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# Leaf Gas Exchange, Photon Capture and Light Harvest in *Aldina heterophylla* along a Vegetation Gradient in the Amazon Rainforest

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## Abstract

**Adaptation along environmental gradients is presumed to induce physiological and biochemical leaf changes in plant species. In this paper, we report how leaf gas exchange, photon capture and light harvest for photosynthesis in *Aldina heterophylla* change along a vegetation gradient from low stature open vegetation on extremely nutrient-poor white sand (Campina, CP), through intermediate closet type (Campinarana, CR) to tall closed rain forest (RF). The pigment concentrations did not differ between the CP, CR and RF habitats. The performance index for the photosynthesis (PIABS) of individuals in RF and CP was approximately 30% higher than that in CR individuals. This species showed similar potential rates of photosynthesis in the different vegetation types; however, the dark respiration rates were higher in CP. Our results indicate that the differences in the leaves and soil nitrogen concentrations are not enough to change the levels of gas exchange. Other environmental features may be driving the observed morphological features in this gradient, in particular, the tree height.**

## Keywords

**Chloroplast Pigments, Dark Respiration, Net Photosynthesis, Physiological Plasticity, Tropical Forest, Biomass Accumulation**

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## 1. Introduction

The Campina vegetation type is made up of low-stature tree species that are associated with white poor sand soil along the Rio Negro river basin within the Brazilian Amazon. This forest type normally shows a transition to a tall evergreen forest along an edaphic environmental gradient as sand soil patches transition into more fertile soils (**Figure 1**).

Along this gradient, environmental traits, such as the air temperature, air relative humidity, soil nutrients available, irradiance at the soil surface and other differences (**Table 1**), can define the distribution of the plants.

Photosynthetic characteristics could be unique to each plant and differ between plants that occur in habitats with different soil characteristics, with a result of few species occurring along an entire gradient. Many studies have demonstrated the differences of leaf gas exchange between contrasting habitats [1]-[6]; however, few studies have reported on the behavior and functional variations of leaves and the photosynthetic characteristics of species that occur along a vegetation gradient, which are the result of multiple environmental factors.

The features of the vegetation gradient, especially poor, sandy soil, have been shown to influence plant functional traits, such as the xylem structure, LMA and construction cost [7]-[9], but it is unknown how the plant physiological features can change along this gradient.

The total nitrogen concentration in soil has traditionally been reported as the main factor in biomass accumulation because it increases photosynthesis as a large fraction of the leaf nitrogen is invested in the photosynthetic apparatus and leaf nitrogen contents reflect the amounts of photosynthetic proteins [10] [11]. In general, the growth of plants at higher irradiance and nitrogen availabilities includes a higher net photosynthesis and nitrogen leaf concentration [12]. Although the relationship between nitrogen and photosynthesis is generally significantly positive and linear, sometimes it can be curvilinear. In this situation, the efficient use of photosynthetic nitrogen decreases with increasing leaf nitrogen concentration [13].

Though we found variations within each species, the consensus is that nitrogen limitations reduce the synthe-


**Figure 1.** Photo illustration of Campina vegetation (A), Campinarana (B) and Rain Forest (C) in the Biological Reserve of Campina INPA, Manaus-AM.

**Table 1.** Predominant differences in environmental features and vegetation structure of the vegetation gradient in the Biological Reserve of Campina of National Institute for Research in the Amazon.

| | CP | CR | RF |
|---|---|---|---|
| [1]Vegetation type | Small patches of trees and shrubs (±3 m high) that are surrounded by areas of bare soil (white sand) | Uniform vegetation, with no open areas, on the white sand with trees and shrubs ± 12 m high | Rain Forest with high DAP and high trees ± 25 m high |
| [2]Irradiance at the soil surface | Total-open vegetation | High irradiance | Low irradiance |
| [3]Air temperature (annual mean) | 22˚C - 38˚C | 20˚C - 29˚C | 23˚C - 27˚C |
| [3]Air humidity (annual mean) | 81% - 90% | 91% - 93% | 91% - 97% |
| [4]Soil particle size composition at 10 - 20 cm depth | 98% sand, 1.2% clay | 96% sand, 3.6% clay | 80% sand, 11.6% clay |
| [4]Total nitrogen ($mg \cdot g^{-1}$)10 - 20 cm depth | 0.37 | 0.45 | 0.6 |
| [4]Carbon/Nitrongen | 13 | 20 | 19 |
| [4] $pH_{H_2O}$ | 3.4 | 3.9 | 4.2 |

[1][15], [2][16]. [3][17], [4][18].

sis of proteins by the photosynthetic apparatus, which can decrease the absorption of light and the subsequent-conversion of excitation energy to chemical energy via Photosystem II (PSII). Chlorophyll fluorescence measurements could be used to rapidly and non-invasively estimate the quantum efficiency of electron transport through PSII in leaves [14]. The investigation of the fluorescence of chlorophyll *a* and analysis of gas exchange can, in a complementary manner, shed some light on the ability of plants to acclimate along vegetation gradients. We investigated the degree of physiological plasticity in *A. heterophylla* across three different vegetation types and the corresponding soil and microclimate differences. The specific hypotheses were as follows:

1) Gas exchange features of *A. heterophylla* leaves change along the vegetation gradient.

