cultivated worldwide, with an estimated production of 113 million tons in an area of 5.6 million hectares (FAOSTAT, 2017). In Brazil, this fruit crop has significant importance, as banana is the fruit with the second largest volume of production in Brazil, only behind orange (IBGE, 2018).

The banana plant is highly sensitive to salinity, requiring irrigation water electrical conductivity values lower than 1.0 dS m^{-1} for adequate development (Gondim et al., 2006). The use of irrigation water with salt contents above the limit tolerated by the crop can cause significant reduction in its growth, which may lead to severe losses in the final production.

Salinity is one of the abiotic stresses that most compromise agricultural production, especially in arid and semiarid regions, where the problems caused by salinization are even more accentuated due to the characteristics of the soil, climatic conditions and the inadequate irrigation management performed by some irrigators, which can thus favor the accumulation of salts in soil and water (Cova et al., 2016).

Salt stress can induce physiological modifications and compromise crop growth, development and production (Freire et al., 2014). In susceptible species, it is manifested by severe reductions in growth, water and photosynthetic exchange activities, disorders in membrane permeability and ionic balance (Cavalcante et al., 2010), besides damaging the stomatal conductance and/or diffusive resistance, transpiration and leaf temperature (Nascimento et al., 2011; Taiz et al., 2017). The increase in the concentration of salts in the soil solution causes the water to become increasingly less available to plants and, once the soil has reduced osmotic potential in the solution, water extraction by plants is substantially affected (Alves et al., 2011; Iqbal et al., 2014).

Studies have been conducted with saline water for banana cultivation, reporting morpho-physiological and yield alterations (Silva et al., 2009; Willadino et al., 2011; Silva Junior et al., 2012). However, most studies are focused on assessing the effects of salinity considering the banana tree in its initial stage (Silva Junior et al., 2016) and some cultivars were not adequately evaluated. Therefore, the objective of this study was to evaluate the effect of the application of irrigation water salinity on physiological and growth variables in banana cultivars.

MATERIAL AND METHODS

The study was carried out in the experimental field of Embrapa Mandioca e Fruticultura Tropical in the municipality of Cruz das Almas, BA, Brazil (12° 40' 19" S, 39° 06' 23" W; 225 m of altitude). The climate of the region is classified as humid to sub-humid, with annual average air relative humidity and air temperature of 80% and 24 °C, respectively, and average annual rainfall of 1.170 mm, with variation between 900 and 1.300 mm (Lima Filho et al., 2013) in a sandy clay loam soil, whose chemical and physico-hydraulic attributes are presented in Table 1.

The experiment was set up in a completely randomized design in a split-plot scheme. The plots were used to evaluate four irrigation water salinity (0.5; 2.0; 3.0 and 4.0 dS m^{-1}), equivalent to four soil solution salinity (0.86; 2.28; 3.25 and 4.58 dS m^{-1}), and four banana cultivars in the subplots (Prata Anã, BRS Platina, Princesa and Pacovan), with three repetitions. The salinity in the soil solution were measured by reading of the soil solution, extracted with soil solution extractors and read in a benchtop conductivity meter. The values remained constant throughout the experiment.

The field experiment consisted of 48 plastic pots with 100 L volume of each, height of 1.0 m and diameter of 0.60 m, used as drainage lysimeters, distributed at spacing of 2.0 x 2.5 m. Each pot was cultivated with one banana seedling, which was already acclimated, totaling 48 plants in the area, 12 plants of each cultivar. A drainage system consisting of 8.0 L of crushed stone nº 0 and a perforated 32-mm-diameter PVC pipe connected to a 20-mm-diameter valve was installed at the bottom of each pot.

The water used in the irrigations (Table 1) was prepared by adding sodium chloride (NaCl) and calcium chloride (CaCl$_2$) in sufficient quantities to obtain electrical conductivities with ionic proportions equivalent to 3:2, respectively for Na$^+$ and Ca^{2+}. The salts were added to obtain salinity of 2.0, 3.0 and 4.0 dS m^{-1}, since the salinity of 0.5 dS m^{-1} was the control treatment, with no addition of salts.

