ISSN 1807-1929
Revista Brasileira de Engenharia Agrícola e Ambiental
v.22, n.9, p.628-633, 2018
Campina Grande, PB, UAEA/UFCG – http://www.agriambi.com.br

DOI: http://dx.doi.org/10.1590/1807-1929/agriambi.v22n9p628-633

Gas exchanges and photochemical efficiency of West Indian cherry cultivated with saline water and potassium fertilization

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Key words:
Malpighia emarginata
saline stress
physiology
fruit crops

ABSTRACT
The problem of the lack of adequate water resources for agriculture has intensified in recent years, making it necessary to use waters with relatively high concentration of salts for the irrigation of crops all over the world. The objective of this study was to evaluate the influence of potassium (K) fertilization as a salt stress attenuator on gas exchanges and photochemical efficiency of West Indian cherry. The crop was cultivated under greenhouse conditions in the municipality of Campina Grande, PB, in lysimeters filled with 250 kg of sandy loam soil. Treatments were distributed in randomized blocks, in a 2 x 4 factorial scheme, with two levels of electrical conductivity of irrigation water - ECw (0.8 and 3.8 dS m^{-1}) and four K doses (50, 75, 100 and 125% of recommendation), in which the dose corresponding to 100% was equal to 19.8 g of K_2O, with three replicates and one plant per repetition. Seedlings of the West Indian cherry cultivar BRS 366-Jaburu, grafted onto a locally developed rootstock from the clonal garden of the EMBRAPA Tropical Agroindustry, Pacajus, CE, were used. Irrigation with salinized water (electrical conductivity of 3.8 dS m^{-1}) compromised the gas exchange and the photochemical efficiency of West Indian cherry plants. Potassium fertilization was not efficient at reducing the stress caused by water salinity on West Indian cherry plants.

Palavras-chave:
Malpighia emarginata
estresse salino
fisiologia
fruticultura

Trocas gasosas e eficiência fotoquímica da aceroleira cultivada com águas salinas e adubação potássica

RESUMO
O problema da falta de recursos hídricos adequados para agricultura vem se intensificando nos últimos anos, tornando-se necessário o uso de águas com relativamente alta concentração de sais para a irrigação das culturas por todo o mundo. Com isso, objetivou-se avaliar a influência da adubação potássica como atenuador do estresse salino sobre as trocas gasosas e eficiência fotoquímica da aceroleira. O cultivo das aceroleiras foi realizado em condição de casa de vegetação no município de Campina Grande, PB, em lisímetros preenchidos com 250 kg de solo de textura franco-arenosa. Os tratamentos foram distribuídos em blocos casualizados, em esquema fatorial 2 x 4, sendo dois níveis de condutividade elétrica da água de irrigação - CEa (0,8 e 3,8 dS m^{-1}) e quatro doses de potássio (50, 75; 100 e 125% da recomendação), sendo a dose correspondente a 100% igual a 19,8 g de K_2O por planta ano^{-1}, com três repetições e uma planta por repetição. Foram utilizadas mudas de acerolas cultivar BRS 366-Jaburu, enxertadas sobre o porta-enxerto Crioulo proveniente do jardim de sementes da EMBRAPA Agroindústria Tropical, em Pacajus, CE. A irrigação com água salina (condutividade elétrica de 3,8 dS m^{-1}) comprometeu as trocas gasosas e a eficiência fotoquímica das plantas de acerola. A adubação potássica não foi eficiente na redução do estresse promovido pela salinidade da água em plantas de aceroleira.

Ref. 185978 – Received 30 Sept, 2017 • Accepted 22 Mar, 2018 • Published 25 Jul, 2018

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Introduction

West Indian cherry (*Malpighia emarginata* D.C.) is a fruit crop that has stood out as an excellent natural source of vitamin C, besides anthocyanins and carotenoids, compounds with beneficial effects on human health due to their recognized antioxidant action (Maciel et al., 2010). According to Freitas et al. (2006), Brazil is the largest producer, consumer and exporter of this fruit in the world, and its Northeast region stands out as the region were West Indian cherry is best adapted to, due to the conditions of soil and climate.

