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Evaluation of NaCl Tolerance in the Physical Reduction of *Jatropha Curcus* L. Seedlings

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Abstract

Jatropha curcas L. is an important bio-fuel crop however, it's tolerance to salinity especially with reference to changes in physical characteristics has been hardly studied. This work aimed to evaluate *Jatropha. curcas* L. tolerance to salinity stress using physical growth patterns, leaf shedding and mineral nutrient deposition. *Jatropha curcas* L. seedlings were grown under four different levels of NaCl concentration: 0 (control), 25, 50 and 100 mM under greenhouse conditions. Harvesting was done when average transpiration of each treatment was less than 50% and 75% as compared to the control.

Results showed a significant variation in transpiration rate among the salinity treatments and control. A gradual reduction in the biomass yield of seedlings with increasing concentration of NaCl was observed. Reference to the result of IC_{50}, the seedlings were tolerant to NaCl irrigation up to 54 mM. Additionally, seedling stems and roots accumulated large amounts of Na and K; a large amount of K particularly accumulated in the stem part, and was likely responsible for the low Na/K ratio observed in the stem. Defoliation however occurred with even irrigation of as low NaCl concentration as 25 mM. Thus, we report that *Jatropha curcas* L. is highly sensitive to Na accumulation, especially in the root-zone.

Keywords: *Jatropha curcas* L., salt tolerance, growth, defoliation, nutrient concentration

1. Introduction

Jatropha curcas L. (Linnaeus) is a multi-purpose small tree/bush under the family *Euphorbiaceae*. It is native to tropical America, but now grown in several parts of the tropics and sub tropics in Africa and Asia (Openshaw, 2000). Recently *J. curcas* has been receiving attention as a source of good quality bio-fuel from its seeds; other traditional uses of this plant include: use as a hedge to protect fields, green manure, oil-soap production and medicinal goods (Kumar & Sharma, 2008). It has characteristics of high temperature tolerance and can grow under low fertility and moisture

conditions (Augustus, Jayabalan & Seiler, 2002). In addition, it is rarely troubled by pests and diseases and will grow on a wide range of annual rainfall from 200 to over 1500mm (Openshaw, 2000). Thus, it is claimed that it can grow on degraded or un-vegetated sites in arid and semi-arid areas (Augustus *et al.*, 2002). *Jatropha curcas* L has also been thought to be beneficial in environmental remediation of degraded land and in terms of job creation around *Jatropha curcas* L plantations (Francis, Edinger & Becker, 2005). Cultivation of the plant in arid and semi-arid regions requires intensive irrigation and fertilization which may cause salinization due to progressive salt accumulation in the soils as a consequence of the salt dissolved in the irrigation water and associated high evapotranspiration rates (Chaves, Flexas & Pinheiro, 2009). It is claimed that 6% of the world's land and 30% of the world's irrigated areas already suffer from salinity related problems (Munns & Tester, 2008). Agriculture in such regions will increasingly be forced to utilize marginal or salinity water, which pose risks of soil salinization and yield reduction (Paranychianakis & Chartzoulakis, 2005).

Salinity is the major environmental factor limiting the growth and productivity of crops (Allakhverdiev, Sakamoto, Nishiyama, Inaba & Murata, 2000). Under salinity conditions, water uptake by plants is inhibited due to low soil water potential; plants also suffer physiological and biochemical disturbances and oxidative stress from toxic effects of Na^+ and Cl^-; there's also a decrease in internal availability of nutrients due to changes in the absorption of essential nutrients (Hasegawa, Bressan, Zhu & Bohnert, 2000, Zhu, 2001, Munns, 2002).

E.N. Silva *et al*. revealed that *J. curcas* plants exhibited avoidance to salinity stress conditions by closing stomata (Silva, Ribeiro, Ferreira-Silva, Viégas & Silveira, 2010). Kumar *et al*. had also reported that NaCl induced an oxidative stress in callus (Kumar, Pamidimarri, Kaur, Boricha & Reddy, 2008). Patel, Panchal, Pandey and Pandey (2010b) discussed changes in tissues and whole-plant accumulation patterns of nutrients, as well as possible mechanisms for avoidance of sodium toxicity in response to salinity. According to Díaz-López *et al.* (2012), their work suggested that this crop showed a moderate tolerance to salinity using growth and Salt-Tolerance index (ST-index). Although there are several research studies on the reduction of biomass as a result of salinity stress above mentioned, no study focused on specific physical reduction effects, such as leaf shedding. This study therefore, aimed to evaluate *J. curcas* salt tolerance by observing growth, nutrient deposition and defoliation.

