



Physiological and morphological responses of young mahogany (*Swietenia macrophylla* King) plants to drought

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ABSTRACT

Young mahogany (*Swietenia macrophylla* King) plants were grown under either well-watered (pre-dawn leaf water potential, Ψ_{pd} , ca. -0.40 MPa) or drought (Ψ_{pd} , ca. -3.52 MPa) conditions to examine some physiological strategies that allow the maintenance of leaf turgor. In well-watered plants, stomatal conductance (g_s) was nearly constant ($440 \text{ mmol m}^{-2} \text{ s}^{-1}$) between 7:00 and 13:00 h. This was accomplished by significant increases in transpiration (E) and apparent total hydraulic conductance (K_T), in which averages were higher at 13:00 h. From 13:00 to 17:00 h, g_s , E , and K_T decreased sharply, reaching their lower values at 17:00 h. In these plants, significant increases in height (116%), stem diameter (50%) and leaf area (200%) were registered over the experimental period (20 days). Analyses of linear regression between g_s or E and leaf-to-air vapor pressure deficit (Δ_w) were not significant. In water-stressed plants, g_s and E were higher at 7:00 h and lower from 9:00 to 17:00 h, while K_T was higher in early morning (7:00 h) and in late afternoon (17:00 h) than between 9:00 and 15:00 h. Moreover, both g_s and E decreased potentially ($P < 0.001$) with the diurnal increases on Δ_w . Drought also decreased leaf and leaflet numbers and reduced total leaf area, but had no effect on stem height and diameter. Leaf proline was higher (ca. 400%, between 13:00 and 15:00 h) in water-stressed plants, suggesting osmotic adjustment under drought. Twelve hours after resumption of irrigation, Ψ_{pd} was similar ($P > 0.05$) between well-watered and drought-stressed plants, suggesting an ability of plants to recover turgor after stress cessation. Altogether, our data support the hypothesis that young mahogany plants have the ability to satisfactorily tolerate or postpone drought.

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1. Introduction

During the past few years, reforestation projects have become increasingly important in the Brazilian Amazon region. Among a large variety of woody species native to the Amazon basin and under high exploitation pressure, mahogany (*Swietenia macrophylla* King) receives special attention because of its high commercial value. However, this species is still poorly studied in relation to its management in plantations and its physiological responses to environmental stresses.

Much of the Amazon region is prone to relatively long periods of dry season, when rates of evapotranspiration, by far, exceed rainfall. Besides, deforestation is believed to have already altered the regional climate, increasing monthly mean air temperatures

and altering rainfall patterns (Victoria et al., 1998; Marengo et al., 2000; Chagnon et al., 2004). The predicted increment on deforestation rates (Soares-Filho et al., 2006) as well as warmer temperatures and decreased precipitation during dry months, could manifest in longer and perhaps, more severe dry seasons for the Amazon region. Therefore, drought is an important stress factor limiting seedlings growth in the Amazon.

The effects of water deficit have been reported for a large number of angiosperms and gymnosperms, resulting on considerable changes in plant physiology, morphology and overall biochemical processes (Asada, 1999; Silva and Lemos Filho, 2001; Mittler, 2002; Dünisch et al., 2003; Pinheiro et al., 2004, 2005; Carvalho, 2005; Santos et al., 2006; Cernusak et al., 2007; Chaves et al., 2008). In terms of water relations, the mechanisms controlling water loss through stomata (transpiration) seem to be an efficient process to provide (or to maintain) leaf turgor under drought. This includes stomatal responses to soil water potential and fluctuating environmental conditions, in special to air

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humidity and temperature, radiation, and vapor pressure difference – VPD (Oren et al., 1999; Buckley and Mott, 2002; Mediavilla and Escudero, 2004; Sellin and Kupper, 2004; Pinheiro et al., 2005; Romero and Botia, 2006). Under drought, the maintenance of leaf turgor may also be achieved by the way of osmotic adjustment in response to the accumulation of proline, sucrose, soluble carbohydrates, glycinebetaine, and other solutes in cytoplasm improving water uptake from drying soil (Moghaieb et al., 2004; Carvalho, 2005).

In trees, drought effects should be more significant in the initial stages of plant growth (e.g., during the first year of cultivation) because of the shallow root system. This idea is in line with Dünisch et al. (2003), who found more negative leaf water potential in young (one-year-old) *Carapa guianensis* plants than in five- and eight-year-old plants. Although leaf water potential is not an index of stress severity, it is quite possible that more negative water potential may be associated with lower relative water content. As a consequence, plants could attenuate drought effects by increasing root system, as observed in drought tolerant clones of *Coffea canephora* (Pinheiro et al., 2005).

In this work, we aimed to evaluate the extent of the physiological and morphological responses of young plants of mahogany to short-term (20 days) drought. For this purpose, the diurnal-course of leaf water potential and gas exchange and its relationship to environmental conditions (in special to the fluctuating leaf-to-air vapor pressure difference) were evaluated when pre-dawn leaf water potential (Ψ_{pd}) of drought-stressed plants were around -3.5 MPa, simulating drought conditions that may occur in the Amazon region (Dünisch et al., 2003). The effects of drought in the leaf content of proline and in the above-ground morphology were also studied.