In the Northeast region, West Indian cherry production reaches 22,964 tons in an area of 7,237 ha, which represents about 70% of the Brazilian production (Caetano, 2010). Thus, the cultivation of West Indian cherry becomes highly promising in the Northeast region, having good prospects for the fruit grower market. Despite the good adaptability of West Indian cherry, the Brazilian semi-arid region poses risks to its cultivation, due to the scarcity of water resources, where the available sources of water for irrigation commonly have high concentrations of salts, especially sodium (Jiang et al., 2012; Souza et al., 2017).

The excess of salts in the soil causes reduction in the osmotic potential, resulting in water deficit in the crops. This deficit causes stomatal closure, limiting CO_2 assimilation and transpiration in plants, and consequently reduces their photosynthetic rate (Silva et al., 2010). In addition, plants subjected to saline stress undergo changes in the functional state of the thylakoid membranes of chloroplasts, which cause alterations in the characteristics of the fluorescence signals (Chaum & Kirmanee, 2011; Silva et al., 2011a; Freire et al., 2014), thus diminishing growth and yield (Sousa et al., 2012).

As a consequence, in an attempt to mitigate the deleterious effects caused on crops by the excess salts present in irrigation water and/or soil, some management practices have been developed. These practices include fertilization based on the use of fertilizers which favor the acquisition of nutrients by plants under saline conditions (Silva et al., 2011b). Works conducted by Gurgel et al. (2005, 2010) concluded that potassium (K) fertilization led to a decrease in the deleterious effects of irrigation with high-salinity water on the melon crop.

The beneficial effect of potassium is due to the fact that it is vital for photosynthesis, so that situations of deficiency cause reduction in the photosynthetic rate and increase in respiration. In addition, this nutrient favors the formation and translocation of carbohydrates, improving fruit quality (Araújo et al., 2012). Although the benefits of K fertilization are known, studies providing evidence of its mitigating action under conditions of saline stress on plants, mainly in West Indian cherry, are still scarce in the literature. Therefore, this study aimed to evaluate the influence of K fertilization as a mitigating factor of saline stress on gas exchanges and photochemical efficiency of West Indian cherry.

Material and Methods

The experiment was carried out under greenhouse conditions from July 2016 to July 2017, in recipients adapted as drainage lysimeters, at the Center of Technology and Natural Resources of the Federal University of Campina Grande (CTRN/UFCG), located in the municipality of Campina Grande - PB, Brazil (7° 15' 18" S; 35° 52' 28" W; ~550 m).

The experimental design was in randomized blocks, with three replicates, using a 2 x 4 factorial scheme, and treatments consisted of two levels of irrigation water electrical conductivity - ECw (0.8 and 3.8 dS m^{-1}) and four K doses (50, 75, 100 and 125% of recommendation). The 100% dose corresponded to 19.8 g of K_2O $plant^{-1}$ $year^{-1}$ (Musser, 1995).

The lysimeters were filled using a 1-kg layer of crushed stone no. 0, followed by 250 kg of a sandy loam eutrophic Regolithic Neosol, properly pounded to break up clods, from the rural area of the municipality of Esperança, PB, whose chemical and physical characteristics were obtained according to the methodologies proposed by Claessen (1997): Ca^{2+} = 9.07 $cmol_c$ kg^{-1}; Mg^{2+} = 2.78 $cmol_c$ kg^{-1}; Na^+ = 1.64 $cmol_c$ kg^{-1}; K^+ = 0.23 $cmol_c$ kg^{-1}; H^++Al^{3+} = 8.61 $cmol_c$ kg^{-1}; Al^{3+} = 0 $cmol_c$ kg^{-1}; CEC = 22.33 $cmol_c$ kg^{-1}; organic matter = 2.93 dag kg^{-1}; P = 39.8 mg kg^{-1}; pH in water (1:2.5) = 5.58; electrical conductivity of the saturation extract = 2.15 dS m^{-1}; SAR = 0.67 (mmol $L^{-1})^{0.5}$; exchangeable sodium percentage = 7.34%; sand = 659.9 g kg^{-1}; silt =161.2 g kg^{-1}; clay = 178.9g kg^{-1}; water content at 33.42 and 1519.5 kPa = 25.91 and 12.96 dag kg^{-1}, respectively.