2) Characteristics of absorption and light processing will indicate in which habitat type *A. heterophylla* will be most vulnerable to environmental stress.

## 2. Material and Methods

### 2.1. Study Sites

The study was conducted at the Biological Reserve of Campina of the National Institute for Research in the Amazon, Amazonas State, Brazil (2˚34'S; 60˚02'W) between March and June in 2008. In this area, there is a vegetation gradient of Campina (CP), Campinarana (CR) and Rainforest (RF), with different environmental features and vegetation structures (**Table 1**). Campina (open low-stature heath forest) are "islands of vegetation with xeromorphic features" on white sand [15] and are characterized by open vegetation, low plant diversity and high endemism on spodsolic soils (**Figure 1**). Campina grade into Campinarana (dense heath forest, CR), which are characterized by arborescent vegetation with a few emergent trees and an understory with a high density of trees (DBH < 10) that also grow on spodsols with a leached, sandy layer. They differ from Campina in that they have a continuous and uniform canopy and large amounts of undecomposed litter (**Figure 1**) [15]. The third vegetation type found along the gradient is tall rain forest (RF). RF occurs primarily on oxisols with varying sand content and experiences less litter accumulation and higher decomposition rates than does CR (**Figure 1**).

The climate of the Reserve, according to Köppen classification, is Ami, with a dry season from May to October and a rainy season occurring from November to April [19].

### 2.2. The Study Species

*Aldina heterophylla* Spruce ex Benth (Fabaceae) is a woody shrub or tree belonging to the subfamily Papilionoideae, tribe Swartziaeae, which shows no evidence of root nodules or nitrogen fixation but can show arbuscular mycorrhizas [20] [21]. It usually has compound leaves (varying from two to five leaflets, most commonly found with three) or may be unifoliate, such as in the CP [20] [22]. In the CP, *A. heterophylla* often forms islands of vegetation in CP, where it has the highest importance value index and can reach up to 3 meters high [17] (**Figure 1**). *A. heterophylla* is a canopy species in the other vegetation types, reaching an average of 8 to 10 meters high in CR and approximately 20 to 25 meters high in the RF.

### 2.3. Determination of Chloroplast Pigment Concentrations

The terminal leaflet of fully expanded leaves was used for the extraction of pigments. Three leaves of 10 individuals of *A. heterophylla* in CP, CR and RF were collected in the morning and placed in a cooler until transport to the laboratory. The concentration of chlorophyll *a*, chlorophyll *b* and carotenoids were extracted using acetone as described in [23] and quantified using a spectrophotometer (Ultrospec 2100 pro UV/visible—Amersham Biosciences). From the concentrations of chlorophylls and carotenoids, the concentration of the total chlorophyll (chlorophyll *a* + chlorophyll *b*), chlorophyll *a*/chlorophyll *b* (Chl *a*:*b*) and total chlorophyll/carotenoids ($Chl_{tot}$: Car) were calculated. The chloroplastic pigment contents were calculated based on the area ($\mu mol \cdot cm^{-2}$), and we used the equations described by [24].

### 2.4. Determination of Chlorophyll *a* Fluorescence

Chlorophyll *a* fluorescence was determined by a portable fluorometer (Plant Efficiency Analyzer-MK2-9600—Hansatech, Norfolk, UK) between 0800 and 1100 hours in 10 individuals per vegetation type and 10 mature terminal leaflets per individual. The leaflets selected were subjected to a period of 30 minutes of dark

adaptation, after which they were exposed to a pulse of saturating light at an intensity of 3000 μmol·m$^{-2}$·s$^{-1}$ issued by six diodes (wavelength 650 nm) for 5 s in the central region of the adaxial side of the lamina. For each leaf, the following variables of fluorescence were obtained: the initial fluorescence at 0 seconds ($F_0$), fluorescence $F_{50\mu s}$ (considered $F_0$ in this study), $F_{100\mu s}$, $F_{300\mu s}$, $F_{2ms}$, $F_{30ms}$, maximum fluorescence ($F_m$) and variable fluorescence ($F_v = F_m - F_0$).

From these data, we calculated the parameters shown in **Table 2**.