2. Material and Methods

2.1. Planting Materials and Experimental Conditions

J. curcas seeds imported from Tanzania were sown and grown for three weeks in small black tray nursery beds under greenhouse conditions at Tottori University, Japan. The seedlings were then transplanted into bigger plastic pots (20 cm height, 3.0 L) in sandy soil (soil characteristics are shown in Uzoma *et al*. 2011). The soils were mixed with chemical fertilizer (Total N: 0.44, P: 2.75, K: 0.57 g pot^{-1}). Eighty four days after sowing, NaCl irrigation was done at the following rates: 0 (control), 25, 50 and 100 mM. The pots were kept at field capacity using the corresponding NaCl irrigation water (volumetric soil water content; 8%). The pots were covered with aluminum foil to prevent evaporation from the soil surface. The amount of transpiration of each treatment was monitored by weighing pots bi-hourly from 6:00 to 18:00 every day. After measuring the final weight each day, NaCl irrigation was done to replace the lost water. Fallen leaves were promptly collected from each seedling and kept in sampling bags till the end of the cultivation cycle. Air temperature was maintained at 19－31 °C and relative air humidity at more than 55%.

2.2. Growth, Nutrient Deposition, and Harvesting of Samples

Sample harvesting was done at two levels, when average transpiration rate per hour at the peak of each treatment was less than 50%, and 75% as compared to that of the control. Inhibition rate (IR) at the 50 and 75% transpiration rates is denoted by $IR_{50\%}$ and $IR_{75\%}$ respectively. The final harvesting was carried on three seedlings for all treatments on Day 61; detailed harvesting patterns are shown in Table 1. The height and trunk diameter of each seedling were recorded at each harvesting time. Harvested seedlings were then separated into leaves, fallen leaves, stems (including branches), and root parts. All parts were then oven-dried at 80 °C for 48 h and biomass weighed. Water use efficiency (WUE; g dry biomass L^{-1}) was calculated as total dry weight per amount of cumulated transpiration for each seedling (Begg & Turner, 1976). All treatments were replicated three times.

The dried biomass parts were ground into fine powder, sieved and stored for various analytical procedures. For nutrient deposition, the powder (50 mg) was digested with a mixture of 1 mL 99% H_2O_2 and 1 mL of concentrated H_2SO_4 under heating. Total nutrients Na and K were determined using standard procedures on atomic absorption spectrometry (Z-2300, Hitachi, Tokyo, Japan).

2.3. Evaluation of Nacl Tolerance

Assessment of the seedling tolerance to salinity was done according to procedures suggested by Maas and Hoffman (1977), Steppuhn, van Genuchten and Grieve (2005a), and Díaz-López *et al.* (2012), Relative total dry weight (the total dry weight after NaCl irrigation treatment － the total dry weight before NaCl irrigation treatment) is denoted as yield (Y). Relative yield (Yr) was determined using the formula; Yr = Y / Ym where Ym is the relative total dry weight in 0 mM (control). After the calculation of Yr according to Díaz-López *et al.* (2012), an exponential model was used to evaluate the NaCl tolerance. $Yr = a * e^{b*ECi}$ where, ECi is the electrical conductivity of irrigation water, 'a' and 'b' are constants. Additionally, Salt-Tolerance index (ST-index) was calculated, and used as an indicator for inherent tolerance of the crop to root-zone salinity (Maas & Hoffman, 1977, Steppuhn *et al.*, 2005a). ST－index = EC_{i50} (1 + b) where, EC_{i50} is the electrical conductivity of irrigation water that reduces the yield to 50% of the maximum yield.