The bottom of each lysimeter was connected to a 4-mm-diameter hose to drain the leachate into a container, in order to evaluate the drained water and determine water consumption by plants. The tip of the drain inside the pot was involved with a nonwoven geotextile (Bidim OP 30) to avoid clogging by soil particles.

The experiment used West Indian cherry seedlings, cultivar BRS 366-Jaburu, grafted onto locally developed rootstocks from the Embrapa Tropical Agroindustry, Pacajus, CE. At transplantation, seedlings were 240 days old. During the acclimation period in the greenhouse, plants were irrigated with low-salinity water (0.8 dS m^{-1}). The cultivar BRS 366-Jaburu stands out for its high yield (57 t ha^{-1}) and production of ascorbic acid (2,648 mg $100g^{-1}$). These plants are about 1.87 m tall with crown diameter of 2.18 m. Fruits are shiny when ripe, weighing on average 4 to 5 g when unripe, adequate stage to obtain vitamin C, and 6 to 7 g after ripening (EMBRAPA, 2012).

Prior to seedling transplantation, the soil was brought to field capacity using the respective water of each treatment. After transplantation, irrigation was carried out every day by applying in each lysimeter a water volume to maintain the soil near field capacity, and the applied volume was determined according to the water requirement of the plants, estimated by the water balance: volume applied minus the volume drained in the previous irrigation, plus a leaching fraction of 0.10, applied at intervals of 30 days.

Water with electrical conductivity of 3.8 dS m^{-1} was prepared by dissolving the salts NaCl, $CaCl_2.2H_2O$ and $MgCl_2.6H_2O$, at equivalent proportion of 7:2:1, respectively, in public-supply water (ECw = 1.40 dS m^{-1}) in the municipality of Campina Grande, PB, based on the relationship between ECw and concentration of salts (10 * $mmol_c$ L^{-1} = ECw dS m^{-1}), according to Richards (1954). On the other hand, the ECw level of 0.8 dS m^{-1} was prepared by mixing rainwater (ECw = 0.02 dS m^{-1}) in the municipal-supply water (ECw = 1.40 dS m^{-1}).

Fertilization with phosphorus, potassium and nitrogen was carried out as recommended by Musser (1995), applying the equivalent to 250, 33 and 53 g $plant^{-1}$ $year^{-1}$, respectively, of single superphosphate, potassium chloride and urea. Phosphorus was applied all at planting. Nitrogen and potassium were split into 12 portions, applied monthly. In order to prevent probable deficiencies in micronutrients, West Indian cherry plants received every week 5 L of solution containing 1.5 g L^{-1} of Ubyfol [N (15%); P_2O_5 (15%); K_2O (15%); Ca (1%); Mg (1.4%); S (2.7%); Zn (0.5%); B (0.05%); Fe (0.5%); Mn (0.05%); Cu (0.5%); Mo (0.02%)].

