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Morpho-physiology and oil yield of castor bean (*Ricinus communis* L.) as a function of salinity and the cationic nature of irrigation water

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Abstract

This study aimed to evaluate the morpho-physiology and oil yield of castor bean (cultivar 'BRS Energia') as a function of irrigation water salinity and cationic nature. The experiment was carried out using an Ultisol of sandy loam texture, in Campina Grande, Paraíba, Brazil. Six combinations of cations and water electrical conductivity (ECw) were studied (S_1 - Control; S_2 - Na^+; S_3 - Ca^{2+}; S_4 - Na^++Ca^{2+}; S_5 - K^+ and S_6 - Na^++Ca^{2+}+Mg^{2+}), in a randomized block design, with four replicates and five plants in each plot. Plants in the control treatment (S_1) were irrigated using water of 0.6 dS m^{-1}, while the other treatments were irrigated using water of 4.5 dS m^{-1}, prepared using chloride salts of different cations. Gas exchanges, growth and oil yield of castor bean were more affected by ECw variation than by the cationic nature of the water. Among the tested cations, potassium promoted the highest values of gas exchange and growth except in the flowering and fruiting stages. Oil yield is the most sensitive variable to the cationic nature of irrigation water.

Keywords: irrigation, *Ricinus communis* L., salinity.
Abbreviations: *A*_CO_2 assimilation rate; AW_available water; Ci_internal CO_2 concentration; DAS_ days after sowing; *E*_transpiration; ECw_electrical conductivity of water; EICI_ instantaneous carboxylation efficiency; *gs*_stomatal conductance; LA_leaf area; NL_number of leaves; OC_oil content; OM_organic matter; OY_oil yield; PH_plant height; SAR_sodium adsorption ratio; SD_stem diameter; SL_sandy loam; WUE_intrinsic water use efficiency.

Introduction

Castor bean (*Ricinus communis* L.) belongs to the *Euphorbiaceae* family, with potential for economic exploitation in the northeast region of Brazil, due to its xerophilic and heliophilic characteristics. It is an oil seed crop of significant socioeconomic value, with products and byproducts used in castor oil industry and agriculture (Ribeiro et al., 2009). Its oil, or ricin, extracted by seed pressing, contains 90% of ricinoleic fatty acid, which gives it singular characteristics, allowing wide industrial use (Amorim Neto et al., 2001). Under adverse climatic conditions, especially in the semiarid regions, high temperatures, low rainfall, irregular distribution of rains and intense evaporation are common in most months of the year, and irrigation is the best way to guarantee agricultural production (Nobre et al., 2011). However, the composition of the water available in this region varies depending on the total salt concentration, local geology and type of the water source, and waters rich in sodium, calcium, magnesium and, in some cases, potassium are commonly found (Medeiros et al., 2003).

The use of water and/or soils with salt problems can limit crop growth and production due to the reduction of the osmotic potential of the soil solution, which can also cause ion toxicity, nutritional imbalance or both, because of the excessive accumulation of certain ions in plant tissues, especially chloride and sodium (Flowers, 2004). However, the use of these waters in irrigation depends on certain factors, including salt type and concentration, exposure time, crop phenological stage, edaphoclimatic factors and the interaction between them (Ashraf and Harris, 2004).

Quantitative and qualitative depletion of water resources have led to the search for alternatives for a more efficient use of water and the rational use of the commonly called low-quality waters, since the use of saline water in agriculture is almost mandatory in semiarid regions (Alves et al., 2011). Thus, many authors have studied the effects of irrigation water with different salinity levels on the cultivation of castor bean (Campos et al., 2009; Soares et al., 2012; Alves et al., 2012; Nobre et al., 2013; Santos et al., 2013; Lima et al., 2014a); however, information on the effect of water with different cationic nature is still scarce.

Since most crops cultivated in the region are classified as sensitive to moderately sensitive to salinity, there arises the need for characterizing these waters, in order to establish an adequate management of crop, soil and irrigation systems (Medeiros et al., 2003), especially for the castor bean cultivar 'BRS Energia'. Thus, it becomes imperative to develop new studies on the effects of different types of cations in the irrigation water on castor bean, under the conditions of the semiarid region in Brazil. In this context, the morpho-physiology and oil yield of the castor bean cultivar 'BRS Energia' were evaluated as a function of irrigation water salinity and cationic nature.

Results and Discussion

Effect of irrigation water salinity and cationic nature on castor bean physiology

According to the F test (Table 1), there was significant effect of water salinity on stomatal conductance, transpiration, internal CO_2 concentration, CO_2 assimilation rate, intrinsic water use efficiency and instantaneous carboxylation efficiency of castor bean. According to the comparison of means for stomatal conductance (*gs*) (Fig 1A), there was significant difference between the treatments, and plants irrigated with low-salinity water (Control) and water containing K (S_5) were statistically superior to plants in the S_2 treatment, while plants in the treatments S_1, S_3, S_4, S_5 and S_6 did not differ statistically. The lowest *gs* values were observed in plants irrigated with water containing Na (S_2). There was significant difference for *gs* (Table 1) in the contrasts of S_1 *vs* the other treatments (S_2; S_3; S_4; S_5 and S_6), S_2 *vs* S_5 and S_5 *vs* the other salts (S_2; S_3; S_4 and S_6). According to the estimate of the means (Table 1), plants irrigated with water of ECw=0.6 dS m^{-1} showed an increase of 0.09 mol of H_2O m^{-2} s^{-1} for *gs*, compared with plants under high salinity water – ECw=4.5 dS m^{-1} (S_2; S_3; S_4; S_5 and S_6). Such decrease of *gs* in the treatments under the highest saline level can be related to the change in the osmotic potential and, consequently, to the reduction of water availability in plant tissues. Thus, possibly there was stomatal closure and a consequent reduction of the normal CO_2 flow towards the carboxylation site (Tezara et al., 2005).

When water containing Na was used in the irrigation (S_2), *gs* decreased by 0.13 mol of H_2O m^{-2} s^{-1}, compared with the treatment S_5 (K). In the comparison between S_5 and the other treatments, at the same level of ECw (S_2; S_3; S_4 and S_6), *gs* increased by 0.10 mol of H_2O m^{-2} s^{-1}. The lowest decrease in *gs* (Fig 1 A) when plants were irrigated with water containing K, in relation to the other types of saline waters, can be associated with the functions of K^+ in plant metabolism, since this macronutrient participates in the maintenance of the ionic equilibrium and cell turgor, through the control of stomatal opening and closure (Gurgel et al., 2010). As observed for *gs*, leaf transpiration (*E*) also differed statistically as a function of the types of salts in the irrigation water (Fig 1B), especially in the treatments Control and S_5, with the highest mean values of *E* (2.72 and 2.69 mmol of H_2O m^{-2} s^{-1}, respectively). However, the treatments S_1, S_3, S_4, S_5 and S_6 did not differ with respect to *E*. According to the estimate of the mean for *E* (Table 1), there was an increase of 0.53 mmol of H_2O m^{-2} s^{-1} when plants irrigated with waters of 0.6 dS m^{-1} and 4.5 dS m^{-1} were compared. On the other hand, the S_2 treatment significantly affected *E*, showing a decrease of 0.54 mmol of H_2O m^{-2} s^{-1} when compared to S_6 (Table 1).

