GROWTH OF *J. curcas* SEEDLINGS UNDER WATER DEFICIT CONDITION

CRESCIMENTO DE MUDAS DE *Jatropha curcas* EM CONDIÇÃO DE DÉFICIT HÍDRICO

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ABSTRACT: The elucidation of drought tolerance mechanisms in plants may facilitate the commercial exploitation of the species *J. curcas* in semi-arid regions. This study was designed to evaluate the growth and drought tolerance strategy of *J. curcas* plants grown under different water regimes. The work was carried out on an outdoor bench in full sunlight at Goiás State University, following the completely randomized experiment design with a 2x2 factorial scheme (two wild populations of *J. curcas*, naturally found in the states of São Paulo and Goiás, and two water supply regimes: 0% and 100% of evapotranspiration) and five replications. First a mixture of soil, sand and manure was prepared at the ratio of 3:1:0.5, respectively. The plants were irrigated daily, and at 60 days of age they were subjected to 10 days of water deficit and then rehydrated for five days. Under water deficit condition the *J. curcas* plants decreased the shoot growth, adjusted the leaf area to reduce the transpiration rate and increased the root system growth. *J. curcas* plants delay dehydration as a strategy to tolerate water deficit, for which the species reduces the transpiration rate and sustains tissue hydration using the water stored in the succulent stem.

KEYWORDS: Drought. Renewable energy. Oilseed.

INTRODUCTION

The predicted shortage of fossil fuels coupled with the growing concern for the environment has intensified the search for renewable energy sources. Therefore, it is critical to develop appropriate technologies and identify raw materials to be used as alternative energy sources, like biodiesel, with minimal damage to the environment (FREITAS et al., 2012; MATOS et al., 2009).

Brazil features great potential for biofuel production in most of its territory given the soil and climate characteristics, biodiversity (several potential biofuel-producing species adapted to different climates and biomes), availability of land and manpower, as well as proven expertise in the field of agricultural science (DIAS et al., 2008). In addition, Brazilian conditions perfectly meet cropping requirements, as the country is provided with abundant water and sunlight. Vegetable oils have been widely studied as candidates for renewable energy programs because they provide for decentralized generation of energy.

The main raw materials used for biodiesel production in Brazil are soybeans, animal fat and cotton, contributing 73.92%, 21.21% and 2.45% respectively, whereas other materials account for only 2.42% of this production (ANP 2014). There is a need, therefore, to diversify raw materials for biodiesel production through the introduction of promising species such as *J. curcas* L.

J. curcas (Euphorbiaceae) is an oilseed species originating from Central America, considered a rustic plant adapted to different environmental conditions. It is a species with great economic potential, particularly because its seeds constitute the raw material for the production of biodiesel oil, a feature that has contributed to increasing the commercial exploitation of its culture (DIAS et al., 2007).

The *J. curcas* plant develops under diverse climatic conditions, from very dry to moist tropical regions, tolerating rainfall levels between 600 and 1,500 mm year^{-1}. It is a xerophytic species with deciduous leaves and tolerance to lack of water; it can survive with 200 mm of annual rainfall up to three consecutive years of drought, slowing down growth in such periods. Since *J. curcas* oil can be used for the production of biodiesel, broad prospects open up for the expansion of planting areas for this crop in arid and semi-arid regions. Yet, commercial plantations in Brazil are still insignificant due to modest scientific knowledge (MATOS et al., 2014).

Little is known about the biochemistry and physiology of *J. curcas*, there are no defined cultivars and some agronomic aspects still need further research, such as tolerance to drought. Climate changes have increased water shortage periods in different regions of the world, and

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drought constitutes the main cause of low productivity of cultures worldwide. Researches on water deficit in *J. curcas* have focused on plants older than six months. Notwithstanding, experiments with young plants are relevant for assessing the plant performance at field planting age. Given the need for this kind of information for the production of *J. curcas* plants, as well as for better understanding of the attributes for drought tolerance, which will enable commercial exploitation under conditions of low water availability, the present study was designed to evaluate the growth and drought tolerance strategy of *J. curcas* plants grown under different water regimes.

