iForest
Biogeosciences and Forestry

Research Article
doi: 10.3832/ifor2813-011
vol. 12, pp. 61-68

Gas exchange, biomass allocation and water-use efficiency in response to elevated CO_2 and drought in andiroba (*Carapa surinamensis*, Meliaceae)

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Prolonged droughts are predicted for some parts of the Amazon; however, it is still unclear how Amazonian trees will respond to water stress under the ongoing increase in CO_2 concentration. The aim of this study was to assess the effect of elevated CO_2 (eCO_2) and drought on photosynthetic rates, water-use efficiency, and biomass allocation in andiroba (*Carapa surinamensis*). The plants were grown in pots at ambient (400 ppm CO_2) and eCO_2 (700 ppm) at two water regimes, soil at 50% field capacity, FC (drought) and soil at 100% FC for 163 days. We measured light saturated photosynthesis on a mass basis ($A_{sat\text{-}mass}$), stomatal conductance to CO_2 on a mass basis ($g_{sCO2\text{-}mass}$), whole-plant water-use efficiency (WUE_P), biomass accumulation, specific leaf area (SLA) and total leaf area. At eCO_2, $A_{sat\text{-}mass}$ increased 28% in well-watered plants and 93% under drought, whereas $g_{sCO2\text{-}mass}$ declined 39% in well-watered plants at eCO_2, with no effect of drought on $g_{sCO2\text{-}mass}$ at eCO_2. The total biomass gain improved 73% at eCO_2 and over CO_2 levels it was reduced (54%) by drought. WUE_P improved (188%) at eCO_2 in well-watered plants and 262% under drought. SLA declined 23% at eCO_2, but the effect of drought on SLA was null. On the contrary, total leaf area was greatly reduced (67%) by drought, but it was not affected by eCO_2. The large increase in total biomass and the substantial improvement in WUE_P under eCO_2, and the sharp decline in leaf area under water stress widen our knowledge on the physiology of this important species for the forest management of large areas in the Amazon region.

Keywords: Carboxylation Efficiency, Nonstructural Carbohydrates, Specific Leaf Area, Shoot-root Ratio, Tree Growth

Introduction

The tropical rainforest of the Amazon basin covers about 5.1 × 10^6 km^2 (out of 6.915 × 10^6 km^2 of the total basin area, which extends across several countries) and stores about 86 Pg of carbon in above and below ground biomass (Saatchi et al. 2007, Grace 2016), *i.e.*, 21.8% of total carbon (393.4 Pg) stored in above and below ground biomass of land vegetation (Pan et al. 2013). The Amazon rainforest is also important for recycling through transpiration about 50% of total precipitation of the Amazon region (Salati & Vose 1984) and for its outstanding biodiversity. It has been anticipated that atmospheric CO_2 concentration can reach 800 ppm by the end of the 21st century (Bellasio et al. 2018) and that changes in rainfall distributions can lead to an increase in frequency and severity of droughts, particularly in Eastern Amazon (Duffy et al. 2015). Although a mild dry season has little effect on growth rates of Amazonian trees in the central Amazon (Dias & Marenco 2016, Camargo & Marenco 2017), it has been observed a decline in the amount of carbon stored in above ground biomass per unit land area and an increase in tree mortality under severe drought (Grace 2016).

The most common response observed in free-air CO_2-enrichment is an increase in photosynthetic rates (Ainsworth & Rogers 2007, Way et al. 2015) and in several vascular plants there is also an improvement in net primary productivity (Nowak et al. 2004), but the effect of long-term exposure to elevated CO_2 concentration (eCO_2) on tropical trees still remains to be elucidated. Stomatal conductance (g_s) often declines at eCO_2 (Ainsworth & Rogers 2007), which contributes to improve water-use efficiency (WUE) and biomass gain of plants under CO_2-enrichment (Leakey et al. 2009, 2012). Plant WUE (WUE_P), the ratio between total biomass gain and total evaporation of water from the plant (Lambers et al. 2008) is a parameter often used to assess plant response to water stress. At the leaf level WUE describes the ratio between net photosynthetic rate (*A*) and leaf transpiration (*E*), and both photosynthesis and transpiration are often closely related to stomatal conductance (Marenco et al. 2001, Lambers et al. 2008). Besides g_s, other factors such as biomass allocation to plant organs, whole-plant respiration and variation in boundary layer conductance affect WUE_P, and hence it is not uncommon

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Received: Apr 14, 2018 - Accepted: Nov 14, 2018

Citation: Oliveira MF, Marenco RA (2019). Gas exchange, biomass allocation and water-use efficiency in response to elevated CO_2 and drought in andiroba (*Carapa surinamensis*, Meliaceae). iForest 12: 61-68. - doi: 10.3832/ifor2813-011 [online 2019-01-24]

Communicated by: Rossella Guerrieri

to find discrepancy between WUE_P and A/E trends (Lambers et al. 2008).

In the Brazilian Legal Amazon (5,016,136 km² – Grace 2016) large areas has been deforested over the past decades and hence there are extensive areas available for rehabilitation. It has been estimated that about 120,000 km² could be restored or reforested by 2030 (Brazil-MMA 2016). Several species, including andiroba (*Carapa*, a genus that includes 11 species in the Neotropics – Kenfack 2011) have been tested to evaluate their performance in reforestation projects or forestry systems (Brienza et al. 2008, Souza et al. 2008). Andiroba is a promising species because it produces wood of good quality and a multipurpose oil (carapa oil) can be extracted from their seeds (Kenfack 2011). This is particularly important in (agro)forestry systems, as non-timber forest products can generate additional income while the tree is not large enough for timber harvesting (Klimas et al. 2012). In the Neotropics, the *Carapa* tree (andiroba in Brazil) occurs in lowland forests from Central America and the Caribbean to South America (Kenfack 2011). At sapling stage it can grow about 1-2 cm yr^{-1} in diameter and 1-1.5 m yr^{-1} in height (Dunisch et al. 2002, Camargo & Marenco 2012). Canopy trees of this species can reach up to 2.0 m in diameter and 60 m in height (Fournier 2002). *Carapa* is more tolerant to *Hypsipyla grandella* than mahogany – *Swietenia macrophylla* (Souza et al. 2008), thus it can be used in plantations to replace the highly prized mahogany, which highlights the importance of andiroba in forest management project. The aim of this study was to assess the effect of eCO_2 and drought on photosynthesis, water-use efficiency, and biomass gain in saplings of *Carapa surinamensis*. In this experiment we hypothesized that subjecting the plants to eCO_2 increases biomass gain and photosynthetic rates, and decreases stomatal conductance, which leads to an increase in water-use efficiency.

Materials and methods

Experimental setup and plant material

The experiment was conducted at the National Institute for Research in the Amazon – INPA (03° 05′29″ S, 59° 59′ 35″ W), Manaus, Brazil under greenhouse and growth chamber conditions. Seeds of andiroba (*Carapa surinamensis* Miq, Meliaceae) were germinated in vermiculite and 15 days after emergence, the seedlings were transplanted to large pots (20 cm diameter and 26 cm deep, about 8 L capacity) containing 7 kg of substrate, a mixture of soil forest of the first 20 cm, amended with mulched material and 5 g kg^{-1} (soil) of 10-10-10 (NPK) fertilizer. We used large pots to avoid root restriction during the experimental period. Once the plants had reached 60 cm in height (about six months after emergence) the saplings were randomly sorted into four treatments: two CO_2 levels, ambient CO_2 (400 ppm) and eCO_2 (700 ppm) and two water regimes, soil kept at 50% field capacity – FC (hereafter referred to as the drought condition) and 100% FC. The experiment was run for 163 days, from July 14 to December 23, 2015.

Before initiating the experiment and for 30 days, we measured the light intensity (photosynthetically active radiation – PAR) inside the greenhouse, which turned out to be a mean of about 200 µmol m^{-2} s^{-1} over a 12-h period. Furthermore, because of the latitudinal location (-3.091°), incoming irradiance remains rather constant over the year. Then we set up the growth chamber to emulate the mean light intensity in the greenhouse over a 12-h photoperiod.

