

## Ecophysiological performance of eight *Jatropha curcas* L. provenances cultivated in Tunisia

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### Abstract

Bio-diesel crops are presented as a potential tool to mitigate global warming. However, these crops are often highly water consuming, which limits their use in semi-arid areas. In this respect, the *Jatropha* is considered by many researchers as the most appropriate species in these dry conditions. The aim of our investigation was to study the possibility of its use in Tunisia regarding its behavior in semi-arid area of the north-west region. Eight provenances of *Jatropha curcas* L. introduced from Brazilia (5), Surinam (1), Mozambic (1) and Tanzania (1) were compared on the basis of their ecophysiological performance. Results showed variability in photosynthesis, leaf transpiration, chlorophyll content and leaf growth between accessions during the growing season. Average photosynthesis and leaf transpiration values ranged from 7 to 13  $\mu\text{mol m}^{-2} \text{s}^{-1}$  and from 2.5 to 3.5  $\text{mmol m}^{-2} \text{s}^{-1}$ , respectively. Specific dry matter per unit leaf area varied from 50  $\text{g m}^{-2}$  to 90  $\text{g m}^{-2}$ . Provenances from the Mozambic and Pranà with the lowest biomass per unit leaf area and high photosynthetic capacity are more likely to offer greater productivity in semi-arid zone.

**Key words:** Biodiesel crops, irrigated jatropha, semi-arid areas, genetic diversity, global warming

### Introduction

One of the causes of the global food crisis and the rising agricultural product's prices is increased area under bio-diesel crops at the expense of food crops (Sander *et al.*, 2010). This crisis coincides with the climate change phenomenon and the encouragement of substitutes for fossil fuels is considered as the main driver of the global warming (Ajay and Sudkher, 2000). However, there are limits for the intensification of bio diesel crops and their impact on the environment, especially because of excessive use of water, fertilizers and pesticides (Openshow, 2008). Compared to oil palm which is currently the main source of biodiesel, jatropha seems better because palm oil invades the good lands, at the expense of food crops and biodiversity. Moreover, jatropha can grow in semi-arid lands and even help fight against desertification. *Jatropha* is an evergreen shrub that can reach 5 m in height and lives more than 50 years; its seed contains 40% of oils (Martinez *et al.*, 2006; Kumer and Sharma, 2008). And after esterification, a fuel of molecular weight close to that of oil is produced.

Maes *et al.* (2009a) describe jatropha as a soft wood species with high water content of stem and trunk and low specific area. Achten *et al.* (2010) confirmed that the species develops a set of mechanisms for coping with water stress, such as, regulating stomata conductance; stopping stem and twig growth and reducing total leaf area. Therefore, jatropha appears to be a potential substitute for the semi-arid regions such as Tunisia (Saddem, 2009). In this perspective, an experimental protocol including eight provenances of jatropha was set up by the National Research Institute of Rural Engineering, Water and Forests in collaboration with the General Directorate of Forests and the AGROILS Company in different agro-ecological sites.

The objectives of our research were to understand the behavior and the performances of jatropha in terms of water and carbon use on one hand; and to quantify the leaf mass area as a criteria of classification of the tested provenances on the other hand.

### Materials and methods

**Experimental site and plant material:** The experimental field is situated in semi-arid zone near Nabeul (N=36°27', E=10°42', Alt=29 m) in a sandy soil. Plant material was represented by eight jatropha provenances from Brasilia, Surinam, Mozambic and Tanzania (Table 1). Seedlings were planted in randomized complete blocks with five replications per ecotype with a density of 2 x 3 m between rows. Plants were drip irrigated and average height was 1.3 m.