MATERIAL AND METHODS

The experiment was carried out at Goiás State University, Ipameri Unity (Lat. 17º 43' 19" S, Long. 48º 09' 35" W, Alt. 773 m), Ipameri, Goiás. The completely randomized experiment design was used with a 2x2 factorial scheme (two wild populations of *J. curcas*, one being naturally found in the state of São Paulo and the other in Goiás, and two water supply regimes: 0% and 100% of the daily evapotranspiration) and five replications.

Firstly, a mixture of soil, sand and cattle manure was prepared at the ratio of 3:1:0.5, respectively. After the mixture was analyzed, fertilization and pH correction were carried out according to technical recommendations for the species (LAVIOLA and DIAS et al., 2008). The seeds were sown in plastic pots containing 8 kg of soil.

The seedlings were irrigated daily during 60 days with a water volume corresponding to the daily evapotranspiration, estimated according to recommendations in Allen et al. (2006). The plants were subjected to two treatments: compulsory 10-day water deficit (0% of daily evapotranspiraton) and subsequent 5-day rehydration in the first group and daily irrigation (100% of daily evapotranspiraton) in the second group. The treatments were designed to simulate a summer period in the field soon after the planting of seedlings, when they go on without water for a while and are, then, irrigated for analysis of their recoverability potential. Assessments occurred between 7:00 and 10:00 am and were done when the plants completed 10 days of drought and five days of rehydration.

Growth Variables

The plant height from neck to tip was measured with a graduated scale. The stem diameter was measured with a digital pachymeter at the base of the plant stem. The number of leaves was determined by counting all the leaves in each plant. The total biomass and root mass ratio (RMR) were measured by obtaining the dry weight. The leaf area was obtained by calculating the length and width of each leaf using the equation proposed by Severino et al. (2007).

Relative Water Content and Leaf Transpiration

To obtain the relative water content, six leaf disks with 12-mm diameter were extracted, weighed and placed to saturate in Petri dishes with distilled water for six hours. Then the disks were weighed again, dried at 70° C for 72 hours, and then the dry weight in grams was obtained. The relative water content was calculated using the formula proposed by Turner (1981). The daily transpiration was gravimetrically estimated, by determining the difference between the pots weight at one-hour intervals from 07:00 to 18:00 h, in accordance with Cavatte et al. (2012).

Photosynthetic Pigments

In order to determine the concentration of chlorophylls (Cl *a*+*b*) and total carotenoids, leaf discs with known area were extracted (third pair of fully expanded leaves) and placed in glasses containing dimethylsulfoxide (DMSO). Subsequently, extraction was performed in water bath at 65° C for three hours. Aliquots were obtained for spectrophotometric reading at 490, 646 and 663 nm. The contents of chlorophyll (Cl *a*+*b*) were determined according to the equation proposed by Wellburn (1994).

Statistical Procedures

The variables were subjected to variance analysis following the completely randomized experiment design with five replications. The parcel corresponded to a usable plant. The data were processed rehydration considering it as a single treatment to make it possible to make a comparison with treatments of 0 and 100% of the daily evaporation. The differences between the treatment averages were analyzed through the Newman-Keuls test at 5% probability using the SISVAR 5.3 software (FERREIRA, 2011).

RESULTS

The variance analysis indicated no significant difference between the populations of *J. curcas* (P > 0.05). In relation to the different water supplies, a significant difference could be observed in most variables analyzed.

The plant height (Figure 1A) was on average 25% higher in the irrigated plants compared to those subjected to water deficit. The stem diameter was on average 32% longer in the irrigated plants compared to those subjected to water deficit (Figure 1B). The rehydrated plants showed intermediate values. The number of leaves was on average 33% higher in the plants irrigated daily compared to those under water deficit (Figure 1C). The rehydrated plants showed intermediate values of plant height, stem diameter and number of leaves over the plants under drought and and irrigated daily. Despite the slight variation, there was no statistical difference in total biomass between the treated plants (Figure 1D).