2.4. Statistical Analysis

Each experimental data were subjected to statistical analyses using R version 2.12.0 (The R Foundation for Statistical Computing). Treatment differences were assessed using ANOVA at $p < 0.05$ level of significance, while treatment means were separated using Tukey's multiple-range test. Linear and curve regression analyses were used to check for trends in the residuals.

3. Results

3.1. Harvesting Time

Growth rate under $IR_{50\%}$, 100 mM treatment was highest, reaching harvesting at day 4, followed by 25 and 50 mM (Table 1). However, there were no significant differences in the amount of NaCl accumulated at that time. The harvesting time in $IR_{75\%}$, 100 mM treatment followed a similar tendency as $IR_{50\%}$ above; additionally, NaCl irrigation induced significant differences in the amount of accumulated NaCl at that time. On the last harvesting day (Day 61), 100mM treatment showed the highest level of accumulated NaCl; this increased with NaCl concentration, and was significantly higher as compared to the other treatments.

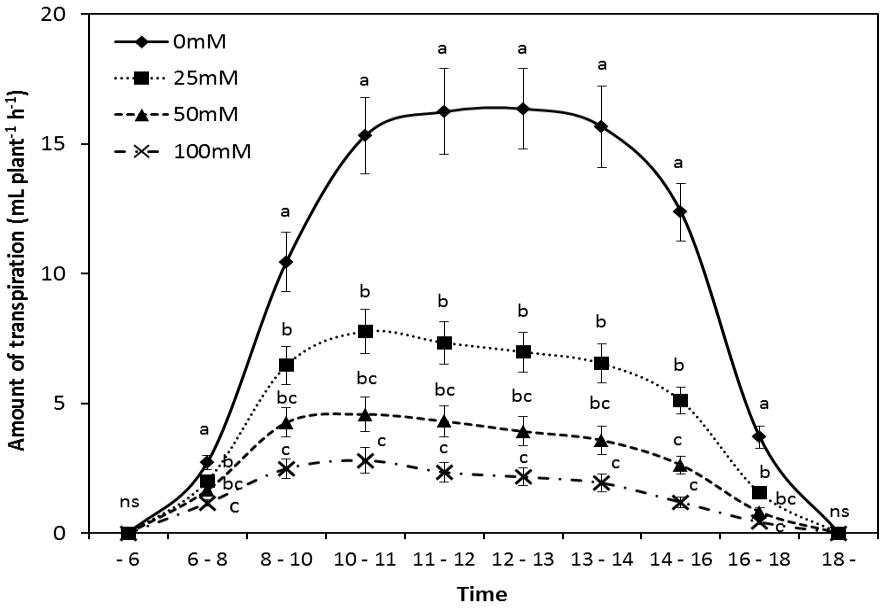
Table 1. Harvesting days and amount of cumulated NaCl in irrigation water

Havest time	Treatment (mM)	Harvest day	NaCl (g)
$IR_{50\%}$	25	13	2.31 a
	50	5	1.46 a
	100	4	1.66 a
	L		ns
$IR_{75\%}$	25	55	5.50 a
	50	20	4.41 b
	100	5	1.83 c
	L		***
Final harvest	0	61	0.00 d
	25	61	5.26 c
	50	61	6.53 b
	100	61	7.28 a
	L		**

Note: **and*** indicate significant differences at $P< 0.05$ and 0.001, and ns indicates not significantly different. Means with the different letters are significantly different by Tukey's multiple range tests ($P< 0.05$). L indicates linear regression analysis.

3.2. Transpiration Rate

The highest transpiration rate was 16.36 ± 1.56 mL at 12-13 o'clock in 0 mM treatment (Figure 1); in addition, this treatment was significantly different from other treatments at 6-18 o'clock, though there were no significant differences before 6 and after 18 o'clock. The time zone of 10-11 o'clock showed the highest amount of transpiration, 7.77 ± 0.87, 4.58 ± 0.68, 2.80 ± 0.50 mL h^{-1} respectively in the plants irrigated with 25, 50 and 100 mM NaCl treatments. There were significant differences between 25 mM and 100 mM treatments at 6-18 o'clock, though 50 mM treatment had no significant difference with 100 mM treatment throughout.