2. Methods

2.1. General

This research was carried out in a greenhouse, in Belém-PA (01°28'03"S, 48°29'18"W), north of Brazil. Mahogany (*S. macrophylla* King; Meliaceae) seedlings were selected according to their uniformity in relation to stem height and number of leaves and leaflets. The selected seedlings were planted in 16 L pots (one plant per pot), filled with 20 kg of yellow loam Latosol, adjusted to appropriate acidity and macro- and micronutrient contents using 1000 mL of Hoagland nutrient solution per pot (Hoagland and Arnon, 1950), fractionated into four applications (250 mL per application) every three days. Five days after the last application of nutrient solution, leaf fertilization with 0.05 g urea per plant was done. Irrigation was performed daily to maintain soil near field capacity by replacing evapotranspired water, estimated by weighing each pot just prior to watering. When 11-month-old plants were separated into two groups (water regimes): one continued to receive regular irrigation (control plants), and in the other watering was withheld (drought-stressed plants) until Ψ_{pd} of drought-stressed plants reached -3.52 ± 0.07 MPa, when physiological measurements and sampling for leaf proline were performed. Excepting for Ψ_{pd} , determined from 04:30 to 05:30 h, environmental and physiological assessments were taken throughout the day (7:00, 9:00, 11:00, 13:00, 15:00 and 17:00 h) over five consecutive days, and the average for each hour was used for plotting. After this period, all plants were irrigated (at about 17:00 h) and Ψ_{pd} was evaluated 12 h latter to examine the ability of drought-stressed plants to adjust pre-dawn leaf water potential to control level. Morphological analyses were performed at the moment of water regimes differentiation (Day 0) and twenty days from withholding irrigation (Day 20, $\Psi_{pd} = -3.52 \pm 0.07$ MPa). The experiment was arranged in a completely randomized design, consisting of two treatments (well-watered plants, Control; and drought-stressed

plants, Drought-stress), with ten replicates per treatment. A single plant per pot was considered as a replicate. Six plants were used for physiological evaluations; while the four remaining plants (completely intact) were used for morphological measurements and plant recovery, avoiding possible errors due to manipulation during physiological measurements.

2.2. Environmental measurements

Diurnal air temperature (T_{air}) and relative air humidity (RH) were obtained using a thermohygrometer (m5203, Incoterm Ind., Porto Alegre, Brazil) placed inside the greenhouse, and leaf-to-air vapor pressure deficit (Δ_w) was estimated according to Landsberg (1986), considering both air and leaf temperatures data. Photosynthetically active radiation (PAR) was measured with a quantum sensor attached to a steady-state porometer chamber. Measurements were performed in parallel with leaf water potential (Ψ_x) and gas exchange assessments.

2.3. Leaf water potential

Pre-dawn leaf water potential (Ψ_{pd}) and leaf-water potential at each time of day (Ψ_x) were evaluated as described by Pinheiro et al. (2008), using a Scholander-type pressure chamber (Model 670; PMS Instrument Co., Albany, OR, USA). As sample, one leaflet of the third leaf-pair from the apices was selected from six different replicates per water regime treatment.

2.4. Stomatal conductance, transpiration and apparent total hydraulic conductance

The diurnal-course of stomatal conductance to water vapor (g_s), transpiration (E), and leaf temperature (T_{leaf}) were evaluated using a portable steady-state porometer (Li-1600; LiCor Bioscience, Lincoln, NE, USA). During measurements, leaf chamber conditions tracked ambient air temperatures ranging from 25 to 32 °C. Apparent total hydraulic conductance [$K_T = (g_s \times \Delta_w) / (\Psi_{pd} - \Psi_x)$] was calculated using Ψ_{pd} to approximate soil water potential, and g_s and Δ_w were evaluated at the same time as Ψ_x (Hubbard et al., 1999; Donovan et al., 2000). Two healthy, mature leaflets from a single leaf, at the second or third pair from the apices were measured per plant.

2.5. Leaf proline content

Leaflet discs (ca. 1 g) were collected from a single leaf at the second or third pair from the apices and immediately dried at 72 °C for 48 h. After grounding, an aliquot of 50 mg (dry mass basis, DM) was vigorously homogenized in 5 mL distilled water, following incubation at 100 °C for 30 min. After centrifugation at $450 \times g$ for 20 min, the supernatant was reserved and the extraction was repeated twice. From the combined supernatants, an aliquot of 1 mL was used for determinations, as described by Bates et al. (1973). The absorbance of the sample was obtained at 520 nm, using a spectrophotometer (Genesys™ 10series, Thermo Electron Co. Madison, WI, USA). Proline content was calculated from standard curve, using purified proline as a standard. Results were expressed in $\mu\text{mol proline g}^{-1}$ DM.

2.6. Above-ground morphology

The total number of mature leaves and leaflets were directly counted, and plant height was determined as the length of the stem. The diameter of the stem was measured with a manual caliper (Model 3545605128; Vonder Co., Curitiba, PR, Brazil) through measurements performed at the basal, middle, and apical

sections of the stem. An average of measurements was used for treatment comparisons. Total leaf area was computed as total area of leaflets per plant using a portable leaf area meter (Model AM300; ADC Bioscientific Ltd., Hoddesdon, UK).

2.7. Statistical analyses

Physiological variables were assessed throughout the day (7:00, 9:00, 11:00, 13:00, 15:00 and 17:00 h) over five different days and averages for each time and (per replicate) were used for analysis. Data were tested for homogeneity of variance (Levene's test) and no transformation was necessary. Analysis of linear correlation was performed to evaluate possible relations between environmental and physiological variables with g_s or E and correlation coefficients were tested for significance by Student's t -test. The relationships between $g_s \times \Delta_w$ and $E \times \Delta_w$ were tested using regression analysis (F -test, $P \leq 0.05$). Data for morphological variables, total leaf area and leaf water potential after resuming irrigation, were subjected to analysis of variance and tested for significance by Newman–Keuls' test ($P \leq 0.05$).

3. Results

3.1. Environmental conditions

The environmental conditions recorded during the experiment represented a typical day in the tropical region where this study was carried out. RH decreased from 92% at 7:00 h to 58% at 15:00 h, followed by a slight increment up to 70% at 17:00 h (Fig. 1a). Similar diurnal-courses of T_{air} and T_{leaf} were observed, with maximum temperatures around midday (32–34 °C) (Fig. 1a). PAR was minimum at early morning (7:00 h) and late afternoon (17:00 h), with the maximum value at 13:00 h (Fig. 1b). Δ_w was ca. 0.3 kPa at 7:00 h (minimum value) and 1.92 kPa at 15:00 h (maximum value) (Fig. 1b).