Irrigation was performed using a drip system, which had pressure-compensating emitters with flow rate of 4 L h^{-1}. After planting the seedlings, soil moisture was monitored with two TDR probes installed in each pot, at a 0.10 m distance from the pseudostem and at depths of 0.20 and 0.40 m. The water volume replaced in each irrigation was calculated from Eq. 1.

$$VR = \left(\theta_{fc} - \theta_a \right) V_s \, 1000 \qquad (1)$$

where:

VR - volume of replacement, L;
θ_{fc} - moisture at field capacity, m^3 m^{-3};
θ_a - actual moisture at the moment of irrigation, m^3 m^{-3}; and,
V_s - volume of soil occupied by the plant, m^3.

Table 1. Physico-hydraulic and chemical characterization of the soil and water used in the experiment

pH in H$_2$O	EC (dS m^{-1})	K	P	Ca	Mg	Ca+Mg	Al	Na	H+Al	SB	CEC	V (%)	OM (g kg^{-1})
		(mg dm^{-3})				(cmol$_c$ dm^{-3} (Soil))							
6.60	0.86	0.70	65	3.50	1.30	4.80	0	0.05	1.21	5.54	6.75	82	18.42
		(mg L^{-1} (Water))											
6.50	0.50	27	0.05	28.40	17	45.17	-	28	-	-	-	-	-

Depth (m)	Soil water retention (cm^3 cm^{-3})					
	0.01MPa	0.033MPa	0.1 MPa	0.3MPa	1.5 MPa	Available water
0-0.20	0.23	0.21	0.21	0.19	0.18	0.05
0.20-0.40	0.31	0.24	0.24	0.23	0.22	0.09

SB - Sum of bases; CEC - Cation exchanged capacity; V - Base saturation; OM - Organic matter

In the vegetative growth stage (150 days after transplanting-DAT), stomatal conductance (g_s) measurements were performed using the Delta-T AP4 porometer, previously calibrated, at times of 9, 11 and 15 h, in all plants. The relative transpiration in the treatments under salt stress was evaluated by covering the soil surface with a plastic tarpaulin for a 6-day period, along the vegetative growth stage of the plants (150-156 DAT) and, during this period, the water loss exclusively by transpiration was evaluated for 24 h based on the difference of moisture in the soil profile (Eqs. 2 and 3), as well as the water extraction by plants in the 150-156 DAT period, according to Eq. 4.

$$TR = \theta_j - \theta_{j+1} + I + D_r \qquad (2)$$

where:

TR   - transpired water depth, mm;

θ_j - water depth stored in the soil volume before irrigation, mm;

θ_{j+1} - water depth stored in the soil volume after 24 h, mm;

I   - water depth infiltrated in the period (j) - (j+1), mm; and,

D_r - water depth drained in the period (j) - (j+1), mm.

The water depth stored in the soil profile at a given time was calculated by Eq. 3:

$$L = \int_0^{0.4} \theta d_z \qquad (3)$$

where:

L   - water depth stored in the soil profile, m;

dz   - soil depth, m; and,

θ - volumetric soil moisture

Water extraction in the soil volume in the period (j) - (j+1) was calculated by Eq. 4:

$$Ext = \int_0^{0.4} \theta_j - \int_0^{0.4} \theta_{j+1} \qquad (4)$$

where:

Ext   - water extraction in the 0-0.40 m layer, m;

θ_j - water depth stored at the time after irrigation, m; and,

θ_{j+1} - water depth stored in the soil before the next irrigation, m.

The transpiration of plants under saline water application (Ts) was compared with the average of plants in the control treatment (Tns), i.e., relative to the lysimeters which received water with EC of 0.5 dS m^{-1}, generating the relative transpiration adapted from Sinclair (1986) (Eq. 5).

$$RT = \frac{Ts}{Tns} \qquad (5)$$

where:

RT   - relative transpiration;

Ts   - transpiration of treatments under stress, mm d^{-1};

Tns   - transpiration of the treatment under no stress, mm d^{-1}.