However, in the contrast S_2 *vs* S_5, *E* was significantly influenced and decreased by 0.88 mmol of H_2O m^{-2} s^{-1} in S_2. In addition, the transpiration of plants in the S_5 treatment was significantly affected, compared with S_2, S_3, S_4 and S_6, and showed an increase of 0.62 mmol of H_2O m^{-2} s^{-1} (Table 1).

The reduction of *E* in the treatments S_2, S_3, S_4 and S_6 (Fig 1B) is mainly due to the partial stomatal closure (Fig 1A), a strategy of plants to avoid excessive dehydration of guard cells or a consequence of water imbalance on the leaf epidermis, a mechanism that leads to the decrease of CO_2 diffusion and, consequently, the reduction of the CO_2 assimilation rate (Ribeiro et al., 2009, Machado et al., 2010).

As to the internal CO_2 concentration (Ci), the highest values were observed in plants under S_2, S_3, S_4 and S_6 treatments, which were statistically higher than those observed in plants of S_1 and S_5, according to the test of comparison of means (Fig 2A). Castor bean plants irrigated with low-salinity water (0.6 dS m^{-1}) showed decrease of 72.71 μmol m^{-2} s^{-1} in Ci, compared with those receiving water of 4.5 dS m^{-1} (S_2; S_3; S_4; S_5 and S_6); while plants irrigated with water containing Na (S_2) increased Ci by 33.81 μmol m^{-2} s^{-1} when compared with those under S_3 (Table 1). Similarly, Ci increased by 30.18 μmol m^{-2} s^{-1} in S_2, compared with plants in the treatment S_6 (Table 1). However, the Ci in S_2 was 85.43 μmol m^{-2} s^{-1} lower than in S_5. On the other hand, there was a reduction of 61.51 μmol m^{-2} s^{-1} in Ci when S_5 was compared with the other types of salt in the irrigation water (S_2; S_3; S_4 and S_6).

It should be pointed out that higher Ci (S_2; S_3; S_4 and S_6) means that the carbon entering the leaf mesophyll cells was not being metabolized by the photosynthetic apparatus. In addition, such increase in Ci is an indication that there was no restriction for CO_2 absorption by the plant; however, when it reached the mesophyll cells, the process of fixation during the carboxylation phase was compromised (Habermann et al., 2003). The different types of water salinity also had significant influence on *A* (Fig 2B), and plants under low-salinity water (S_1) and receiving water containing K (S_5) were statistically superior to the other treatments (S_2, S_3, S_4 and S_6). According to the estimate of the mean for the contrasts (Table 1), the CO_2 assimilation rate increased by 9.92 μmol m^{-2} s^{-1} in plants subjected to water of 0.6 dS m^{-1}, in comparison to those receiving water of 4.5 dS m^{-1} (S_2; S_3; S_4; S_5 and S_6).

For the contrast S_2 *vs* S_6 (Table 3), when irrigation water containing Na was used (S_2), plants showed a reduction of 4.52 μmol m^{-2} s^{-1} in the CO_2 assimilation rate, compared with S_6. For S_2 *vs* S_5 (Table 1), plants irrigated with water containing Na (S_2) showed a decrease of 13.69 μmol m^{-2} s^{-1} in *A* compared with those in S_5. On the other hand, when plants were irrigated with S_5 (K), *A* increased by 11.41 μmol m^{-2} s^{-1}, compared with the other treatments under ECw = 4.5 dS m^{-1} (S_2; S_3; S_4 and S_6). Based on the results above, it can be inferred that the reduction of the CO_2 assimilation rate is a reflex of the decrease in transpiration and stomatal conductance, and the highest effect was due to the variation of water salinity (from 0.6 to 4.5 dS m^{-1}). Likewise, Lima et al. (2014a) observed reduction in the CO_2 assimilation rate of castor bean as a function of irrigation water salinity (ECw from 0.3 to 3.9 dS m^{-1}). A significant difference was observed between the intrinsic WUE values of the studied treatments (Table 1) and, according to the means comparison test (Fig 3A), WUE was statistically higher in the treatment using low-salinity water (S_1) and water containing K (S_5), in comparison to the other treatments (S_2; S_3; S_4 and S_6). However, no significant differences were observed when plants under these treatments were compared. According to the contrasts (Table 1), WUE increased by 2.96 (μmol m^{-2} s^{-1}) (mol H_2O m^{-2} s^{-1})$^{-1}$ in plants subjected to ECw = 0.6 dS m^{-1}

Table 1. Summary of F test and estimated means of different contrasts (ŷ) related to the stomatal conductance (*gs*), transpiration (*E*), internal CO_2 concentration (Ci), CO_2 assimilation rate (*A*), intrinsic water use efficiency (WUE) and instantaneous carboxylation efficiency (EICI) of castor bean cv. 'BRS Energia' irrigated with water of different types of salinity at 80 days after sowing.

SV	F test					
	gs	*E*	Ci	*A*	WUE	EICI
Blocks	ns	ns	ns	ns	ns	ns
Types of Salinity	*	*	**	**	**	*
$\hat{y}_1$	*	*	**	**	**	*
$\hat{y}_2$	ns	ns	*	ns	*	ns
$\hat{y}_3$	ns	*	*	*	*	ns
$\hat{y}_4$	*	*	**	**	**	*
$\hat{y}_5$	*	*	**	**	**	*
CV	14.41	14.94	6.60	19.13	14.69	18.71
	Mean estimate					
$\hat{y}_1$	0.09	0.53	-72.71	9.92	2.96	0.05
$\hat{y}_2$	ns	ns	33.81	ns	-1.17	ns
$\hat{y}_3$	ns	-0.54	30.18	-4.52	-1.35	ns
$\hat{y}_4$	-0.13	-0.88	85.43	-13.69	-4.05	-0.06
$\hat{y}_5$	0.10	0.62	-61.51	11.41	3.26	0.05

$\hat{y}_1$ (S_1 vs S_2; S_3; S_4; S_5; S_6); $\hat{y}_2$ (S_2 vs S_3); $\hat{y}_3$ (S_2 vs S_6); $\hat{y}_4$ (S_2 vs S_5); $\hat{y}_5$ (S_5 vs S_2; S_3; S_4; S_6);
S_1=Control; S_2= Na^+; S_3=Ca^{2+}; S_4=Na^++Ca^{2+}; S_5=K^+; S_6=Na^++Ca^{2+}+Mg^{2+}; SV – Source of variation; DF - Degree of freedom; CV – Coefficient of variation; (*) and (**) Significant at 0.05 and 0.01 probability; (ns) not significant.