**Measurements and statistical analysis:** Five trees per ecotype and twelve leaves per tree, three per direction (north, south, east and west) were chosen for stomatal conductance (Gs), leaf transpiration (LT) and photosynthesis (PN) monitoring during two typical days within a growing season, 26 June 2009 and 2 Sept, 2009. These parameters were measured using a photosynthesis analyzer chamber Li-Cor (Nebraska, USA) and measurements

Table 1. List of provenances of *J. curcas* and number of trees cultivated in the experimental field near Nabeul

Code	Provenance	Number of trees
P1	Arusha – Tanzania	98
P2	Mozambic	73
P3	Pranà – Brazilia	70
P4	Morte de Minas – Brazilia	75
P5	Mato Grosso – Brazilia	66
P6	Regiao Sureste – Brazilia	50
P7	Vale do Fequitinhonla – Brazilia	49
P8	Surinam	21

were performed after a stabilization time of 30 min. Chlorophyll content index (CCI) was measured using a chlorophyll meter (CCM200, Opti-Sciences) on September, 2. Leaf length, width and area of collected leaves were taken using an ADC (Ejalkamp) area meter. For dry leaf matter, the same leaves were oven dried at 70 °C for 48 h and weighed after stabilization of the dry weight by a precision balance 0.1 g. The leaf mass area LMA (g/m<sup>2</sup>) of each sample was obtained by the ratio (dry matter/leaf area).

Statistical analysis was performed with ANOVA and means were compared using Newman and Keuls test at  $P=0.01$ .

## Results

**Climatic conditions during the 2009 growing season:** There was a shortage of precipitation during the months of May, June, July and August. This deficit was associated with a high evaporation (170 mm) during August. The water deficit was compensated by daily drip irrigation adopting the values of maximum evapotranspiration. Maximum temperature reached 40 °C on a daily scale, minimum temperature varied between 5 to 21 °C (Fig.1). The cumulative daily temperatures during the growing season were 4361 degree-day from May to October.

**Leaf photosynthesis, transpiration and chlorophyll content:** The data of photosynthesis were often characterized by a strong variability (Fig. 2) and mean values of PN were consistent with values observed in C<sub>3</sub> metabolism species (Holl *et al.*, 2007). Average values measured on June 26 were significantly higher

than those measured on September 2, they ranged from 7  $\mu\text{mol m}^{-2} \text{s}^{-1}$  (P1) to 12  $\mu\text{mol m}^{-2} \text{s}^{-1}$  (P6 and P8). However, we noted that provenances assimilated more carbon on 26 June did not match on 2 Sept, 2009.

Similarly, the values of leaf transpiration were quite variable with averages ranging from 2.5 to 3.5  $\text{mmol m}^{-2} \text{s}^{-1}$ . The values measured in late June were significantly higher than those of September (Fig. 3). Provenances with the highest leaf transpiration assimilated more carbon and *vice-versa*.

Chlorophyll content index varied from a minimum level of CCI=21 for provenances P2 and P4 to a maximum level of CCI=28 for provenance P1 (Fig. 4). Although differences were not significant, it may be noted that provenances P1 and P4, respectively showed the highest and the lowest photosynthesis levels in September.

**Leaf growth and leaf mass area:** Leaf growth is illustrated by the final leaf length (LL) and width (LW) that showed some variability between provenances (Fig. 5). LW varied from 36 (P6) to 43 cm (P8), while LL varied from 33 cm (P1) to 37 cm (P3 and P8). The ratio LL/LW ranged from 0.87 (P1) to 0.96 (P2)