Plants subjected to the ambient CO_2 treatment were kept under greenhouse conditions, whereas those subjected to eCO_2 were kept in a growth chamber (TPC-19®, Biochambers Inc., Winnipeg, Canada; working area of 1.72 m² and 1.52 m height). In the growth chamber CO_2 was kept constant (700 ppm); we used a 12-h photoperiod with PAR of 200 µmol m^{-2} s^{-1}, the light was turned off at night; day/night temperature and relative humidity were 27/25 °C and 80-90%, respectively. We also monitored the light and temperature conditions inside the greenhouse with specific sensors (Li-190 SA®, Li-Cor, Lincoln, USA and Humitter 50Y®, Vaisala, Oyj, Finland) connected to a datalogger (Li-1400®, Li-Cor, Lincoln, USA), which was set to record data at 15-min intervals. On randomly selected days we monitored the CO_2 concentration in the greenhouse with an infrared gas analyzer (LI-6400®, Lincoln, USA) with empty chamber.

The water volume the soil could hold was recorded and used to calculate soil water content (SWC) at 50% and 100% FC. At these water contents, soil moisture percentages, measured with a probe (MPM160B®, ICT International, Armidale, Australia), were 21% (v/v, soil at 50% FC) and 31% (v/v, soil at 100% FC). For further information we also measured the predawn leaf water potential of the plants with a Sholander pressure chamber (1505D®, PMS Instrument Company, Albany, USA). Once a week and during the experimental period the plants in the greenhouse and growth chamber were randomly moved to avoid position effects. At the end of the experimental period we measured gas exchange (data were collected just once), and determined dry matter (DM) of stems (W_S), roots (W_R) and leaves (W_L), total plant DM (W_T), shoot/root ratio (SRR), leaf area (A_L), specific leaf area (SLA, leaf area to leaf mass ratio), consumptive use of water (CUW – total amount of water consumed by a plant), whole-plant water-use efficiency (WUE_P), and leaf total nonstructural carbohydrates (TNC).

Gas exchange parameters

At the end of the experimental period, gas exchange parameters were measured with a portable gas exchange system (LI-6400XT®, Li-Cor, Lincoln, USA). The measurements were carried out between 08:00 and 14:00 in two fully expanded leaves per plant, after a stabilization period of about 10 min at ambient and eCO_2 and the light intensity used for measurements. We measured stomatal conductance to CO_2 (g_{sCO2}), light saturated photosynthesis (A_{sat}), C_i/C_a (intercellular CO_2 to ambient CO_2 ratio), carboxylation efficiency (CE, the initial slope of the A/C_i regression line – Evans & Seemann 1984), leaf respiration in the light (R_L), and dark respiration (R_D). A_{sat} and g_{sCO2} were measured at 1000 µmol m^{-2} s^{-1}, ambient temperature (27 °C), relative humidity of 70 ± 5%, and CO_2 concentration of 400 ppm (ambient CO_2 treatment) and 700 ppm (eCO_2 treatment). We also determined light and CO_2-saturated photosynthesis (A_{max}) at a $[CO_2]$ of 2000 ppm (C_a), as previously described (Marenco et al. 2017). To obtain $A_{sat\text{-}mass}$, $A_{max\text{-}mass}$, CE_{mass} and $g_{sCO2\text{-}mass}$, photosynthetic rates, CE and g_{sCO2} per unit area were converted to a mass basis by multiplying them by SLA. For CE, we used the conventional unit (*i.e.*, µbar CO_2 instead of µmol CO_2); thus CE_{mass} was expressed in mmol g^{-1} s^{-1} bar^{-1}. R_L was determined at low light intensity (10-75 µmol m^{-2} s^{-1}) as the intercept of the A/PAR regression line, whereas R_D was determined as the absolute value of A in the dark (PAR = 0) and at the same ambient conditions as those used for measuring $A_{sat\text{-}mass}$; both R_L and R_D were expressed on a mass basis (*i.e.*, $R_{L\text{-}mass}$ and $R_{D\text{-}mass}$).

Biomass allocation

At the end of the experimental period (163 days = t_{163}), we determined the dry matter of leaves, stems and roots by oven-drying the plant material at 72 °C to constant mass. We also determined the specific leaf area (SLA). Leaf area (A_L) was measured with an area meter (LI-3000®, Li-Cor, Lincoln, USA). At the beginning (t_0) and at the end of the experiment (t_{163}), we measured plant height, H (from ground level to the apical bud). We also measured plant diameter (D) at 6.0 cm from the ground with digital calipers.

The biomass gain (ΔB) of plant organs (ΔW_L, ΔW_S and ΔW_R) and leaf area gain (ΔA_L) during the experimental period was calculated as the difference between the biomass of plant organs recorded at the end of the experimental period (*i.e.*, at t = 163 by harvesting the plants) and that estimated at the beginning of the experiment (initial biomass at t_0). To estimate the biomass at t_0, additional plants (n = 30 plants) were produced and harvested to generate allometric equations and estimate: (i) the total biomass, W_T (g) = $10.157 \cdot \exp 0.1692D$ (r^2 = 0.95), where D (in mm) denotes the plant diameter at 6.0 cm from the base of the plant; (ii) the stem biomass, W_S (g) = $2.4298 \cdot \exp 0.1909D$ (r^2= 0.94); and (iii) root biomass, W_R (g) = $1.3277 \cdot \exp 0.1988D$ (r^2 = 0.88). Leaf biomass (W_L) was calculated as the aggregate biomass of individual leaflets: W_{LL} (g) = -0.2052 + 0.04L_L (r^2 = 0.85), where L_L (in cm) stands for leaflet length, n = 100 leaflets from 30 plants. Leaf area (A_L) at the beginning of the experiment was calculated as the aggregate area of individual leaflets: A_{LL} (cm²) = -47.117 + 8.5452L_L (r^2= 0.89). These equations allowed us to estimate A_L and the biomass gain (ΔB) of plant organs throughout the experimental period (163 days).

The consumptive use of water (CUW)

The CUW during the experimental period was obtained by daily recording the amount of water consumed by the plant. During the whole experimental period (163 days) each potted plant was weighed (at 07:00-08:00, accuracy of 1 g) and the amount of water consumed recorded and restored to keep the soil at its target water content (50% FC or 100% FC). For rewatering, we did not take into account the mass change due to daily plant growth because in comparison with the amount of water lost by total evaporation from the plant, the biomass gain was very low (< 1%). To avoid evaporation from the soil surface the pot was covered with a plastic bag sealed to the base of the plant. Thus, all water lost from each pot was assumed to come only from plant evaporation.

Whole-plant water-use efficiency (WUE_P)

WUE_P was calculated as the ratio of the whole-plant biomass gain (ΔW_T) to CUW. That is, $WUE_P = \Delta W_T$ / CUW, where ΔW_T denotes the difference between W_T at t_{163} and W_T at t_0 (*i.e.*, $\Delta W_T = W_{Tt163} - W_{Tt0}$). The relative growth rates (RGR) was calculated as follows: RGR (g g^{-1} day^{-1}) = [ln W_{Tt163} – ln W_{Tt0}]/($t_{163} - t_0$), where W_{Tt163} and W_{Tt0} denote the plant biomass (in grams) at the indicated times, and ($t_{163} - t_0$) denotes the time interval ($t_{163} - t_0$ = 163 days).

Total nonstructural carbohydrates (TNC)

Leaf total nonstructural carbohydrate content was measured at the end of the experimental period. Nonstructural carbohydrates were extracted in boiling ethanol (80% v/v), and purified with chloroform. Starch in the ethanol-soluble residue was hydrolyzed in 0.5 M NaOH and the precipitate removed by centrifugation (15 min at 1000×g). Glucose content in the sample was measured spectrophotometrically at 490 nm after reaction with phenol-sulfuric acid, and the sugar content determined using a glucose standard.

Statistical analysis

We used a split-plot experimental design, with CO_2 levels (400 and 700 ppm) as whole plots and water regimes (soil at 50 and 100% FC) as subplots, with five replications (plants). Data were subjected to analysis of variance (ANOVA) and the *post-hoc* Fisher's LSD test (p = 0.05) was used for mean separation. Biomass, CUW, CE_{mass} and TNC data were $\log_{10}$ or $\log_{(10+1)}$-transformed before conducting the ANOVA. Statistical analyzes were performed using the software package STATISTICA® ver. 7.0 (StatSoft Inc., Tulsa, OK, USA).