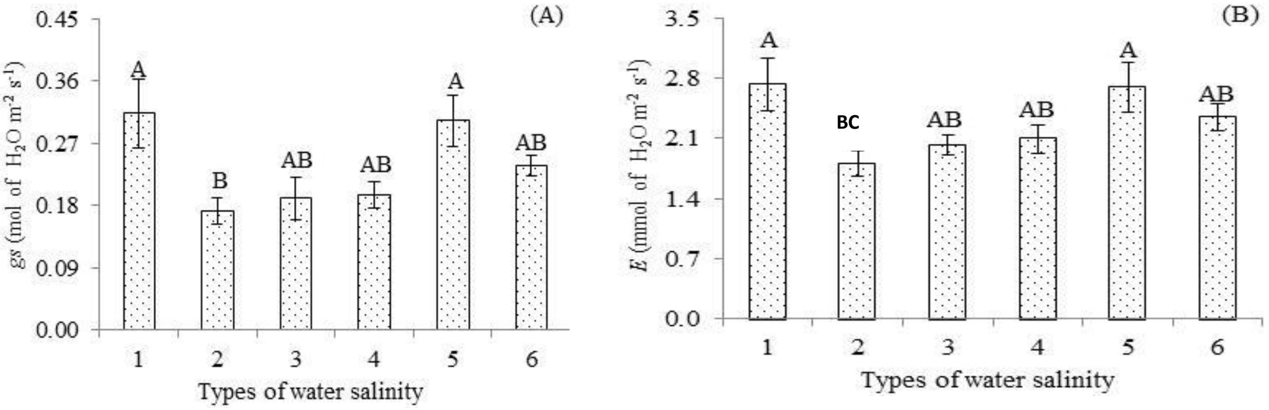

Fig 1. Stomatal conductance - *gs* (A) and transpiration - *E* (B) of castor bean cv. 'BRS Energia', depending on the types of irrigation water salinity at 80 days after sowing. S_1 = Control; S_2= Na^+; S_3 = Ca^{2+}; S_4 = Na^++Ca^{2+}; S_5 = K^+; S_6 = Na^++Ca^{2+}+Mg^{2+}; Bars represent the standard error of the mean (n = 4). Means followed by different letters differ significantly at 0.05 probabilty level.

compared with those under ECw = 4.5 dS m^{-1}. The intrinsic water use efficiency in S_2 was 1.17 (μmol m^{-2} s^{-1}) (mol H_2O m^{-2} s^{-1})$^{-1}$ lower than in S_3. Plants receiving water containing Na (S_2) showed a reduction of 1.35 (μmol m^{-2} s^{-1}) (mol H_2O m^{-2} s^{-1})$^{-1}$ in WUE compared with those irrigated using water containing Na+Ca+Mg (S_6). Plants in the S_2 treatment showed a reduction of 4.05 (μmol m^{-2} s^{-1}) (mol H_2O m^{-2} s^{-1})$^{-1}$ in WUE, compared with those receiving water containing K (S_5). Plants in S_5 showed an increment of 3.26 (μmol m^{-2} s^{-1}) (mol H_2O m^{-2} s^{-1})$^{-1}$ in WUE, in comparison to the treatments with other types of salt (S_2; S_3; S_4 and S_6). The decrease in WUE observed in the treatments S_2, S_3, S_4 and S_6 (Fig 3A) is due to the decrease in stomatal conductance (Fig 1A) and transpiration (Fig 1B), because when *gs* is limited in order to reduce water loss (*E*), the CO_2 inflow to the cell and the CO_2 assimilation rate also decrease. It should be noted that WUE relates the amount of carbon fixed by the plant per unit of water lost in this process (Taiz and Zeiger, 2013). Thus, it is important to absorb as much CO_2 as possible with the minimum water loss, a fact observed in plants irrigated with low-salinity water (S_1) and water containing K (S_5). As to the instantaneous carboxylation efficiency (EICI) (Fig 3B), plants irrigated with low-salinity water (S_1) and water containing K (S_5) were statistically superior to plants under the other types of salinity (S_2; S_3; S_4 and S_6). However, these treatments did not differ significantly. According to the estimate of the mean (Table 1), EICI increased by 0.05 [(μmol m^{-2} s^{-1}) (μmol mol^{-1}]$^{-1}$ in plants under low-salinity water (0.6 dS m^{-1}), compared with those under ECw of 4.5 dS m^{-1} (S_2; S_3; S_4; S_5; S_6). For the contrasts S_2 *vs* S_3 and S_2 *vs* S_6, there was no significant effect on EICI. On the other hand, significant effect was observed for S_2 *vs* S_5 and S_5 *vs* S_2; S_3; S_4 and S_6. EICI decreased by 0.06 [(μmol m^{-2} s^{-1}) (μmol mol^{-1}]$^{-1}$ in plants in S_2, compared with those receiving water containing K (S_5) (Table 1). Plants subjected to the treatment S_5 tended to increase EICI (Table 1), with mean

Table 2. Summary of F test and estimated means of different contrasts (ŷ) related to the plant height (PH), stem diameter (SD), number of leaves (NL) and leaf area (LA), oil content (OC) and oil yield (OY) of castor bean cv. 'BRS Energia' irrigated with water of different types of salinity at 80 days after sowing.

SV	F test					
	PH	SD	NL	LA	OC	OY
Blocks	ns	ns	ns	ns	ns	ns
Types of Salinity	**	**	**	**	**	**
$\hat{y}_1$	**	**	**	*	*	**
$\hat{y}_2$	ns	ns	ns	ns	ns	ns
$\hat{y}_3$	ns	ns	ns	ns	ns	ns
$\hat{y}_4$	*	*	*	*	**	**
$\hat{y}_5$	*	*	**	*	*	*
CV	6.76	9.69	18.46	15.46	7.73	14.01
	Mean estimate					
$\hat{y}_1$	25.69	7.75	18.77	11125.86	9.05	49.68
$\hat{y}_2$	ns	ns	ns	ns	ns	ns
$\hat{y}_3$	ns	ns	ns	ns	ns	ns
$\hat{y}_4$	-9.12	-3.55	-10.50	-1757.00	12.61	11.06
$\hat{y}_5$	7.41	3.15	9.87	1457.39	5.25	-3.39

$\hat{y}_1$ (S_1 vs S_2; S_3; S_4; S_5; S_6); $\hat{y}_2$ (S_2 vs S_3); $\hat{y}_3$ (S_2 vs S_6); $\hat{y}_4$ (S_2 vs S_5); $\hat{y}_5$ (S_5 vs S_2; S_3; S_4; S_6);
S_1= Control; S_2= Na^+; S_3=Ca^{2+}; S_4=Na^++Ca^{2+}; S_5=K^+; S_6=Na^++Ca^{2+}+Mg^{2+}; SV – Source of variation; DF- Degree of freedom; CV – Coefficient of variation; (*) and (**) Significant at 0.05 and 0.01 probability; (ns) not significant.

