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Water relations and gas exchange in castor bean irrigated with saline water of distinct cationic nature

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Aiming to evaluate the water relations and gas exchange in castor bean cv. BRS Energia under salinity and cationic nature of irrigation water, an experiment was conducted under controlled conditions in an Ultisol Eutrophic of sandy loam texture. Treatments consisted of six combinations of water salinity (S_1 – Control (supply water ECw=0.6 dS m⁻¹); S_2 - Na⁺, S_3 - Ca²⁺, S_4 - Na⁺+ Ca²⁺, S_5 - K⁺ and S_6 - Na⁺+ Ca²⁺ + Mg²⁺all prepared with chloride salt of respective cation and having ECw=4.5 dS m⁻¹, distributed in a randomized block design with four replications. The use of saline water of distinct cationic nature affected the water relations of the castor bean, the least deleterious effects were observed in plants irrigated with potassic water The negative effects of the cationic nature of irrigation water were more evident in gas exchange at 40 days after sowing (DAS), the highest values for stomatal condutance, transpiration rate, assimialation of CO₂, instantaneous carboxylation efficiency and intrinsic water use efficiency were observed in case of potassium ion. The castor bean 'BRS Energia' is more sensitive to the presence of sodium ions in the irrigation water, both in terms of water relations as well as for gas exchange.

Key words: Ricinus communis L., salt stress, semiarid.

INTRODUCTION

The demand for vegetable oils is growing every year and the global interest in oilchemical industry is high, given its application in diverse forms. In this context, the castor bean (*Ricinus communis* L.) stands out as a distinct crop since oil extracted from its seeds has excellent properties of wide use as an industrial input due to the fact that in many of its applications, it can not be replaced by other vegetable oils (Santos and Kouri, 2007). This crop is

*Corresponding author. E-mail: geovanisoareslima@gmail.com Author(s) agree that this article remain permanently open access under the terms of the <u>Creative Commons Attribution</u> <u>License 4.0 International License</u> unique source of ricinoleic acid (over 85% of oil, which may be used for the manufacture of surfactants, coatings, lubrificants, fungicides, pharmaceuticals and cosmetics and a variety of other products (Pinheiro et al., 2008; Babita et al., 2010).

In the semi-arid region of Northeast Brazil, the uneven distribution of rainfall during the year makes irrigation as an indispensable practice for crop production. However, most of the surface reservoirs (small and medium size and ponds) and ground water (wells) has high salt content, and present variability in their ionic composition (Suassuna and Audry, 1990). Use of these water sources may, depending on its constitution, change the physiological and biochemical functions of plants resulting in disturbances in water relations and changes in the absorption and metabolism of essential plant nutrients (Amorim et al., 2010).

Furthermore, the use of water with different ionic compositions may cause varying degrees of stress to plants or alter the physical and chemical properties of the soil (Aquino et al., 2007) however, the degree of severity which components influence with these plant development depends on factors such as plant species, cultivar, growth stage and the ionic composition of the water (Sousa et al., 2012). Among the physiological indices affected by salinity stand outs the CO2 assimilation rate, transpiration, stomatal conductance and internal CO₂ concentration, which can be inhibited by accumulation of Na⁺ and/or Cl⁻ in chloroplasts, affecting biochemical and photochemical processes involved in photosynthesis (Taiz and Zeiger, 2013).

Associated with this is the interferences of salts in the water relations of the plants that are reflected in osmotic potential, relative water content and saturation water deficit in leaf, in addition to the ion toxicity and imbalance of nutrient absorption causing widespread reduction of growth with serious damage in productivity (Ahmed and Moritani, 2010). Thus, despite the existence of genetic variability for tolerance to salinity, physiological and biochemical mechanisms that contribute to this tolerance are still poorly understood, mainly under different cationic nature of water (Mansour et al., 2003), especially in castor bean crop cv. BRS Energia.

Considering understanding of changes in water relations and physiological responses of plants grown under the influence of abiotic stress, such as saline, can provide important tools to aid in the management of species irrigated with saline waters, this study proposed to evaluate the water relations and gas exchange in the castor bean cv. BRS Energia irrigated with saline water of distinct cationic nature.

MATERIALS AND METHODS

The experiment was carried out during the period of November 2013 to February 2014 under greenhouse conditions 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, with geographic coordinates 7°15'18 " S, 35°52'28 " W and mean altitude of 550 m. Treatments consisted of six combinations of water salinity (S1 - Control; S2 - Na⁺, S3 - Ca²⁺, S_4 - Na⁺+ Ca²⁺, S_5 - K⁺ and S_6 - Na⁺+ Ca²⁺ + Mg²⁺), in a randomized block design with four replications each constituted of five plants, totaling 120 experimental units. Plants in the control treatment (S₁) were irrigated with water of electrical conductivity (ECw) of 0.6 dS m^{-1} while in other treatments (S₂, S₃, S₄, S₅ and S₆) were irrigated with ECw = 4.5 dS m^{-1} . For the preparation of water, chloride salt of respective cation was used maintaining equivalent ratio of 1:1 in S₄ $(Na^+: Ca^{2+})$ and 7:2:1 in S₆ $(Na^+: Ca^{2+}: Mg^{2+})$, respectively. In Table 1 the characteristics of water used in S1 (control) treatment are presented. Seeds of castor bean cultivar BRS Energia were used, the plants of genotype present crop cycle of 120-150 days, semiindehiscent fruits, oil content in seeds on average 48% and productivity of approximately 1.800 kg ha⁻¹ (Silva et al., 2009).

Plastic recipients of 100 L capacity (height 50 cm, diameter at bottom 30 cm and superior 33 cm) were used, these were filled with 2 kg of crushed stone (size zero), 54 kg of homogenized soil (without clods) followed by 76 kg of the same soil mixed with humus to attain 1% organic matter, in the total soil volume. The soil used in the study was an Ultisol Eutrophic, collected in a depth of 0 to 30 cm in the district of São José da Mata (Campina Grande-PB). The chemical and physical characteristics of soil (Table 2) were determined according to the methodologies proposed by Claessen (1997). The recipient was adopted as lysimeter having two openings at the bottom to allow drainage of excess water which was collected in plastic bottles allowing estimation of the water consumed by the plant using equation of water balance.

On the basis of soil analysis correction of pH was performed according to Ribeiro et al. (1999) by adding 49.25 g of lime to the soil of each lysimeter (130 kg soil), amount required for neutralization of A^{3+} and raising Ca^{2+} and Mg^{2+} saturation to 70%. After liming the soil presented the following chemical characteristics: Ca^{2+} ; Mg^{2+} ; Na^+ ; K^+ ; H^+ ; Al^{3+} and CEC = 1.14 ,1.36, 0.30, 0.14 , 0.11 , 0 and 3.05 cmol_c kg⁻¹, respectively; Organic matter = 1.08 dag kg⁻¹; P = 47.80 mg kg⁻¹ and pH soil: water (1: 2.5) = 6.42.