Results

In the greenhouse relative humidity was 70-80% and mean temperature 27.5 °C (ranging from 26 °C at night to 29 °C at midday); mean PAR ranged from 185 to 215 µmol m^{-2} s^{-1} (over a 12-h period) and mean ambient CO_2 concentration was 410 ± 17 ppm (day/night of 400/420 ppm). Over the experimental period, mean predawn leaf water potentials were -0.20 MPa in well-watered plants and -0.34 MPa in those subjected water stress.

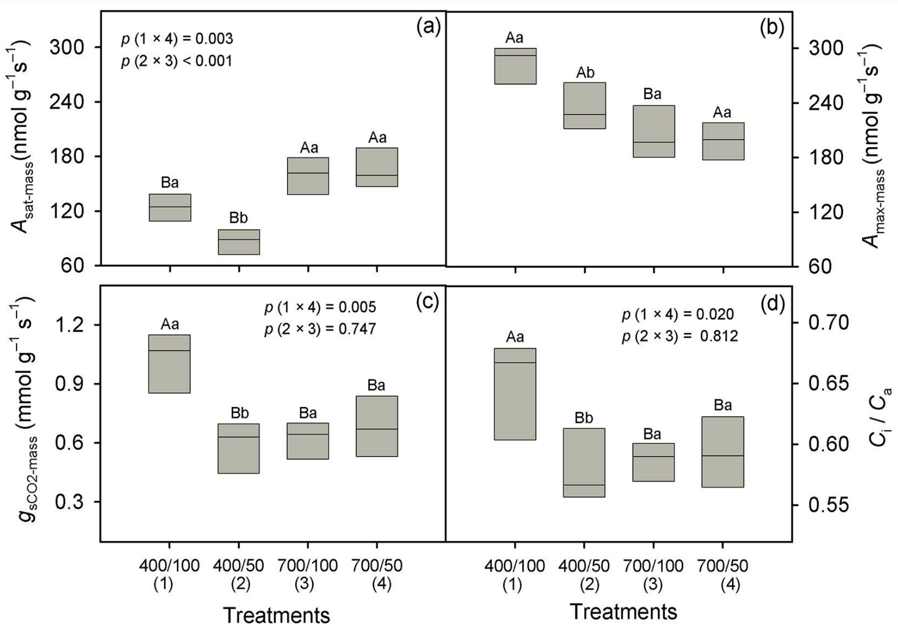
Gas exchange

Taking well-watered plants grown at ambient CO_2 as a baseline, $A_{sat\text{-}mass}$ increased 28% (124 to 159 nmol g^{-1} s^{-1}) under eCO_2 in well-watered plants, and 93% (86-166 nmol g^{-1} s^{-1}) in plants submitted to water stress (Fig. 1a). Across water regimes, $A_{max\text{-}mass}$ was 22% lower at eCO_2 (259 against 202 nmol g^{-1} s^{-1}, p = 0.002 – Fig. 1b, Tab. 1), and over CO_2 levels, $A_{max\text{-}mass}$ declined 11% under drought (244 against 217 nmol g^{-1} s^{-1}, p = 0.039 – Fig. 1b, Tab. 1). Values of $g_{sCO2\text{-}mass}$ declined 43% under drought at ambient CO_2 (1.01 to 0.58 mmol g^{-1} s^{-1}), and 39% in well-watered plants at eCO_2 (1.01 to 0.62 mmol g^{-1} s^{-1}, p = 0.001 – Fig. 1c). However, there was no effect of drought on $g_{sCO2\text{-}mass}$ at eCO_2 (p = 0.22– Fig. 1c). There was no difference in C_i/C_a values between water regimes at eCO_2 (mean of 0.59, p = 0.57 – Fig. 1d). At ambient CO_2, however, C_i/C_a slightly decreased (11%) under drought (p = 0.006 – Fig. 1c), but this decline was not strong enough to affect C_i, which only varied in response to the CO_2 treatment. Mean C_i values were 399 ppm at eCO_2 and 240 ppm at ambient CO_2 (Tab. 2). In both CO_2 environments, we found no effect of water stress on CE_{mass} (p = 0.20 – Tab. 1), but over water regimes it was lower at eCO_2 (p = 0.003 – Fig. 2a). $R_{L\text{-}mass}$ was not affected by drought or eCO_2 (p > 0.05 – Tab. 1), but on average $R_{D\text{-}mass}$ was lower at eCO_2 (Tab. 2).

Biomass allocation, water-use efficiency and TNC

Growth in diameter was greater under eCO_2 than in ambient CO_2 conditions (p = 0.001), whereas height growth was enhanced at ambient CO_2 (p = 0.012 – Tab. 1, Tab. 2). Irrespective of the CO_2 treatment, growth rates in diameter and height were greater in well-watered plants than under drought (p ≤ 0.009 – Tab. 1, Tab. 2). CO_2-enrichment did not affect ΔW_L (p = 0.18 – Tab.

Fig. 1 - Light saturated photosynthesis on a mass basis ($A_{sat\text{-}mass}$, panel A), light and CO_2-saturated photosynthesis on a mass basis ($A_{max\text{-}mass}$, panel B); stomatal conductance to CO_2 on a mass basis ($g_{sCO2\text{-}mass}$, panel C) and intercellular to ambient CO_2 ratio (C_i/C_a, panel D) in *Carapa surinamensis* grown at 400 and 700 ppm of CO_2 and two water regimes, soil at 50% FC and 100% FC. Significant differences between boxes are indicated with different small letters within a CO_2 level and different capital letters within a water regime (Fisher LSD test at p ≤ 0.05). The boundaries of the box indicate the 25th and 75th percentile, and the solid line within the box denotes the median. The mean value for each box is available in the appendix table (Tab. 1A). The p values of the comparisons 1 × 4 and 2 × 3 (meaning of numerals is shown in the x-axis of the panel) are given when the interaction (CO_2 level × water regime) is significant (p ≤ 0.05, Tab. 1).



Tab. 1 - Values of *F* (with *p* values in parenthesis) are given for the effect of CO_2 concentrations (CO_2) and water regimes (water) on evaluated parameters. Abbreviations are indicated in the abbreviation's section.

Parameters	CO_2	Water	CO_2 × Water
$A_{sat\text{-}mass}$ (nmol g^{-1} s^{-1})	55.98 (<0.001)	2.52 (0.151)	5.69 (0.044)
$A_{max\text{-}mass}$ (nmol g^{-1} s^{-1})	19.17 (0.002)	6.06 (0.039)	3.03 (0.120)
C_i/C_a (unitless)	2.18 (0.178)	5.61 (0.045)	8.56 (0.019)
C_i (ppm)	288.14 (<0.001)	1.26 (0.294)	2.71 (0.138)
$g_{sCO2\text{-}mass}$(mmol g^{-1} s^{-1})	7.99 (0.022)	4.59 (0.065)	8.48 (0.019)
$R_{L\text{-}mass}$(nmol g^{-1} s^{-1})	1.03 (0.339)	0.44 (0.526)	0.0004 (0.984)
$R_{D\text{-}mass}$(nmol g^{-1} s^{-1})	17.65 (0.003)	0.18 (0.681)	5.76 (0.043)
Log CE_{mass} (mmol g^{-1} s^{-1} bar^{-1})	18.04 (0.003)	1.95 (0.200)	3.06 (0.118)
Log TNC (mg g^{-1})	20.06 (0.002)	0.56 (0.476)	2.46 (0.156)
HGR (cm day^{-1})	10.60 (0.012)	23.21 (0.001)	2.99 (0.122)
Log DGR (cm day^{-1})	24.54 (0.001)	11.62 (0.009)	0.26 (0.621)
SLA (m^2 kg^{-1})	116.26 (<0.001)	1.24 (0.297)	0.25 (0.629)
ΔA_L (m^2 per plant)	0.15 (0.705)	34.66 (<0.001)	1.09 (0.326)
Log ΔW_L (g per plant)	2.10 (0.185)	35.69 (<0.001)	0.25 (0.629)
Log ΔW_S (g per plant)	18.99 (0.002)	16.63 (0.004)	0.41 (0.540)
Log ΔW_R (g per plant)	38.36 (<0.001)	4.68 (0.063)	0.02 (0.883)
Log SRR (unitless)	23.30 (0.001)	1.61 (0.240)	0.14 (0.715)
RGR (g kg^{-1} day^{-1})	24.25 (0.001)	22.32 (0.001)	0.04 (0.855)
Log ΔW_T (g per plant)	19.36 (0.002)	18.60 (0.003)	0.25 (0.626)
Log CUW [kg (water) per plant]	15.99 (0.004)	59.06 (<0.001)	0.04 (0.844)
WUE_P[g (DM) kg^{-1}(water)]	200.19 (<0.001)	13.21 (0.007)	8.93 (0.017)