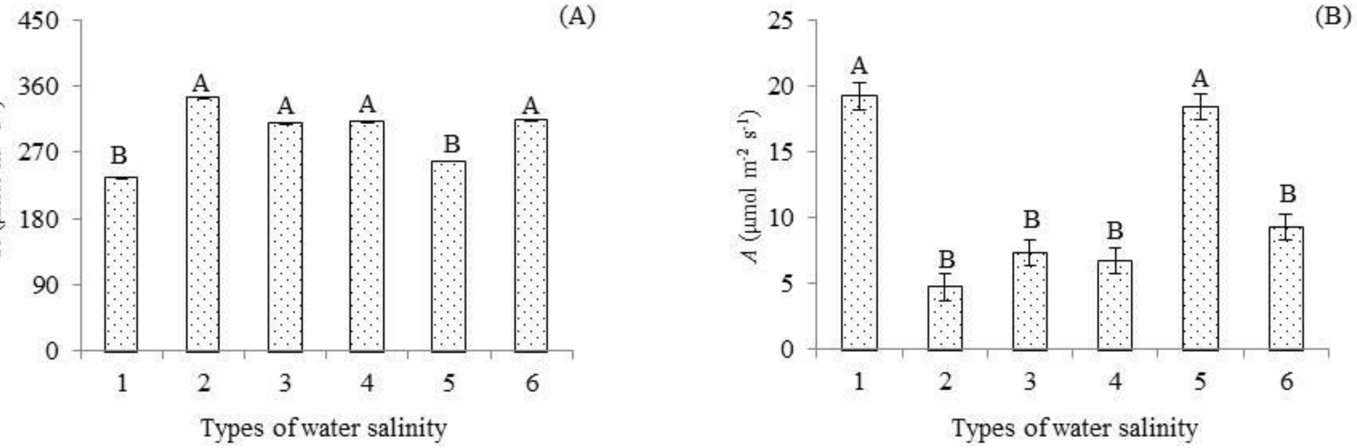

Fig 2. Internal CO_2 concentration – Ci (A) and CO_2 assimilation rate - *A* (B) of castor bean cv. 'BRS Energia', depending on the types of irrigation water salinity at 80 days after sowing. S_1 = Control; S_2= Na^+; S_3 = Ca^{2+}; S_4 = Na^++Ca^{2+}; S_5 = K^+; S_6 = Na^++Ca^{2+}+Mg^{2+}; Bars represent the standard error of the mean (n = 4). Means followed by different letters differ significantly at 0.05 probabilty level.

value of 0.05 [(µmol m^{-2} s^{-1}) (µmol mol^{-1})]$^{-1}$, higher than in the other treatments (S_2; S_3; S_4 and S_6). The results suggest that this higher decrease in EICI in the treatments S_2, S_3, S_4 and S_6 is a reflex of the low CO_2 assimilation rates (Fig 2B), in relation to the CO_2 content in the substomatal chamber (Fig 2A). Machado et al. (2005) reported that the instantaneous carboxylation efficiency is closely related to the CO_2 assimilation rate and the intracellular CO_2 concentration.

Effect of irrigation water salinity and cationic nature on growth and oil yield

Based on the summary of the F test (Table 2), there was significant effect (p<0.01) of the different types of water salinity on PH, SD, NL, LA, OC and OY of castor bean. Lima et al. (2014b), studying the influence of irrigation with saline water simulated with NaCl (ECw from 0.3 to 3.9 dSm^{-1}, also observed significant effect on plant height, stem diameter, number of leaves and leaf area at 60 DAS. According to the means comparison test for PH (Fig 4A), plants irrigated with low-salinity water (S_1) statistically differed (p<0.01) from those irrigated with high-salinity water and different ionic compositions (S_2; S_3; S_4; S_5 and S_6). The treatment S_5 differed significantly regarding PH from treatments with Na (S_2) and Ca (S_3), which did not differ significantly from S_4 and S_6. Based on the contrasts of means (Table 2), PH varied significantly (p<0.01) and increased by 25.69 cm in the treatment with ECw of 0.6 dS m^{-1} (S_1), compared with those under 4.5 dS m^{-1} (S_2; S_3; S_4; S_5 and S_6). These results agree with Lima et al. (2014b), who observed that the increase in irrigation water salinity (ECw from 0.3 to 3.9 dS m^{-1}) reduced PH of the castor bean cultivar 'BRS Energia' at 30, 60 and 120 DAS. This tendency for a decrease in PH under saline-stress conditions can be attributed to the fact that, in order to adjust osmotically, plants spend a certain amount of energy in the accumulation of sugars, organic acids and ions in the vacuole, which could be used for growth (Santos et al., 2012). As to the treatments S_2 *vs* S_3 and S_2 *vs* S_6, there was no significant effect on PH. Therefore, the results suggest that Na, Ca and Mg acted similarly on plant growth. On the other hand, there was significant effect for S_2 *vs* S_5 and S_5 *vs* (S_2; S_3; S_4 and S_6) and, according to the estimate of the mean (Table 2), plants

Table 3. Chemical characteristics of the water used in the experiment.

Types of Salinity	Ca^{2+}	Mg^{2+}	Na^+	K^+	HCO_3^-	Cl^-	ECw	pH	SAR
			(mmol$_c$ L^{-1})				dS m^{-1}		(mmol L^{-1})$^{0.5}$
Control*	1.19	1.58	2.83	0.10	1.45	4.22	0.60	7.23	2.41
Na^+	6.12	3.50	33.10	0.16	6.40	57.00	4.50	7.40	15.11
Ca^{2+}	31.37	15.25	5.22	0.18	3.40	38.75	4.50	7.35	1.08
$Na^+ + Ca^{2+}$	17.75	5.00	20.22	0.16	3.80	41.25	4.50	7.38	6.00
K^+	1.75	6.87	5.59	22.28	5.00	38.00	4.50	7.75	1.29
$Na^+ + Ca^{2+} + Mg^{2+}$	11.00	9.00	25.74	0.14	2.60	39.75	4.50	7.40	8.14

*Obtained by dilution of the water of supply system (Campina Grande, Paraiba) with rain water; ECw - electrical conductivity of water; SAR - sodium adsorption ratio; CO_3^{2-}= absent.