Gas exchanges were evaluated in the middle third of the crown of West Indian cherry plants during the transition from flowering to fruiting (at 180 days after transplanting), based on the following parameters: stomatal conductance (gs - mol H_2O m^{-2} s^{-1}), CO_2 assimilation rate (A) (μmol m^{-2} s^{-1}), transpiration (E) (mmol H_2O m^{-2} s^{-1}) and internal CO_2 concentration (Ci) (μmol mol^{-1}) using a portable infrared gas analyzer (IRGA), model LCPro+ Portable Photosynthesis System®. After collection, these data were used to quantify instantaneous water use efficiency (WUEi) (A/E) [(μmol m^{-2} s^{-1}) (mmol H_2O m^{-2} s^{-1})$^{-1}$] and intrinsic carboxylation efficiency (EiCi) (A/Ci) [(μmol m^{-2} s^{-1}) (μmol mol^{-1})$^{-1}$]. Initial fluorescence (Fo), maximum fluorescence (Fm), variable fluorescence (Fv) and potential quantum efficiency (Fv/Fm) were also measured in leaves pre-adapted to dark using leaf clips for 30 min, between 7 and 8 a.m., using a modulated fluorometer Plant Efficiency Analyser – PEA II®.

The data were subjected to analysis of variance by F test and, when significant, means were compared by Tukey test at 0.05 probability level for water salinity levels and regression analysis was conducted for K doses, using the statistical program SISVAR-ESAL (Ferreira, 2011).

Results and Discussion

There was significant influence ($p < 0.05$) of the interaction between levels of irrigation water salinity and K fertilization on variable fluorescence (Fv). The other parameters of gas exchanges and chlorophyll a fluorescence were only significantly influenced ($p < 0.05$) by the irrigation water salinity, except the instantaneous water use efficiency (WUEi), which was not significantly affected by any of the factors studied. Potassium doses significantly influenced ($p < 0.05$) only the variable fluorescence (Table 1).

Irrigation water salinity of 3.8 dS m^{-1} negatively affected gas exchanges in West Indian cherry, leading to accentuated reductions of the order of 47.58, 33.14, 41.03 and 56.75% in stomatal conductance, transpiration, CO_2 assimilation rate and instantaneous carboxylation efficiency, respectively, in comparison to the ECw level of 0.8 dS m^{-1} (Figures 1A, B, C and E). However, internal CO_2 concentration increased by 38.03% as a function of the increment in ECw levels from 0.8 to 3.8 dS m^{-1} (Figure 1D). According to Freire et al. (2014), such increment in Ci inside the leaves indicates that CO_2 is not being used for the synthesis of sugars in the photosynthetic process, suggesting that probably a non-stomatal factor was interfering with this process, which can be confirmed by the reduction in the photosynthetic rate of plants irrigated with 3.8 dS m^{-1} water (Figure 2A). Despite the drastic reduction in stomatal conductance, and consequently in transpiration, it did not influence the instantaneous water use efficiency or internal CO_2 concentration, since there was no reduction in CO_2 influx in plants irrigated with high-salinity water.