Note: Bars represent standard errors (n = 3); 'means' with different letters are significantly different; 'ns' indicates non-significant difference by Turkey's multiple range tests ($P< 0.05$).

Figure 1. Seedling transpiration rate during different time zones

3.3. Growth Rate

In $IR_{50\%}$ at harvest, height and trunk diameter showed no significant differences among the NaCl irrigation treatments (Table 2). However, the leaf dry weight had a strong correlation with salinity level at that time. In $IR_{75\%}$, dry weight was significantly affected by salinity stress in all parts except leaves. At the end of cultivation, highest total dry weight yield and cumulative transpiration was observed in 0 mM treatment, though the minimum value for water use efficiency (WUE) was also observed in this treatment. Throughout the entire experiment, specific leaf area (SLA) tended to decrease with cultivation time. Cumulative transpiration showed high correlation with level of salinity at all harvesting times. All parameters measured strongly correlated with salinity stress as cultivation time progressed.

Table 2. Seedling growth and yield characteristics, and water-use dynamics during cultivation

Harvest time	Treatment	Height	Trunk diameter	Dry weight				SLA	Amount of cumulated	WUE
				Leaf	Stem	Root	Total			
	(mM)	(cm)	(mm)	(g)				($m^2\ g^{-1}$)	transpiration (L)	($g\ DW\ L^{-1}$)
$IR_{50\%}$	25	43.03 a	22.23 a	7.14 a	19.88 a	5.61 a	32.63 a	0.32 b	1.51 a	9.60 a
	50	44.00 a	22.03 a	6.45 a	15.90 ab	4.22 a	26.57 ab	0.37 a	0.48 b	18.30 a
	100	39.07 a	22.43 a	5.12 b	13.86 b	4.30 a	23.29 b	0.35 ab	0.28 b	19.58 a
	L	*	ns	**	*	ns	*	ns	**	ns
$IR_{75\%}$	25	45.40 a	27.75 a	6.67 a	30.08 a	12.17 a	48.92 a	0.31 b	3.46 a	8.95 a
	50	42.27 a	21.50 b	6.16 a	18.07 b	5.31 b	29.54 b	0.40 a	1.39 b	8.36 a
	100	38.13 a	19.03 b	5.52 a	10.63 c	3.30 b	19.45 b	0.38 ab	0.32 c	4.71 a
	L	*	**	ns	***	**	***	ns	***	ns
Final harvest	0	46.53 a	30.00 a	8.73 a	37.29 a	15.65 a	61.67 a	0.19 b	6.85 a	6.39 c
	25	46.63 a	26.78 ab	8.24 a	28.36 b	11.23 b	47.83 b	0.22 ab	3.30 b	9.08 b
	50	45.73 ab	24.83 b	7.37 a	23.21 c	9.11 bc	39.70 c	0.25 a	2.01 c	10.91 ab
	100	43.37 b	23.02 b	6.49 a	17.05 d	7.12 c	30.66 d	0.25 a	1.08 d	11.68 a
	L	**	***	*	***	***	***	**	***	***

Note: Values are the mean of 3 samples; *, ** and *** indicate significant differences at $P< 0.05$, 0.01 and 0.001 and ns indicates not significantly different. Means with the different letters are significantly different by Tukey's multiple range tests ($P< 0.05$). L indicates linear regression analysis.

3.4. Growth Inhibition

At the final harvesting time, $IC_{50\%}$ (Inhibitory concentration of 50% dry weight - half as compared to that under the control) occurred at 54 mM NaCl irrigation treatment (Figure 2). EC_{i50} (EC of NaCl reducing yield by 50% of the maximum possible yield) and the salt-tolerance index (ST-index) were 5.63 dS m^{-1} and 6.23 dSm^{-1} respectively (Figure 3).