1) and, over CO_2 levels, ΔW_L was reduced by 59% under drought stress (46.8 *vs.* 19.0 g per plant, $p < 0.001$ – Fig. 3a). The ΔW_S was greater (83%) at eCO_2 (63.2 *vs.* 34.5 g per plant over water regimes), but it was significantly reduced (52%) by drought over CO_2 levels (66.1 *vs.* 31.6 g per plant, p = 0.004 – Fig. 3b, Tab. 1). Over water regimes, ΔW_R more than doubled under eCO_2 (30.9 *vs.* 10.3 g per plant, $p < 0.001$ – Tab. 1), and across CO_2 levels it was reduced (51%) by drought (27.6 *vs.* 13.6 g per plant, p = 0.06 – Fig. 3c, Tab. 1). Mean SRR declined under eCO_2 from 6.9 to 3.5 (p = 0.001), with no effect of water regimes (p = 0.24 – Fig. 3d, Tab. 1).

Over water regimes RGR was improved (51%) at eCO_2 (7.55 *vs.* 5.01 g kg^{-1} day^{-1}), and across CO_2 levels, it declined (32%) under drought (7.50 *vs.* 5.07 g kg^{-1} day^{-1}, p = 0.001 – Fig. 4a, Tab. 1). Total leaf area gain (ΔA_L) was not affected by CO_2-enrichment (p = 0.70), but across CO_2 levels, it was lower under drought (0.71 *vs.* 0.23 m^2 per plant, $p < 0.001$ – Tab. 1, Tab. 2). Although the plants were a little taller under ambient conditions, across water regimes ΔW_T was 73% greater at eCO_2 (129.8 *vs.* 75.0 g per plant, p = 0.002 – Fig. 4b, Tab. 1) and, over CO_2 levels, it was reduced 54% by drought (140.5 *vs.* 64.2 g per plant, p = 0.003 – Fig. 4b). One can see in this figure that CO_2 enrichment greatly mitigates the effect of drought, as ΔW_T doubled under drought at eCO_2 (85.5 *vs.* 43.0 g per plant).

Across water regimes, CUW was reduced (43%) under eCO_2 (27.2 *vs.* 15.5 kg [water] per plant over the whole study period) and across CO_2 levels it was 63% lower under drought (31.2 *vs.* 11.5 kg [water] per plant, $p < 0.001$ – Fig. 4c, Tab. 1). Therefore, the WUE_p was substantially improved by CO_2-enrichment, particularly under drought (Fig. 4d). At eCO_2, it was improved by 188% in well-watered plants and by 262% under drought. However, WUE_p was not improved by drought at ambient CO_2 (p = 0.66, Fig. 4d). Across water regimes, SLA declined (23%) at eCO_2 (20.4 *vs.* 15.7 m^2 kg^{-1}, $p < 0.001$ – Tab. 1, Tab. 2) with no effect of soil water content on SLA (p = 0.29). Like-

Tab. 2 - Height growth rate (HGR), diameter growth rate (DGR), specific leaf area (SLA), gain of leaf area (ΔA_L), intercellular CO_2 concentration (C_i), and leaf respiration in the light ($R_{L\text{-}mass}$) and leaf dark respiration on a mass basis ($R_{D\text{-}mass}$) in *Carapa surinamensis* at two water regimes (soil at 50% and 100% FC) and two CO_2 levels (400 ppm and eCO_2 -700 ppm). Within rows, significant differences between mean values are indicated with different small letters within a CO_2 level and different capital letters within a water regime (Fisher's LSD test, $p \leq 0.05$). Each value represents the mean (± SD) of five plants (n = 5). In the last four columns and within rows, significant differences between means of CO_2 levels over water regimes and between means of water regimes over CO_2 levels are indicated with different capital letters (Fisher's LSD test, $p \leq 0.05$, n = 10).

Parameter	400 ppm		700 ppm		400 ppm	700 ppm	100% FC	50% FC
	100% FC(1)	50% FC(2)	100% FC(3)	50% FC(4)				
HGR (cm day^{-1})	0.22 ± 0.08 ^{Aa}	0.08 ± 0.02 ^{Ab}	0.13 ± 0.04 ^{Ba}	0.06 ± 0.02 ^{Ab}	0.15 ^{A}	0.09 ^{B}	0.18 ^{A}	0.07 ^{B}
DGR (mm day^{-1})	0.03 ± 0.01 ^{Ba}	0.02 ± 0.01 ^{Ba}	0.05 ± 0.02 ^{Aa}	0.04 ± 0.01 ^{Ab}	0.03 ^{B}	0.05 ^{A}	0.04 ^{A}	0.03 ^{B}
SLA (m^2 kg^{-1})	19.9 ± 0.8 ^{Aa}	20.9 ± 1.4 ^{Aa}	15.5 ± 1.6 ^{Ba}	15.9 ± 0.9 ^{Ba}	20.4 ^{A}	15.7 ^{B}	17.7 ^{A}	18.4 ^{A}
ΔA_L (m^2 per plant)	0.78 ± 0.35 ^{Aa}	0.21 ± 0.09 ^{Ab}	0.65 ± 0.23 ^{Aa}	0.25 ± 0.12 ^{Ab}	0.49 ^{A}	0.45 ^{A}	0.71 ^{A}	0.23 ^{B}
C_i (ppm)	253 ± 15 ^{Ba}	227 ± 15 ^{Ba}	396 ± 25 ^{Aa}	401 ± 26 ^{Aa}	240 ^{B}	399 ^{A}	325 ^{A}	314 ^{A}
$R_{L\text{-}mass}$ (nmol g^{-1} s^{-1})	3.45 ± 1.42 ^{Aa}	3.73 ± 0.88 ^{Aa}	3.03 ± 0.29 ^{Aa}	3.32 ± 0.83 ^{Aa}	3.59 ^{A}	3.17 ^{A}	3.24 ^{A}	3.53 ^{A}
$R_{D\text{-}mass}$ (nmol g^{-1} s^{-1})	8.15 ± 1.09 ^{Aa}	7.45 ± 0.61 ^{Aa}	5.49 ± 0.65 ^{Ba}	5.98 ± 1.04 ^{Aa}	7.80 ^{A}	5.73 ^{B}	6.82 ^{A}	6.71 ^{A}

Fig. 2 - Carboxylation efficiency of Rubisco on a mass basis (CE_{mass}, panel a) and total nonstructural carbohydrates (TNC, panel b) in *Carapa surinamensis* grown at 400 and 700 ppm of CO_2 and two water regimes, soil at 50% FC and 100% FC. Significant differences between boxes are indicated with different small letters within a CO_2 level and different capital letters within a water regime (Fisher's LSD test, $p \leq 0.05$). Further information is described in Fig. 1.