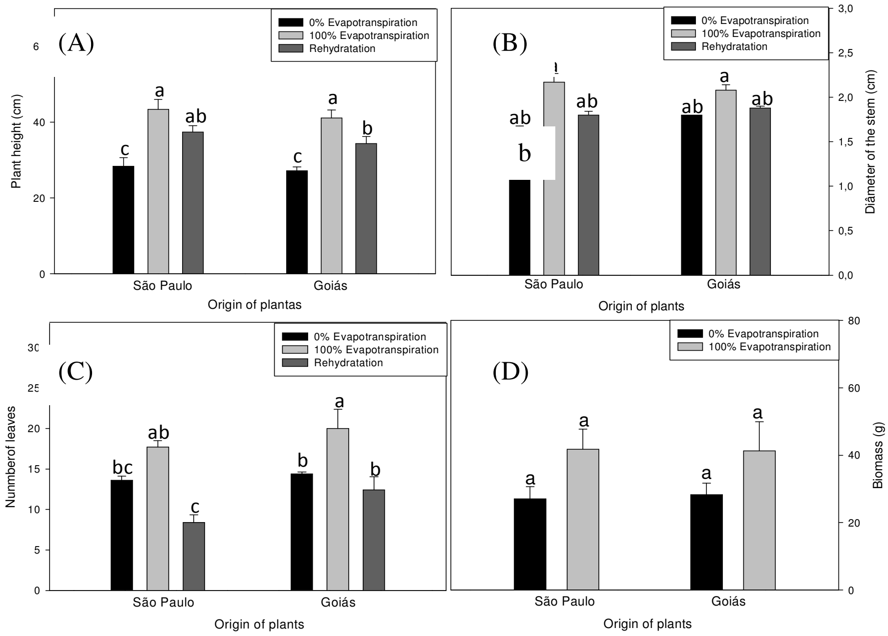

Figure 1. Plant height (A), stem diameter (B), number of leaves (C) and plant biomass (D) of *J. curcas* naturally found in the states of São Paulo and Goiás and treated with different water regimes. The values represent the average ± standard error (n = 5). The bars followed by the same letter do not differ at 5% probability according to Newman-Keuls test. Ipameri, GO, 2013.

The total chlorophyll content was quite similar between treatments, showing no statistical difference (Figure 2A). The total carotenoid content was on average 13% higher in the plants under water deficit in relation to those irrigated daily (Figure 2B). The rehydrated plants presented higher carotenoid concentrations.

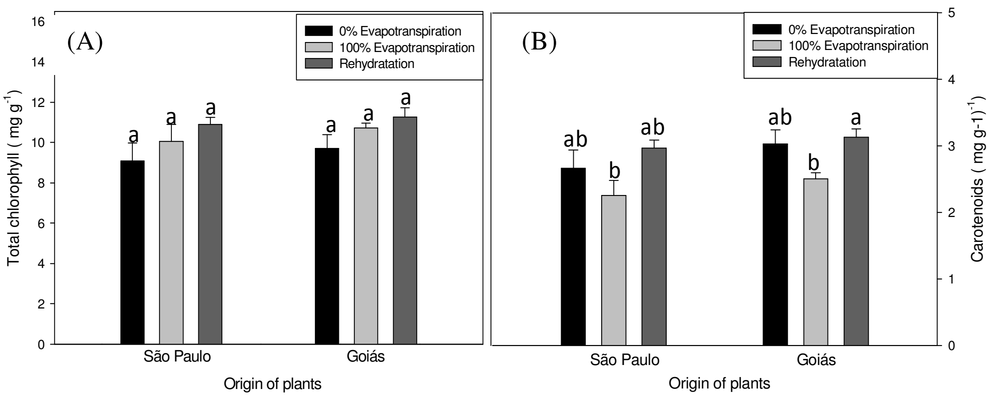

Figure 2. Chlorophyll (*a*+*b*) (A) and total carotenoids (B) of the *J. curcas* plants naturally found in the states of São Paulo and Goiás and treated with different water regimes. Statistics as shown in Figure 1. Ipameri, GO, 2013.