3.2. Diurnal-course of physiological variables

For control plants, slight variation in Ψ_x was observed throughout the day, with higher values at 7:00 (–1.03 MPa) and at 17:00 h (–0.83 MPa), and the lower ones from 9:00 to 15:00 h (Fig. 2a). For these plants, stomatal conductance was sustained at high rates (ca. 440 mmol m⁻² s⁻¹) throughout the morning, and values did not differ significantly until 13:00 h. However, considerable decrease in g_s was observed from 15:00 to 17:00 h (Fig. 2b). Regarding the diurnal-course of transpiration, control plants showed low E at 7:00 h (1.97 mmol m⁻² s⁻¹) and at 17:00 h (1.34 mmol m⁻² s⁻¹), with maximum values at 13:00 h (7.57 mmol m⁻² s⁻¹) (Fig. 2c). A similar trend was observed for K_T (Fig. 2d).

Ψ_x of drought-stressed plants was always low, regardless of the time of day. This response was expected since the effects of drought were evaluated when Ψ_{pd} of drought-stressed plants were around –3.5 MPa. However, differently from control plants, no significant changes in Ψ_x were observed throughout the day, varying from –3.92 MPa at 7:00 h to –3.43 MPa at 17:00 h (Fig. 2a). Drought caused considerable decreases in g_s and values were higher (274.5 mmol m⁻² s⁻¹) at early morning (7:00 h) and completely suppressed from 9:00 to 17:00 h, reaching averages around 3.75 mmol m⁻² s⁻¹ (Fig. 2b). Decreased g_s caused expressive limitations in E , which observed values were lower than 2.0 mmol m⁻² s⁻¹ regardless of the time of the day (Fig. 2c). For K_T , the diurnal-courses of control and drought-stressed plants showed an opposite trend. In drought-stressed plants, K_T was higher at early morning (0.56 mmol m⁻² s⁻¹ MPa⁻¹), followed by significant

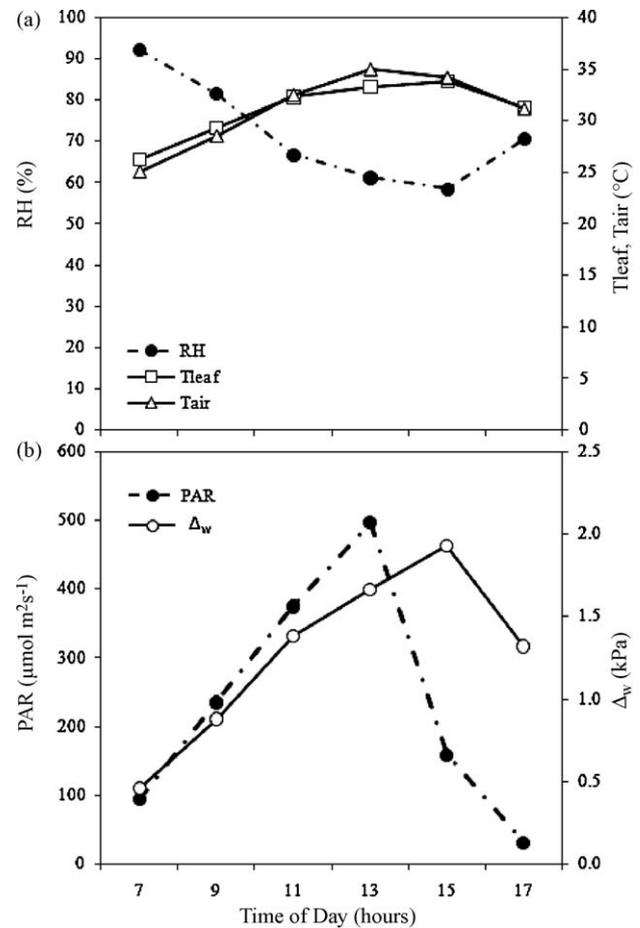


Fig. 1. Diurnal-course of relative air humidity (RH, a), leaf temperature (T_{leaf} , a), air temperature (T_{air} , a), photosynthetically active radiation (PAR, b), and leaf-to-air vapor pressure deficit (Δ_w , b) during measurements of leaf gas exchange throughout the day. Data for each variable at each time of the day are the mean of six replicates measured on five consecutive days \pm standard error.

decrease between 9:00 and 15:00 h. In the late afternoon (17:00 h), K_T did not differ from that recorded at 7:00 h (Fig. 2d).

3.3. Correlation analysis

In control plants, g_s was positively correlated to PAR ($P < 0.001$), E , Ψ_x , and K_T ($P < 0.05$), and negatively correlated with Δ_w ($P < 0.05$). No significant correlation was observed between g_s and others variables (Table 1). In drought-stressed plants g_s was positively correlated with RH, E , and K_T , and negatively correlated with PAR, Δ_w , T_{air} , and T_{leaf} . For these plants, non-significant correlation between g_s and Ψ_x was found (Table 1). Excepting to RH, in which E was negatively correlated, transpiration of control plants was positively correlated with PAR, T_{air} ($P < 0.001$), Δ_w , T_{leaf} ($P < 0.01$), Ψ_x and K_T ($P < 0.05$). In drought-stressed plants, positive correlations between $E \times RH$ and $E \times K_T$ and negative correlations between $E \times PAR$, $E \times \Delta_w$, $E \times T_{air}$, and $E \times T_{leaf}$ were found. For these plants, E was not correlated with Ψ_x (Table 1). These data pointed that in water-stressed plants both g_s and E were potentially responsive to diurnal fluctuations on Δ_w (Fig. 3). However, such relations were not significant in control plants. Thus, for drought-stressed plants, the increment on Δ_w , as observed from 7:00 to 9:00 h (0.52–1.03 kPa, respectively), caused 85% decrease in g_s and 73% decrease in E (as estimated by the regression analysis equations, see Fig. 3). Afterwards, additional increments registered on Δ_w (between 11 and 15 h) did not lead to further reductions in g_s or E (Fig. 3). At least in part, this response