Different irrigation waters were prepared using the chloride salt of respective ion using the water of the local supply system. The amount of salt to be added was determined according to the equation recommended by Richards (1954), taking into account the relationship between ECw and the concentration of salt (10*mmol L⁻¹ = 1 dS m⁻¹). The salts used in the preparation of irrigation water had a purity above 99%. Before seeding, volume of water required to reach the field capacity of soil was determined by capillary saturation method followed by free drainage. After the soil was raised to the field capacity by addition of respective water, sowing was performed by placing ten seeds of castor bean cv. BRS Energia in each recipient, at a depth of 5 cm, and distributed equidistantly.Ten days after sowing (DAS) thinning was carried out to maintain one plant per recipient.

The soil was kept at field capacity with daily irrigation, applying volume of respective treatment according to the water consumed by plants, estimated by water balance: volume of water applied minus the volume of water drained in the previous irrigation, plus leaching fraction of 0.10, on the basis of studies conducted previously (Nobre et al., 2013; Lima et al., 2014).

Fertilization was carried out as per recommendations of Novais et al. (1991) using 40.62 g of potassium nitrate and 75 g of monoammonium phosphate, corresponding to 100, 150 and 300 mg kg⁻¹ of N , K₂O and P₂O₅, respectively, as top dressing in four applications at intervals of ten days, beginning at 15 DAS. In order to meet the possible micronutrient deficiencies 5 L of solution containing 2.5 g L⁻¹ ubyfol (N -15, P₂O₅-15, K₂O -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%) was applied by foliar spraying at 30 and 60 DAS.

The phytosanitary treatments were carried out during the assay

Table 1. Chemical characteristics of the supply water used in the control treatment.

Ca ²⁺	Mg ²⁺	Na⁺	a ⁺ K ⁺ HCO ₃		CO3	CI	F our d0 m ⁻¹		CAD (man al 1 -1)0,5	
			(mmol _c L ⁻¹	ECW dS m	рп	SAR (IIIIIIOI L)				
1.19	1.58	8 2.83 0.10 1.45		1.45	0.00	4.22	0.60	7.23	2.41	

ECw-electrical conductivity SAR - sodium adsorption ratio.

Table 2. Chemical and physical characteristics of the soil used in the experiment before application treatment.

Chemical characteristics												
	О.М.	Р	K⁺	Na⁺	Ca ⁺²	Mg ⁺²	AI ⁺³	H⁺				
рн _{ps}	dag kg⁻¹	(mg kg ⁻¹)										
5.10	0.34	20.09	0.07	0.05	0.40	1.30	0.04	1.74				
	Physical characteristics											
Size fraction (g kg ⁻¹)				Water content (kPa)			Total parasity	Density				
Sand	Silt	Clay	class	33.42	1519.5 dag kg ⁻¹		m ³ m ⁻³	(kg dm⁻³)				
856.10	110.7	33.20	SL	6.72	1.62	5.10	0.49	1.54				

 pH_{PS} - pH of saturated paste; O.M- organic matter: determined by wet digestion Walkley-Black method; Ca²⁺ and Mg²⁺ extracted with KCl 1 mol L⁻¹ at pH 7.0; Na⁺ and K⁺ extracted with NH₄ OAc 1 mol L⁻¹ at pH 7.0; SL – sandy loam; AW – available water.

period, consisting of manual weeding at weekly interval, scarification of surface soil before each irrigation event and staking of plants in the flowering stage, in order to prevent lodging. In addition, insecticide of neonicotinoid chemical group, triazole fungicide chemical group and acaricide belonging to abamectin chemical group were applied at a dose of 5.4; 7.0 to 3.5 g L^{-1} , respectively.

The effects of different treatments on castor bean crop were observed at 20 and 40 DAS, by determining the osmotic potential of leaf (ψ s), electrolyte leakage in the membrane (ELM), relative water content (RWC), water saturation deficit (WSD), stomatal conductance (*gs*), transpiration (*E*), CO₂ assimilation rate (*A*), internal CO₂ concentration (CI), instantaneous carboxylation efficiency (EICI) and intrinsic water use efficiency (WUE). To determine the osmotic potential of castor bean, leaves of the plants were collected from middle third part, placed in plastic bags and stored at 5 °C; to extract the cell exudate, samples were placed in tubes for centrifugation at 10000 rpm for 10 minutes; the freezing point of the samples was measured by reading aliquots of 5 mL in osmometer microprocessor (PZL 1000) determining in this way, the osmolality of the sample in H₂O mOsm kg⁻¹ being converted in MPa, as per recommendation of Bagatta et al. (2008), by Equation (1):

$$\psi s (MPa) = -C \left(\frac{mOsmol}{kg}\right) x \ 2.58 \ x 10^{-3}$$
⁽¹⁾

Wherein: ψ s (MPa) - leaf osmotic potential; C - sample osmolality observed in the osmometer.

In order to assess the capacity of rupture of cell membrane under saline stress, leakage of electrolyte was determined in the cell membrane. For the purpose, 3rd leaf of the shoot from apex was collected, 10 discs of 113 mm², were washed with distilled water in order to remove other electrolytes adhered to the leaves, after words discs were placed in beaker with 50 ml of double distilled water and closed tightly with foil. The beakers were kept at 25°C for 90 min and proceeded initial determination of electrical conductivity (C); later the beaker was taken to the oven with forced air

ventilation and submitted to temperature of 80°C for 90 min, when the measurement of the final electrical conductivity (Cf) was performed. Thus, leakage of electrolyte in the cell membrane was obtained according to Scott Campos and Thu Phan Thi (1997), using Equation (2):

$$ELM = \frac{Ci}{Cf} \times 100$$
(2)

Wherein: ELM – electrolyte leakage in the membrane (%); Ci –initial electrical conductivity (dS m^{-1}); Cf- final electrical conductivity (dS m^{-1}).

For assessing the water status of the plant the relative water content (RWC) in the leaf blade was determined; three fully expanded leaves located in the upper third part of the plant were used. The determination of RWC was performed according to the methodology of Weatherley (1950), using Equation (3):

RWC (%) =
$$\frac{(FW - DM)}{(TM - DM)} \times 100$$
 (3)

Wherein: RWC- relative water content (%); FW- fresh weight of leaf (g); TM-turgid mass (g); DM- dry mass (g);

The water saturation deficit (WSD) is an excellent indicator of the water balance of the plant as it represents the amount of water it needs to reach saturation. In this regard, WSD was determined according to the methodology described by Taiz and Zeiger (2013) according to Equation (4):

WSD (%) =
$$\frac{(TM - FW)}{(TM - DM)} \times 100$$
(4)

Wherein: WSD - water saturation deficit (%); FW - fresh weight of leaf (g); TM - turgid mass (g); DM - dry mass (g).