The transpiration rate was on average 40% higher in the plants irrigated daily compared to those under water deficit (Figure 3A). The relative water content had little variation between treatments, showing no statistical difference (Figure 3B). The leaf area was on average 41% larger in the irrigated plants compared to those under water deficit (Figure 3C). The rehydrated plants showed values even lower than those for plants under water deficit. Despite the slight variation, there was no statistical difference in root mass ratio between the treated plants (Figure 3D).

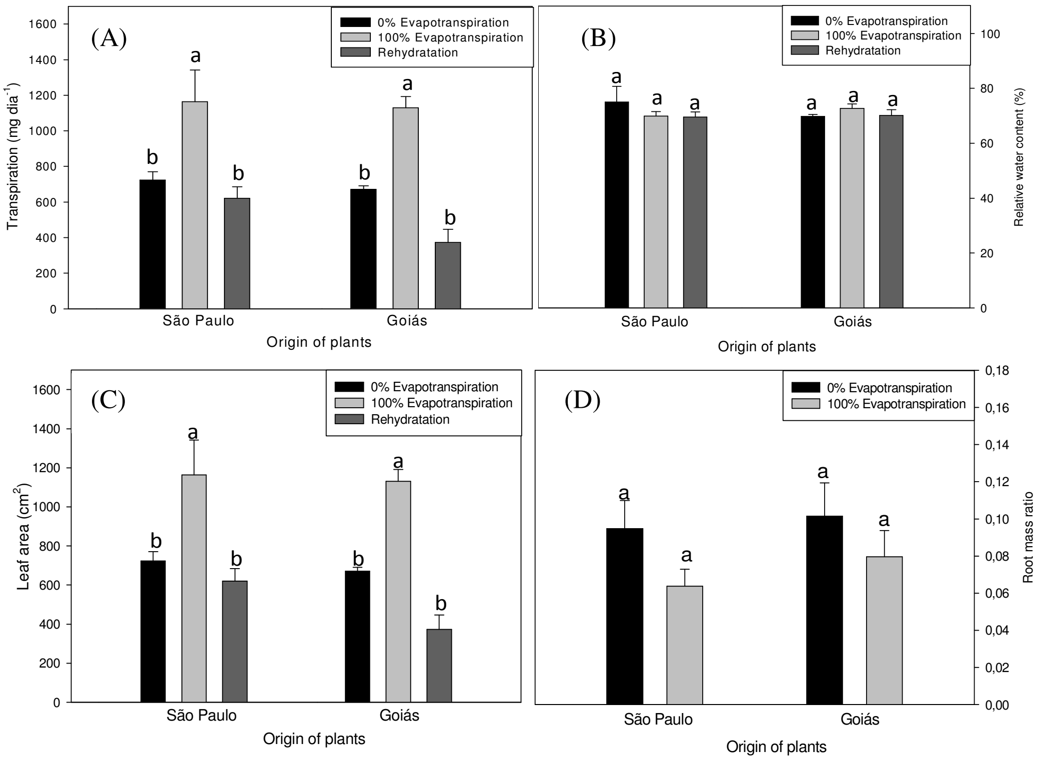

Figure 3. Leaf transpiration (A), relative water content (B), leaf area (C) and root mass ratio (D) of *J. curcas* plants naturally found in the states of São Paulo and Goiás and treated with different water regimes. Statistics as shown in Figure 1. Ipameri, GO, 2013.

DISCUSSION

Although *J. curcas* oil is recognized as ideal for the production of biodiesel, which can partially replace conventional diesel in a sustainable way, the species remains in need for scientific information. A reliable evaluation of the species performance requires agricultural, climatic and physiological knowledge, which is not yet available (TRABUCCO et al., 2010).