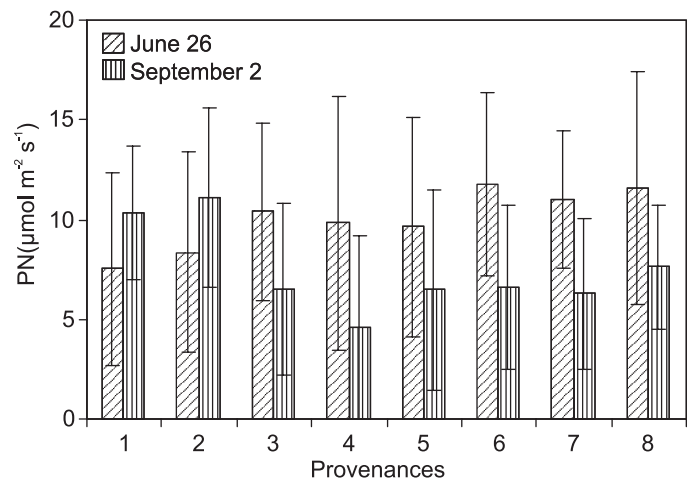


Fig. 2. Net photosynthesis (mean and standard deviation) measured on June 26 ( $T_a = 28\text{ }^\circ\text{C}$  and  $\text{PAR} = 1700\ \mu\text{E. m}^{-2} \text{s}^{-1}$ ) and September 2, 2009 ( $T_a = 32\text{ }^\circ\text{C}$  and  $\text{PAR} = 1400\ \mu\text{E. m}^{-2} \text{s}^{-1}$ ) for eight provenances of *J. curcas* cultivated in the experimental field near Nabeul.

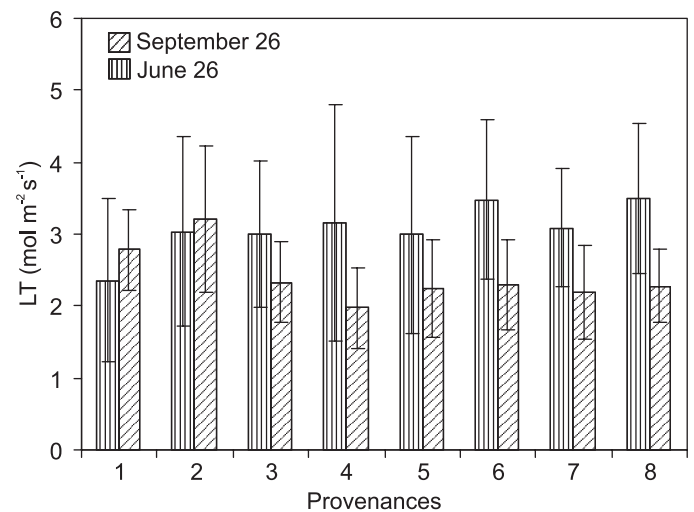


Fig. 3. Leaf transpiration, LT (mean and standard deviation) on June 26 ( $T_a = 28\text{ }^\circ\text{C}$  and  $\text{PAR} = 1700\ \mu\text{E. m}^{-2} \text{s}^{-1}$ ) and September 2, 2009 ( $T_a = 32\text{ }^\circ\text{C}$  and  $\text{PAR} = 1400\ \mu\text{E. m}^{-2} \text{s}^{-1}$ ) for eight provenances of *J. curcas* cultivated in the experimental field near Nabeul.

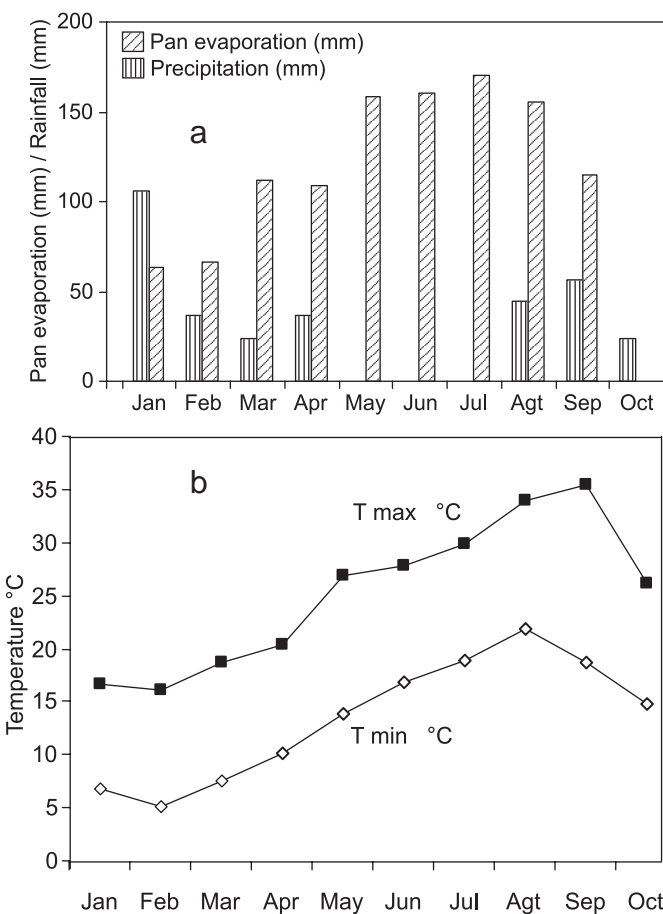


Fig. 1. Monthly changes of pan evaporation and rainfall (a) and min and max temperatures (b) measured in a nearby meteorological station from January to October 2009.

showing a wide variability in leaves' shape of the species between *jatropha* provenances.