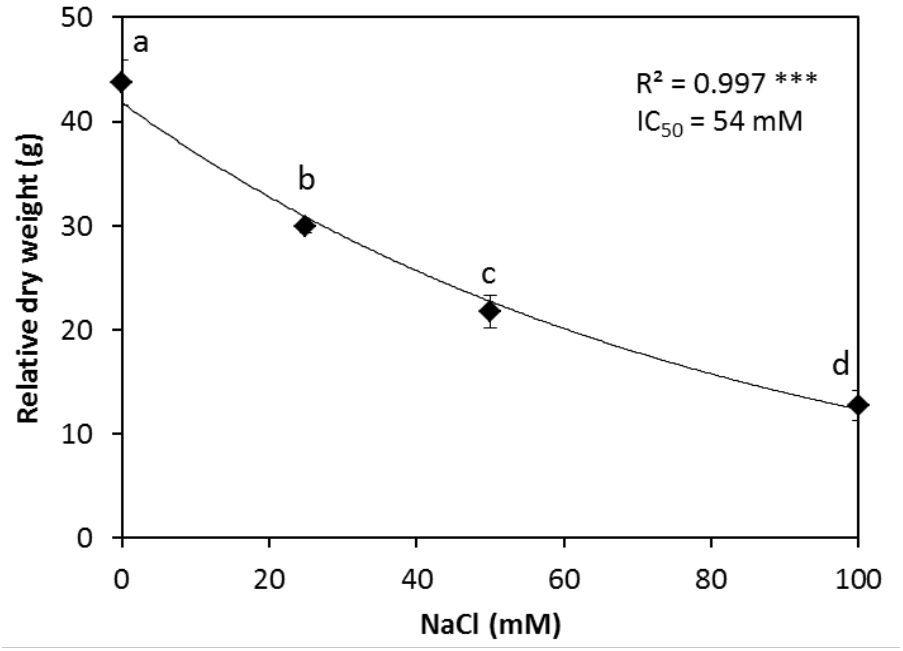
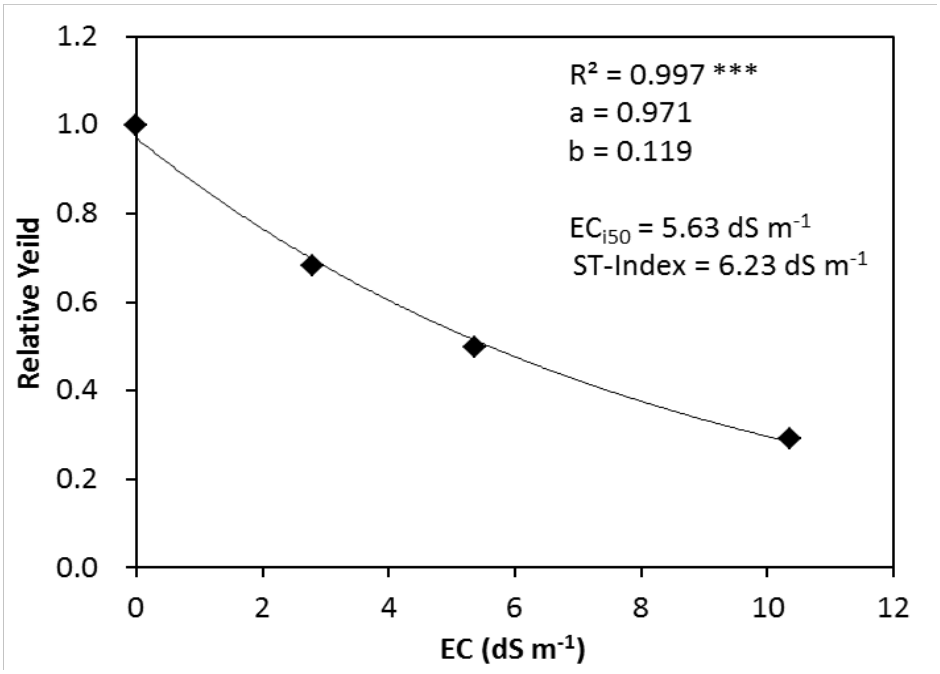


Figure 2. Relative biomass yield as affected by NaCl irrigation at final harvest



Note: *** indicates significantly different at $P< 0.001$.Bars represent standard errors (n = 3). Means with the different letters are significantly different by Tukey's test ($P< 0.05$). "a" is a constant that reflects the shape of the curve, and "b" is always negative and defines the intensity of the model.