Leaf relative water content (RWC) at 150 DAT was obtained according to the equation proposed by Rodríguez-Gamir et al. (2010) (Eq. 6).

$$RWC = \frac{FM - DM}{TM - DM} \qquad (6)$$

where:

RWC - relative water content, %;

FM   - fresh mass of leaf disc, g;

DM   - dry mass of leaf disc, g; and,

TM   - turgid mass of leaf disc, g.

Leaf turgor pressure was determined using Wiltmeter° device and estimated from Eq. 7, proposed by Calbo et al. (2010). The evaluation was performed at 150 DAT using samples of leaves from the middle third of the third leaf of the banana plant, collected at 5:00 h in the morning.

$$\Psi P = P_y \cdot f_w \qquad (7)$$

where:

ΨP - turgor pressure, kPa;

P_y - pressure read on the device's manometer, kPa; and,

f_w - wiltmeter factor.

The free proline concentration in the leaf tissue of each cultivar at 150 DAT was determined according to the methodology of Bates et al. (1973). The amount of free proline was obtained by standard curve. The air vapor pressure deficit (VPD) was calculated from the air temperature values, used to determine the water vapor saturation pressure in the air, as proposed by Tetens (1930) (Eq. 8).

$$VPD = e_s - e_a \qquad (8)$$

where:

VPD - air vapor pressure deficit, kPa;

e_s - air saturation vapor pressure, kPa; and,

e_a - partial vapor pressure, kPa.

Plant growth was evaluated monthly, until the flowering period, by measurements of the following variables: plant height, pseudostem diameter at 0.10 m from the soil, number of leaves and leaf area. Leaf area was estimated according to the methodology proposed by Moreira (1999).

The obtained data were subjected to analysis of variance and Tukey's means comparison test for cultivars at p ≤ 0.05. While regression analysis was used for electrical conductivity of irrigation water. The statistical program Sisvar was used (Ferreira, 2011).

RESULTS AND DISCUSSION

Stomatal conductance (g_s) was influenced (p ≤ 0.05) by the salinity of irrigation water, with no difference among

cultivars. Stomatal conductance values were reduced as the salinity of irrigation water increased. The rate of reduction in stomatal conductance was 40.22 mmol m^2 s^{-1} per unit of dS m^{-1} of electrical conductivity (EC) (Figure 1).

The reduction of stomatal conductance with the increase in EC of the water applied to the soil was possibly due to the increased resistance to CO_2 diffusion, caused by the high levels of irrigation water salinity in the soil solution (Silveira et al., 2010; Silva et al., 2011; Kusvuran, 2012).

The increase in the level of salts in the soil solution causes a reduction in soil water potential, which is usually higher than the root water potential. Thus, there is a reduction in the water potential gradient in the soil-root direction, with an increase in the soil water retention tension, thus compromising the absorption by roots. The reduction in stomatal conductance is a mechanism of tolerance to salinity, which reduces water losses through the ostiole and minimizes the occurrence of desiccation of the plant (Silva et al., 2014; Rodrigues et al., 2014).

The higher stomatal conductance were found in plants under application of water with EC of 0.5 dS m^{-1} in all three times during the day (Table 2). The application of water with EC equal to or above 2.0 dS m^{-1} contributes to the reduction in the total soil water potential and rises up the soil-air potential gradient. Therefore, it intensifies the impact on stomatal closure, by reducing stomatal conductance, regardless of the time, compared to the application of non-saline water of 0.5 dS m^{-1}.

The increase in the salinity level in irrigation water to 2.0 dS m^{-1} resulted in a reduction in plant stomatal conductance of 36.6% (at 11:00 h), and of 49.9% in the highest saline level (EC 4.0 dS m^{-1}), when compared to the lower salinity level (EC 0.5 dS m^{-1}). The reduction of stomatal opening with the

increase of salinity level causes a reduction in the CO_2 uptake in the photosynthetic process, which reduces considerably the net photosynthesis rate by plants, negatively affecting the growth and development of banana crop (Silva Junior et al., 2012).