## 2.5. Determination of Gas Exchange

The net photosynthetic rate responses to irradiance ($P_n$-I) (light curves) were determined by measuring the instantaneous $CO_2$ gas exchange with an infrared gas analyzer—IRGA (LI-6400, LI-COR Biosciences). The measurements were made on fully expanded terminal leaflets that were located in the upper crown in full sun between 0800 and 1100 hours. The leaves were accessed using temporary towers assembled next to each plant for this purpose. To measure the light response of net photosynthesis, we used 11 levels of irradiance inside the leaf chamber (Photosynthetic Photon Flux Density (PPFD): 0, 25, 50, 75, 100, 250, 500, 750, 1000, 1500, 2000 μmol·m$^{-2}$·s$^{-1}$) in ascending order. The LI-6400 IRGA was fixed at a rate of flow of 400 μmol·s$^{-1}$ and was fitted with a leaf chamber for measuring the concentration of $CO_2$, temperature and $H_2O$ vapor at approximately 385 ± 1 μmol·mol$^{-1}$, 31˚C ± 1˚C and 21 ± 1 mmol·mol$^{-1}$, respectively. The VPD was maintained at 2.02 ± 0.3. Before the start of the measurements, the leaves were subjected to an irradiance of 1000 μmol·m$^{-2}$·s$^{-1}$ for a period of 5 to 10 min for the adaptation of the leaf to the measuring chamber. The exponential equation model (Equation (1)) was used to fit the photosynthetic response to a light intensity curve [25]:

$$P_n = P_{nmax} - \left( \frac{P_{nmax} + R_d}{e^{\alpha I / P_{nmax}} + R_d} \right) \quad (1)$$

where:

I = irradiance (PPFD);

$P_n$ = net photosynthetic $CO_2$ assimilation rate (μmol $CO_2$ m$^{-2}$·s$^{-1}$);

$P_{nmax}$ = maximum net photosynthetic $CO_2$ assimilation rate;

$R_d$ = respiration in the dark-adapted leaves (μmol $CO_2$ m$^{-2}$·s$^{-1}$), *i.e.*, when I = 0 μmol $CO^2$ m$^{-2}$·s$^{-1}$;

$\alpha$ = apparent quantum yield.

In this work, only $P_{nmax}$ and $\alpha$ were estimated by the model. The light compensation irradiance (Ic; PPFD when $P_n = 0$) was calculated by the formula $I_c = R_d/\alpha$, and the light saturation irradiance ($I_s$; PPFD when $P_{nsat}$ = 90% $P_{nmax}$) was estimated as (Equation (2)):

**Table 2.** Detailing the data obtained and calculated from the fluorescence of chlorophyll *a*, and parameters used in this text (modified from [14]).

| *Data extracted from the recorded fluorescence of chlorophyll a* | | |
|---|---|---|
| Fluorescence at 50 μs | $F_0$ | |
| Maximal fluorescence, when all Photosystem II reaction centers are closed | $F_m$ | |
| Variable fluorescence | $F_v$ | |
| Maximum quantum yield of primary photochemistry | $F_v/F_m$ | |
| *The especific fluxes expressed per reaction centre* (*RC*) | | |
| Absorption per RC | ABS/RC | $=[(TR_0/RC)/(TR_0/ABS)]$ |
| Trapping at time zero, per RC | $TR_0/RC$ | = (MO/VJ) |
| Dissipation at time zero, per RC | $DI_0/RC$ | $=[(ABS/RC) - (TR_0/RC)]$ |
| Electron transport at time zero, per RC | $ET_0/RC$ | $=[(TR_0/RC)\ (ET_0/TR_0)]$ |
| *The yields* | | |
| Maximum quantum yield of primary photochemistry | $\varphi_{Po}$ $(TR_0/ABS)$ | $=F_v/F_m = 1 - (\ F_{50\mu s}/F_m)$ |
| Maximum quantum yield of non photochemical deexcitation | $\varphi_{Do}$ $(DI_0/ABS)$ | $=DIO/ABS = 1 - \varphi P_0 = (F_{50\mu s}/F_m)$ |
| Probability that a trapped exciton moves an electron further than $Q_A^-$ | $\Psi_o$ $(ET_0/TR_0)$ | = 1 − VJ |
| *Vitality indexes* | | |
| Performance Index | $PI_{ABS}$ | $=(RC/ABS)[\varphi_{P0}/(1 - \varphi_{P0})][\Psi_0/(1 - \Psi_0)$ |

$$I_s = \left(\frac{P_{nmax} + R_d}{\alpha}\right) \ln\left(\frac{P_{nmax} + R_d}{0.1P_{nmax}}\right) \quad (2)$$

The quantity of irradiance that exceeded the amount of light that plants were able to use in the photosyn thetic process (φ) was calculated by [26]: $\varphi = (I/P_n) * \alpha$.

## 2.6. Determination of Soil and Plant Nitrogen Content

Ten composite samples for each vegetation type, close to the individuals of *A. heterophylla* that were used in the physiological study, were collected at depths of 0 - 10 cm, 10 - 20 cm and 20 - 40 cm. The dried samples were ground in a Wiley mill and passed through 100-mesh screen. The total nitrogen was determined using a semimicro Kjeldahl digestion method [27].