Figure 3. Relative yield as influenced by NaCl irrigation at final harvest

Table 3. Na and K of concentration and Na/K in different parts of *J. curcas* seedlings at different harvest times

Part	Harvest time	Treatment (mM)	Na (g kg^{-1} DW)	K (g kg^{-1} DW)	Na/K
Leaf	$IR_{50\%}$	25	9.88 a	17.00 a	0.63 a
		50	7.83 b	17.86 a	0.56 a
		100	7.66 b	16.15 a	0.60 a
		L	**	ns	ns
	$IR_{75\%}$	25	20.25 a	11.57 b	1.86 a
		50	13.18 b	14.62 ab	0.90 b
		100	8.00 c	17.59 a	0.50 b
		L	***	*	***
	Final harvest	0	3.07 c	8.75 b	0.41 b
		25	19.92 b	11.96 ab	1.66 a
		50	22.28 ab	12.69 ab	1.76 a
		100	25.28 a	15.10 a	1.68 a
		L	**	**	*
Stem	$IR_{50\%}$	25	7.08 a	20.19 a	0.35 a
		50	3.29 b	16.57 a	0.20 b
		100	4.63 b	21.41 a	0.22 b
		L	ns	ns	**
	$IR_{75\%}$	25	12.63 a	17.17 a	0.73 a
		50	13.25 a	22.43 a	0.59 b
		100	5.69 b	23.15 a	0.25 c
		L	**	ns	***
	Final harvest	0	0.31 b	9.68 c	0.03 b
		25	9.96 a	15.42 bc	0.62 a
		50	13.90 a	21.82 ab	0.61 a
		100	16.87 a	25.02 a	0.66 a
		L	***	***	***
Root	$IR_{50\%}$	25	10.13 a	14.72 ab	0.68 a
		50	10.53 a	13.67 b	0.77 a
		100	12.77 a	17.58 a	0.73 a
		L	ns	*	ns
	$IR_{75\%}$	25	11.58 a	8.40 b	1.38 a
		50	12.42 a	11.45 ab	1.08 b
		100	11.88 a	14.43 a	0.83 c
		L	ns	**	***
	Final harvest	0	2.23 c	10.56 a	0.21 b
		25	11.23 b	8.56 b	1.31 a
		50	14.06 ab	8.56 b	1.64 a
		100	16.13 a	8.45 b	1.93 a
		L	***	*	***

Note: *, ** and *** indicate significant differences at $P< 0.05$, 0.01 and 0.001 and ns indicates not significantly different.

3.5. Mineral Nutrient Deposition

At $IR_{50\%}$ and $IR_{75\%}$, at same transpiration inhibition rate, highest leaf Na concentration was found in the 25 mM treatment, and this was significantly different from other treatments (Table 3). Leaf Na concentration was generally high at all harvesting times. Meanwhile, K was predominant in the stem, thus Na/K ratio was lower as compared to other parts.

3.6. Defoliation

Table 4 shows soil and leaf cumulated Na concentration under the different NaCl treatments at the time of 1st, 2nd and 3rd defoliation. After onset of initial defoliation, seedlings continued to shed leaves in all treatments. The number of fallen leaves increased with soil and leaf cumulated Na concentration; initial defoliation of the 1st – 3rd leaves however, appeared uncorrelated to soil and leaf Na concentration. Amount of Na in soil (irrigation water) and fallen leaves in *J. curcas* seedlings at the time of 1st, 2nd and 3rd leaf falling showed no significant difference under different NaCl irrigation treatment.

Table 4. Soil and leaf Na concentration under the different NaCl treatments at the time of the 1st , 2nd and 3rd defoliation

Treatment	1st		2nd		3rd	
(mM)	Soil (g)	Leaf (mg Dw)	Soil (g)	Leaf (mg DW)	Soil (g)	Leaf (mg DW)
25	0.94 a	2.40 a	1.23 a	3.12 a	1.33 a	3.39 a
50	1.03 a	2.61 a	1.70 a	4.32 a	1.79 a	4.55 a
100	1.31 a	3.32 a	1.34 a	4.76 a	1.87 a	4.76 a
L	ns	ns	ns	ns	ns	ns

Note: ns indicates not significantly different. Means with the different letter are significantly different by Tukey's multiple range tests ($P< 0.05$). L indicates linear regression analysis.