Results in Fig. 6 showed clearly that provenances P4, P6 and P8 were able to produce more biomass per unit leaf area with LMA up to 90 g/m<sup>2</sup>, while provenances P1, P2 and P3 gave the lowest LMA of 50 g/m<sup>2</sup> and provenances P5 and P7 were intermediates. Finally, results illustrated by Fig. 6 showed three groups of provenances according to their biomass productivity (LMA). G1 (P4, P6, P8) with 90 g/m<sup>2</sup>, G2 (P5, P7) with 70 g/m<sup>2</sup> and G3 (P1, P2, P3) with 50 g/m<sup>2</sup>.

### Discussion

**Climate conditions and stomatal conductance effects:** Differences in the phenology, such as bud setting, maximum leaf area index, flowering and senescence may be causing differences in behavior between provenances according to their origins (Rao *et al.*, 2008). The variability of photosynthetic capacity may be related to a wide genetic variability of the species *J. curcas* as shown in previous studies reported by Kaushik *et al.* (2007) and Ram *et al.* (2008).

Regarding the climatic conditions, we noted that June was dry, with medium temperatures ( $T_{max} = 28\text{ }^{\circ}\text{C}$ ) while September was rainy with highest temperatures ( $T_{max} = 35\text{ }^{\circ}\text{C}$ ). This could partly explain the differences in measured photosynthesis and transpiration between the two periods. Maes *et al.* (2009) showed some disparity of photosynthesis according to climate parameters.

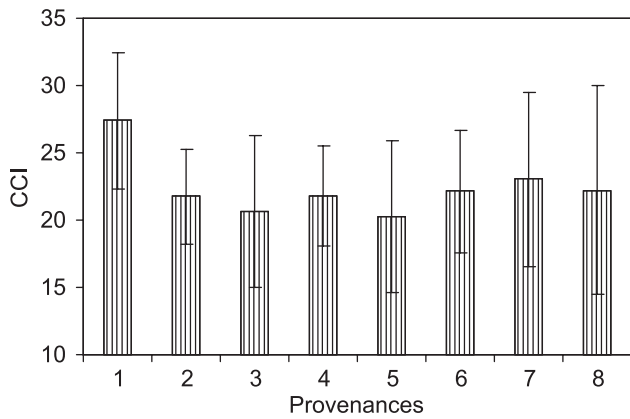


Fig. 4. Chlorophyll content index (CCI) measured in September for eight provenances of *J. curcas* cultivated at Nabeul

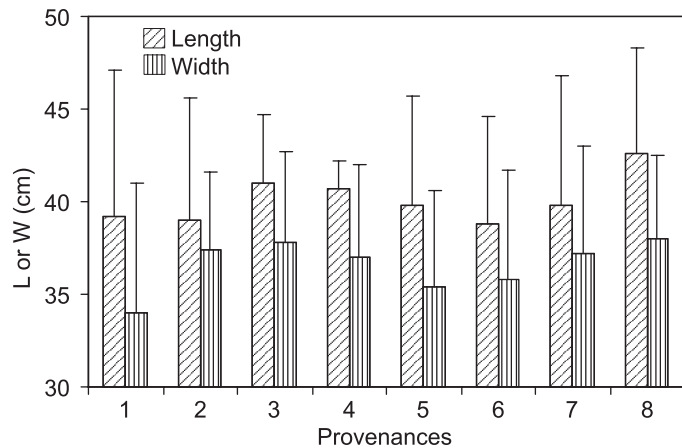


Fig. 5. Final leaf length and width (white) measured for eight provenances of *J. curcas* cultivated in the experimental field near Nabeul.