The foliar nitrogen content was measured from 10 fully expanded terminal leaflets from 10 plants in each of the three vegetation types. The samples were oven dried at 65˚C and milled. Approximately 0.1 g was digested according to [28], and the total nitrogen was determined using the Kjeldahl method [29].

## 2.7 Experimental Design and Statistical Analysis

The sampling design was completely randomized with three treatments. Ten samples were collected from RF, CR and CP habitats for all analyses, with the exception of the gas exchange analyses, which were conducted on 5 random samples from each habitat. For this work, 10 individuals were selected from each vegetation type, taking into consideration the uniformity of height and distance that ensures the independence of individuals. Therefore, in the experiment, 15 and 30 trees were analyzed, depending upon the dependent variable. The data were subjected to an analysis of variance (ANOVA), and the means were compared by the Tukey test ($P \leq 0.05$). The gas exchange results were subjected to regression analysis, and the curves of $P_n$-I were adjusted using an exponential equation model.

# 3. Results and Discussion

The pigment concentrations ranged from 1.40 to 2.62 μmol·cm$^{-2}$, and there were no differences between CP, CR and RF habitats, most likely as a result of high variance in the leaf pigment concentrations (**Figure 2**).

The Chl *a*, *b* contents in CP, CR and RF were indicative of a high degradation rate of these pigments. In addition, the low values of $Chl_{tot}$/Car may indicate a high activity of xanthophyll cycle in these plants showing acclimation to high irradiance [5] [29]-[31].

Chloroplast pigments concentrations could be a indicative of different mechanisms for adaptation to different irradiance conditions [32], but the high intraspecific variation in the chloroplastid pigment concentration does not provide evidence of it being related to some trait for light capture and dissipation. In addition, low concentrations of chloroplast pigments in all vegetation types may be an indication of the low nutrient availability in CP, CR and FR sites. Plants living in environments with a low nitrogen availability and high irradiance, such as tropical forests, may be more susceptible to photo damage; however, the development of photoprotectors, such as chloroplast pigments, tends to be energetically expensive and slower [33]. Moreover, plants growing in nitrogen-poor environments with high levels of irradiance stored a significant amount of their nitrogen in non-photosynthetic proteins [34]. Low values of Chl *a*/*b* have been positively correlated with low rates of light-collecting complex Chl-PSII proteins (LHCII) [35]. Similar results was observed in [36], with Chl *a*/*b* varying from 3 to 3.3 in adult leaves. Thus, plants growing in oligotrophic systems have lower LHCII, indicating a combined response of high irradiance and low nutrient content of these areas [37].

However, the chlorophyll *a* fluorescence showed differences in the energy-processing strategies between trees found in the CP, CR and RF habitat types. The values of variable fluorescence and maximum fluorescence ($F_v/F_m$), which is a measure of the maximum quantum efficiency of PSII, in the three vegetation types was close to the optimal values of 0.83, indicating that the PSII of *A. heterophylla* leaves was not stressed [38], and it did not differ between habitat types (**Table 3**).

Others species studied by [39] showed similar results of $F_v/F_m$ at midday, varying from 0.79 to 0.80. This result suggested that Campina species are well-adapted to the poor nutrients conditions and excessive irradiance.

The results of $F_0$, $F_v$ and $F_m$ suggest optimal light use in *A. heterophylla* leaves, showing no signs of stress,