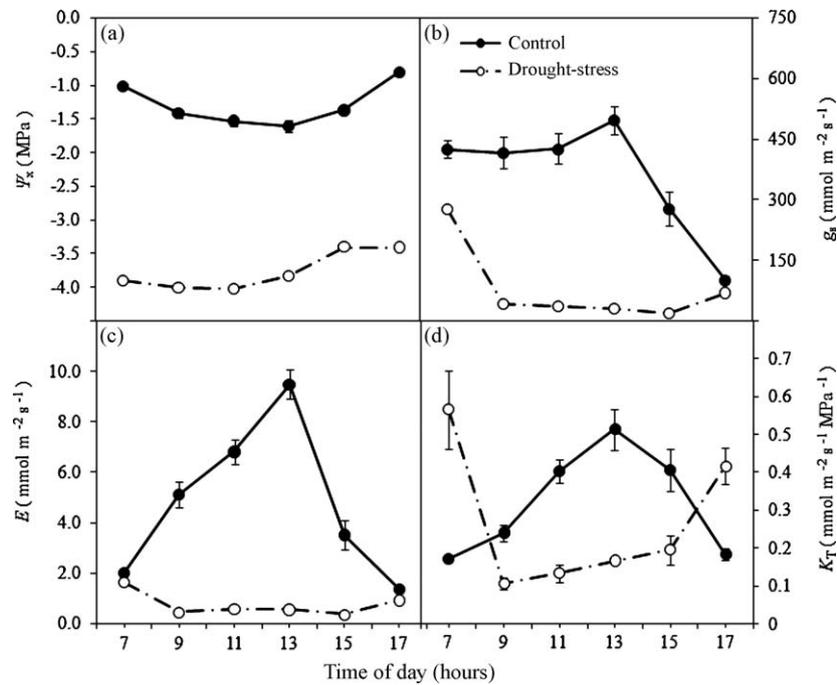


Fig. 2. Diurnal-course of leaf water potential (Ψ_x , a), stomatal conductance to water vapor (g_s , b), transpiration (E , c), and apparent total hydraulic conductance (K_T , d) in well-watered (Control, closed symbol and full line) and water-stressed (Drought-stress, opened symbol and dashed line) mahogany plants. Data for each variable at each time of the day are the mean of six replicates measured on five consecutive days \pm standard error.

was expected since an expressive correlation between g_s and E (0.95, $P < 0.001$) was evidenced in drought-stressed plants (Table 1).

3.4. Leaf proline content

Until 9:00 h, no significant difference in proline (Pro) was observed between treatments (Fig. 4). From 11:00 h on, drought-stressed plants showed an increase of about 220% in Pro, being higher (400%) between 13:00 and 15:00 h. Control plants did not show significant changes in Pro throughout the day (Fig. 4).

3.5. Morphological variables, total leaf area and recovery of leaf water potential

Morphological variables are shown in Table 2. At the beginning of the experiment (Day 0) no differences were observed between treatments, suggesting uniformity in above-ground morphology. Throughout the experimental period, both leaf and leaflet numbers remained unchanged in control plants; however, there was a 200% increase in leaf area, implying continued leaf expansion. Furthermore, control plants showed significant increases in height (116%) and stem diameter (50%). Comparisons between treatments on Day 20 evidenced 30% and 42% decrease in leaf and leaflet numbers

in drought-stressed plants, which was due to leaf (leaflet) abscission. As a consequence, no increment in total leaf area was observed in drought-stressed plants. Drought did not influence plant height, but 20 days after withholding irrigation, stem diameter was around 20% lower in drought-stressed plants. Therefore, secondary growth was limited under drought. Twelve hours after resuming irrigation, no difference in Ψ_{pd} was observed between treatments (Table 3).

4. Discussion

In well-watered plants, the decrease in Ψ_x observed between 9 and 15 h suggests a possible imbalance between water uptake from the soil and water loss through transpiration (Maier-Maercker, 1999; Mott and Franks, 2001). According to Meinzer (2003), when the soil water potential surrounding the roots remain constant the fluctuations in Ψ_x are determined by variation in E and K_T . Hence, in control plants the high rates of g_s recorded until 13:00 h as well as increasing Δ_w throughout the morning favored high E , resulting in the observed midday depression in Ψ_x . Such idea is supported by the close and positive correlation coefficients found between $E \times \Delta_w$, $E \times \Psi_x$ and $E \times K_T$. Moreover, greater variations in Ψ_x might have been attenuated by the high K_T observed from 11:00 to 15:00 h. This is supported by the fact that

Table 1
Correlation coefficients between physiological and environmental variables in well-watered (Control) and water-stressed (Drought-stress) mahogany plants.

Treatment	Physiological and environmental variables								
		RH ^a	PAR	Δ_w	T_{air}	T_{leaf}	E	Ψ_x	K_T
Control	g_s	0.14	0.64***	-0.29*	-0.06	-0.24	0.67*	0.61*	0.54*
	E	-0.58***	0.86***	0.44**	0.66***	0.51**	-	0.84*	0.87*
Drought-stress	g_s	0.82***	-0.46**	-0.78***	-0.83***	-0.86***	0.95*	0.19	0.71*
	E	0.73***	-0.49**	-0.74***	-0.74***	-0.79***	-	0.08	0.78*

Asterisks indicates significant Pearson's coefficient at * $P < 0.05$, ** $P < 0.01$ or *** $P < 0.001$

^a Abbreviations: stomatal conductance (g_s), transpiration (E), relative air humidity (RH), photosynthetic active radiation (PAR), leaf-to-air vapor pressure difference (Δ_w), air temperature (T_{air}), leaf temperature (T_{leaf}), leaf water potential (Ψ_x), and apparent total hydraulic conductance (K_T).