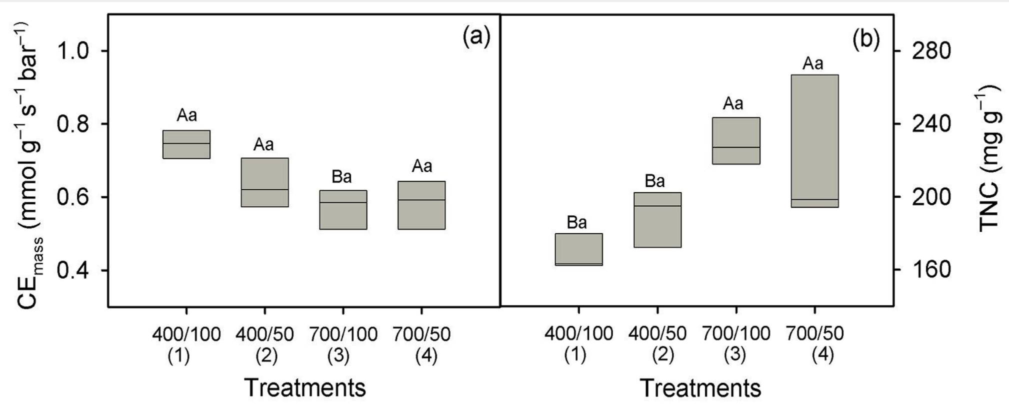


Fig. 3 - Gain of leaf dry matter (ΔW_L, panel a), stem dry matter (ΔW_S, panel b), root dry matter (ΔW_R, panel c), and shoot/root ratio (SRR, panel d) in *Carapa surinamensis* grown at 400 and 700 ppm of CO_2 and two water regimes, soil at 50% FC and 100% FC. Significant differences between boxes are indicated with different small letters within a CO_2 level and different capital letters within a water regime (Fisher's LSD test, $p \leq 0.05$). Further information is described in Fig. 1.

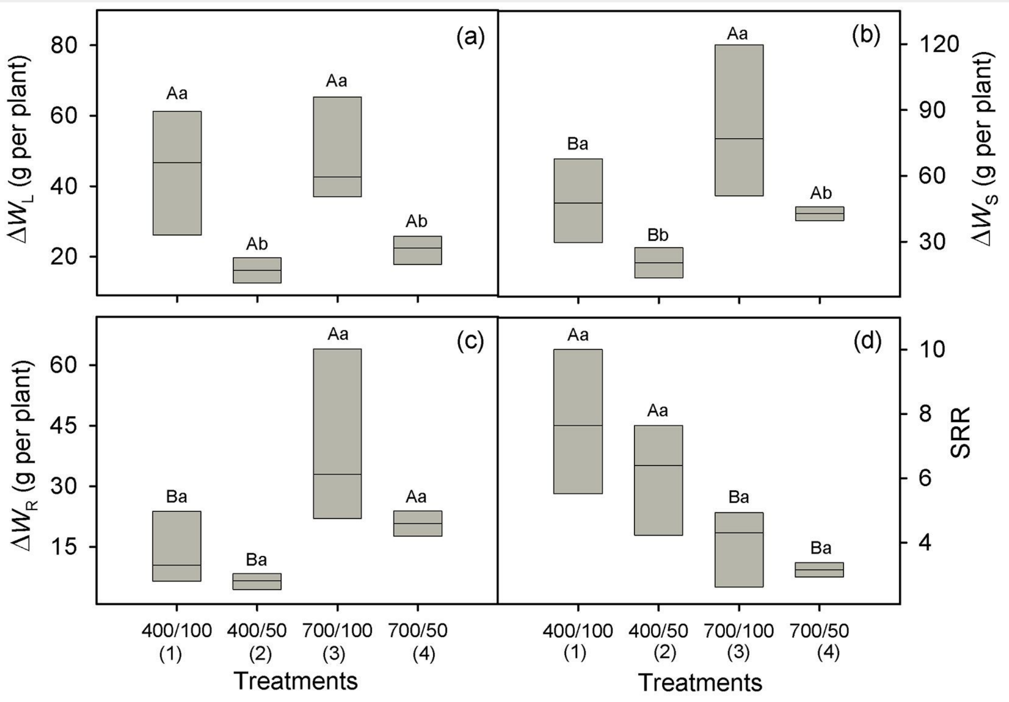
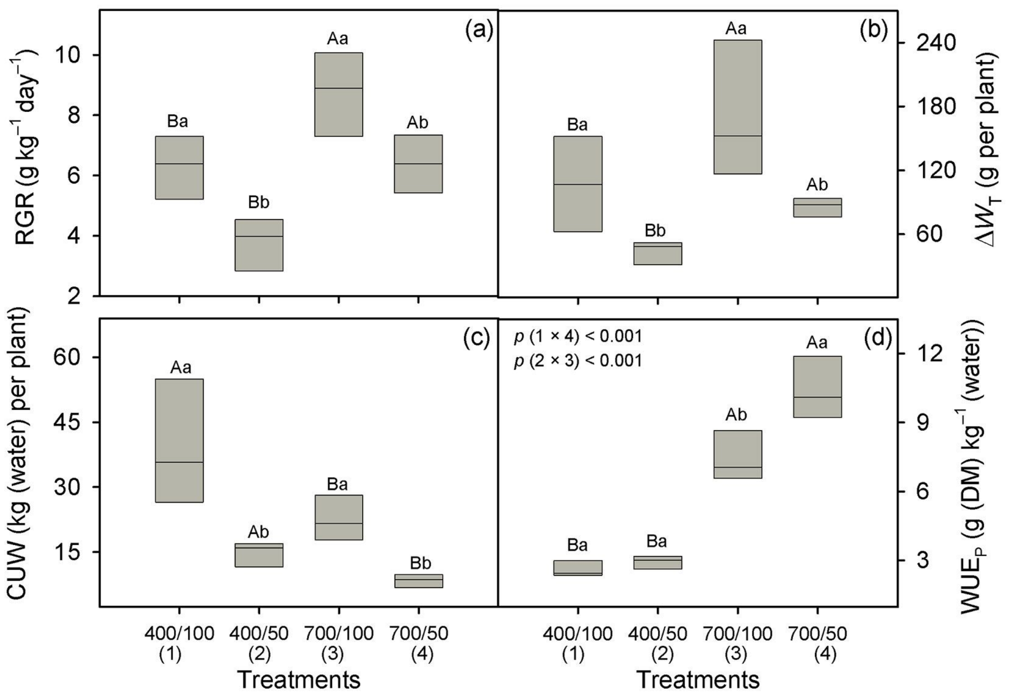


Fig. 4 - Relative growth rate (RGR, panel a), gain of total dry matter (ΔW_T, panel b), consumptive use of water (CUW, panel c) and whole-plant water-use efficiency (WUE_P, panel d) in *Carapa surinamensis* grown at 400 and 700 ppm of CO_2 and two water regimes, soil at 50% FC and 100% FC. Significant differences between boxes are indicated with different small letters within a CO_2 level and different capital letters within a water regime (Fisher's LSD test, $p \leq 0.05$). Further information is described in Fig. 1.



wise, the TNC content was only affected by CO_2 enrichment, with an increase of 27% under eCO_2 (179 *vs.* 227 mg g^{-1}, p = 0.002 – Fig. 2b, Tab. 1).

Discussion

Irrespective of the water regime, $A_{sat\text{-}mass}$ was greater in plants subjected to eCO_2 than in those kept at ambient CO_2 conditions, which indicates that the new leaves flushed during the experimental period did not experience down-regulation of photosynthetic at sustained eCO_2. Because the C_i values were higher at eCO_2 (240 ppm at ambient CO_2 against 399 ppm at eCO_2), this suggests that at ambient CO_2 the Rubisco carboxylation rate was limited by the diffusion of CO_2 to the carboxylation sites (Lambers et al. 2008, Leakey et al. 2012). Improvement of carbon assimilation under CO_2 enrichment is in accordance with the findings reported by others (Curtis & Wang 1998, Nowak et al. 2004, Ainsworth & Long 2005, Ainsworth & Rogers 2007). In well-watered plants we found a rise of 28% in $A_{sat\text{-}mass}$ under eCO_2, but that improvement was greater (93%) in plants submitted to drought. The decline of $A_{sat\text{-}mass}$ under drought at ambient CO_2 can be the result of the combined effect of diffusive (g_s and mesophyll conductance) and non-diffusive (biochemical) limitations of photosynthesis (Cornic et al. 1992, Parry et al. 2002, Flexas et al. 2012). One can see in Fig. 1a,c that the decrease in $A_{sat\text{-}mass}$ at ambient CO_2 under water stress was associated with a drop in $g_{sCO2\text{-}mass}$. It has been proposed that the decline in photosynthetic rate under mild drought (leaf water potential above -0.9 MPa) is most of time associated with a decline in stomatal conductance (Cornic et al. 1992). The increase of $A_{sat\text{-}mass}$ at eCO_2 concurs with the results reported by Ainsworth & Long (2005) and Nowak et al. (2004) who found that photosynthesis increases by about 30-50% under eCO_2. Kelly et al. (2016) also reported an improvement in photosynthetic rates and a decline in the C_i/C_a ratio in response to CO_2-enrichment, which is in agreement with the results we found in this study (Fig. 1d).