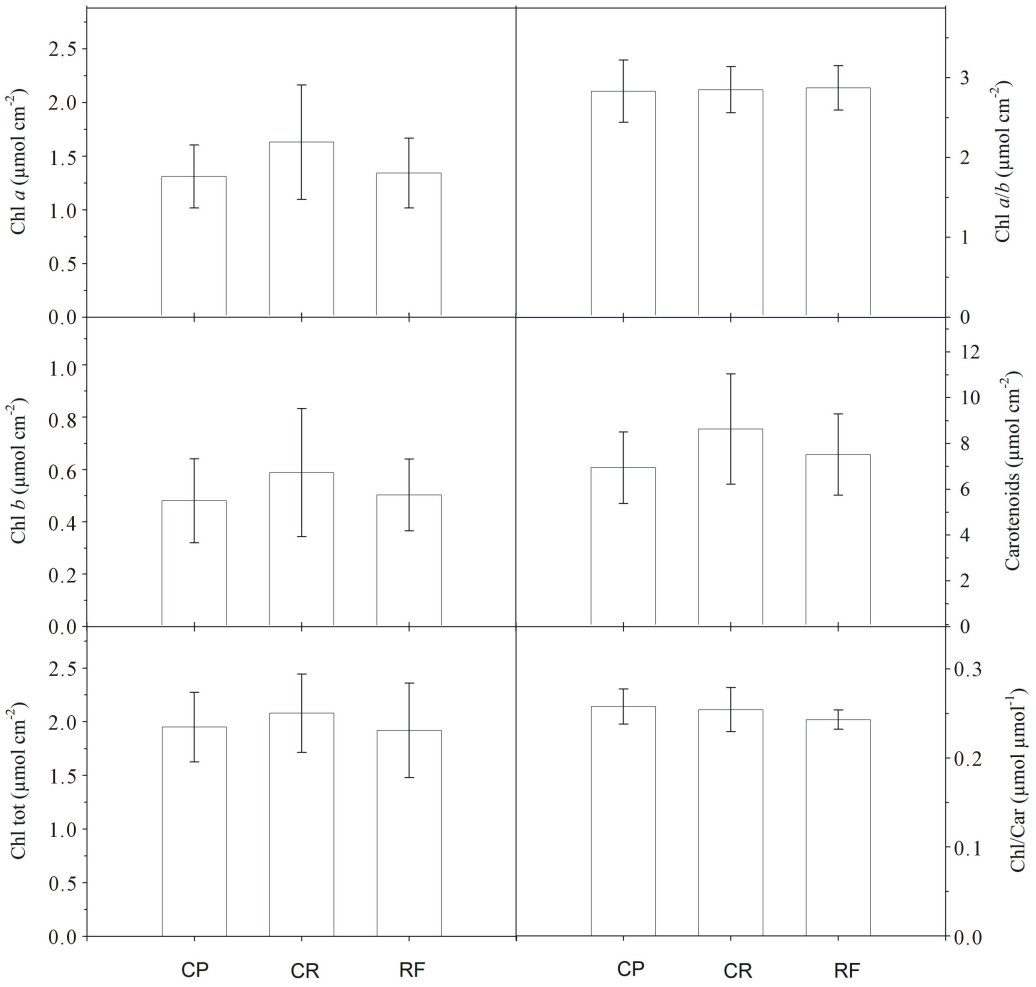


**Figure 2.** Chloroplast Pigment contents in Campina (CP), Campinarana (CR) and Rain Forest (RF). The values represent the average of 10 individuals (± SD).

**Table 3.** Leaf specific flow expressed by the reaction center (RC) of *Aldina heterophylla* in Campina (CP), Campinarana (CR) and Rain Forest (RF). The values represent the average of the readings of ten individuals (± SD).

| | CP | CR | RF |
|---|---|---|---|
| $F_0$ | 175.7 ± 7.3 a | 184.2 ± 12.7 a | 174.8 ± 5.7 a |
| $F_m$ | 1120.1 ± 48.2 a | 1131.1 ± 65.6 a | 1173.7 ± 99.9 a |
| $F_v$ | 944.4 ± 47.5 a | 946.9 ± 57.6 a | 998.9 ± 95.1 a |
| $F_v/F_m$ | 0.84 ± 0.01 ab | 0.83 ± 0.01 b | 0.85 ± 0.01 a |
| ABS/RC | 1.39 ± 0.10 ab | 1.49 ± 0.09 b | 1.34 ± 0.07 a |
| $TR_0/RC$ | 1.14 ± 0.08 a | 1.21 ± 0.06 b | 1.12 ± 0.06 a |
| $DI_0/RC$ | 0.25 ± 0.03 b | 0.28 ± 0.03 a | 0.23 ± 0.02 b |
| $ET_0/RC$ | 0.71 ± 0.06 ab | 0.70 ± 0.06 a | 0.69 ± 0.05 b |
| $TR_0/ABS$ | 0.83 ± 0.01 ab | 0.82 ± 0.01 b | 0.83 ± 0.01 a |
| $DI_0/ABS$ | 0.18 ± 0.01 ab | 0.19 ± 0.01 a | 0.17 ± 0.01 b |
| $ET_0/TR_0$ | 0.63 ± 0.04 a | 0.57 ± 0.05 a | 0.62 ± 0.02 a |
| $PI_{ABS}$ | 6.24 ± 1.4 a | 4.87 ± 1.04 b | 6.20 ± 0.67 a |

*Means followed by same letter do not defer to each other by Tukey test at 5% significance level. % related to differences between compared values that more differ between treatments.

even in conditions of high irradiance, low water availability and nutrition. However, some studies have shown that for some species, $F_v/F_m$ is not sensitive to abiotic stresses [40]-[44], highlighting the advantage of using other calculated parameters derived from the rapid growth of fluorescence and assessing the functionality of PSII compared with an assessment of only the $F_v/F_m$. In this study, it did not provide significant evidence of differentiation in the photochemical performance of PSII in *A. heterophylla* in response to environmental differences.