With air temperature increase between 9:00h e 11:00h, there is a gradual increase in the vapor pressure deficit until it reaches a maximum value. The water potential gradient between the soil and the boundary layer of air on the leaf surface is maximum at this moment, so that the soil cannot meet the demanded flow, resulting in partial stomatal closure or reduction of stomatal conductance, which is accentuated under conditions of increased salinity (Munns & Tester, 2008; Taiz et al., 2017).

All cultivars had linear reduction of water extraction with the increase in EC, and the largest decrease occurred in the cv. Princesa (0.701 mm d^{-1} dS^{-1}), followed by BRS Platina, Prata Anã and Pacovan (0.393 mm $d^{-1}dS^{-1}$), (Figure 2A). The cultivars showed different levels of sensitivity in terms of soil water extraction, consistent with the relative transpiration, which followed the same behavior.

There was difference in water extraction between the cultivars for all EC values, except for 0.5 dS m^{-1} (Table 3). The cultivar BRS Platina showed higher soil water extraction compared to Prata Anã (EC 2.0 dS m^{-1}) and to Prata Anã (EC 3.0 dS m^{-1}). The cultivar Princesa had lower water extraction rate under irrigation with 4.0 dS m^{-1}.

The average relative transpiration (RT) of banana cultivars was not influenced by the cultivars and was reduced with the increase in the salinity level of irrigation water for all of them.
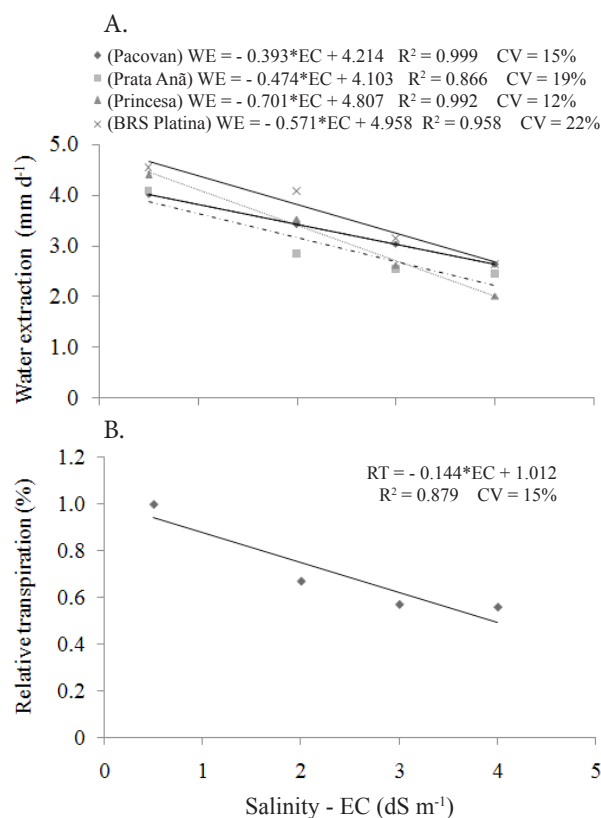

$$gs = -40.228*EC + 276.1$$
$$R^2 = 0.792 \quad CV = 24\%$$
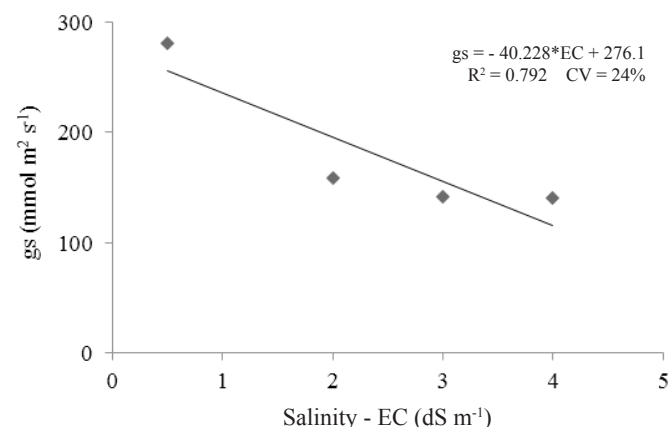
* Significant at p ≤ 0.05 by F test

Figure 1. Stomatal conductance (gs) in banana cultivars as a function of irrigation water salinity (EC)