Stomatal conductance (gs) (mol of H₂O m⁻² s⁻¹), transpiration (*E*) (mmol of H₂O m⁻² s⁻¹), the CO₂ assimilation rate (*A*) (μ mol m⁻² s⁻¹)

Table 3. Summary of analysis of variance and estimated means of different contrasts (\hat{y}) related to the osmotic potential of leaf (ψ s), electrolyte leakage in the membrane (ELM), relative water content (RWC) and water saturation deficit (WSD) of castor bean cv. BRS Energia irrigated with water of different types of salinity, at 20 and 40 days after sowing (DAS).

	F test										
01/	ų	IS	EL	.M ¹	R	NC	WSD ²				
SV	Days after sowing										
	20	40	20	40	20	40	20	40			
Block	ns	ns	ns	*	ns	ns	ns	ns			
Types of Salinity	*	*	**	*	**	**	**	*			
Ŷ1	*	*	*	*	*	*	*	*			
ŷ ₂	ns	ns	*	*	**	ns	**	ns			
ŷ ₃	ns	ns	ns	ns	**	ns	**	ns			
Ŷ4	ns	7ns	ns	ns	**	*	**	*			
ŷ ₅	ns	ns	*	*	*	*	**	*			
CV	3.46	6.18	12.42	11.33	6.46	11.48	12.91	24.53			
			Mean estimate								
Ŷ1	-0.15	-0.23	-5.00	-1.71	11.46	20.01	-11.46	20.01			
Ŷ2	ns	ns	5.96	3.01	-30.14	ns	30.14	-8.25			
ŷз	ns	ns	ns	ns	-27.36	ns	27.36	-10.45			
ŷ 4	ns	ns	ns	ns	-30.51	-29.65	-30.51	-29.65			
Ŷ5	ns	ns	5.70	1.62	9.69	23.77	-9.96	23.77			

 \hat{y}_1 (S₁ vs S₂, S₃, S₄, S₅, S₆); \hat{y}_2 (S₂ vs S₃); \hat{y}_3 (S₂ vs S₆); \hat{y}_4 (S₂ vs S₅); \hat{y}_5 (S₅ vs S₂, S₃, S₄, S₆); VS – variation source; CV – Coefficient of variation; (*) and (**) Significant at 0.05 and 0.01 probability; (ns) not significant; ¹statistical analysis after data transformation in \sqrt{X} ; ²statistical analysis after data transformation in $\sqrt{X + 1}$

and the internal CO₂ concentration (μ mol m⁻² s⁻¹) (CI) were also evaluated in the third leaf from the apex, using portable photosynthesis measurement "LCPro +" equipment from ADC BioScientific Ltda. These data were used to quantified the intrinsic water use efficiency (WUE) (A/E) [(μ mol m⁻² s⁻¹) (mol H₂O m⁻² s⁻¹)⁻¹] and the instantaneous efficiency of carboxylation (A/Ci) [(μ mol m⁻² s⁻¹) (μ mol mol⁻¹]⁻¹(Jaimez et al., 2005).

The data were evaluated by analysis of variance following 'F' test; when significant, mean comparison test (Tukey at 0.05 probability) and the contrasts between the treatment means were performed using the SISVAR-ESAL statistical software. For each treatment apart from mean, standard error was also calculated. The contrasts were defined as follows: \hat{y}_1 (S₁ vs S₂, S₃, S₄, S₅, S₆); \hat{y}_2 (S₂ vs S₃); \hat{y}_3 (S₂ vs S₆); \hat{y}_4 (S₂ vs S₅); \hat{y}_5 (S₅ vs S₂, S₃, S₄, S₅). Given the normality of residues from the present study observed through the high coefficient of variation values (CV> 20%) (Tables 3 to 5), it was necessary to perform exploratory data analysis with transformation of data in \sqrt{X} and $\sqrt{X+1}$.

RESULTS AND DISCUSSION

Analysis of variance (Table 3) indicated significant effect of salinity on osmotic potential of leaf, electrolyte leakage, relative water content and water saturation deficit of castor bean at 20 and 40 DAS. Comparison of means for the osmotic potential of leaves of castor bean at 20 DAS (Figure 1A), indicated that plants irrigated with water of low electrical condutvity (S₁) reached the highest ψ s and differed significantly (p<0.05) from those receiving S₂, S₄, S₅, S₆ and comparing the means of these treatments, it is observed that the plants irrigated with water having sodium (S_2, S_4, S_6) and potassium (S_5) ion in its composition, had the lowest values in the energy status of the water in the leaves.

At 40 DAS significant effect (p<0.05) was observed in leaf osmotic potential (Figure 1B) in plants irrigated with water of lowest salinity (S₁) as compared to the S₂, S₄ and S₅ treatments, however, there were no significant differences compared to plants irrigated with the S₃ and S₆ treatments. According to comparison of means, the lowest values for the leaf osmotic potential occurred in plants S₂ followed by plants of treatments S₅ and S₄.

Comparing the data of 20 DAS with that of 40 DAS, a greater reduction in osmotic potential of leaf in the latter period is observed. This response suggests that the decrease in ψ s is a function of exposure time to salt stress, in order to maintain a high water potential in tissues. This decrease in leaf osmotic potential can be considered an adaptive strategy of the species in relation to increased salt concentration in the soil solution enabling the hydration of plant tissues and slowing the damaging processes caused by water deficit (Santos et al., 2012; Coelho et al., 2014).

In assessing the contrasts of the means for the osmotic potential of leaf at 20 and 40 DAS (Table 3) it is noticed that the plants irrigated with ECw = 0.6 dS m⁻¹ (S₁) differed significantly in relation to those irrigated with ECw of 4.5 dS m⁻¹ (S₂, S₃, S₄, S₅, S₆). In both periods the

	F test											
01/	gs ¹		E		А		CI		EICI		WUE	
5V	Days after sowing											
	20	40	20	40	20	40	20	40	20 ¹	40	20	40
Block	ns	*	*	*	ns	*	**	**	ns	*	*	**
Types of Salinity	ns	*	ns	ns	**	**	**	ns	**	*	*	*
Ŷ1	ns	*	ns	*	*	**	*	ns	*	*	*	*
ŷ ₂	ns	ns	ns	*	*	*	ns	ns	*	*	ns	*
ŷ ₃	ns	ns	ns	ns	ns	*	*	ns	*	*	ns	*
ŷ 4	ns	ns	*	*	*	**	*	ns	*	*	ns	*
ŷ 5	ns	ns	*	ns	*	*	*	ns	*	*	ns	*
CV	21.55	17.37	19.51	13.83	13.81	17.95	6.91	5.28	17.76	10.56	18.91	16.52
						Mean es	stimate					
ŷ ₁	ns	0.84	ns	0.90	8.61	6.55	-30.00	ns	0.07	0.02	0.84	0.77
ŷ ₂	ns	ns	ns	-0.66	-7.17	-5.71	ns	ns	-0.04	-0.02	ns	-1.44
ŷ ₃	ns	ns	ns	ns	ns	4.79	-17.00	ns	-0.02	-0.02	ns	-1.27
ŷ 4	ns	ns	-2.80	-0.55	-9.06	-8.64	24.00	ns	-0.06	-0.03	ns	-2.40
ŷ 5	ns	ns	2.41	ns	6.45	5.51	-22.00	ns	0.05	0.02	ns	1.62

Table 4. Summary of analysis of variance and estimated means related to the stomatal conductance (*gs*), transpiration (*E*), assimilation rate of CO₂ (*A*), internal concentration of CO₂ (CI), instantaneous efficiency of carboxylation (EICI) and intrinsic water use efficiency (WUE) of castor bean plants irrigated with water of different types of salinity at 20 and 40 days after sowing.