The specific fluxes expressed per reaction center (RC) show some differences between the CR vegetation type and the CP and RF vegetation types. The ABS/RC, which represents the absorption of light per RC and is correlated with the PSII antenna complex size, was 11% higher in CR than in CP and RF. The TR/RC, which is the trapping at time zero per RC, was higher in the CR than it was in the CP and the RF (**Table 3**), and the dissipation at time zero for RC was higher in the CR.

The performance index ($P_{IABS}$), combines three parameters relevant to photosynthetic activity: the density of reaction centers (expressed on an absorption basis), the quantum yield of primary photochemistry, and the ability to feed electrons into the electron chain between PSII and PSI. The $P_{IABS}$ showed similar results in individuals from RF and CP, with an average of 27% and 28% higher than the CR individuals, respectively (**Table 3**).

From this detailed investigation, using variables such as the performance index ($P_{IABS}$), we can confirm that individuals in the CR showed lower performance for the processing of high irradiance in the canopy. The $P_{IABS}$ results indicate that individuals of *A. heterophylla* in CR are most likely experiencing situations of photoinhibition more frequently than are those in CP or RF.

Although the individuals located in the CR showed a higher transfer of electrons per reaction center ($TR_0/RC$), these individuals had a reduced probability of transferring this excitation to move an electron beyond quinone A ($QA^-$) ($ET_0/TR_0$), indicating a low processing capacity of the amount of energy received at the reaction center. This result could be related to susceptibility to photoinhibition [41] [42] [45].

When compared with CP and CR, the RF microclimate showed the most favorable conditions for plant growth and biomass accumulation, with a lower light incidence inside the forest, soil that was rich in organic matter and clay and a high air relative humidity. Moreover, CP showed higher temperatures, poorer soil and a lower air relative humidity, and the $P_{IABS}$ result was similar to that found in RF. Therefore, the differences between CP and RF are obvious, but the main feature that differs between CR of CP is the forest density. This higher density of plants can increase the competition of water and nutrients in this area. Thus, with less water available, the photo damage could be more frequent, resulting in lower $P_{IABS}$. As shown by [46], during droughts, plants in open areas (sun) have more opportunity to rehydrate overnight than do plants in enclosed areas (shade), which have more competition. Therefore, we suggest that the low availability of water is most likely responsible for the decrease of $P_{IABS}$ in CR.

In addition, individuals in CR are potentially more subject to water stress as a result of a more open and lower canopy than RF, resulting in lower humidity. The soil in CR is also sandy, with a low water retention capacity, reflecting the dependence of the fluctuation of groundwater or rain to water availability [18].

The results of chlorophyll *a* fluorescence are similar to those of species adapted to extreme conditions of drought and high irradiance, such as desert species, but the magnitude is lower. This results suggests that water could be the factor that most influences the performance index of PSII for *A. heterophylla* in this vegetation gradient.

The differences found in $P_{IABS}$ showed no direct relationship with the results found for carbon assimilation ($P_{nmax}$), shown in **Figure 3** and **Table 5**, which showed no differences for these results.

The gas exchange results, such as the maximum photosynthesis ($P_{max}$), the transpiration with irradiance of 2000 $\mu mol \cdot m^{-2} \cdot s^{-1}$ ($E_{2000}$), the conductance to irradiance of 2000 $\mu mol \cdot m^{-2} \cdot s^{-1}$ ($gs_{2000}$), the apparent quantum yield ($\alpha$) irradiance and the irradiance of saturation ($I_s$), were similar for all vegetation types (**Table 4**). In contrast, the dark respiration rates were 50% lower in the RF than in CR, which was 34% lower than that found in CP (**Table 4**).

The soil nitrogen content was generally highest in RF and lowest in CP. At the depth of 0 - 10 cm, there were no differences between CR ($0.61 \pm 0.5$ $mg \cdot g^{-1}$) and RF ($0.83 \pm 0.5$ $mg \cdot g^{-1}$), but CP ($0.38 \pm 0.7$ $mg \cdot g^{-1}$) was significantly lower. The leaf nitrogen content was 19% lower in CP ($17.85 \pm 1.0$ $mg \cdot g^{-1}$) than it was in CR ($21.21 \pm 3.0$ $mg \cdot g^{-1}$) and RF ($21.27 \pm 1.0$ $mg \cdot g^{-1}$) (**Table 5**). CP species are characterized by high C/N, that reflect the low availability of N required for growth, was showed by [36].