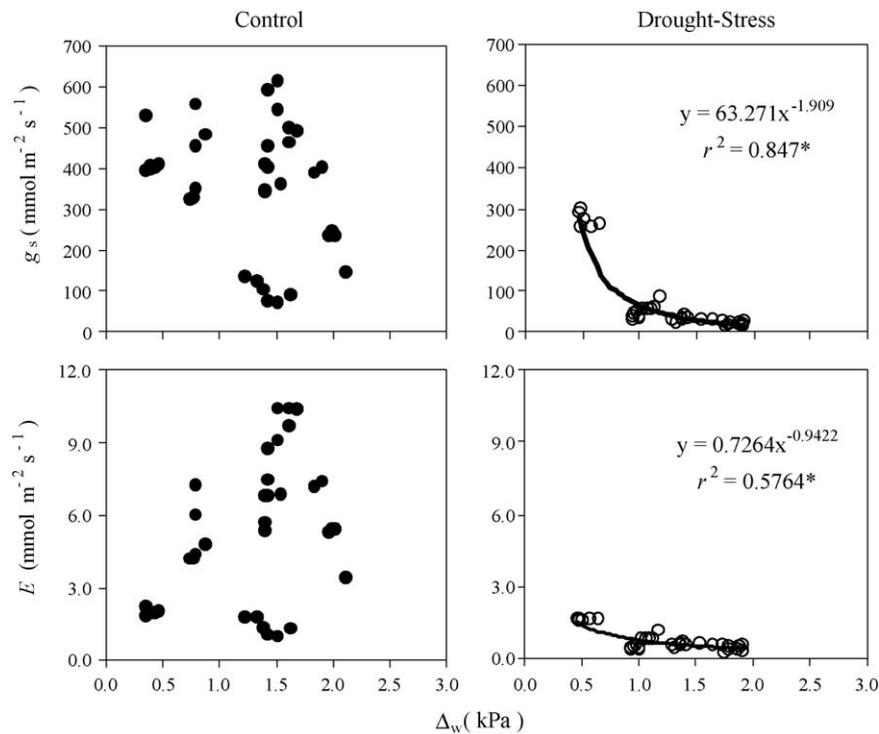


Fig. 3. Stomatal conductance to water vapor (g_s) and transpiration (E) in relation to diurnal-course of leaf-to-air vapor pressure deficit (Δ_w) in well-watered (Control, closed symbol) and water-stressed (Drought-stress, opened symbol) mahogany plants. Data for each variable in each time of the day are the mean of six replicates measured on five consecutive days. Regression analyses for each variable and treatment are followed by the respective equations in the graph, and asterisks indicate significant coefficients of determination (F 's test, $P < 0.05$).

on full-irrigated soils, high K_T allows continuous water transport from soil to leaves, as previously reported for drought tolerant genotypes of *C. canephora* under water deficit conditions (i.e., $\Psi_{pd} = -3.5$ MPa) (Pinheiro et al., 2005). When considered together, our data suggest that the maintenance of leaf turgor throughout the day (indirectly estimated as the diurnal variations in Ψ_x) was granted by increasing K_T , regardless of the high g_s and E over the morning hours. Also, the maintenance of g_s at high rates for longer periods might allow greater CO_2 influx, favoring net photosynthesis. At least in part, this explain the considerable increases in plant height and stem diameter in control plants, despite the experiment had been evaluated for a relatively short period (20 days).

For some Amazonian woody species, evaluated over the wet and dry seasons, the maintenance of minimum Ψ_x throughout the day (or during the period of stress) was achieved by the way of an isohydric mechanism controlling stomatal aperture (Fisher et al., 2006). Apparently, isohydric mechanism does not explain diurnal variations in g_s of drought-stressed mahogany plants, since no significant correlation between Ψ_x and g_s was found. Moreover, Ψ_x at midday was substantially lower under drought conditions despite of the expressive reductions in g_s and E . By contrast, Tardieu and Simonneau (1998) described a typical anisohydric

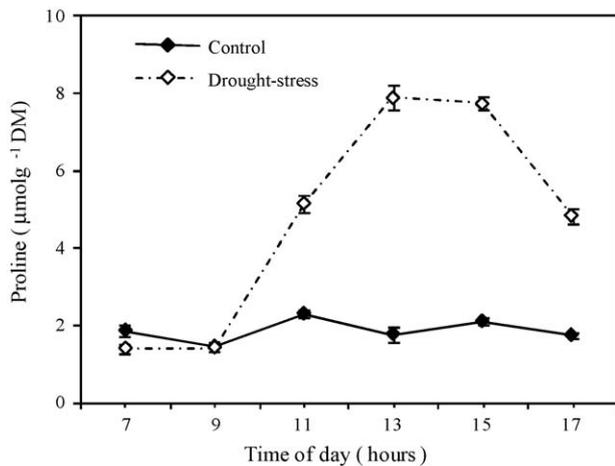


Fig. 4. Leaf proline content in well-watered (Control, closed symbol) and water-stressed (Drought-stress, opened symbol) mahogany plants. Data for each variable at each time of the day are the mean of six replicates measured on five consecutive days \pm standard error.

Table 2

Morphological variables in well-watered (Control) and water-stressed (Drought-stress) mahogany plants.

Variable	Treatment	Days after water regime differentiation	
		0	20
Leaf number (unit)	Control	16.00 \pm 0.36 Aa*	17.83 \pm 0.48 Aa
	Drought-stress	16.66 \pm 0.33 Aa	12.83 \pm 1.01 Bb
Leaflet number (unit)	Control	84.33 \pm 3.61 Aa	91.83 \pm 3.31 Aa
	Drought-stress	87.83 \pm 1.37 Aa	61.83 \pm 3.13 Bb
Total leaf area (m ²)	Control	0.084 \pm 0.008 Aa	0.172 \pm 0.018 Ba
	Drought-stress	0.078 \pm 0.011 Aa	0.086 \pm 0.009 Ab
Stem height (m)	Control	0.40 \pm 0.05 Aa	0.86 \pm 0.18 Ba
	Drought-stress	0.39 \pm 0.05 Aa	0.79 \pm 0.16 Bb
Stem diameter (mm)	Control	6.35 \pm 0.10 Aa	9.50 \pm 0.10 Ba
	Drought-stress	6.40 \pm 0.10 Aa	8.00 \pm 0.20 Bb

* Different capital letters denote significant differences between means for each variable into the same water regime as measured in different experimental days (time effect), and different small letters denote significant differences between means for each variable from different water regimes as evaluated at the same experimental day (treatment effect) by Newman-Keuls' test ($P < 0.05$). Values are means (\pm standard error), $n = 4$.