 \hat{y}_1 (S₁ vs S₂, S₃, S₄, S₅, S₆); \hat{y}_2 (S₂vs S₃); \hat{y}_3 (S₂vs S₆); \hat{y}_4 (S₂vs S₅); \hat{y}_5 (S₅vs S₂, S₃, S₄, S₆); VS – variation source; CV – Coefficient of variation; (*) and (**) Significant at 0.05 and 0.01 probability; (ns) not significant; ¹statistical analysis after the transformation of data in \sqrt{X}

data indicated that the plants when irrigated with water of 0.6 dS m⁻¹, increased their Ψ s by -0.15 and -0.23 MPa in reference to plants irrigated with ECw of 4.5 dS m⁻¹, respectively, at 20 and 40 DAS. However, analysing the other contrasts there were no significant changes in the Ψ s at any time of evaluation. In this context, it can be inferred that the different cations present in the irrigation water affected Ψ s in a similar way.

For the electrolyte leakage measured at 20 DAS it is observed through the comparison of means (Figure 1C) that plants under irrigation with water of low salinity (S_1) and predominant in calcium (S_3) had the smallest leakage in membrane (10.52 and 11.34%, respectively) showing minor damage

to the integrity of the leaf membrane. This result is justified by the role of calcium in the integrity of cell membrane, being a constituent of the cell wall (pectates), participating in the regulation of functionality of the cell membrane, and activation of several enzyme systems (Mengel and Kirkby, 2000). But when the plants were irrigated with S₅ treatment (potassium), largest electrolyte leakage (20.51%) was observed, though statistically not different from plants that were submitted to S₂ (Na), S₄ (Na+Ca) and S₆ (Na+Ca+Mg), indicating occurrence of injuries to cell membrane. Increasing the concentration of electrolytes in leaf cells may represent a mechanism to prevent desiccation of the tissue due to the reduction of the osmotic component of leaf water potential (Fioreze et al., 2013).

Similar to that observed at 20 DAS, electrolyte leakage obtained at 40 DAS (Figure 1D) differed significantly due to the application of different types of salts, the largest highlighting values (12.84 and 12.81%) were observed in plants irrigated with water prepared using NaCl (S_2) and KCl (S_5). It is noted when plants were irrigated with water having calcium in its composition (S_3) or with low concentration of salt (S_1), the lowest values were obtained for electrolyte leakage. It appears from the results obtained for ELM, the plants irrigated with water consisting of potassium, showed the greatest permeability of the



Figure 1. Osmotic potential of leaf - ψ s of (A and B), and leakage of electrolytes in the leaves (C and D) of castor bean at 20 and 40 days after sowing, depending on the types of irrigation water salinity.

membrane with consequent release of ions, that is, greater loss of integrity and destabilization of the cell membrane. The rupture of cell membrane occurs through the release of electrolytes in higher levels of stress due to increased amounts of reactive oxygen species (superoxide) free radical and lysing enzymes, that result in the disruption and increased permeability of membranes and often irreversible damage to the organelles and molecules present within the cells (Alonso et al., 1997).

The contrasts of means for the leaf electrolyte leakage at 20 and 40 DAS, are presented in Table 3, comparing the data of plants irrigated with ECw of 0.6 dS m⁻¹ in relation to those under ECw = 4.5 dS m⁻¹, significant effect was found at both stages. On the basis of estimated means (Table 3) a decrease in electrolyte leakage of 5.0 and 1.17% was observed in plants irrigated with low salinity water (S₁) compared to plants subjected to ECw of 4.5 dS m⁻¹, respectively, at 20 and 40 DAS.

The higher ELM observed in plants under the high level of electrical conductivity, is the result of stress to which the plants were exposed because of high salt concentrations found in irrigation water which caused changes in water status of the plant due to the decrease in osmotic potential of leaf (Figure 1A and B). In a manner similar to that observed for S₁ versus other treatments, S₃ vs. S₂ also presented significant

differences observing increase in leakage of electrolyte of 3.01 to 5.96% at 20 and 40 DAS, respectively; however S_2 , when confronted with S_6 and S_5 did not influence significantly the ELM in any study period (Table 3).

This response may be related to the decrease in Ψ s (Figure 1A and B) due to the excess salts present in irrigation water because the effect of cationic nature of water occurred in similar way. While the S₅ versus S₂, S₃, S₄ and S₆ showed significant effect on the ELM indicating, through the mean estimate (Table 3) that irrigation water with potassium increased by 5.70 and 1.62% ELM compared to the other treatments (S₂, S₃, S₄ and S_6) at 20 and 40 DAS, respectively. The increase in leakage of leaf electrolytes, especially in plants irrigated with potassic water (S_5) , may have occurred because of this element does not participate in the composition of any organic compound (structural function) in plant metabolism (Rodrigues et al., 2009), therefore is easily released and/or redistributed to the different organs of the plant.

Analysing the relative water content (RWC) of leaf obtained at 20 DAS (Figure 2A), it is observed that the plants irrigated with sodic water (S_2) showed lower RWC (42%), differing from the other treatments. On the other hand, plants subjected to irrigation treatment under S_1 , S_3 , S_4 S_5 and S_6 were statistically similar. The reduction in RWC of leaf observed in the plants irrigated with sodium water, can be related to an increase in damage caused to



Figure 2. Relative water content - RWC (A and B), and water saturation deficit -WSD (C and D) of leaf of castor bean at 20 and 40 days after sowing, depending on the types of irrigation water salinity.

cell membranes (Figure 1C and D), because the increase in the leaching of ions affect the cell water potential and therefore promoted loss of cellular turgor (Mansour and Barbosa, 2000). This restriction in the leaf water status, observed in the present study by reduction in RWC affects the absorption of nutrients by the roots and therefore growth and development of plants are seriously impaired.

Comparison by mean test at 40 DAS (Figure 2B) showed that the castor bean plants irrigated with low salinity water (S_1) and water salinized by potassium (S_5) had a significantly higher RWC, averaging respectively 98.23 and 97.24%, whereas plants irrigated with other treatments (S_2 , S_3 ; S_4 and S_6) had respectively 67.58, 75.85, 72.36 and 78.04%. It should be underscored that the effects of different types of salinity were more evident in plants irrigated with water treatments S_2 and S_3 (Figure 2B). When results are compared, a decrease of 30.50, 21.99, 25.58 and 19.74% occurs in plants irrigated with treatments S_2 , S_3 ; S_4 and S_6 , when contrasted to those irrigated with potassium water.