The photosynthetic rates were not sensitive to nitrogen concentrations in the soil, which are lower in CP and

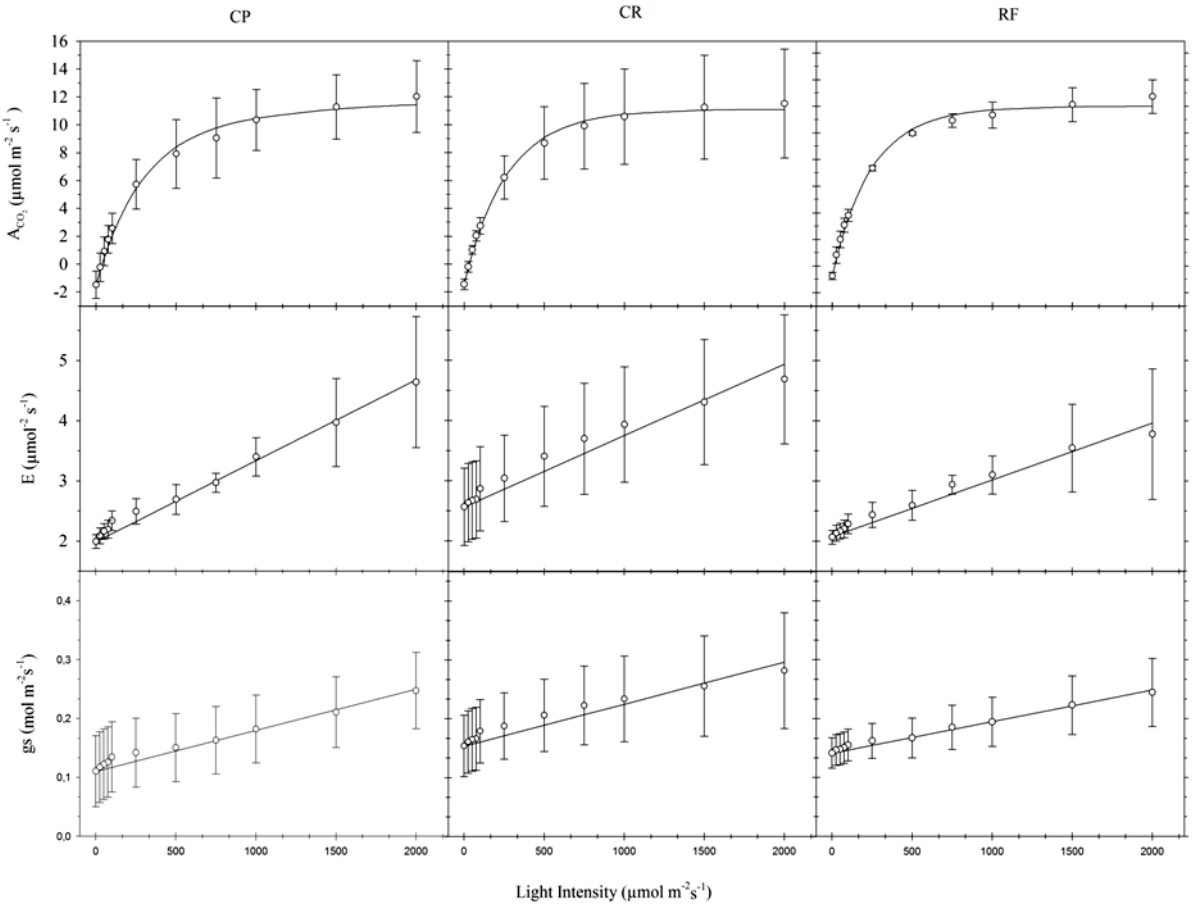


**Figure 3.** The relationship between light intensity (PPFD) and net photosynthesis, leaf transpiration and stomatal condutance in leaves of *A. heterophylla* in Campina (CP), Campinarana (CR) and Rain Forest (RF). The values represent the average of the readings of five individuals (± SD). The lines represent the exponential regression obtained for each treatment.

**Table 4.** Leaf photosynthetic parameters of *A. heterophylla* in Campina (CP), Campinarana (CR) and Rain Forest (RF). The values represent the average of the readings of five individuals (± SD).

| | CP | CR | RF |
|---|---|---|---|
| $P_{nmax}$ (µmol·m$^{-2}$·s$^{-1}$) | 11.3 ± 2.6 a | 12.6 ± 3.9 a | 12.0 ± 1.3 a |
| $Rd_{31}$ (µmol·m$^{-2}$·s$^{-1}$) | 2.3 ± 0.3 a | 1.5 ± 0.3 b | 0.8 ± 0.3 c |
| $\alpha$ (mol $CO_2$ mol$^{-1}$ PAR) | 0.048 ± 0.01 a | 0.049 ± 0.01 a | 0.054 ± 0.01 a |
| $I_c$ (µmol·m$^{-2}$·s$^{-1}$) | 41.6 ± 12.9 a | 29.6 ± 8.3 a | 14.7 ± 7.2 b |
| $I_s$ (µmol·m$^{-2}$·s$^{-1}$) | 666.2 ± 92 a | 617.9 ± 66 a | 574.0 ± 151 a |
| $\varphi_{2000}$ (µmol·m$^{-2}$·s$^{-1}$) | 8.7 ± 1.9 a | 8.0 ± 1.0 a | 9.2 ± 2.3 a |
| $E_{2000}$ (mmol·m$^{-2}$·s$^{-1}$) | 4.8 ± 0.5 a | 4.9 ± 1.1 a | 4.0 ± 1.1 a |
| $gs_{2000}$ (mmol·m$^{-2}$·s$^{-1}$) | 266 ± 55 a | 295 ± 98 a | 248 ± 57 a |

*Means followed by same letter do not differ by Tukey test at 5% significance.

increase as the vegetation changes to RF. Other tropical species also demonstrated sensitivity to the N concentration but were more efficient in their use of light [47].