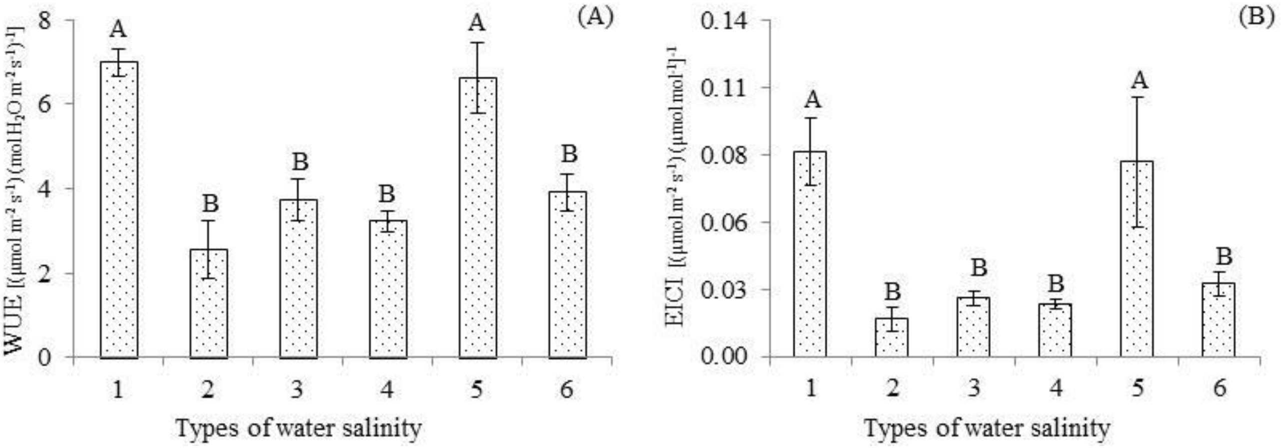

Fig 3. Intrinsic water use efficiency – WUE (A) and instantaneous carboxylation efficiency EICI (B) of castor bean cv. 'BRS Energia', depending on the types of irrigation water salinity at 80 days after sowing. S_1 = Control; S_2= Na^+; S_3 = Ca^{2+}; S_4 = Na^++Ca^{2+}; S_5 = K^+; S_6 = Na^++Ca^{2+}+Mg^{2+}; Bars represent the standard error of the mean (n = 4). Means followed by different letters differ significantly at 0.05 probabilty level.

Table 4. Chemical and physical characteristics of the soil used in the experiment before application of treatments.

Chemical characteristics									
pH$_{ps}$	M.O dag kg^{-1}	P (mg kg^{-1})	K^+	Na^+	Ca^{2+}	Mg^{2+}	Al^{3+}	H^+	ECse (dS m^{-1})
			....		(cmol$_c$ kg^{-1})	....			
5.10	0.34	20.09	0.07	0.05	0.40	1.30	0.04	1.74	0.16

Physical characteristics									
Size fraction (g kg^{-1})			Textural class	Water content (kPa)		AW	Total porosity m^{3}m^{-3}	BD (kg dm^{-3})	DP (kg dm^{-3})
Sand	Silt	Clay		33.42	1519.5 dag kg^{-1}				
856.10	110.70	33.20	SL	6.72	1.62	5.10	0.49	1.54	2.72

pH$_{PS}$- pH of saturated paste; O.M – organic matter: determined by wet digestion Walkley-Black method; $Ca^{2+}$and Mg^{2+}extracted with KCl 1 mol L^{-1} at pH 7.0; Na^+ and K^+ extracted with NH_4 OAc 1 mol L^{-1} at pH 7.0; SL –sandy loam; AW –available water; BD – Bulk density; DP-density particles; ECse-electrical conductivity of the saturation extract.

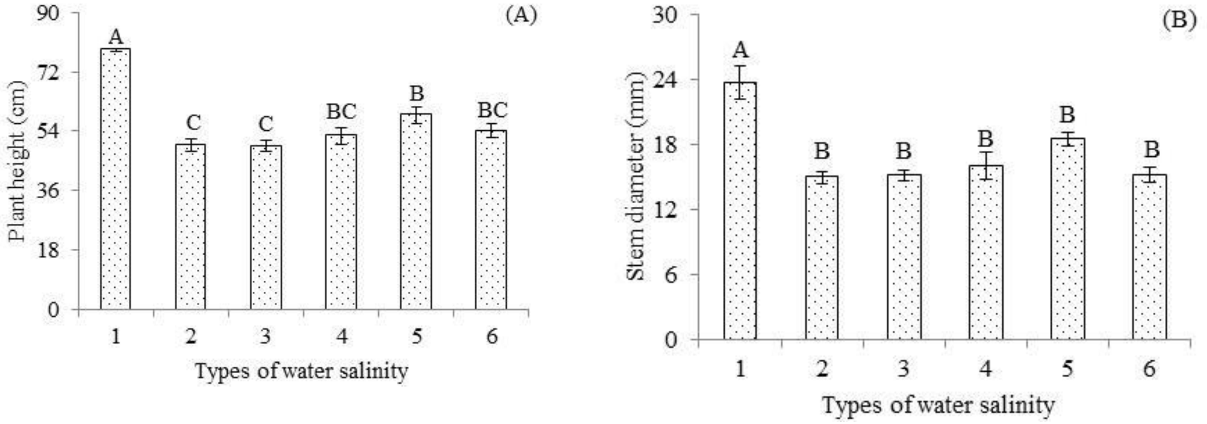

Fig 4. Plant height (A) and stem diameter (B) of castor bean cv. 'BRS Energia', depending on the types of irrigation water salinity at 80 days after sowing. S_1 = Control; S_2= Na^+; S_3 = Ca^{2+}; S_4 = Na^++Ca^{2+}; S_5 = K^+; S_6 = Na^++Ca^{2+}+Mg^{2+}; Bars represent the standard error of the mean (n = 4). Means followed by different letters differ significantly at 0.05 probabilty level.