As mentioned earlier, decrease in the RWC of leaf in plants under saline stress induced by different types of salt (S_2 , S_3 , S_4 and S_6) is an adaptive mechanism developed by the plants, even though the absorption of water from the soil occurs in low proportions (Garcia et al., 2009). Further, water deficiency, caused by osmotic effect, characterizes physiological drought and brings about morphological and anatomical changes in the plants with an imbalance in water absorption and in transpiration rates (Santana et al., 2011).

Contrasts of means for RWC presented in Table 3 show that plants irrigated with low ECw (0.6 dS m⁻¹) have significant differences compared to treatment with water at 4.5 dS m⁻¹ and estimated means at 20 and 40 DAS indicated an increase of 11.46 and 20.01%, when plants were irrigated with water of ECw=0.6 dS m⁻¹. In the case of treatment S₂ versus S₃ and S₂ versus S₆, the differences were significant at 20 DAS and during this period, RWC decreased respectively by 30.14 and 27.36% in plants irrigated with treatment S₂.

However, when S_2 versus S_5 is analysed, there is significant influence on RWC in both the periods under analysis, the decrease in treatment S_2 being 30.51 and 29.65%, respectively at 20 and 40 DAS. Table 3 shows that the RWC of plants irrigated with S_5 was affected significantly when compared to treatments S_2 , S_3 , S_4 and S_6 and estimate of mean showed an increase of 9.69 and 23.77%, respectively, at 20 and 40 DAS in plants irrigated with S_5 . Results in current study demonstrate a greater decrease in RWC of leaf with the cationic traits of water particularly the plants irrigated with sodic water. Water saturation deficit (WSD) of castor bean plants assessed at 20 DAS was affected significantly (p<0.05) by irrigation water of different types of salinity.

Comparison by mean test (Figure 2C) showed that WSD of plants irrigated with water salinized by sodium (S_2) was significantly higher than other treatments (S_1, S_3, S_2)

 S_4 , S_5 and S_6). Comparison of results in Figure 2A and C reveal inverse relation between RWC and WSD. Increase in WSD may be due to the high osmotic level of the soil solution which affects the availability of water for the plants. Its absorption becomes more difficult, with the occurrence of low turgor pressure and elongation of cells (Jácome et al., 2003).

The behavior of the results of water saturation deficit of the castor bean plant at 40 DAS is different from that at 20 DAS. When means of treatments are compared (Figure 2D), the plants irrigated with low ECw (S₁) and with potassium-induced salinity (S₅) showed the lowest means, differing statistically from those irrigated by treatments S₂, S₃ and S₄. However, there was no significant difference with regard to plants irrigated with S₆. Means of treatments at 40 DAS revealed a smaller WSD in all the treatments (Figure 2D) compared to 20 DAS (Figure 2C) possibly because of osmotic adjustment in the castor bean plants.

Data of estimates of means (Table 3) demonstrate that there was a reduction of 11.46 and 20.01% in WSD in plants under control treatment compared to those under $ECw=4.5 \text{ dS m}^{-1}$ at 20 and 40 DAS respectively. Lowest WSD in plants at the low water salinity (0.6 dS m⁻¹) may be explained by the high foliar osmotic potential (Figures 1A and B), due to decrease in electrolyte leakage (Figure 1C and D) and higher RWC in leaves (Figures 2A and B) in the two periods under analysis.

Contrast between treatments S_2 versus S_3 (Table 3) demonstrated that, when sodic water was used for irrigation, the plants had 30% higher WSD compared to S_3 (calcium) while at 40 DAS, plants in S_3 had 8.25% increase in WSD when compared to those with S_2 . When treatment S_2 is compared to S_6 at 20 DAS, one may note that the plants irrigated with former had a 27.36% increase in WSD compared to those irrigated with S_6 , however, at 40 DAS, the plants suffered 10.45% WSD when compared to those with water salinity induced by Na+Ca+Mg.

In the case of treatments S_2 versus S_5 , the plants irrigated with sodic water showed an increase of 30.51 and 29.65% in WSD, respectively at 20 and 40 DAS. On the other hand, there was a 9.96% decrease in plants irrigated with S_5 at 20 DAS when compared to treatments (S_2 , S_3 , S_4 and S_6). A different trend was verified at 40 DAS observed by estimated means (Table 3), namely, that the use of water with potassium caused a 23.77% decrease in WSD in relation to treatments S_2 , S_3 , S_4 and S_6 .

Based on the summary of F test (Table 4), there was a significant effect of the different types of water salinity on the assimilation rate of CO_2 , instantaneous efficiency of carboxylation and intrinsic efficiency in the use of water, at 20 and 40 DAS. However, internal concentration of CO_2 and stomatal conductance were affected significantly only at 20 and 40 DAS respectively. The aforementioned table also shows that transpiration rate was not affected

at any time during the study period.

At 20 DAS gs was not affected due to different types of salt, whereas at 40 DAS irrigation with saline water caused significant changes in stomatal conductance (Figure 3A and B). Comparison of means for gs at 40 DAS (Figure 3B) demonstrates that plants irrigated with low salinity water (control) showed gs statistically higher than those of treatments S_2 , S_3 , S_4 and S_6 which did not differ statistically neither among themselves nor compared to S₅. The decrease of stomatal conductance of the plants irrigated with water of the treatments S_2 , S_3 , S_4 and S_6 respectively with regard to S_1 , is related to the change in osmotic potential and, consequently, to the reduction in water availability in their tissues. Stomatal closure probably occurred, with the subsequent decrease of the normal flux of CO_2 towards the carboxylation site. The latter brought about changes between the appropriate equilibrium and the transport of electrons, carbon metabolism and ATP and NADPH consumption which together made inefficient photosystem II and compromised significantly the production of photosynthates (Tezara et al., 2005).

Table 4 shows the results of the contrasts of means with regard to stomatal conductance measured at 20 and 40 DAS. Significant effect for gs occurred only at 40 DAS. It should be emphasized that in castor bean plants irrigated with low salinity water (ECw=0.6 dS m⁻¹) gs increased by 0.84 mol of $H_2O \text{ m}^{-2} \text{ s}^{-1}$ when compared to those irrigated with water at ECw=4.5 dS m⁻¹ (S₂, S₃, S₄, S_5 and S_6). Therefore, the most negative result on gs is the result of the cationic nature of the irrigation water due to excess of salt in the soil solution which causes changes in the osmotic-water potential directly affecting water absorption by the plants. On the other hand, the lack of significant effect (p>0.05) at 20 DAS for all treatments under analysis suggests that stomatal conductance under saline conditions varies according to the development of the plant and depends on the duration of exposure to stress and the age of the plant (Freitas et al., 2013).