Table 2. Stomatal conductance (gs) and vapor pressure deficit (VPD) in banana cultivars subjected to irrigation water salinity at three different hours of day

Time VPD/ Hour (kPa)	Stomatal conductance (mmol m^2 s^{-1})			
	Salinity levels of irrigation water (dS m^{-1})			
	0.5	2.0	3.0	4.0
9:00 1.41	204.08 bA	160.37 aB	116.53 aC	102.87 bC
11:00 2.18	281.08 aA	178.01 aB	141.75 aC	140.91 aC
15:00 1.61	93.54 cA	69.18 bB	72.38 bB	80.36 cB

Means followed by the same letter, lowercase in the column and uppercase in the row do not differ statistically by Tukey test at p ≤ 0.05

A.

* (Pacovan) WE = - 0.393*EC + 4.214 $R^2 = 0.999$ CV = 15%
■ (Prata Anã) WE = - 0.474*EC + 4.103 $R^2 = 0.866$ CV = 19%
▲ (Princesa) WE = - 0.701*EC + 4.807 $R^2 = 0.992$ CV = 12%
× (BRS Platina) WE = - 0.571*EC + 4.958 $R^2 = 0.958$ CV = 22%

B.

$$RT = -0.144*EC + 1.012$$
$$R^2 = 0.879 \quad CV = 15\%$$

* Significant at p ≤ 0.05 by F test

Figure 2. Soil water extraction -WE (A) and relative transpiration - RT (B) of banana cultivars as a function of irrigation water salinity (EC)

Table 3. Mean soil water extraction of banana cultivars irrigated with salinity levels in irrigation water

Cultivars	Mean soil water extraction (mm d⁻¹)			
	Salinity levels of irrigation water (dS m⁻¹)			
	0.5	2.0	3.0	4.0
Pacovan	4.02 a	3.42 ab	3.04 a	2.64 a
Prata Anã	4.09 a	2.84 b	2.53 b	2.45 a
Princesa	4.41 a	3.53 ab	2.62 b	2.01 b
BRS Platina	4.55 a	4.08 a	3.15 a	2.63 a

Means followed by the same letter in the column do not differ statistically by Tukey test at p ≤ 0.05

The rate of reduction of RT was 14.4% for unit increase of 1.0 dS m⁻¹ in the electrical conductivity of irrigation water (Figure 2B).

The behavior of RT is a response of the reductions in stomatal conductance and total soil water potential as the EC increased. Stomatal conductance is responsible for the input and output flows of CO_2 and water through the stomata, and the smaller the opening, the greater the stomatal resistance, resulting in a decrease of transpiration (Munns & Tester, 2008; Taiz et al., 2017), consequently leading to losses in biometric growth and biomass by plants (Willadino et al., 2011).

The mean relative water content in the leaf (RWC) did not differ between cultivars under application of 0.5 dS m⁻¹ electrical water conductivity. From 2.0 dS m⁻¹, the means decreased with no consistency that could allow a regression model to be fitted. At EC between 2.0 and 3.0 dS m⁻¹, the means of RWC that decreased the most were those of "Pacovan" and "Princesa" (Table 4).

With the application of irrigation water with EC of 4.0 dS m⁻¹, only the cultivar Prata Anã maintained the mean relative water content in the leaf above 90%, which represents a reduction in RWC of only 1.37% compared to the mean obtained with the application of 0.5 dS m⁻¹ water. This result indicates that the cultivar Prata Anã was able to maintain its RWC without alterations even under increased water salinity in the soil, which possibly reflects, compared to the other cultivars, the activation of mechanisms of physiological adaptation against the adversity in soil water availability (Taiz et al., 2017). The reduction of RWC in many crops is a recurrent action when there is an increase in soil salinity (Ferraz et al., 2015).