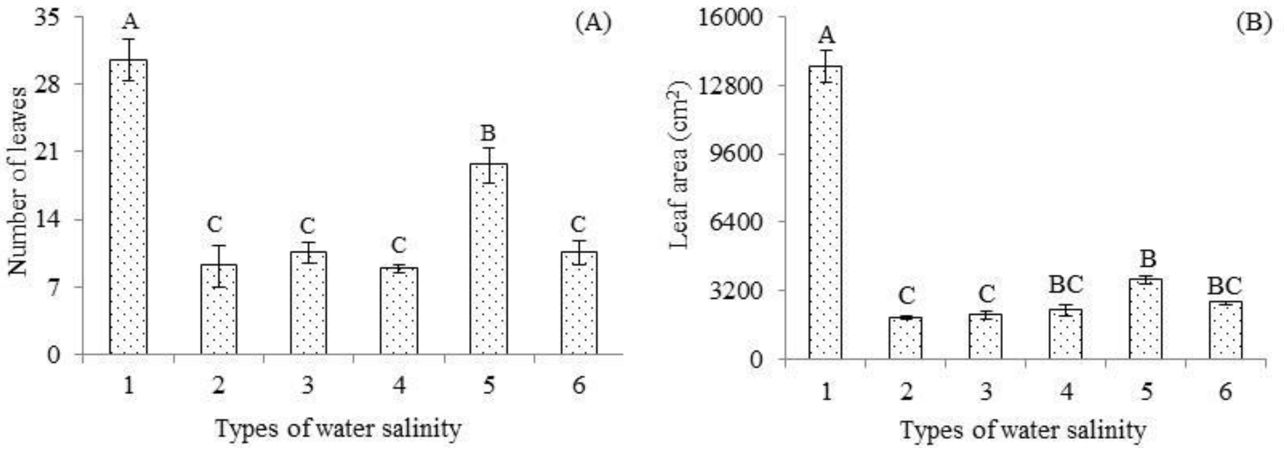

Fig 5. Number of leaves (NL) and leaf area (LA) of castor bean cv. 'BRS Energia', depending on the types of irrigation water salinity at 80 days after sowing. S_1 = Control; S_2= Na^+; S_3 = Ca^{2+}; S_4 = Na^++Ca^{2+}; S_5 = K^+; S_6 = Na^++Ca^{2+}+Mg^{2+}; Bars represent the standard error of the mean (n = 4). Means followed by different letters differ significantly at 0.05 probabilty level.

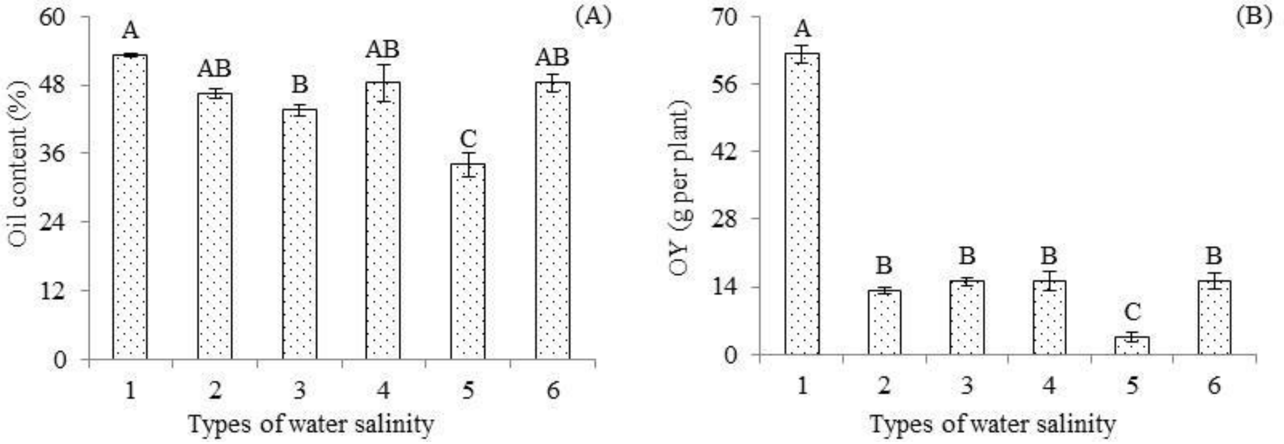

Fig 6. Oil content (A) and oil yield - OY (B) of castor bean cv. 'BRS Energia', depending on the types of irrigation water salinity at 100 days after sowing. S_1 = Control; S_2= Na^+; S_3 = Ca^{2+}; S_4 = Na^++Ca^{2+}; S_5 = K^+; S_6 = Na^++Ca^{2+}+Mg^{2+}; Bars represent the standard error of the mean (n = 4). Means followed by different letters differ significantly at 0.05 probabilty level.

irrigated using water containing Na (S_2) showed PH 9.12 cm lower than those subjected to water containing K (S_5), while plants in the treatment S_5 tended to have higher PH, with mean value of 7.41 cm, compared with plants in the other treatments with ECw = 4.5 dS m^{-1} (S_2; S_3; S_4 and S_6). In spite of that, the highest PH observed for the treatment with water containing K (S_5) is justified by the importance of this nutrient in plant metabolism, since it acts in the regulation of the osmotic potential of the cell, the balance of negative charges of the organic acids inside the cells and the balance of anions absorbed by the roots (Bernardi et al., 2010). According to the comparison of means for stem diameter (Fig 4B), as observed for PH (Fig 4A), there was a significant difference ($p<0.05$) between the control treatment (S_1) and the others (S_2; S_3; S_4; S_5 and S_6), which did not differ ($p>0.05$) statistically. Like PH (Table 2), significant effects were observed for SD when plants subjected to water of 0.6 dS m^{-1} (S_1) were compared with those under 4.5 dS m^{-1} (S_2; S_3; S_4; S_5 and S_6); SD was 7.75 mm higher in S_1 compared with the others. Alves et al. (2012), studying the influence of irrigation water of ECw from 0.6 to 4.6 dS m^{-1} on the growth of the castor bean cultivar 'BRS Energia', observed a reduction of 3.32 mm in SD at 40 DAS. According to Table 2, plants in the S_2 treatment were not significantly different ($p > 0.05$) from those in S_3 and S_6. Thus, it can be inferred that irrigation with different types of salt (Na^+, Ca^{+2}, Mg^{+2}) caused similar effects on SD, but plants in S_2 were significantly different ($p<0.05$) from those in S_5, with SD value 3.55 mm lower. SD increased by 3.15 mm in the comparison of S_5 *vs* S_2, S_3, S4 and S_6. Based on the means comparison test for the number of leaves (Fig 5A), there was significant ($p < 0.05$) difference between the studied salts and, as expected, the treatment S_1 was superior to the others (S_2; S_3; S_4; S_5 and S_6). On the other hand, when the data were compared as a function of the types of salt in the irrigation water, the use of water containing K had significant and positive influence on NL and differed from S_2, S_3, S_4 and S_6. According to the estimate of the mean for the number of leaves (Table 2), plants receiving water of 0.6 dS m^{-1} (S_1) showed higher NL, exceeding in 18.77 the mean observed for plants under 4.5 dS m^{-1} (S_2; S_3; S_4; S_5 and S_6). However, in the comparison of the treatments S_2 *vs* S_3 and S_2 *vs* S_6 (Table 2), there was no significant ($p > 0.05$) influence on NL. On the other hand, NL was statistically lower when water containing Na (S_2) was used in the irrigation, with a decrease of 10.50 leaves compared with the plants in S_5. Plants subjected to irrigation using water containing K (S_5) showed an increment of 9.87 in NL compared with the other treatments under ECw = 4.5 dS m^{-1} (S_2, S_3,S_4, S_6) (Fig 5A). Thus, under saline stress conditions, morphological changes in the plants are common, which lead to the reduction of the transpiring surface as an alternative to maintain adequate water absorption, and the reduction in NL is among these adaptations (Oliveira et al., 2010), a fact also observed by Alves et al. (2012) and Lima et al. (2014a).