Transpiration (E) had partially the same behavior as stomatal conductance at 20 DAS (Figure 3C). No significant effect was observed for the different types of salinity in irrigation water and with regard to plants irrigated with low salinity water (control). In other words, irrigation water with Na⁺, Ca²⁺, Na⁺+Ca²⁺, K⁺, Na⁺+Ca²⁺+Mg²⁺ in the initial stages of the development of the castor bean plant did not affect the plant's physiology with regard to transpiration since as the stomatal conductance was not affected (Figure 3A) but there were significant differences (p<0.05) in measures at 40 DAS which suggest that the effect of the different salts (Na⁺, Ca^{2+} , Na^++Ca^{2+} , K^+ , $Na^++Ca^{2+}+Mg^{2+}$) on E intensified as stress time was prolonged. These results demonstrate that stress effect caused by different types of salinity in castor bean plants may differ during the crop cycle.

The partial closing of the stomata caused by different



1= Control, 2= Na⁺, 3= Ca²⁺, 4=Na⁺+Ca²⁺, 5=K⁺, 6=Na⁺+Ca²⁺+Mg²⁺

Figure 3. Stomatal conductance -gs (A and B), and transpiration -E (C and D) of the castor bean plant at 20 and 40 days after sowing, depending on the types of irrigation water salinity.

types of salinity in the water at 40 DAS (Figure 3B) also influenced the rates of transpiration of the castor bean during the same evaluation period. When the plant means of the treatments were compared, it may be seen that castor bean plants irrigated with lower ECw (S1) had a higher E (3.85 mmol H₂O m⁻² s⁻¹), significantly differing (p<0.05) only from those irrigated with treatment S₂. It may also be observed (Figure 3D) that castor bean plants irrigated with water of the treatments S2, S3, S4, S5 and S_6 , had a 34.37,17.15, 21.41, 20.06 and 24.33% reduction in transpiration when compared with plants irrigated with low salinity water. When different treatments were evaluated, there was a greater reduction in the E of plants irrigated with water containing only sodium (Figure 3D) in its composition. This is due to decrease in osmotic potential (Figure 1B), and the increase in the water saturation deficit (Figure 2D) and to the decrease in stomatal conductance (Figure 3B) at 40 DAS. Moreover, decrease in transpiration may also have been caused by the toxic effects of the salts absorbed by the plants, by the low capacity of osmotic adjustment of the crops and/or reduction of total potencial of water caused by the increase of saline concentration (Silva et al., 2011).

Contrasts of transpiration means revealed that there was a significant alteration in E at 20 DAS when S₂

versus S_5 and S_5 versus the other salts (S_2 , S_3 , S_4 and S_6) were compared (Table 4). Estimate of mean (Table 4) shows that E decreased by 2.80 mmol of H₂O m⁻² s⁻¹ with regard to treatment S_5 when plants were irrigated with S_2 water containing sodium. When mean obtained in S₅ is contrasted by comparing plants irrigated with water containing other types of salt, a reduction of 2.41 mmol of $H_2O m^{-2} s^{-1}$ occured. It has been verified that, at 40 DAS (Table 4), when the plants were irrigated with water of ECw=0.6 dS m⁻¹, there was an increase of 0.90 mmol of $H_2O \text{ m}^{-2}\text{s}^{-1}$ in *E* compared to plants irrigated with ECw = 4.5 dS m⁻¹. According to data of mean estimates (Table 4), there was a reduction of 0.66 and 0.55 mmol of H₂O m^{-2} s⁻¹ in plants irrigated with treatment S₂ when compared to those irrigated with S_3 and S_5 , respectively. As a general rule, the above-mentioned results showed that water containing sodium ion affects transpiration regardless of its proportion in irrigation water.

Assimilation rate of CO_2 was significantly affected (p<0.05) by different types of water salinity at 20 DAS, whilst comparison of means (Figure 4A) showed that CO_2 assimilation rate of plants irrigated with S₁ water of low salinity and with potassium (S₅) was significantly different (p<0.05) when compared to plants irrigated with treatments S₂ and S₆. However, no significant difference (p.>0.05) was found when *A* of S₁ and S₅ were compared



Figure 4. CO_2 assimilation rate -A (A and B), and internal concentration of CO_2 -CI (C and D) of the castor bean plant at 20 and 40 days after sowing, depending on the types of irrigation water salinity.

with treatments S₃ and S₄. It may also be perceived (Figure 4A) that castor bean plants at 20 DAS irrigated with water of treatments S₁ and S₅ had the higher CO₂ assimilation rates (35.56 and 32.12 μ mol m⁻² s⁻¹) compared to other treatments (S₂, S₃, S₄ and S₆), following the same trend observed for stomatal conductance. These rates are satisfactory since the castor bean is a C_3 plant in which the CO_2 assimilation rates varies between 10 and 20 µmol m⁻² s⁻¹ (Taiz and Zeiger, 2013). On the other hand, decrease in CO2 assimilation rate especially in plants irrigated with sodic water may have been caused by the reduction of stomatal conductance (Figure 3A), transpiration (Figure 3C) and increase of CO₂ internal concentration (Figure 4C). This reveals low efficiency in the use of CO₂ introduced into the cell.

In the case of CO₂ assimilation rate at 40 DAS (Figure 4B), comparison of mean test showed that the highest rate (17.02 µmol m⁻² s⁻¹) occurred in plants irrigated with low ECw (S₁), which was statistically higher than the rates observed in those irrigated with treatments S₂, S₃; S₄ and S₆. However, there was no significant difference in the *A* of plants under treatment S₅ compared to S₁.There was 63.45, 29.84, 51.52 and 35.25% decrease in *A* when means of treatments S₂, S₃, S₄ and S₆ were compared to

control. Data demonstrate (Figures 4A and B) that decrease in CO_2 assimilation rate is associated with age and imposition of stress period in the crop. In fact, plants irrigated with sodium water were greatly affected in the two periods under analysis.

The observed behavior of *A* corroborates with data on stomatal conductance (Figure 3B) and transpiration (Figure 3D) and suggests that the decrease in the photosynthesis rate may have been caused by the partial closure of the stomata, associated to the osmotic effects of salinity and ionic toxicity on the metabolism (Bezerra et al., 2005). Further, decrease in CO_2 assimilation rate, caused by the closure of the stomata as a response to the low potential of soil water due to high concentration, is also a determining factor in growth and productivity of crops (Gurgel et al., 2003).

Table 4 shows the contrasts of means for CO_2 assimilation rate. There was no significant difference (p>0.05) for S₂ versus S₆ at 20 DAS; estimates of means (Table 4) revealed that plants irrigated with low water conductivity (0.6 dS m⁻¹) had an increase in *A* of 8.61 and 6,55 µmol m⁻² s⁻¹ at 20 and 40 DAS, respectively, when compared to those irrigated with ECw of 4.5 dS m⁻¹(S₂, S₃, S₄, S₅ and S₆). On the other hand, a decrease of 7.17 and 5.71 µmol m⁻² s⁻¹ respectively at 20 and 40 DAS in

the CO_2 assimilation rate occurred when the variable was analysed in plants irrigated with sodium water (S₂) and in those irrigated with calcium (S₃).