There was a slight decline (17%) of CE_{mass} under elevated eCO_2 (Fig. 2a), which can be ascribed to a decrease in SLA (Tab. 2), as leaf thickness inversely increased with SLA (Lambers et al. 2008). However, it is apparent that this small decline in CE_{mass} did not impair $A_{sat\text{-}mass}$ which increased at eCO_2. This concurs with the result of a meta-analysis carried out by Ainsworth & Long (2005) who found just a slight decline (6%) in the maximum carboxylation rate of Rubisco. Both $R_{L\text{-}mass}$ and $R_{D\text{-}mass}$ tended to decrease at eCO_2 (11-20%) but the effect of eCO_2 was only significant for dark respiration (Tab. 1). This is in agreement with the result reported by Curtis & Wang (1998), who found a small decrease (18%) in $R_{D\text{-}mass}$ under eCO_2. The decrease in leaf respiration can be ascribed, at least in part, to a decrease in SLA under eCO_2. Much of the discrepancy on the effect of eCO_2 on leaf respiration can be attributed to the difficulty in measuring this parameter with gas exchange techniques (Leakey et al. 2009). We found an increase (27%) in TNC under eCO_2, and it has been suggested that the accumulation of leaf carbohydrates at eCO_2 can reduce the expression of genes coding for photosynthetic enzymes (Córdoba et al. 2017), but in this study we did not found evidence of photosynthetic acclimation.

Subjecting the plants to eCO_2 mitigates the negative effect induced by low water availability and, on average, $A_{sat\text{-}mass}$ increased 54% at eCO_2. However, contrary to expectation $A_{max\text{-}mass}$ declined at eCO_2 in well-watered plants and under drought at ambient CO_2. The decline of $A_{max\text{-}mass}$ under ambient CO_2 can be attributed, at least partially, to a reduction in $g_{sCO2\text{-}mass}$ (Fig. 1c), but an effect of mesophyll conductance cannot be ruled out (Flexas et al. 2012). In fact, in several species there is a close correlation between mesophyll conductance and photosynthetic rate (Lauteri et al. 1997, Singsaas et al. 2004, Bahar et al. 2018). It has been reported that an increase in TNC under eCO_2 can lead to down-regulation of photosynthesis (Leakey et al. 2012, Fatichi et al. 2014). Although $A_{sat\text{-}mass}$ was not reduced by eCO_2 (Fig. 1a), down-regulation of $A_{max\text{-}mass}$ in response to an increase in TNC cannot be entirely disregarded. It is important to note, however, that $A_{max\text{-}mass}$ was measured at a C_a value of 2000 ppm (mean C_i of about 1100 ppm) against a C_a of 700 ppm (C_i of 399 ppm) used for measuring A_{sat} at eCO_2. Kitao et al. (2015) found that mesophyll conductance exponentially declines with increasing C_i. Furthermore, Leakey et al. (2012) suggested that the apparent down-regulation of carboxylation rates at eCO_2 can also be caused by a decrease in mesophyll conductance. Thus, it seems plausible to suggest that the decline of $A_{max\text{-}mass}$ at eCO_2 was caused by a decline of mesophyll conductance and, to a lesser extent, by the increase in TNC at eCO_2.

In well-watered plants, $g_{sCO2\text{-}mass}$ was lower under eCO_2 which is not unexpected, as the most common response is a decrease of stomatal conductance under eCO_2 (Curtis & Wang 1998, Ainsworth & Long 2005, Leakey et al. 2009, 2012). One can see in Fig. 1c that $g_{sCO2\text{-}mass}$ did not decline under drought at eCO_2, which leads to similar C_i/C_a values under CO_2 enrichment (Fig. 1d). This shows that stomata were sensitive to drought at ambient CO_2, but rather insensitive at eCO_2, which suggests some stomatal acclimation (a physiological change triggered by a new environmental condition) to eCO_2 (Morison 1998). Yan et al. (2017) found that under drought, stomatal conductance was dependent on xylem-abscisic acid concentration at ambient CO_2, whereas it was insensitive to abscisic acid and predominantly regulated by leaf turgor at eCO_2. We show that subjecting the plants to eCO_2 negates the effect of drought on stomatal conductance, and consequently water stress did not lead to a reduction in $A_{sat\text{-}mass}$ under eCO_2. Even when predawn leaf water potential only slightly declined under drought (-0.20 to -0.34 MPa), there was a sharp decline of $g_{sCO2\text{-}mass}$ at ambient CO_2 under drought, which suggests that andiroba is rather sensitive to small changes in leaf water potential (Camargo & Marenco 2012).

Besides the effect of eCO_2 on $g_{sCO2\text{-}mass}$, the pronounced reduction in total leaf area under water stress also contributed to reduce CUW under drought (Fig. 4c). This occurs because of the negative effect of water stress on leaf production, as cell division and leaf expansion are greatly reduced under drought (Bradford & Hsiao 1982, Tardieu et al. 2011). At eCO_2 the drop in CUW essentially mirrored the drop in ΔW_L, as $g_{sCO2\text{-}mass}$ did no decline under drought at eCO_2. At ambient CO_2, however, the drop in CUW reflected both the decline in ΔW_L and $g_{sCO2\text{-}mass}$. Besides, in the greenhouse the air was slightly drier (10% lower RH) and a little (1.5 °C) warmer than in the growth chamber, which could also have had a small effect on CUW and thereby on WUE_P at ambient CO_2.

WUE_P was greatly improved by submitting the plants to eCO_2 (Fig. 4d), because of the combined effect of a decline in CUW and an increase in ΔW_T. The enhancement of WUE_P at eCO_2 is agreement with the results reported by others (Kauwe et al. 2013, Kelly et al. 2016). Cernusak et al. (2011) found that WUE_P of seedlings of *Swietenia macrophylla* and *Ormosia macrocalyx* (tropical trees) can improve from 54% (*S. macrophylla*) to 91% (*O. macrocalyx*) in response to eCO_2, which shows that WUE_P can exhibit large variation between species even at the same experimental conditions. It is worth noting, that $g_{sCO2\text{-}mass}$ and WUE_P followed different trends. For example, $g_{sCO2\text{-}mass}$ declined under drought at ambient CO_2, whereas WUE_P remained unchanged in that condition (Fig. 4d). This can be explained if we take into account that, besides stomatal conductance, WUE_P also depend on other factors including biomass allocation to plant organs, leaf morphology, plant respiration and canopy boundary layer conductance, which ultimately leads to difference between A/E and WUE_P (Lambers et al. 2008, Leakey et al. 2012).