According to the comparison of means for leaf area (Fig 5B), plants irrigated with low-salinity water (S_1) showed the highest LA compared with the other treatments, as NL and the other analysed variables. Also, the LA of plants in the treatment S_5 was significantly different only from those in S_2 and S_3. In addition, the value obtained for LA in the treatments S_4, S_5 and S_6 did not differ. The estimate of the mean for LA (Table 2) shows an expressive increase in LA of plants under low-salinity water (0.6 dS m^{-1}), exceeding the mean value in 11125.86 cm^2, compared with those irrigated with water of 4.5 dS m^{-1}. In addition, no significant influence on LA was observed when plants irrigated with water containing Na (S_2) were compared with those receiving water containing Ca (S_3) and Na+Ca+Mg (S_6).

Still according to Table 2, LA was significantly affected in the plants in S_2, with a decrease of 1757.00 cm^2 when compared with plants in S_5. However, in the comparison of the treatment S_5 *vs* the others (S_2, S_3, S_4, S_6), LA increased by 1457.39 cm^2, i.e., a reduction lower than that previously observed for the treatments S_2 *vs* S_5. Thus, the decreases in LA, especially in plants irrigated with water containing Na are reflections of the high accumulation of the ions Na^+ and Cl- in the soil solution, at levels that alter cell ionic balance. Also, the reductions in the number of leaves (Fig 5A) and leaf area (Fig 5B) in this study are an example of probable adaptations of plants cultivated under saline stress conditions. According to the means comparison test for oil content (Fig 6A), plants in S_1 did not differ ($p>0.05$) from those receiving water containing Na (S_2), Na+Ca (S_4) and Na+Ca+Mg (S_6). However, significant difference for OC was observed between the treatment using water with K (S_5) and the others (S_1, S_2, S_3, S_4 and S_6). The contrasts of means for OC (Table 2) showed no significant difference ($p > 0.05$) for S_2 *vs* S_3 and S_2 *vs* S_6. Based on the estimate of the mean (Table 2), plants irrigated with low-salinity water (0.6 dS m^{-1}) showed an increment of 9.05% in OC, when compared with those under ECw of 4.5 dS m^{-1} (S_2; S_3; S_4; S_5 and S_6). On the other hand, when this variable was analyzed between plants receiving water with Na (S_2) and K (S_5), OC increased by 12.61%. The oil content was 5.25% lower in S_5 compared with the other treatments with ECw = 4.5 dS m^{-1} (S_2; S_3; S_4 and S_6).

As for oil yield (Fig 6B), plants in the control treatment (S_1) showed OY significantly higher compared with plants under other types of water salinity. Also, significant differences for OY were found between plants subjected to water containing K (S_5) and those subjected to water containing Na (S_2), Ca (S_3), Na+Ca (S_4) and Na+Ca+Mg (S_6). Significant influence ($p < 0.05$) on OY was observed in plants irrigated using water with ECw of 0.6 dS m^{-1} compared with those under 4.5 dS m^{-1}, according to the analysis of variance of the studied contrasts. The highest OY was observed in plants under low-salinity water (0.6 dS m^{-1}), exceeding the mean value in 49.68 g $plant^{-1}$, in relation to those irrigated with ECw of 4.5 dS m^{-1} (Table 2).

Still according to Table 2, OY in S_2 was 11.06 g $plant^{-1}$ higher than in S_5. Plants in S_5 showed a decrease of 3.39 g $plant^{-1}$ in OY, compared with those under different types of cations (S_2; S_3; S_4 and S_6). In general, among the studied cations, a lower OY was observed in plants irrigated with water containing K (S_5). Although this result corroborates the data obtained for the oil content in the seeds (Fig 6A), it is difficult to explain when data of gas exchange and growth variables are taken into account, since plants irrigated with water containing K (S_5) showed the highest values for these variables.

Considering that OY is obtained by multiplying the oil content by the mass of seeds, the mass of seeds in the S_5 treatment was 20 times lower than in the other treatments – S_2; S_3; S_4 and S_6. Thus, this expressive decrease in OY for plants in S_5 may be attributed to the fact that K^+ is absorbed in amounts higher than the necessary, so called “luxury consumption”. Under these conditions, the excess of K^+ may have interfered negatively with the absorption of the other cations by the crops, especially those competing for the same absorption sites in root tissues (Meurer, 2006), inhibiting the absorption of Ca^{2+} and Mg^{2+} (Marschner,1995). Therefore, under Ca^{2+} deficiency, flowering and yield were severely affected, because pollen tube formation and pollen grain germination depend on the presence of Ca^{+2} (Beyoung, 1965).

Materials and Methods

Localization, experimental procedure, treatments and plant material

The experiment was carried out in a greenhouse using drainage lysimeters from November 2013 to February 2014, at the Center of Technology and Natural Resources of the Federal University of Campina Grande (CTRN/UFCG), located in the municipality of Campina Grande-Paraíba, Brazil (07°15’18’’ S; 35°52’28’’ W; 550 m). The treatments consisted of six types of salinity (S_1 - Control; S_2 - Na^+; S_3 - Ca^{2+}; S_4 - Na^+ + Ca^{2+}; S_5 - K^+ and S_6 - Na^+ + Ca^{2+} + Mg^{2+}), maintaining the proportions of 1:1 and 7:2:1 for Na+Ca and Na+Ca+Mg, respectively. Plants in the control treatment (S_1) were irrigated using water with electrical conductivity (ECw) of 0.6 dS m^{-1}, while the other treatments (S_2; S_3; S_4; S_5 and S_6) received water of ECw = 4.5 dS m^{-1}, prepared with chloride salts of different cations (Table 3). The experiment was set in a randomized block design with six treatments and four replicates, totaling 24 experimental plots, each one with five plants. The castor bean cultivar ‘BRS Energia’ was used in the experiment, for its strong genetic material, precocity (cycle from 120 to 150 days), short size, semi-indehiscent fruits, average oil content of 48% in the seeds and average yield of 1,800 kg ha^{-1} (Silva et al., 2009).]