When the effect of treatment S_2 is compared to that of S_6 (Table 4), an increase of 4.79 µmol m⁻² s⁻¹ occurred in CO₂ assimilation rate at 40 DAS while it decreased 9.06 and 8.64 $\mu mol~m^{\text{-2}}~s^{\text{-1}},$ respectively at 20 and 40 DAS, in comparison to plants irrigated with potassic (S₅) water. When the behavior of A between S_5 and the other saline waters (S₂, S₃, S₄ and S₆) is evaluated, the estimate of mean (Table 4) reveals that CO₂ assimilation rate, at 20 and 40 DAS, in S₅ (potassic water) increased by 6.45 and 5.51 µmol m⁻² s⁻¹, respectively, with regard to those irrigated with the other cations. It may thus be inferred that the reduction of CO₂ assimilation rate was caused by the decrease in RWC, transpiration and stomatal conductance. The most evident effect in both the periods was observed due to variation in water salinity (ECw from 0.6 to 4.5 dS m⁻¹).

In the case of internal concentration of CO₂ at 20 DAS (Figure 4C), plants irrigated with water S_6 (Na⁺+Ca²⁺+Mg²⁺) had the highest CI (214.90 µmol m⁻² s⁻¹) and exceeded by 25.63, 14.02, 14.20 and 19.61% (p<0.05) of concentration obtained in plants irrigated with treatments S_1 , S_3 , S_4 and S_5 , respectively. Low internal concentration of CO₂ observed in plants under S₁ and S₅ treatments explains the highest values found for the CO₂ assimilation rate (Figure 4A) in these treatments. It underscored that a higher should be internal concentration of CO_2 (S₂ S₃, S₄ and S₆) means that the carbon introduced into the leaf mesophyll cell was not being metabolized by the photosynthesis apparatus due to saline stress to which the plants were exposed. Further, increase in the internal concentration of CO₂ indicated that there was no restriction in the acquisition of CO₂ by the plant. However, when it reached the mesophyll cells, the fixation process during the carboxylation phase was compromised, this may have been caused by the degradation of the photosynthesis apparatus as a response to the process of leaf senescence of the tissues due to stress from excess of salts (Silva et al., 2013).

The internal concentration of CO_2 at 40 DAS was not affected significantly (p>0.05) by the treatments under analysis (Figure 4D). Similarity among treatments suggests that, regardless of the type of salinity, the availability of CO_2 to plants was illimited and shows that the decrease in the photosynthesis process was not only due to restricted stomatal opening but also due to damages to the cell structure responsible for the assimilation of CO_2 . The latter may be due to the reduction of the osmotic potential and by the accumulation of ions beyond the tolerance limit of the castor bean plants (Fernandes et al., 2010).

Table 4 presents contrasts of means for the internal concentration of CO_2 . There was a significant (p<0.05) effect of treatments at 20 DAS. Estimates of means in

Table 4 showed that plants irrigated with water of ECw= 0.6 dS m⁻¹ reduced internal concentration of CO₂ by 30 µmol m⁻² s⁻¹ in comparison to those of ECw=4.5 dS m⁻¹. Increase in CI in plants under ECw=4.5 dS m⁻¹ level was probably due to decrease in stomatal conductance, transpiration and CO₂ assimilation rate, perhaps because of damages in the photosynthesis apparatus due to high internal concentration of CO₂. On the other hand, since the internal concentration of CO₂ of plants irrigated with S₂ did not differ significantly from those under S₃ water, it indicated that salinity caused by sodium and calcium had a similar effect on CI.

Different from the above-mentioned results, the estimate of means (Table 4) at 20 DAS shows that water salinized by sodium (S₂) resulted in a decrease of 17 μ mol m⁻² s⁻¹ in Cl when compared to plants irrigated with S₆ (Na+Ca+Mg). Analysis of the data S₂ versus S₅ registered that castor bean plants irrigated with sodium water (S₂) had an increase of 24 μ mol m⁻² s⁻¹ in the Cl of CO₂ when compared to treatment S₅. Likewise, the use of irrigation water with potassium (S₅) caused a decrease of 22 μ mol m⁻² s⁻¹ in Cl when contrasted to plants with the other types of salt (S₂, S₃, S₄ and S₆).

Since instantaneous efficiency of carboxylation is a physiological parameter with close links to the intracellular concentration of CO2 and to the assimilation rate of CO₂ (Konrad et al., 2005), data showed a significant effect on EICI of the different types of water salinity at 20 DAS (Figure 5A). Comparison of means reveal that plants under treatments S_1 and S_5 had the highest EICI [(0.22 and 0.18 (µmol m⁻² s⁻¹) (µmol mol⁻¹)⁻¹] and did not differ statistically from rates of plants under treatments S₃ and S₄ [0.16 and 0.16 (μ mol m⁻² s⁻¹) (μ mol mol⁻¹)⁻¹]. On the other hand, rate of EICI of castor bean plants irrigated with S1 and S5 differed significantly from rates with treatments S_2 and S_6 . The lowest EICI rate [0.09 (µmol m⁻² s⁻¹) (µmol mol⁻¹)⁻¹] occurred in plants irrigated with S₆ water containing sodium+calcium+ magnesium.

Consequently, increases in the instantaneous efficiency of carboxylation in current analysis, especially in plants with treatments S_1 and S_5 are mainly due to increases in the assimilation rate of CO₂ (Figure 4A) and to decreases in the internal concentration of carbon dioxide (Figure 4C). Further, reduction in EICI rates in treatments S_2 and S_6 may be associated with stomatal and non-stomatal causes due to the osmotic and toxic effects, caused by the accumulation of Na⁺ and Cl⁻ ions in the foliar limbos (Neves, 2008).

The instantaneous efficiency of carboxylation was also significantly affected at 40 DAS by the different types of water salinity. Results of mean test (Figure 5B) showed that the highest rates [(0.06 (μ mol m⁻² s⁻¹) (μ mol mol⁻¹)⁻¹)], with regard to EICI, occurred in water with low salinity (S₁) and potassium (S₅) which differed significantly from treatments S₂ and S₄. However, there were no differences between treatments S₃ and S₆. Above results suggest



Figure 5. Instantaneous efficiency of carboxylation – EICI (A and B), and intrinsic water use efficiency-WUE (C and D) of the castor bean plant at 20 and 40 days after sowing, depending on the types of irrigation water salinity.

that decrease, which was greater in EICI of plants with treatments S_2 and S_4 , is a consequence of the low CO_2 assimilation rates (Figure 4B) with regard to CO_2 in the sub-stomatal chamber (Figure 4D). If the internal concentration of CO_2 increases and there is decrease in the consumption of CO_2 in the chloroplasts due to the reduction in photosynthesis activity, EICI will also decrease.