The means of turgor pressure (ΨP) of leaf cells differed only between the cultivars Prata Anã and BRS Princesa, which had the highest and lowest means of ΨP, respectively, confirming the relationship between RWC and the turgor pressure of leaf cells (Figure 3A). One of the justifications for the reduction in RWC, as well as in turgor pressure, especially

Table 4. Relative water content (RWC) in leaves of banana cultivars subjected to increasing salinity of irrigation water

Cultivars	Relative water content (%)			
	Salinity levels of irrigation water (dS m⁻¹)			
	0.5	2.0	3.0	4.0
Pacovan	91.38 a	90.99 a	85.92 b	82.87 b
Prata Anã	92.19 a	90.54 a	93.23 a	90.82 a
Princesa	92.11 a	83.55 b	85.45 b	85.98 b
BRS Platina	89.18 a	88.55 a	89.00 a	82.94 b

Means followed by the same lowercase letter in the column do not differ statistically by Tukey test at p ≤ 0.05
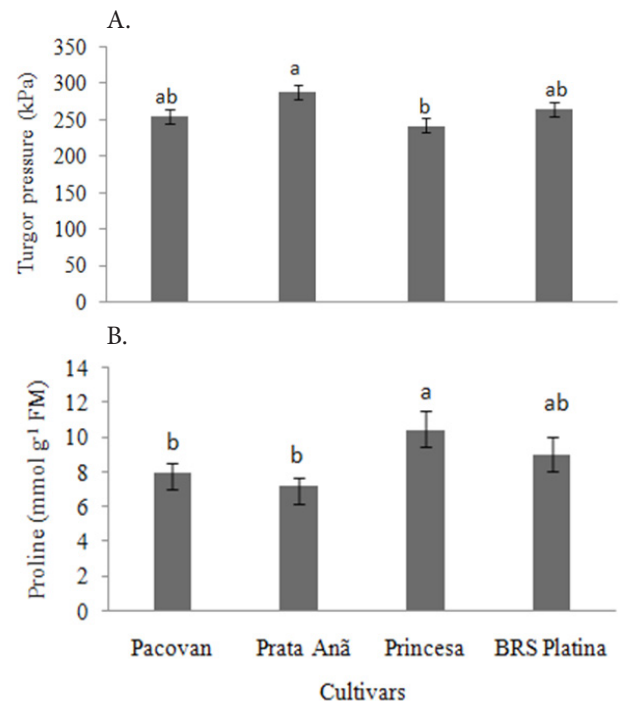
Columns represent the mean and bars represent ± the standard error of the mean; Bars with different letters are significant at p ≤ 0.05 by Tukey test; FM - Fresh matter
Figure 3. Turgor pressure in leaf cells (A) and proline concentration in leaves (B) of banana cultivars subjected to irrigation water salinity

in the cultivar BRS Princesa, may be the accumulation of high contents of Na⁺ and Cl⁻ in the leaves (Silva Junior et al., 2016; Taiz et al., 2017).
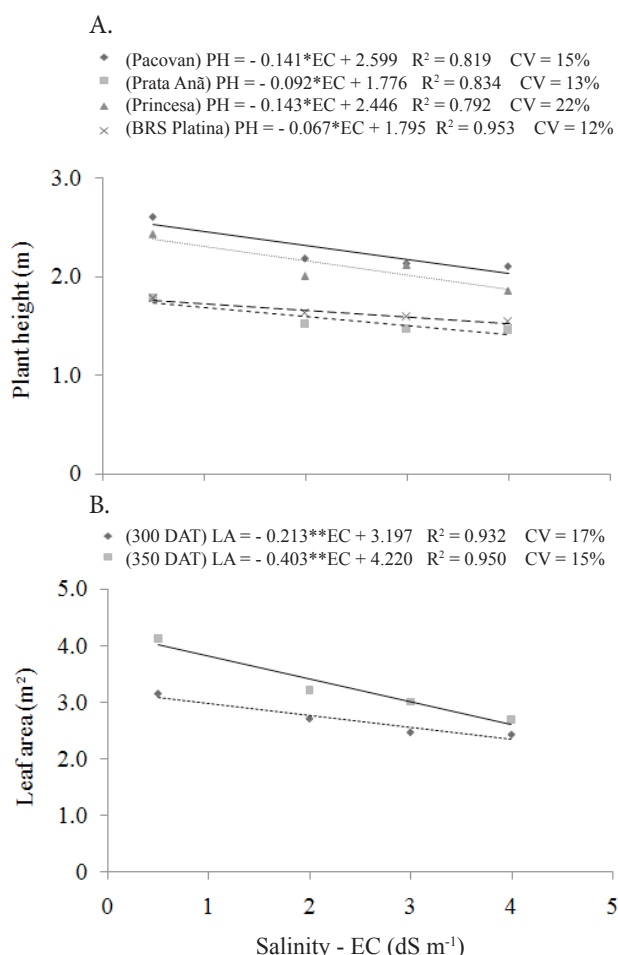
The cultivars influenced the leaf proline concentration, and the highest mean was observed in the cv. Princesa, differing statistically from the cultivars Pacovan and Prata Anã. The mean leaf proline concentration of the cultivar BRS Platina had an intermediate value, not differing statistically from those observed in the other cultivars (Figure 3B).