The ΔW_T increased 73% at eCO_2 and decreased by 54% under drought over CO_2 levels. This is *in tandem* with the increase of $A_{sat\text{-}mass}$ recorded at eCO_2 (28% and 93%, depending on water regime), which resulted in greater amount of carbon allocated to stems and roots. Our ΔW_T values are greater than the above ground biomass gain (20-30%) reported by others (Curtis & Wang 1998, Ainsworth & Long 2005) in plants subjected to eCO_2. This discrepancy can be explained by considering that *Carapa* trees grow at high rates during the juvenile stage (Dunisch et al. 2002, Camargo & Marenco 2012). On the other hand, our results are consistent with those reported by Cernusak et al. (2011), who found that the biomass of well-watered tropical seedlings can increase from about 70% (*Swietenia macrophylla*) to 150% (*Ormosia macrocalyx*) at eCO$_2$. Large biomass allocation to stems and roots suggests that these plant organs can be the predominant sinks of *Carapa* saplings under CO$_2$ enrichment, as ΔW_L remained unaltered at eCO$_2$. In comparison with ambient CO$_2$, more carbon was allocated to roots at eCO$_2$ (Fig. 3c), and due to the absence of an effect of eCO$_2$ on ΔW_L, this ultimately led to a lower SRR. The decline of SRR at eCO$_2$ is rather unexpected, as in several species it remains unchanged under eCO$_2$ (Poorter & Nagel 2000). The drastic reduction in ΔW_T at ambient CO$_2$ under water stress is consistent with the long-term decline in total biomass (250 to 150 Mg(C) ha^{-1}, in 10 yrs) observed under drought (*i.e.*, a 50% reduction in water supply – Grace 2016). RGR increased under eCO$_2$, which concurs with the finding reported by Poorter & Nagel (2000). We found that the amount of carbon stored as TNC increased only 27% at eCO$_2$, *i.e.*, less than the starch increase of 60-80% reported by Nowak et al. (2004), which is consistent with the large fraction of biomass accumulated in stems and roots. SLA was greatly decreased under CO$_2$-enrichment. This is in accordance with the rise in TNC often found at eCO$_2$ (Curtis & Wang 1998, Ainsworth & Long 2005).

Conclusions

We postulated that plants under CO$_2$-enrichment enhance $A_{sat\text{-}mass}$, biomass accumulation and WUE$_P$, and that eCO$_2$ leads to a reduction of stomatal conductance, which was supported by data. However, surpassing our expectations, submitting the plants to eCO$_2$ entirely nullifies the negative effect of drought on $A_{sat\text{-}mass}$ and $g_{sCO2\text{-}mass}$, and improves total biomass accumulation, but unexpectedly CO$_2$-enrichment leads to a decline in $A_{max\text{-}mass}$. Total leaf area was greatly reduced by drought, but the absence of a positive effect of eCO$_2$ on leaf area production was unexpected. These findings widen our understanding of the effect of eCO$_2$ and water stress on the physiology of *Carapa*, an important species of multipurpose use, particularly in the Amazon region. It should be acknowledged, however, that several factors can affect the performance of mature trees in their natural environment (*e.g.*, light, nutrient and water availability, and the capacity of the root system to explore the soil for water and nutrients), which suggests caution in extrapolating results from green-house experiments to forest ecosystems.

List of abbreviations

(A): net photosynthetic rate; (A_L): total leaf area; ($A_{sat\text{-}mass}$): light saturated photosynthesis on a mass basis; ($A_{max\text{-}mass}$): light and CO$_2$-saturated photosynthesis on a mass basis; (B): biomass; (ΔB): biomass gain over the experimental period; (C_a): ambient CO$_2$ concentration; (CE$_{mass}$): carboxylation efficiency on a mass basis; (C_i): intercellular CO$_2$ concentration; (CUW): consumptive use of water (amount of water consumed by a plant); (DM): dry matter; (eCO$_2$): elevated CO$_2$ concentration; ($g_{sCO2\text{-}mass}$): stomatal conductance to CO$_2$ on a mass basis; (Log): logarithm; (PAR): photosynthetically active radiation; ($R_{L\text{-}mass}$): leaf respiration in the light on a mass basis; ($R_{D\text{-}mass}$): dark respiration on a mass basis; (RGR): relative growth rate; (SLA): specific leaf area; (SRR): shoot/root ratio; (TNC): total nonstructural carbohydrate; (W_L): leaf DM; (W_S): stem DM; (W_R): root DM; (W_T): total DM; (WUE$_P$): whole-plant water-use efficiency.

Acknowledgments

The authors acknowledge the Brazilian Ministry of Science, Technology, Innovations and Communications (MCTIC/INPA PRJ 15.120), and the National Council for Scientific and Technological Development (CNPq, 302041/2015-0) for grant (to RAM) and scholarship to MFO. We thank the anonymous reviewers for their useful comments and suggestions.

Author contributions

MFO collected data and conducted statistical analysis and RAM supervised the experimental work and wrote the article with contributions of the first author.

References

Ainsworth EA, Long SP (2005). What have we learned from 15 years of free-air CO$_2$ enrichment (FACE)? A meta-analytic review of the responses of photosynthesis, canopy properties and plant production to rising CO$_2$. New Phytologist 165: 351-372. - doi: 10.1111/j.1469-8137.2004.01224.x

Ainsworth EA, Rogers A (2007). The response of photosynthesis and stomatal conductance to rising [CO$_2$]: mechanisms and environmental interactions. Plant, Cell and Environment 30: 258-270. - doi: 10.1111/j.1365-3040.2007.01641.x

Bahar NH, Hayes L, Scafaro AP, Atkin OK, Evans JR (2018). Mesophyll conductance does not contribute to greater photosynthetic rate per unit nitrogen in temperate compared with tropical evergreen wet-forest tree leaves. New Phytologist 218: 492-505. - doi: 10.1111/nph.15031

Bellasio C, Quirk J, Beerling DJ (2018). Stomatal and non-stomatal limitations in savanna trees and C4 grasses grown at low, ambient and high atmospheric CO$_2$. Plant Science 274: 181-192. - doi: 10.1016/j.plantsci.2018.05.028

Bradford KJ, Hsiao TC (1982). Physiological responses to moderate water stress. In: "Physiological Plant Ecology II - Water Relation and Carbon Assimilation" (Lange OL, Nobel PS, Osmond CB, Ziegler H eds). Springer-Verlag, Heidelberg, Germany, pp. 263-324. - doi: 10.1007/978-3-642-68150-9_10

Brazil-MMA (2016). Nota técnica 01/2016/DEMC/SMCQ/MMA [Technical note 01/2016/DEMC/SMCQ-MMA]. Ministério do Meio Ambiente MMA, Brasília, Brazil, pp. 1-6. [online] URL: http://www.mma.gov.br/images/arquivos/clima/convencao/indc/NotaTecnicaMMA_RatificacaoAcordoParis.pdf

Brienza S, Pereira JF, Yared JAG, Mourão M, Gonçalves DA, Galeão RR (2008). Recuperação de áreas degradadas com base em sistema de produção florestal energético-madeireiro: indicadores de custos, produtividade e renda [Land reclamation based on production forestry system: cost indicators, productivity and incomes]. Amazônia Ciência e Desenvolvimento 4: 197-219. [in Portuguese]

Camargo MAB, Marenco RA (2012). Growth, leaf and stomatal traits of crabwood (*Carapa guianensis* Aubl.) in central Amazonia. Revista Árvore 36: 07-16. - doi: 10.1590/S0100-67622012000100002

Camargo MAB, Marenco RA (2017). Tree growth over three years in response to monthly rainfall in central Amazonia. Dendrobiology 78: 10-17. - doi: 10.12657/denbio.078.002

Cernusak LA, Winter K, Martínez C, Correa E, Aranda J, Garcia M, Jaramillo C, Turner BL (2011). Responses of legume *versus* non legume tropical tree seedlings to elevated CO$_2$ concentration. Plant Physiology 157: 372-385. - doi: 10.1104/pp.111.182436

Córdoba J, Pérez P, Morcuende R, Molina-Cano JL, Martinez-Carrasco R (2017). Acclimation to elevated CO$_2$ is improved by low Rubisco and carbohydrate content, and enhanced Rubisco transcripts in the G132 barley mutant. Environmental and Experimental Botany 137: 36-48. - doi: 10.1016/j.envexpbot.2017.02.005

Cornic G, Ghashghaie GCJ, Genty B, Briantais JM (1992). Leaf photosynthesis is resistant to a mild drought stress. Photosynthetica 27: 295-309. [online] URL: http://www.researchgate.net/publication/229076628

Curtis PS, Wang X (1998). A meta-analysis of elevated CO$_2$ effects on woody plant mass, form, and physiology. Oecologia 113: 299-313. - doi: 10.1007/s004420050381

Dias DP, Marenco RA (2016). Tree growth, wood and bark water content of 28 Amazonian tree species in response to variations in rainfall and wood density. iForest - Biogeosciences and Forestry 9: 445-451. - doi: 10.3832/ifor1676-008