Based on results of contrast of means for EICI (Table 4), a significant effect occurred among all the treatments evaluated at 20 and 40 DAS. Estimate of means showed that castor bean plants irrigated with water at the low electrical conductivity (0.6 dS m⁻¹) increased EICI by 0.07 and 0.02 [(μ mol m⁻² s⁻¹) (μ mol mol⁻¹)⁻¹] respectively at 20 and 40 DAS when compared to those with ECw of 4.5 dS m⁻¹ (S₂, S₃, S₄, S₅ and S₆). Plants irrigated with water salinized with sodium (S₂) EICI decreased by 0.04 and 0.02 [(μ mol m⁻² s⁻¹) (μ mol mol⁻¹)⁻¹] in comparison to plants irrigated with treatment S₃ at 20 and 40 DAS, respectively.

Data from estimate of mean (Table 4) revealed that the use of water rich in sodium decreased EICI by 0.02 and 0.02 [(μ mol m⁻² s⁻¹) (μ mol mol⁻¹)⁻¹] when compared to plants irrigated with treatment S₆ in the two periods analysed (20 and 40 DAS). However, EICI decreases by

0.06 and 0.03 [(μ mol m⁻² s⁻¹) (μ mol mol⁻¹)⁻¹] in plants irrigated with S₂ with regard to mean of plants irrigated with S₅ at 20 and 40 DAS, respectively. On the other hand, EICI increased by 0.05 and 0.02 [(μ mol m⁻² s⁻¹) (μ mol mol⁻¹)⁻¹] when treatment S₅ was compared to the other types of salts in irrigation water (S₂; S₃; S₄ and S₆). As a rule, decrease in EICI, especially in plants irrigated with different types of cations (S₂; S₃; S₄ and S₆) is associated with the reduction of CO₂ assimilation rate and to the increase in the internal concentration of CO₂.

The intrinsic water use efficiency at 20 DAS (Figure 5C) did not show significant effect of the treatments under study. However, plants irrigated with S_1 had the highest WUE rate 4.03 [(µmol m⁻² s⁻¹) (mol H₂O m⁻² s⁻¹)⁻¹]. The absence of a significant influence of the different types of salinity on WUE is due to the results of the assimilation rate of CO₂ (Figure 4A) and transpiration (Figure 3A) during the same period of study. Since intrinsic water use efficiency is obtained by the relationship between CO₂ assimilation rate and leaf transpiration, in which rates are related to the amount of carbon fixed by the plant for each unit of water lost (Jaimez et al., 2005), probably the plants which are capable of maintaining a high efficiency in the use of water under saline conditions, have a great tolerance to saline stress. In fact, decrease in water

consumption implies a reduction in the absorption of specific ions thereby avoiding toxicity to the plants (Flowers and Flowers, 2005).

Based on the results of the test of comparison of mean for the intrinsic water use efficiency at 40 DAS (Figure 5D), treatments S_1 and S_5 were statistically superior than S_2 and S_4 , although they did not differ from plants irrigated with treatments S₃ and S₆. Figure 5D shows that the highest rates of intrinsic WUE [4.43and 4.96 (µmol m 2 s⁻¹) (mol H₂O m⁻² s⁻¹)⁻¹] were obtained in plants irrigated by water with the low salinity (S_1) and containing potassium (S_5) , respectively. On the other hand, irrigation with sodic water (S_2) , sodium+calcium (S_4) and sodium+calcium+magnesium (S₆) provided lower WUE rates, respectively with mean values of 2.56, 2.56 and 3.84 [(μ mol m⁻² s⁻¹) (mol H₂O m⁻² s⁻¹)¹]. Decrease in intrinsic water use efficiency in treatments S₂, S₄ and S₆ may be associated to changes in the rates of assimilation of CO₂ and transpiration which possibly occurred because of the low availability of water in the soil, caused by the reduction of osmotic potential due to high concentration of salts. The latter caused the closure of stomata and consequently reduced CO₂ assimilation and transpiration, directly affecting WUE (Willadino and Camara, 2004).

Table 4 demonstrates that at 20 DAS all contrasts except S₁ versus other salts (S₂; S₃; S₄; S₅; S₆) were not significant while at 40 DAS, all were significantly affected. Estimates of mean (Table 4) demonstrated intrinsic WUE increase of 0.84 and 0.77 [(μ mol m⁻² s⁻¹) (mol H₂O m⁻²s⁻¹)⁻¹ ¹] in castor bean plants irrigated at ECw 0.6 dS m⁻¹ compared to those cultivated with saline water of ECw= 4.5 dS m⁻¹ at 20 and 40 DAS respectively. Table 4 also shows that intrinsic water use efficiency in S2 was 1.44 [(umol $m^{-2} s^{-1}$) (mol $H_2O m^{-2} s^{-1}$)⁻¹] lower than that in S_3 . When plants irrigated with sodium water (S_2) were contrasted, there was a reduction of 1.27 [(μ mol m⁻² s⁻¹) (mol H_2O m⁻² s⁻¹)⁻¹] in intrinsic WUE in plants irrigated with S_6 (Na+Ca+Mg) water. Estimate of mean (Table 4) demonstrated that, when S₂ was employed in irrigation water, a decrease of 2.40 [(μ mol m⁻² s⁻¹) (mol H₂O m⁻² s⁻¹) ¹)⁻¹] in intrinsic WUE occurred in comparison to plants with potassium water (S₅). Results show that the use of predominantly sodium water caused a greater deleterious effect on intrinsic WUE. However, plants irrigated with treatment S₅ had an increase of 1.62 [(μ mol m⁻² s⁻¹) (mol $H_2O \text{ m}^{-2} \text{ s}^{-1}$ with regard to the other types of salts (S₂, S_3 , S_4 and S_6). As a rule, among the cations analysed, there was a higher efficiency in plants irrigated with potassium water (S₅). The results obtained corroborate with data for rates of CO₂ assimilation (Figure 4B) and transpiration (Figure 3D) at 40 DAS.

Conclusions

(1) Water and physiological relationships of the castor beancy. BRS Energia plants are more sensitive to

variations in electrical conductivity of irrigation water as compared to the cationic nature of water.

(2) Irrigation with distinct cationic nature of water affects the osmotic potential, electrolyte leakage, relative water content and water saturation deficit of castor bean cv. BR Energia in the studied periods, the lowest deleterious effect in plants were observed with potassium water.

(3) The negative effects of cationic nature of irrigation water are more evident in gas exchanges, especially in stomatal conductance and CO_2 assimilation rate at 40 days after sowing.

(4) Among the ions studied, plants irrigated with potassic water show highest values for stomatal conductance, transpiration, CO_2 assimilation rate, instantaneous efficiency of carboxylation and intrinsic efficiency of water use at 40 DAS.

(5) Castor bean cv. BRS Energia plant is more sensitive to sodium ions in irrigation water with regard to water relationships and gas exchanges.

Conflict of Interest

The authors have not declared any conflict of interest.

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