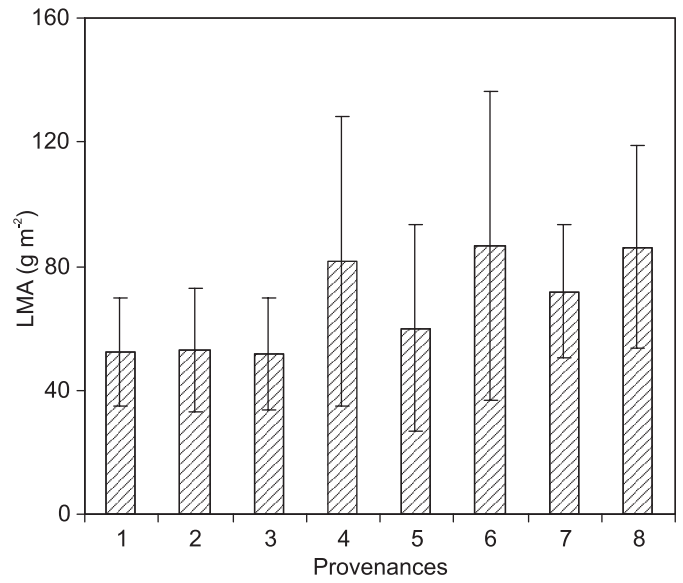


Fig. 6. Changes of final dry matter produced per unit leaf area (LMA, g/m<sup>2</sup>) in *J. curcas* collected from eight provenances grown at Nabeul

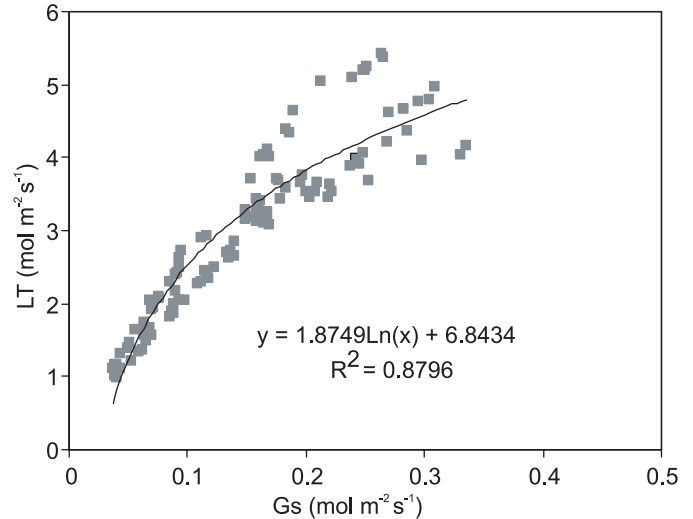


Fig. 7. Relationship between LT and Gs (average of 15 individuals) in June 26, 2009 and September 02, 2009 in *J. curcas* collected from eight provenances grown at Nabeul

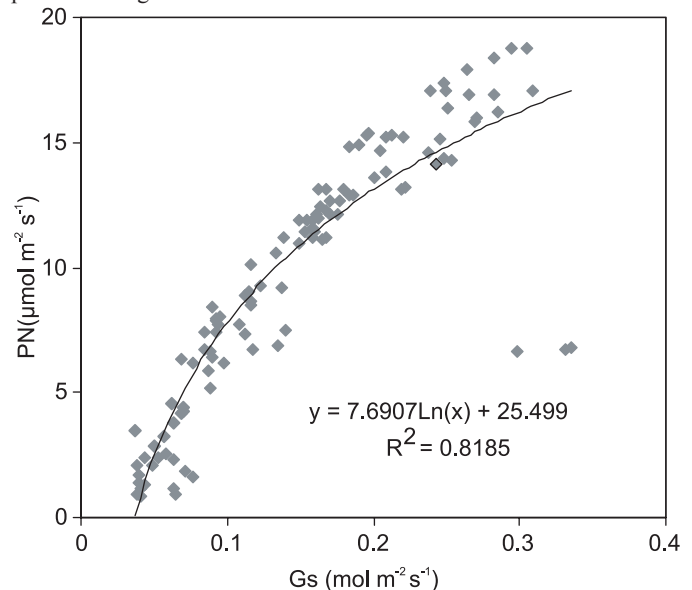


Fig. 8. Relationship between PN and Gs (average of 15 individuals) in June 26, 2009 and September 02, 2009 in *J. curcas* collected from eight provenances grown at Nabeul