4. Discussion

Under high salinity conditions, plants experience water stress resulting from increased amounts of sodium which lowers osmotic potential of soil (Wang *et al.*, 2003). This condition causes plants a general reduction in size and dry matter production (Garg & Gupta, 1997, Taiz & Zeiger, 2006). In this study, seedlings became sensitive to salinity as NaCl accumulated in the root-zone (Table 1). The growth of Kenaf (*Hibiscus cannabinus*) under moderate salt stress is affected primarily through a reduction in elongation of the stem (Curtis & Lauchli, 1986); high concentration of salt induces root growth to slow down or stop elongation (Kramer, 1983, Patel *et al.*, 2010b) and causes reduction in root production (Garg & Gupta, 1997). Results of this study indicate that dry matter yield of all parts decreased as salinity concentration; all parameters except leaf dry weight were correlated with NaCl irrigation as days of treatment increased (Table 2). This result confirmed that the lowest dry matter yield reduction in response to increasing salinity occurs in the leaves (Patel, Jadeja & Pandey, 2010a); On the other hand, SLA (specific leaf area) increased with the increasing salinity solution.

Under saline conditions, plant growth is inhibited/reduced by: osmotic effect (change in plant water status), toxic effect (leading to physiological and biochemical disturbances and oxidative stress) and the nutritional imbalance effect (caused by alterations in absorption of essential nutrients) (Hasegawa *et al.*, 2000, Zhu, 2001, Munns, 2002). Focusing on plant osmotic effect, transpiration rate and water retention decreased under short-term NaCl stress in jute (Chaudhuri & Choudhuri, 1997). In addition, with increasing salt concentration, transpiration rate decreases

significantly in the halophyte *S. salsa* (Lu, Qiu, Lu, Wang & Kuang, 2002). In this study, the inhibition rate of transpiration increased dramatically on treatment with high NaCl concentration despite low amount of cumulated NaCl in the root zone (Figure 1). This observation suggested salt shock occurred in high NaCl treatments arising from high irrigation water salinity (Parida & Das, 2005). On the other hand, transpiration rate and amount of cumulated transpiration of *J.curcas* seedlings under salinity conditions were less than half as compared to the control condition (Figure 1, Table 2). This response by plants to minimize water loss through transpiration and salt loading is a typical mechanism observed in salinity resistant plant species (Steppuhn *et al.*, 2005a). After final harvesting, WUE (water use efficiency) of control was the lowest. Contrarily, highest WUE was recorded in the 100 mM treatment at final harvesting time. This is the reason why transpiration is negatively correlated with the instantaneous water-use efficiency in *J. curcas* seedlings (dos Santos *et al.*, 2013). According to Chaves and Oliveira (2004), at the beginning of water deficit, stomatal conductance decreases faster than the rate of photosynthetic decrease, which causes an increase in the instantaneous water use efficiency, whereas the intrinsic water-use efficiency is negatively correlated with stomatal conductance. Several authors have reported (in different species) that reduction in stomatal conductance caused an increase in the intrinsic water-use efficiency (Aasamaa, Heinsoo & Holm, 2010).

In the present study, IC_{50} of relative dry weight yield (as affected by NaCl) was 54 mM (Figure 2). The relative plant growth under increasingly saline environmental conditions in the root medium is an indicator of salt-stress tolerance (Parida & Das, 2005, Steppuhn, van Genuchten & Grieve, 2005b). *J. curcas* tolerated soil salinity of up to 7.9 dS m^{-1}, therefore this tree species could be categorized as moderately salt tolerant (Patel *et al.*, 2010b). EC_{i50} and ST-index were 5.63 dS m^{-1} and 6.23, respectively (Figure 3). While, *J. curcas* seedlings are able to grow well under up to 4 dS m^{-1} – irrigation water (30 mM NaCl) (Díaz-López *et al.*, 2012), EC_{i50} and ST-index in this study were 10.79 dS m^{-1} and 11.75 respectively. This level of salt tolerance is similar to that of grapefruit (EC_{i50} = 4.59dS m^{-1}, ST-index = 5.54), peanut (EC_{i50} = 4.61dS m^{-1}, ST-index = 6.65) or lemon (EC_{i50} = 5.54dS m^{-1}, ST-index = 6.56) (Steppuhn *et al.*, 2005a).