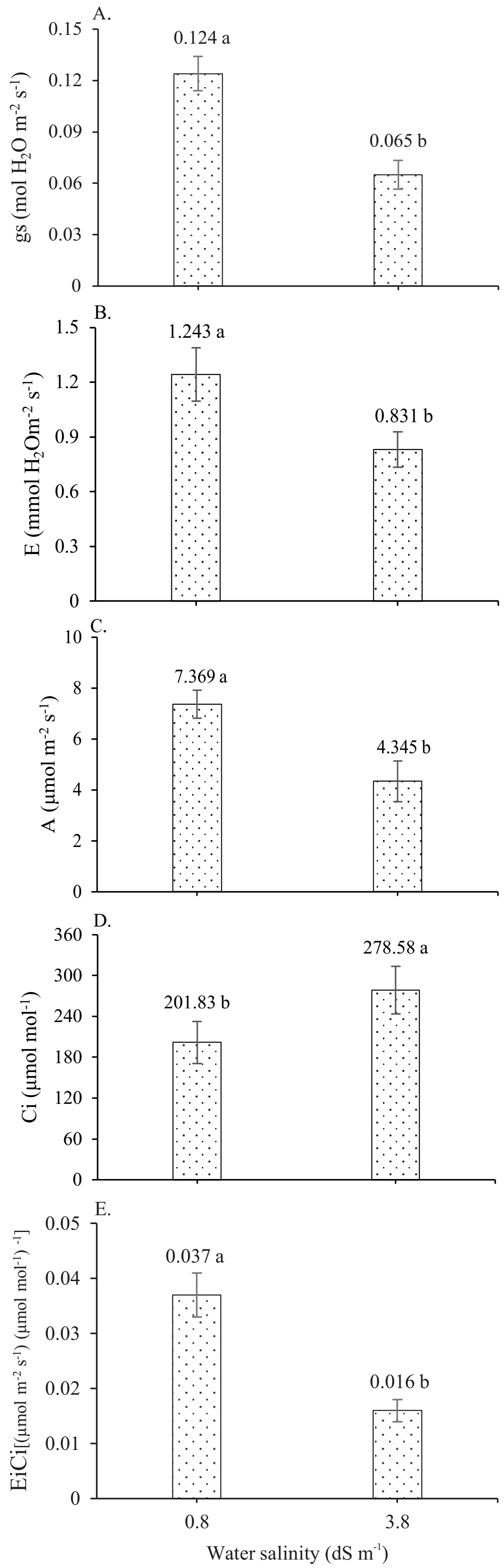
Thus, the reduction in stomatal conductance can be related to the tolerance mechanism of the species, aiming to reduce water loss and consequently the absorption of water and salts from the soil solution, without compromising photosynthetic activity. According to Taiz & Zeiger (2013), plants under salt stress have as the first response to salinity the reduction in stomatal opening, to reduce water loss through the leaves, which is induced by the reduction in the osmotic potential of the soil solution, resulting in higher resistance to water absorption by the roots. Accumulation of salts in the leaves also affects, significantly, essential physiological processes, including gs (Hussain et al., 2012).

The reductions observed in CO_2 assimilation rate and intrinsic carboxylation efficiency coincided with the increase in internal CO_2 concentration of plants cultivated under high salinity (Figures 1C, D and E). Reductions in intrinsic carboxylation efficiency are related to metabolic restrictions in the Calvin cycle, where the received carbon is not being fixed in the carboxylation stage in mesophyll cells (Larcher, 2006; Sousa et al., 2016). According to Taiz & Zeiger (2013), as the stress becomes severe there is a dehydration in mesophyll cells, the mesophyll metabolism is damaged and, consequently, carboxylation efficiency is compromised. This result is an indication that non-stomatal factors act on the

Table 1. Summary of the F test for stomatal conductance (gs), transpiration (E), CO_2 assimilation rate (A), internal CO_2 concentration (Ci), intrinsic carboxylation efficiency (EiCi), instantaneous water use efficiency (WUEi), initial fluorescence (Fo), maximum fluorescence (Fm), variable fluorescence (Fv) and potential quantum efficiency (Fv/Fm) of West Indian cherry plants irrigated with saline water and fertilized with potassium doses at 180 days after transplanting

Source of variation	F test									
	gs	E	A	Ci	EiCi	WUEi	Fo	Fm	Fv	Fv/Fm
Saline levels (SL)	**	**	**	**	**	ns	**	**	**	**
Doses of K (DK)	ns	ns	ns	ns	ns	ns	ns	ns	*	ns
Linear Regression	-	-	-	-	-	-	-	-	*	-
Quadratic Regression	-	-	-	-	-	-	-	-	*	-
Interaction SL*DK	ns	ns	ns	ns	ns	ns	ns	ns	*	ns
Blocks	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
CV (%)	21.91	25.16	22.51	25.15	10.00	36.33	14.76	7.33	11.64	3.86