The budding and development of the leaf primordium depend on the amount of water supplied to the plant. The water deficit reduced its availability for the plant metabolism, such as formation and development of new leaves. Reducing the number and area of leaves is an important strategy for tolerance to water deficit because it reduces the perspiring surface. The adjustment of the total leaf area and leaf number was necessary to withstand the imposed stress and acclimatize to the new condition. The reduction in plant height, stem diameter and total biomass under water deficit conditions is relatively common in *J. curcas* plants (DIAZ - LOPEZ et al., 2012b). Probably the reduced stomatal opening to minimize perspiration compromised the inflow of CO_2 and the production of assimilates for the stem growth. The water deficit stimulated new metabolic and/or structural capabilities mediated by the alteration of gene expression (SHARP et al., 2004).

Despite the lack of statistical difference, there was a slight decrease in the concentration of chlorophyll in plants under water deficit. Possibly the chlorophyll degradation is associated with leaf senescence in plants under water deficit. The presumable higher concentration of carotenoids in treated and rehydrated plants is related to the efficient mechanism for the species photoprotection, as reported by Pompelli et al. (2010).

The reduced transpiration rate in plants deprived from water and then rehydrated associated with similarity in relative water content indicates that the species slowed down dehydration of the plant tissues. The juicy stem functioning as a water buffer associated with the water deficit anticipation mechanism, typical of isohydric plants, contributed to maintaining the high water content in the leaves. The reduction in leaf area helped minimize water losses and maintain the tissues hydrated. In addition, greater input in the root system possibly contributed to maximizing water absorption. Water deficit caused morphological and anatomical changes in plants enough to unbalance the water absorption and transpiration rate. Among the morphological changes, the reduction in leaf number and size was the most significant (DIAZ - LOPEZ et al., 2012a).

Fewer leaves, smaller leaf area, chlorophyll degradation and leaf senescence associated to lower transpiration rate affected the carbon assimilation net rate in plants subjected to water deficit, which resulted in lower biomass accumulation. Nevertheless, one should note that all the plants survived and showed signs of recovery after rehydration. The reduction in the number of leaves, leaf area and other parameters after rehydration was related to the plant short recovery time after the water deficit.

CONCLUSIONS

Under water deficit conditions the *J. curcas* plants slow down the growth of the aerial part, adjust the leaf area to reduce the transpiration rate and increase the growth of the root system.

The strategy of *J. curcas* plants to tolerate water deficit is to delay dehydration, for which the species reduces the transpiration rate and sustains tissue hydration using the water stored in the succulent stem.

RESUMO: A elucidação dos mecanismos de tolerância a seca poderá viabilizar a exploração comercial da espécie *J. curcas* em regiões semi-áridas. Pretendeu-se avaliar o crescimento e a estratégia de tolerância a seca de plantas de *J. curcas* cultivadas sob diferentes regimes hídricos. O trabalho foi conduzido em bancada a pleno sol na Universidade Estadual de Goiás, seguindo o delineamento experimental inteiramente casualizado, em esquema fatorial de 2x2 (duas populações silvestres de *J. crucas* encontradas naturalmente nos estados de São Paulo e Goiás e dois suprimentos hídricos: 0% e 100% da evapotranspiração) e cinco repetições Inicialmente foi preparada uma mistura de solo, areia e esterco na proporção de 3:1:0,5, respectivamente. As plantas foram irrigadas diariamente e aos 60 dias de idade, as mesmas foram submetidas a 10 dias de déficit hídrico e, em seguida, reidratadas por cinco dias. Em condição de déficit hídrico as plantas de *J. curcas* reduzem o crescimento da parte aérea, ajustam a área foliar para reduzir a taxa transpiratória e intensificam o crescimento do sistema radicular. As plantas de *J. curcas* apresentam como estratégia de tolerância ao déficit hídrico, o retardo da desidratação, para tal, a espécie reduz a taxa transpiratória e matem a hidratação dos tecidos utilizando a água armazenada no caule suculento.

PALAVRAS-CHAVE: Seca. Energia renovável. Oleaginosa.

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