Duffy PB, Brando P, Asner GP, Field CB (2015). Projections of future meteorological drought and wet periods in the Amazon. Proceedings of the National Academy of Sciences USA 112: 13172-13177. - doi: 10.1073/pnas.1421010112

Dunisch O, Schwarz T, Neves EJM (2002). Nutrient fluxes and growth of *Carapa guianensis* Aubl. in two plantation systems in the Central Amazon. Forest Ecology and Management 166: 55-68. - doi: 10.1016/S0378-1127(01)00675-2

Evans JR, Seemann JR (1984). Differences between wheat genotypes in specific activity of ribulose-1,5-bisphosphate carboxylase and the relationship to photosynthesis. Plant Physiology 74: 759-765. - doi: 10.1104/pp.74.4.759

Fatichi S, Leuzinger S, Körner C (2014). Moving beyond photosynthesis: from carbon source to sink-driven vegetation modeling. New Phytologist 201: 1086-1095. - doi: 10.1111/nph.12614

Flexas J, Gallé A, Galmés J, Ribas-Carbo M, Medrano H (2012). The response of photosynthesis to soil water stress. In: "Plant Responses to Drought Stress" (Aroca R ed). Springer-Verlag, Berlin, Germany, pp. 129-144. - doi: 10.1007/978-3-642-32653-0_5

Fournier LA (2002). Species description *Carapa guianensis*. In: "Tropical tree seed manual" (Vozzo JA ed). Agricultural Handbook 72, USDA Forest Service, Washington, DC, USA, pp. 360-362.

Grace J (2016). The Amazon carbon balance: an evaluation of methods and results. In: "Interactions between Biosphere, Atmosphere and Human Land Use in the Amazon Basin" (Nagy L, Forsberg BR, Artaxo P eds). Springer-Verlag, Berlin, Germany, pp. 79-100. - doi: 10.1007/978-3-662-49902-3_5

Kauwe MG, Medlyn BE, Zaehle S, Walker AP, Dietze MC, Hickler T, Jain AK, Luo Y, Parton WJ, Prentice IC, Smith B, Thornton PE, Wang S, Wang Y, Warlind D, Weng E, Crous KY, Ellsworth DS, Hanson PJ, Kim H, Warren JM, Oren R, Norby RJ (2013). Forest water use and water use efficiency at elevated CO_2: a model-data intercomparison at two contrasting temperate forest FACE sites. Global Change Biology 19: 1759-1779. - doi: 10.1111/gcb.12164

Kelly JW, Duursma RA, Atwell BJ, Tissue DT, Medlyn BE (2016). Drought x CO_2 interactions in trees: a test of the low-intercellular CO_2 concentration (Ci) mechanism. New Phytologist 209: 1600-1612. - doi: 10.1111/nph.13715

Kenfack D (2011). A synoptic revision of *Carapa* (Meliaceae). Harvard Papers in Botany 16: 171-231. - doi: 10.3100/0.25.016.0201

Kitao M, Yazaki K, Kitaoka S, Fukatsu E, Tobita H, Komatsu M, Maruyama Y, Koike T (2015). Mesophyll conductance in leaves of Japanese white birch (*Betula platyphylla* var. *japonica*) seedlings grown under elevated CO_2 concentration and low N availability. Physiologia Plantarum 155: 435-445. - doi: 10.1111/ppl.12335

Klimas CA, Cropper WP, Kainer KA, Wadt LHO (2012). Viability of combined timber and non-timber harvests for one species: a *Carapa guianensis* case study. Ecological Modelling 246: 147-156. - doi: 10.1016/j.ecolmodel.2012.07.022

Lambers H, Chapin FS, Pons TL (2008). Plant physiological ecology (2nd edn). Springer, New York, USA, pp. 11-254.

Lauteri M, Scartazza A, Guido MC, Brugnoli E (1997). Genetic variation in photosynthetic capacity, carbon isotope discrimination and mesophyll conductance in provenances of *Castanea sativa* adapted to different environments. Functional Ecology 11: 675-683. - doi: 10.1046/j.1365-2435.1997.00140.x

Leakey ADB, Ainsworth EA, Bernacchi CJ, Rogers A, Long SP, Ort DR (2009). Elevated Longed CO_2 effects on plant carbon, nitrogen, and water relations: six important lessons from FACE. Journal of Experimental Botany 60: 2859-2876. - doi: 10.1093/jxb/erp096

Leakey ADB, Ainsworth EA, Bernacchi CJ, Zhu X, Long SP, Ort DR (2012). Photosynthesis in a CO_2-rich atmosphere. In: "Photosynthesis: Plastid Biology, Energy Conversion and Carbon Assimilation" (Eaton-Rye JJ, Tripathy BC, Sharkey TD eds). Springer, Dordrecht, The Netherlands, pp. 733-768. - doi: 10.1007/978-94-007-1579-0_29

Marenco RA, Camargo MAB, Antezana-Vera AS, Oliveira MF (2017). Leaf trait plasticity in six forest tree species of central Amazonia. Photosynthetica 55: 679-688. - doi: 10.1007/s11099-017-0703-6

Marenco RA, Gonçalves JFC, Vieira G (2001). Photosynthesis and leaf nutrient contents in *Ochroma pyramidale* (Bombacaceae). Photosynthetica 39: 539-543. - doi: 10.1023/A:1015699927924

Morison JIL (1998). Stomatal response to increased CO_2 concentration. Journal of Experimental Botany 49: 443-452. - doi: 10.1093/jxb/49.Special_Issue.443

Nowak RS, Ellsworth DS, Smith SD (2004). Functional responses of plants to elevated atmospheric CO_2 - do photosynthetic and productivity data from FACE experiments support early predictions? New Phytologist 162: 253-280. - doi: 10.1111/j.1469-8137.2004.01033.x

Pan Y, Birdsey RA, Phillips OL, Jackson RB (2013). The structure, distribution, and biomass of the world's forests. Annual Review of Ecology, Evolution and Systematics 44: 593-622. - doi: 10.1146/annurev-ecolsys-110512-135914

Parry MAJ, Andralojc P, Khan S, Lea P, Keys AJ (2002). Rubisco activity: effects of drought stress. Annals of Botany 89: 833-839. - doi: 10.1093/aob/mcf103

Poorter H, Nagel O (2000). The role of biomass allocation in the growth response of plants to different levels of light, CO_2, nutrients and water: a quantitative review. Australian Journal of Plant Physiology 27: 595-607. - doi: 10.1071/PP99173_CO

Saatchi SS, Houghton RA, Alvala RCS, Soares JV, Yu Y (2007). Distribution of aboveground live biomass in the Amazon basin. Global Change Biology 13: 816-837. - doi: 10.1111/j.1365-2486.2007.01323.x

Salati E, Vose PB (1984). Amazon Basin: a system in equilibrium. Science 225: 129-138. - doi: 10.1126/science.225.4658.129

Singsaas EL, Ort DR, Delucia EH (2004). Elevated CO_2 effects on mesophyll conductance and its consequences for interpreting photosynthetic physiology. Plant, Cell and Environment 27: 41-50. - doi: 10.1046/j.0016-8025.2003.01123.x

Souza CR, Azevedo CP, Rossi L (2008). Desempenho de espécies florestais para uso múltiplo na Amazônia [Efficiency of forest species for multiple use in Amazonia]. Scientia Forestalis 36: 7-14. [in Portuguese]

Tardieu F, Granier C, Muller B (2011). Water deficit and growth. Co-ordinating processes without an orchestrator? Current Opinion in Plant Biology 14: 283-289. - doi: 10.1016/j.pbi.2011.02.002

Way DA, Oren R, Kroner Y (2015). The space-time continuum: the effects of elevated CO_2 and temperature on trees and the importance of scaling. Plant, Cell and Environment 38: 991-1007. - doi: 10.1111/pce.12527

Yan F, Li X, Liu F (2017). ABA signaling and stomatal control in tomato plants exposure to progressive soil drying under ambient and elevated atmospheric CO_2 concentration. Environmental and Experimental Botany 139: 99-104. - doi: 10.1016/j.envexpbot.2017.04.008

Supplementary Material

Tab. S1 - Means of data used in Figs. 1-4.

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