Although CP and CR are potentially more vulnerable to water stress, the stomatal conductance did not change between the environments.

The photosynthesis response to increase irradiance in CR individuals had a high standard deviation (**Figure 3**), which can indicate the high intraspecific variability of response, *i.e.*, the high sensitivity of these plants to mi-

**Table 5.** Soil Nitrogen content and leaf Nitrogen Content ($mg \cdot g^{-1}$) in *A. heterophylla* in Campina (CP), Campinarara (CR) and Rain Forest (RF). The soil samples represent a range of depth. The leafes values represent the average of the readings of five individuals (± SD).

| | N content ($mg \cdot g^{-1}$) | | |
|---|---|---|---|
| Samples | CP | CR | RF |
| 0 - 10 cm | 0.38 ± 0.7 a | 0.61 ± 0.5 b | 0.83 ± 0.5 b |
| 10 - 20 cm | 0.35 ± 0.6 a | 0.45 ± 0.8 ab | 0.67 ± 0.5 b |
| 20 - 40 cm | 0.26 ± 0.1 a | 0.11 ± 0.1 b | 0.45 ± 0.2 c |
| Leaf | 17.85 ± 1.0 a | 21.21 ± 3.0 b | 21.27 ± 1.0 b |

*Means followed by same letter do not defer to each other by Tukey test at 5% significance level.

croclimatic variations. Therefore, CR is a transitional environment between CP and RF; it is observed that in these areas, the availability of resources is less stable, and there are more frequent microclimatic variations. These less constant environments induce plants to also exhibit less stable metabolic responses, as demonstrated by [4]. This sensitivity to environmental changes can lead to a decrease in the occurrence of this species in the CP, so that predictions of climate change are related to intensity and frequency.

Despite the observed variations of net photosynthesis as described above, the results probably do not varied due same set conditions of leaf chamber for measuring to light, temperature, humidity and $CO_2$ concentration, and the measurements were performed on leaf in full sun. Therefore, even though *A. heterophylla* present differences in net photosynthesis in response to the environment, the ability of instantaneous acclimation observed did not allow the differences between environments.

Among the variables that were significantly different, respiration in dark-adapted leaves at 31˚C ($R_{d31}$) showed the greatest variation between environments. The net photosynthesis did not differ in individuals along the gradient; however, $R_{d31}$ has the highest rates in CP and lower rates in RF. Despite some studies arguing about the light enhancement of dark respiration measured during the day, [48] show that in *Ricinus communis*, this variation is only 3% higher, although the source of carbon changes significantly, with 22% of carbon loss from malate on light-enhanced dark respiration. This 3% light enhancement is not sufficient to match the data of the CP, CR and RF.

The influence of irradiance and temperature on $R_d$ has been observed in other tropical forest species and in individuals of rosewood (*Aniba rosaeodora*), which showed increased rates of $R_d$ treatments in response to increased irradiance [2]. The increase of irradiance and temperature are indicated as the main factors to increase the $R_d$ [49]-[53]. Other work that associated $R_d$ with light and temperature availability was conducted in contrasting vegetation types in Africa between tropical montane rain forest species and an open environmental vegetation species, similar to CP, that showed lower $R_d$ in rain forest than open vegetation types [3]. Furthermore, these authors indicate that these observations could be related to differences in the soil nutrition concentrations. However, in the vegetation gradient studied, despite the differences observed in the soil and leaf nitrogen concentration in RF and CP, we can say that these areas have a nitrogen-poor soil. The lower leaf nitrogen concentrations are not related to the other metabolic aspects, for example, the net photosynthesis. These results indicate that, for a species such as *A. heterophylla* that colonizes environments with different characteristics, high physiological plasticity is expected.

## 4. Conclusions

Individuals of *A. heterophylla* growing in Campinarana are more vulnerable to microclimatic variations, such as water availability, which can be observed mainly by the performance index ($P_{IABS}$) obtained by the fluorescence of chlorophyll *a*, showing a lower processing power of the incident energy.

The differences in the total nitrogen concentration in the soil and leaf and the poor, sandy soil found in CP are not enough to promote changes in the leaf gas exchange of *A. heterophylla*, indicating a high physiological plasticity that permitted the colonization of this species throughout the whole vegetation gradient.

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