The highest proline concentration in the cultivar Princesa associated with higher rate of reduction in water extraction and lower mean of the turgor potential are approached as metabolic disorders induced by the accumulation of salts in the leaves, especially Na⁺ and Cl⁻ (Oliveira et al., 2013), suggesting that this cultivar is more sensitive to salt stress.

Plant height was inhibited with the increase of salinity (EC) for all cultivars. The ranges of reduction in plant height as the irrigation water electrical conductivity increased were higher for the cv. Princesa and lower for the cv. BRS Platina (Figure 4A), a situation that confirm that the cv. "Princesa" is the most sensitive to salts compared to the others, thus reinforcing that banana genotypes may respond differently when subjected to salt stress (Silva Junior et al., 2012).

Reductions in the growth of plants subjected to salinity are directly associated with the reduction in their water absorption due to the decrease in the osmotic potential of the soil solution, caused by the accumulation of soluble salts from irrigation waters, and to the decrease in the absorption of essential nutrients (Munns & Tester, 2008), caused by ionic imbalance (Iqbal et al., 2014).

The average pseudostem diameter and number of leaves

A.

- ♦ (Pacovan) PH = - 0.141*EC + 2.599 R^2 = 0.819   CV = 15%
- ▪ (Prata Anã) PH = - 0.092*EC + 1.776 R^2 = 0.834   CV = 13%
- ▲ (Princesa) PH = - 0.143*EC + 2.446 R^2 = 0.792   CV = 22%
- × (BRS Platina) PH = - 0.067*EC + 1.795 R^2 = 0.953   CV = 12%

B.

- ♦ (300 DAT) LA = - 0.213**EC + 3.197 R^2 = 0.932   CV = 17%
- ▪ (350 DAT) LA = - 0.403**EC + 4.220 R^2 = 0.950   CV = 15%

*, ** - Significant at p ≤ 0.05 and p ≤ 0.01, respectively, by F test

Figure 4. Plant height - PH at 350 days after transplanting (DAT) in banana cultivars (A) and leaf area - LA for all cultivars at 300 and 350 DAT (B), as a function of electrical irrigation water salinity - EC

were not affected by the salinity levels. The leaf area of banana cultivars was affected by the salinity levels (Figure 4B). The rate of reduction in leaf area per salinity level in the evaluated period (350 DAT) corresponded to 0.403 m per unit of dS m^{-1}.

Reduction of leaf area in banana genotypes as a function of the increment in soil salinity levels has been widely reported (Silva et al., 2009; Silva Junior et al., 2012; Willadino et al., 2011), which can possibly be considered as a mechanism of adaptation of the plant to water deficit in soil.

Conclusions

1. The increase in irrigation water salinity inhibited stomatal conductance and transpiration of banana cultivars.

2. Due to the higher capacity for soil water extraction, higher relative water content and higher turgor pressure in the leaves, the cultivars BRS Platina and Prata Anã were the ones with highest tolerance, while Pacovan and Princesa were the most sensitive to the increase in irrigation water salinity.

3. The increase in irrigation water salinity inhibited the growth in plant height in the following order: Princesa > Pacovan > PrataAnã > BRS Platina and leaf area in all banana cultivars.

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