Relationships between PN, LT and Gs show that these parameters are highly correlated (Fig. 7). This strong correlation was also confirmed on *Jatropha* by Ainsworth and Rogers (2007). These correlations indicate that any factor inducing stomata closure, such as water stress or high vapor deficit involves a decrease of PN and LT. This decline is not quite linear, it is small for elevated Gs and *vice versa*. It seems to be more important for PN than for LT (Fig. 8). When Gs decreased from 300 to 100 mmol m<sup>-2</sup> s<sup>-1</sup>, PN decreased from 17.2 to 6.2 μmol m<sup>-2</sup> s<sup>-1</sup> and lost 60 % of its maximum value. At the same time LT dropped from 4.8 to 2.7 mmol m<sup>-2</sup> s<sup>-1</sup> and lost only 43 % of its initial. That means a short water stress could improve the water use efficiency of the species, such result should have a significant impact on the deficit irrigation management of the species in semi-arid area.

**Leaf mass area (LMA) for classification:** The LMA has been expressed as the ratio of accumulated biomass to the leaf area. It has been considered as a key indicator to understand and quantify the degree of stress that the crop is subjected to. Greater the LMA is, more the species or the variety is stressed. LMA was largely used in various forest ecology investigations dealing with changes of water stress and leaf nitrogen content for deciduous tree species (Takahashi and Kohyama, 2005) or to study the consequences of its variation in various environmental ecosystems (Pooter *et al.*, 2009). Shipley (2006) analyzed the relationship between LMA and the relative growth rate of 614 herbaceous and woody species. According to Sharma *et al.* (2009), the LMA can be a suitable parameter of provenances classification.

The present analysis showed an ecotype effect on LMA. There were also significant differences between trees of the same origin of the species *J. curcas*. The Newman-Keuls test was used. Provenances P1, P2 and P3 are those having the lowest LMA, provenances P4 and P8 those with the highest LMA and P5 and P6 were intermediates (Tables 2 and 3). The present investigation revealed differences in intra-specific response of gas exchanges and leaf mass area in *J. curcas*. These features may have important consequences regarding the choice of provenances of this species for cultivation in drought prone areas.

The measured values of photosynthesis and the transpiration in the species *J. curcas* are comparable to usual values of C<sub>3</sub> plants.

Table 2. Variance analysis for LMA of eight provenances of *J. curcas* cultivated in the experiment field near Nabeul

Variable	df	Sum of squares	Mean squares	F value	F <sub>prob</sub>
Provenance	8	24029.27	3432.75	1.92	0.1029
Tree (in provenance)	40	53918.68	1797.28	3.76	<0.0001
Error	114	54439.32	477.53		
Corrected total	151	132387.29			

Table 3. Classification based on Newman-Keuls test for LMA of eight provenances of *J. curcas* cultivated in the experiment field near Nabeul (Group: Newman et Keuls grouping; N: number of plants)

Group	Mean	N	Provenance
A	86.2	20	8
A	81.5	16	4
AB	71.7	20	7
B	63.9	20	6
B	60.0	20	5
B	52.9	20	2
B	52.1	20	1
B	51.5	20	3

Water relations show that leaf transpiration and photosynthesis are strongly controlled by stomata conductance and that there is a disparity between the provenances. According to our results, three groups can be distinguished. P1, P2 and P3 could be more suitable under semi arid climate than P4, P6 and P8; while P5 and P7 could be intermediates.

The LMA is a useful tool for classifying sources and providing information on various provenances to advocate a particular microclimate. Further researches with approaches integrating the whole plant during different stages of the growing season are needed to improve these preliminary results.

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