Table 3

Pre-dawn leaf water potential (Ψ_{pd} , –MPa) of young plants of mahogany under well-watered (Control) and water-limited (Drought-stress) conditions.

Treatment	Days from water regime differentiation		Hours after resuming irrigation
	0	20	
Control	-0.02 ± 0.01 Aa*	-0.05 ± 0.01 Aa	-0.04 ± 0.01 Aa
Drought-stress	-0.04 ± 0.01 Aa	-3.52 ± 0.07 Bb	-0.07 ± 0.01 Aa

* Different capital letters denote significant differences between means for each variable into the same water regime as measured in different experimental days (time effect), and different small letters denote significant differences between means for each variable from different water regimes as evaluated at the same experimental day (treatment effect) by Newman–Keuls' test ($P < 0.05$). Values are means (\pm standard error), $n = 4$.

behavior of stomatal movement in drought-stressed sunflower, in which diurnal-courses of Ψ_x and g_s under drought were similar to those found on drought-stressed mahogany plants. In anisohydric plants, differences between Ψ_x at midday (Ψ_{md}) and Ψ_{pd} ($\Delta\Psi = \Psi_{md} - \Psi_{pd}$) in irrigated and drought-stressed plants remain roughly constant throughout the day (Tardieu and Simonneau, 1998), as observed in this experiment. However, we must be cautious with such inference, because the absence of correlation between Ψ_x and g_s (as observed in drought-stressed plants) *per se* does not necessarily contradict isohydric control of leaf water status. The reason is that when drought develops in isohydric plants, stomata may be quite closed (g_s near to zero) in parallel with continued decreases in Ψ_x . Moreover, since leaf proline content was greatly increased in drought-stressed plants, the apparent homeostasis in Ψ_x throughout the day might have also been achieved by means of osmotic adjustment, as observed in other woody species (DaMatta et al., 2003; Carvalho, 2005).

The magnitude of the decreases in g_s was relatively similar to those reported for other woody species, such as *Mimosa caesalpinifolia* (Silva et al., 2003), *C. canephora* (DaMatta et al., 2003) and *Eucalyptus grandis* (Pita and Pardos, 2001; Almeida and Soares, 2003). In adult mahogany trees, grown under field conditions in Minas Gerais State (southeastern of Brazil), 62% decrease in g_s was observed under mild drought-stress ($\Psi_{pd} = -1.00$ MPa) (Silva and Lemos Filho, 2001). We acknowledge that strict comparisons between plants grown in pots (such as in the present experiment) and under field conditions are limited by various reasons, including plant age, root depth, total and specific leaf area, and environmental conditions determining how drought stress was imposed (duration and intensity of the stress). Nevertheless, the main concurrent result between our data and the ones reported by Silva and Lemos Filho (2001) was the similar magnitude of the decreases in g_s , suggesting that stomatal conductance exert an important role in transpiration control of mahogany.

In general, decreases in g_s under drought conditions limit E and prevent larger fluctuations in Ψ_x , contributing to the maintenance of leaf turgor (Kramer and Boyer, 1995). The control of stomatal aperture can be explained in terms of sensitivity of stomata to increasing VPD (Oren et al., 1999; Tuzet et al., 2003). In many species, g_s decreases exponentially as VPD raises (Monteith, 1995; Bunce, 1996; Matzner and Comstock, 2001). Such behavior was observed in drought-stressed mahogany plants, in which both g_s and E decreased exponentially as increasing Δ_w throughout the day, with no apparent threshold value. According to Monteith (1995) decreases in g_s seem to be linked to sensing of E rather than VPD (Monteith, 1995; Meinzer et al., 1997), which may involve both feedback and feedforward mechanisms of stomatal limitation of transpiration (Meinzer, 2003). On the other hand, homeostasis of Ψ_x by limiting E , as observed in drought-stressed mahogany plants, may also result from the active stomatal coordination of transpiration with variation in the hydraulic properties of the soil-

to-leaf pathway (Meinzer, 2002, 2003) and it likely involves a feedback sensing of changes in water status. Accurate inferences about feedforward or feedback mechanisms controlling g_s are commonly evaluated as the progress of drought-stress, assessed by direct measurements of soil water potential or indirectly, by measuring Ψ_{pd} . Since our data were obtained when mahogany plants were subjected to moderate- to severe-drought ($\Psi_{pd} = -3.5$ MPa), we cannot infer about which mechanism (feedforward or feedback) influenced stomatal response in mahogany. However, considering that K_T was greatly decreased between 9:00 and 15:00 h, and was coincident to lower rates of g_s and E and to higher Δ_w , we can suggest that K_T may have, to some extent, influenced stomatal coordination of transpiration through feedback sensing of changes in water status (Meinzer, 2003). Furthermore, decreasing K_T under limited soil water, as observed in the drought-stressed mahogany plants, is an important trait to avoid excessive cavitation and disturbance to cellular water relations and biochemistry (Jones and Sutherland, 1991; Hubbard et al., 2001). Hence, drought stress experienced by mahogany plants did not result on irreversible damages in water uptake and transport from the soil to above-ground tissues, since drought-stressed plants showed a remarkable ability to recover Ψ_{pd} to control levels, 12 h after irrigation has been resumed.

At least in part, high g_s for longer periods and expressive increment in total leaf area might have allowed improved net CO_2 assimilation at the whole-plant. Also, high K_T might successfully compensate for water loss via E . Hence, both energy supply (ATP from carbohydrates respiration) and minimum turgor pressure to sustain cell division and expansion were granted, favoring the sustained growth observed in control plants during the experimental period. Actually, rapid initial growth under non-limiting soil water is expected in mahogany, since this is a pioneer, large-gap, or late successional tree species (Denslow, 1987; Swaine and Whitmore, 1988).