ns, **, * Respectively not significant, significant at $p < 0.01$ and $p < 0.05$



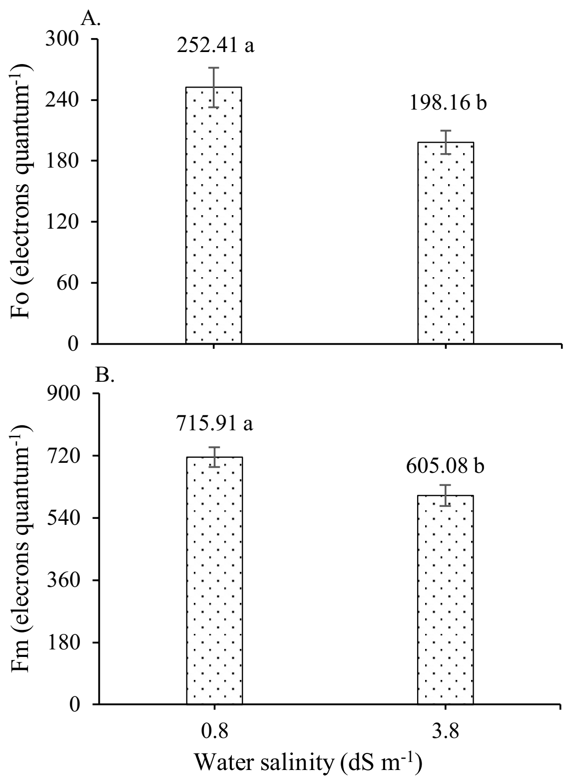
Bars represent the standard error of the mean (n=3). Mean followed by different letter indicates that the treatments differ by Tukey test, $p < 0.05$

Figure 1. Stomatal conductance - gs (A), transpiration - E (B), CO_2 assimilation rate - A (C), internal CO_2 concentration - Ci (D) and intrinsic carboxylation efficiency - CEi (E) of West Indian plants irrigated with saline water at 180 days after transplanting

photosynthetic activity of West Indian cherry, such as low activity of the enzyme Ribulose-1,5-bisphosphate carboxylase/oxygenase (RuBisCO), probably due to the low availability of substrate (ATP and NADPH) for activation and regeneration of the enzyme, which come from the photochemical phase of photosynthesis, leading to reduction in CO_2 carboxylation and increase in its internal concentration (Hussain et al., 2012; Silva et al., 2015; Sá et al., 2017).

Initial fluorescence (Fo) and maximum fluorescence (Fm) of West Indian cherry were reduced by 21.49 and 15.48% as water salinity increased from 0.8 to 3.8 dS m^{-1} respectively (Figures 2A and B). Initial fluorescence is characterized as the minimum fluorescence emitted when all reaction centers, dark adapted, are open (Gorbe & Calatayud, 2012). Maximum fluorescence is an indicator that represents the maximum intensity of the fluorescence emitted, when almost all quinone is reduced and the reaction centers reach their maximum capacity of photochemical reactions (Suassuna et al., 2010; Silva et al., 2015). The reduction in initial and maximum fluorescence indicates reduction in the photochemical capacity of West Indian cherry under saline stress conditions, which combined with the low CO_2 carboxylation efficiency confirms that the low photosynthetic yield of this crop under high salinity results from the damages caused by the excess of salts to the chlorophyll a, reducing its activity and thus the synthesis of ATP and NADPH.

Variable fluorescence in West Indian cherry plants irrigated with low-salinity water (0.8 dS m^{-1}) decreased linearly as K doses increased, with a reduction of 22.78% in the Fv in plants fertilized with 125% of the K recommendation in comparison to those subjected to 50%, which represents a reduction of