The stem and root stored a large amount of Na (probably by multiplying/replicating internal Na content of each organ and each dry weight) when highly concentrated NaCl irrigation water was used or a large amount of NaCl had accumulated in the soil (Table 3). High concentration of Na in the leaves causes plants to close stomata for mitigation of water loss (Naumann, Young & Anderson, 2007, Silva *et al.*, 2010). Besides, high concentration of Na^+ in the leaves decreases the photosynthetic potential (Mott, 1988). On the other hand, K is essential for cell expansion, osmo-regulation and cellular and whole plant homeostasis (Schachtman, Kumar, Schroeder & Marsh, 1997). The role of K in response to salinity is also well documented; Na depresses K uptake (Fox & Guerinot, 1998). An increase in Na content of cells is accompanied by a decrease in K accumulation and differences in Na/K ratio under saline conditions (Cherian & Reddy, 2003). In wheat, the salt tolerance is associated with low rates of Na transportation to shoots with high selectivity for K over Na (Gorham, 1990). The K takes part in many enzymatic activities in plant cell and maintaining Na/K ratio is a key requirement for growth under highly saline conditions (Apse, Aharon, Snedden & Blumwald, 1999). Salinity appeared to inhibit accumulation of K in the leaves, though it increased in the stem part. Thus, Na/K ratio of the stem was about 0.6, which is within the requirements for optimal metabolic efficiency in non-halophytic plants (Greenway & Munns, 1980).This behavior can be associated with Na/K-induced competitive inhibition of the absorption process (Lawlor & Cornic, 2002).

We also observed that EC_{50} of relative dry biomass was 54 mM, although even 25mM Na accumulation in soil and leaves led to leaf shedding (Table 4). The seedlings' response in minimizing transpiration water loss is stomatal closure induced by high Na concentrations in the leaves. Leaf growth inhibition increased with NaCl concentration, and so did the transpiration rate.

These changes could be part of an integrated mechanism of whole-plant acclimation to salt stress (Díaz-López *et al.*, 2012). This behavior would trigger alterations in leaf properties enabling plants to lower self-shade per unit leaf area and create a relatively short path from the stomata to the chloroplasts across which CO_2 diffuses (Flexas, Ribas-Carbó, Diaz-Espejo, Galmés & Medrano, 2008). Plants of this species sequester Na that they absorb in stems and roots and thus minimize the exposure of leaf cells (photosynthetic apparatus) to Na (Patel *et al.*, 2010b).Therefore; the defoliation was caused by NaCl irrigation treatment regardless of the low concentration. In general, defoliation delayed the growth of plants; thus, it could be suggested that there was a risk of seedling biomass reduction due to use of NaCl irrigation water.

5. Conclusion

This study showed that *J. curcas* can grow well in salinity levels of up to 54 mM, thereby falling in the moderately salt tolerant species category. EC_{i50} and ST-index showed that *J. curcas* is more salt tolerant than the typical Mediterranean crops. This plant withstands salt stress by retaining large amounts of Na in stem and roots vacuoles, thereby preserving the leaves. In particular, the low stem Na/K ratio of about 0.6 played an important role in optimizing metabolic efficiency in a non-halophyte plant. The leaf part changed characteristics according to SLA; seedlings minimized leaf-shedding by creating a relatively shorter path from the stomata to chloroplasts across which CO_2 diffuses. Although, *J. curcas* seedling biomass was able to generally tolerate some low levels of salinity, the leaf shedding occurred under even very low Na in the soil and leaves. The defoliation prevents growth of the plants, thus cultivating this plant under saline irrigation water conditions potentially posed a risk of seedling biomass reduction. Therefore, although this plant exhibited salt tolerance through structural adaptation, high levels of salinity are potentially lethal to growth and nutrient deposition. Future research should examine performance of this plant under long-term salinity conditions.

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