The decreased total leaf (leaflet) area in drought-stressed plants could be viewed as a strategy to postpone drought by reducing whole-plant transpiration, with minor effects on plant water-balance. However, reducing total leaf (leaflet) area may also reduce whole-plant net photosynthesis, and above-ground growth. Although growth cessation is commonly observed in drought-stressed plants (Kramer and Boyer, 1995; Kowłowski and Pallardy, 1997), the remarkable increments in stem height and diameter observed in drought-stressed mahogany over the experimental period could, at least in part, be explained by an efficient use of remaining soil water, favoring cell turgor and expansion. Moreover, as in another drought-stressed Meliaceae (*C. guianensis*) seedling from the Amazon region, no significant decreases in net photosynthesis and stomatal conductance were observed until 14 days after withholding irrigation (Gonçalves et al., 2009). Therefore, it could be inferred that maintenance of net photosynthesis in drought-stressed mahogany plants might, to some extent, have contributed to the maintenance of growth.

5. Conclusions

The present data pointed that drought-stressed mahogany plants exhibited an efficient control of E by the way of adjustments in g_s and K_T and by the way of controlling g_s in response to high Δ_w throughout the day. Limitations in whole-plant E were also granted by reducing total leaf area and high accumulation of leaf proline throughout the day was an indicative that osmotic adjustment might have contributed to minor fluctuations in Ψ_x under drought. After stress ceases, mahogany plants were able to recover leaf turgor 12 h after resumption irrigation. Altogether, our data support that mahogany has the ability to satisfactorily tolerate or postpone drought.