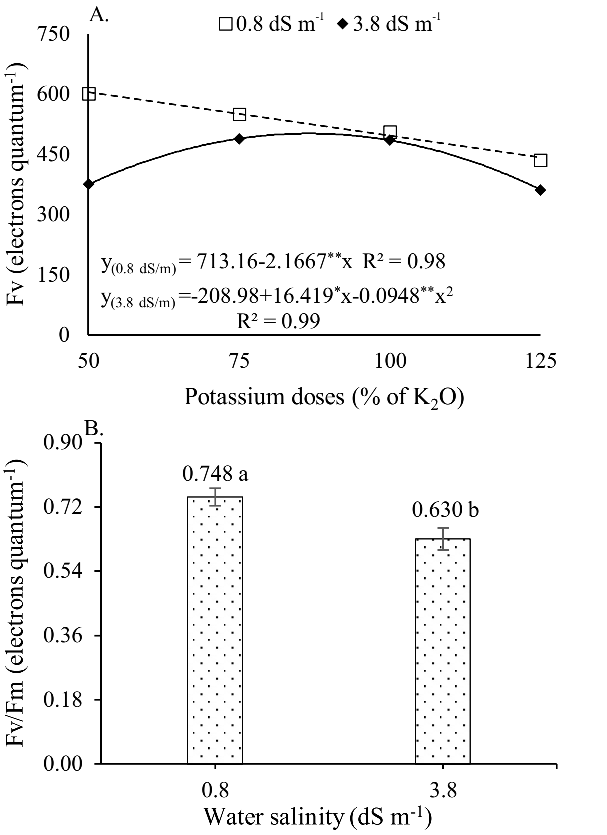


Bars represent the standard error of the mean (n=3). Mean with different letter indicates that the treatments differ by Tukey test, $p < 0.05$

Figure 2. Initial fluorescence - Fo (A) and maximum fluorescence - Fm (B) of West Indian cherry plants irrigated with saline water at 180 days after transplanting

162.50 electrons quantum^{-1} (Figure 3A). In plants irrigated with high-salinity water (EC = 3.8 dS m^{-1}) there was a quadratic behavior for variable fluorescence (Figure 3A), and highest Fv was obtained when plants were fertilized with K dose of 87% (501.93 electrons quantum^{-1}). From this dose on, the increment in K fertilization led to reduction in this variable and corresponded to 362.145 electrons quantum^{-1} at the highest K dose, which represents a decrease of 139.79 electrons quantum^{-1}. Such initial increase in Fv due to the increment in K dose is related to the greater competition of this cation with other ions, such as Na ions, thus reducing the deleterious effects of saline stress on chlorophyll a activity. However, as the dose increased, K may have intensified even more the salt stress.

As previously observed for Fo and Fm (Figures 2A and B), the potential quantum efficiency of photosystem II (Figure 3B) also decreased with the increase in salinity, and irrigation with high-salinity water (3.8 dS m^{-1}) caused reduction in the Fv/Fm ratio from 0.75, in plants irrigated with 0.8 dS m^{-1}, to 0.63 electrons quantum^{-1}. The Fv/Fm ratio reflects the efficiency relative to the absorption of light energy by the antenna complex of the PSII and to its conversion into chemical energy, being considered as a sensitive indicator of photosynthetic performance of the plant, as well as an indicator of disturbances in the photosynthetic system caused by stresses and its reduction consists in the limitation of photochemical activity (Lucena et al., 2012). Fv/Fm values between 0.75 and



Bars represent the standard error of the mean (n=3). Mean with different letter indicates that the treatments differ by Tukey test, $p < 0.05$

Figure 3. Variable fluorescence – Fv (A) as a function of the interaction between irrigation water salinity and potassium doses, and potential quantum efficiency of photosystem II - Fv/Fm (B) of West Indian cherry plants irrigated with saline water at 180 days after transplanting

0.85 electrons quantum^{-1} suggest that the photosynthetic apparatus is intact, as opposed to values lower than 0.75 electrons quantum^{-1} (Reis & Campostrini, 2011; Silva et al., 2015). Therefore, it can be inferred that the photosynthetic apparatus of West Indian cherry plants irrigated with 3.8 dS m^{-1} water (Figure 3B) is compromised by salt stress, since these plants had Fv/Fm values of 0.63 electrons quantum^{-1}.

Conclusions

1. Irrigation using water with electrical conductivity of 3.8 dS m^{-1} compromises gas exchanges and photochemical efficiency of West Indian cherry plants.

2. Potassium fertilization was not efficient at reducing the stress caused by the studied level of water salinity.

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