Establishment and management of the experiment

Plants were cultivated in 100-L drainage lysimeters (height = 50 cm; bottom diameter = 30 cm; top diameter = 33 cm), perforated at the bottom, where a drain of 4 mm of diameter was installed to allow drainage. The tip of the drain inside the lysimeter was involved in a nonwoven geotextile (Bidim OP 30) in order to avoid obstruction by soil material and, outside the lysimeter, a plastic recipient was placed under the drain, in order to collect the drained water and estimate water consumption. The lysimeters were filled with a 2-kg layer of crushed stone, followed by 54 kg of soil material (clod-free and homogenized) and 76 kg of the same soil with humus in order to achieve 1% of organic matter in the entire soil mass. The first 10 cm of each lysimeter remained free in order to facilitate irrigation. The soil material used in the study was collected from the A horizon (0-30 cm layer) of an Ultisol, from the district of São José da Mata (Campina Grande, Paraíba). Before the experiment, the soil was sampled for chemical and hydro-physical characterization (Table 4) at the Laboratory of Irrigation and Salinity of the CTRN/UFCG, according to the methodology proposed by Claessen (1997).

Based on the soil analysis, the acidity was corrected using 49.25 g of dolomitic limestone in each lysimeter (130 kg of soil), to neutralize Al^{3+} and increase the contents of Ca^{2+} and Mg^{2+} to the level of 70%. After liming, the soil showed the following chemical characteristics: Ca^{2+} =1.14 $cmol_c$ kg^{-1}; Mg^{2+} = 1.36 $cmol_c$ kg^{-1}; Na^+ = 0.30 $cmol_c$ kg^{-1}; K^+ = 0.14 $cmol_c$ kg^{-1}; H^+ = 0.11 $cmol_c$ kg^{-1}; CEC = 3.05 $cmol_c$ kg^{-1}; Organic matter = 1.08 dag kg^{-1}; P = 47.80 mg kg^{-1}; pH in water (1:2.5) = 6.42 and ECse=0.20 dS m^{-1}.Different irrigation waters were obtained by the dissolution of sodium, calcium, magnesium and potassium chlorides (99% purity), according to the treatments, in tap water. The amounts were determined taking into consideration the relation between ECw and salt concentration (10*$mmol_c$ L^{-1} = 1 dS m^{-1}) proposed by Richards (1954). After preparation and ECw calibration, using a portable conductivity meter, the waters were stored in plastic recipients, adequately protected in order to avoid evaporation. Before seeding, the volume of water necessary to reach field capacity was added to the soil, according to the treatments. After field capacity was achieved, ten seeds of the castor bean cultivar 'BRS Energia' were equidistantly planted in each lysimeter at a depth of 2 cm. At 10 days after seeding (DAS), thinning was performed in order to leave only one plant per lysimeter. After seeding, the soil was kept close to field capacity by daily irrigations in each lysimeter, according to the treatments, and the volume applied was determined based on crop water demand, estimated by the water balance: volume of water applied minus volume of water drained in the previous irrigation, plus a leaching fraction of 0.10, according to previous studies (Nobre et al., 2013; Lima et al., 2014a). Fertilization with N, K and P was performed according to Novais et al. (1991), using 40.62 g of potassium nitrate and 75 g of monoammonium phosphate, which was equivalent to 100, 150 and 300 mg kg^{-1} of soil of N, K_2O and P_2O_5, respectively. Fertilizers were applied as topdressing, in four applications through fertigation, at intervals of ten days, with the first one at 15 DAS. In order to avoid any possible deficiency of micronutrients, foliar application of 2.5 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%)] was performed at 30 and 60 DAS. The cultural practices during the experiment consisted of weekly manual weeding, superficial soil scarification and staking, after plants reached the flowering stage, in order to avoid lodging. In addition, 5.4 g L^{-1} of insecticides, 7.0 g L^{-1} of fungicide and 3.5 g L^{-1} of acaricide, from the Neonicotinamid, Triazole and Abamectin chemical groups, respectively, were applied.

Traits measured

The effects of the treatments on castor bean physiology were measured at 80 DAS in the fruiting stage, through stomatal conductance (*gs*), transpiration (*E*), CO_2 assimilation rate (*A*), internal CO_2 concentration (Ci), intrinsic water use efficiency (WUE) and instantaneous carboxylation efficiency (EICI). Plant growth was evaluated through plant height (PH), stem diameter (SD), number of leaves (NL) and leaf area (LA).

Racemes were manually harvested from 70 to 100 DAS. Harvest was performed when approximately 90% of the fruits of each raceme reached physiological maturation, and the drying was completed by sun exposure. Castor bean production was evaluated through the oil content (OC) and oil yield (OY) from the seeds of the primary raceme. Stomatal conductance, transpiration, CO_2 assimilation rate and internal CO_2 concentration were evaluated in the third leaf from the plant apex, using the portable device for photosynthesis measurements "LCPro+" (ADC BioScientific Ltda.). Using these data, the water use efficiency (WUE) (*A/E*) and instantaneous carboxylation efficiency (*A/*Ci) were quantified (Jaimez et al., 2005). PH was measured from the plant base to the apical meristem. SD was measured 5 cm distant from the plant base. For NL, only the leaves with minimum length of 3 cm and at least 50% of photosynthetically active leaf area were considered. LA was obtained by measuring the length of the midrib of all the leaves in the plants, based on the methodology described by Severino et al. (2005), according to Eq. 1:

$$\mathrm{LA} = \Sigma 0.26622\ x\mathrm{P}^{2.4248} \qquad (1)$$

where:

LA - total leaf area per plant (cm^2),

P - length of leaf midrib (cm).

After seeds were dried, processed and had their water content corrected to 10%, the oil content was determined by non-destructive method, using a nuclear magnetic resonance (NMR) spectrometer (H^1 Oxford MQA 7005), at the Advanced Chemical Technology Laboratory of Embrapa Cotton, in Campina Grande-PB (American Oil Chemists' Society, 2000). Oil yield per plant was determined using data of oil content and mass of seeds.

Statistical analysis

The results were subjected to analysis of variance by F-test; when significant, the means were compared by Tukey's test and contrasts (ŷ) between treatment means (at 0.05 probability level) were performed using the software SISVAR-ESAL (Ferreira, 2000). For the comparison between treatments, the standard error was calculated for each mean. The contrasts were defined as follows: $\hat{y}_1$ (S_1 vs S_2; S_3; S_4; S_5; S_6), $\hat{y}_2$ (S_2 vs S_3), $\hat{y}_3$ (S_2 vs S_6), $\hat{y}_4$ (S_2 vs S_5) and $\hat{y}_5$ (S_5 vs S_2; S_3; S_4; S_6).

Conclusions

Gas exchanges, growth and oil yield of the castor bean cultivar 'BRS Energia' are more affected by the variation of electrical conductivity in the irrigation water than by its cationic nature; the deleterious effect of saline stress on gas exchange, growth, oil content and oil yield of castor bean varies with the type of cation present in the irrigation water; among the studied ions, potassium promotes the highest values of gas exchange and growth variables; oil yield is the most sensitive variable to the effects of irrigation water salinity and cationic nature; the cultivar 'BRS Energia' is more sensitive to the salinity caused by potassium ions in the irrigation water during the flowering and fruiting stages.

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