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References

- Almeida, A.C., Soares, J.V., 2003. Comparison of water use in *Eucalyptus grandis* plantations and Atlantic rainforest in eastern coast of Brazil. *Revista Árvore* 27, 159–170.
- Asada, K., 1999. The water–water cycle in chloroplasts: scavenging of active oxygen and dissipation of excess photons. *Annual Review of Plant Physiology and Plant Molecular Biology* 50, 601–639.
- Bates, L.S., Waldren, R.P., Teare, I.D., 1973. Rapid determination of free proline for water-stress studies. *Plant and Soil* 39, 205–207.
- Buckley, T.N., Mott, K.A., 2002. Stomatal water relations and the control of hydraulic supply and demand. *Progress in Botany* 63, 309–325.
- Bunce, J.A., 1996. Does transpiration control stomatal responses to water vapour pressure deficit? *Plant Cell and Environment* 19, 131–135.
- Carvalho, C.J.R., 2005. Responses of *Schizolobium amazonicum* [S. *parahyba* var. *amazonicum*] and *Schizolobium parahyba* [*Schizolobium parahybum*] plants to water stress. *Revista Árvore* 29, 907–914.
- Cernusak, L.A., Aranda, J., Marshall, J.D., Winter, K., 2007. Large variation in whole-plant water-use efficiency among tropical tree species. *New Phytologist* 173, 294–305.
- Chagnon, F.J.F., Bras, R.L., Wang, J., 2004. Climatic shift in patterns of shallow clouds over the Amazon. *Geophysical Research Letters* 31, L24212, doi:10.1029/2004GL021188.
- Chaves, A.R.M., Ten-Caten, A., Pinheiro, H.A., Ribeiro, A., DaMatta, F.M., 2008. Seasonal changes in photoprotective mechanisms of leaves from shaded and unshaded field-grown coffee (*Coffea arabica* L.) trees. *Trees* 22, 351–361.
- DaMatta, F.M., Chaves, A.R.M., Pinheiro, H.A., Ducatti, C., Loureiro, M.E., 2003. Drought tolerance of two field-grown clones of *Coffea canephora*. *Plant Science* 164, 111–117.
- Denslow, J.S., 1987. Tropical rainforest gaps and tree species diversity. *Annual Review of Ecology and Systematics* 18, 431–451.
- Donovan, L.A., West, J.B., McLeod, K.W., 2000. *Quercus* species differ in water and nutrient characteristics in a resource-limited fall-line sandhill habitat. *Tree Physiology* 20, 929–936.
- Dünisch, O., Erbreich, M., Eilers, T., 2003. Water balance and water potentials of a monoculture and an enrichment plantation of *Carapa guianensis* Aubl. in the Central Amazon. *Forest Ecology and Management* 172, 355–367.
- Fisher, R.A., Williams, M., Vale, R.L., Costa, A.L., Meir, P., 2006. Evidence from Amazonian forests is consistent with a model of isohydric control of leaf water potential. *Plant Cell and Environment* 29, 151–165.
- Gonçalves, J.F.C., Silva, C.E.M., Guimarães, D.G., 2009. Fotossíntese e potencial hídrico foliar de plantas jovens de andiroba submetidas à deficiência hídrica e a reidratação. *Pesquisa Agropecuária Brasileira* 44, 8–14.
- Hoagland, D.R., Arnon, D.I., 1950. The water-culture method for growing plants without soil. *California Agricultural Experimental Station Circular* 347, 1–32.
- Hubbard, R.M., Bond, B.J., Ryan, M.G., 1999. Evidence that hydraulic conductance limits photosynthesis in old *Pinus ponderosa* trees. *Tree Physiology* 19, 165–172.
- Hubbard, R.M., Ryan, M.G., Stiller, V., Sperry, J.S., 2001. Stomatal conductance and photosynthesis vary linearly with plant hydraulic conductance in ponderosa pine. *Plant Cell and Environment* 24, 113–121.
- Jones, H.G., Sutherland, R.A., 1991. Stomatal control of xylem embolism. *Plant Cell and Environment* 11, 111–121.
- Kramer, P.J., Boyer, J.S., 1995. *Water Relations of Plants and Soils*. Academic Press, San Diego, p. 495.
- Kowlowski, T.T., Pallardy, S.G., 1997. *Physiology of Woody Plants*. Academic Press, San Diego, p. 411.
- Landsberg, J.J., 1986. *Physiological Ecology of Forest Production*. Academic Press, London, p. 198.
- Maier-Maercker, U., 1999. New light on the importance of peristomatal transpiration. *Australian Journal of Plant Physiology* 26, 9–16.
- Marengo, J., Bhatt, U., Cunningham, C., 2000. Decadal and multidecadal variability of climate in the Amazon basin. *International Journal of Climatology* 20, 503–518.
- Matzner, S., Comstock, J., 2001. The temperature dependence of shoot hydraulic resistance: implications for stomatal behavior and hydraulic limitation. *Plant Cell and Environment* 24, 1299–1307.
- Mediavilla, S., Escudero, A., 2004. Stomatal responses to drought of mature trees and seedlings of two co-occurring Mediterranean oaks. *Forest Ecology and Management* 187, 281–294.
- Meinzer, F.C., Hinckley, T.M., Ceulemans, R., 1997. Apparent responses of stomata to transpiration and humidity in a hybrid poplar canopy. *Plant Cell and Environment* 20, 1301–1308.
- Meinzer, F.C., 2002. Co-ordintaion of vapour and liquid phase water transport properties in plants. *Plant Cell and Environment* 25, 265–274.
- Meinzer, F.C., 2003. Functional convergence in plant responses to the environment. *Oecologia* 134, 1–11.
- Mittler, R., 2002. Oxidative stress, antioxidants and stress tolerance. *Trends in Plant Science* 9, 405–410.
- Moghaieb, R.E.A., Saneoka, H., Fujita, K., 2004. Effect of salinity on osmotic adjustment, glycinebetaine accumulation and betaine aldehyde dehydrogenase gene expression in two halophytic plants, *Salicornia europaea* and *Suaeda maritime*. *Plant Science* 166, 1345–1349.
- Monteith, J.L., 1995. A reinterpretation of stomatal responses to humidity. *Plant Cell and Environment* 18, 357–364.
- Mott, K.A., Franks, P.J., 2001. The role of epidermal turgor in stomatal interactions following a local perturbation in humidity. *Plant Cell and Environment* 24, 657–662.
- Oren, R., Sperry, J.S., Katul, G.G., Pataki, D.E., Ewers, B.E., Phillips, N., Schefer, K.V.R., 1999. Survey and synthesis of intra- and interspecific variation in stomatal sensitivity to vapour pressure difference. *Plant Cell and Environment* 22, 1515–1526.
- Pinheiro, H.A., DaMatta, F.M., Chaves, A.R.M., Fontes, E.P.B., Loureiro, M.E., 2004. Drought tolerance in relation to protection against oxidative stress in clones of *Coffea canephora* subjected to long-term drought. *Plant Science* 167, 1307–1314.
- Pinheiro, H.A., DaMatta, F.M., Chaves, A.R.M., Loureiro, M.E., Ducatti, C., 2005. Drought tolerance is associated with rooting depth and stomatal control of water use in clones of *Coffea canephora*. *Annals of Botany* 96, 101–108.
- Pinheiro, H.A., Silva, J.V., Endres, L., Ferreira, V.M., Câmara, C.A., Cabral, F.F., Oliveira, J.F., Carvalho, L.W.T., Santos, J.M., Santos Filho, B.G., 2008. Leaf gas exchange, chloroplastic pigments and dry matter accumulation in castor bean (*Ricinus communis* L.) seedlings subjected to salt stress conditions. *Industrial Crops and Products* 27, 385–392.
- Pita, P., Pardos, J.A., 2001. Growth, leaf morphology, water use and tissue water relations of *Eucalyptus globulus* clones in response to water deficit. *Tree Physiology* 21, 599–607.
- Romero, P., Botia, P., 2006. Diurnal and seasonal patterns of leaf water relations and gas exchange of regulated deficit-irrigated almond trees under semiarid conditions. *Environmental and Experimental Botany* 56, 158–173.
- Santos Jr., U.M., Gonçalves, J.F.C., Feldpausch, T.R., 2006. Growth, leaf nutrient concentration and photosynthetic nutrient use efficiency in tropical tree species planted in degraded areas in central Amazonia. *Forest Ecology and Management* 226, 299–309.
- Sellin, A., Kupper, P., 2004. Within-crown variation in leaf conductance of Norway spruce: effects of irradiance, vapour pressure deficit, leaf water status and plant hydraulic constraints. *Annual of Forest Science* 61, 419–429.
- Silva, B.G., Lemos Filho, J.P.L., 2001. Water relations of woody species in the campus Pampulha/UFMG, Belo Horizonte, MG. *Revista Brasileira de Botânica* 24, 519–525.
- Silva, E.C., Nogueira, R.J.M.C., Neto, A.D.A., Santos, V.F.S., 2003. Stomatal behavior and leaf water potential in three wood species cultivated under water stress. *Acta Botânica Brasílica* 17, 231–246.
- Soares-Filho, B.S., Nepstad, D.C., Curran, L.M., Cerqueira, G.C., Garcia, R.A., Ramos, C.A., Voll, E., McDonald, A., Lefebvre, P., Schlesinger, P., 2006. Modelling conservation in the Amazon basin. *Nature* 440, 520–523.
- Swaine, M.D., Whitmore, T.C., 1988. On the definition of ecological species groups in tropical rain forests. *Vegetatio* 75, 81–86.
- Tardieu, F., Simonneau, T., 1998. Variability among species of stomatal control under fluctuating soil water status and evaporative demand: modeling isohydric and anisohydric behaviors. *Journal of Experimental Botany* 49, 419–432.
- Tuzet, A., Perrier, A., Leuning, R., 2003. A coupled model of stomatal conductance, photosynthesis and transpiration. *Plant Cell and Environment* 26, 1097–1116.
- Victoria, R., Martinelli, L., Moraes, J., Ballester, M., Krusche, A., Pellegrino, G., Almeida, R., Richey, J., 1998. Surface air temperature variations in the Amazon region and its border during this century. *Journal of Climate* 11, 1105–1110.