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Vegetation inside: a comparative analysis of leaf pigments and chlorophyll fluorescence in variegated plants

Vegetação *inside*: uma análise comparativa de pigmentos foliares e fluorescência da clorofila em plantas variegadas

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The characteristic plants of understory environments show varied colors in their leaves, called variegations. These tend to regulate, through their concentrations of leaf pigments, their photosynthetic processes. The objective of this research was to carry out a comparative analysis of leaf pigment concentrations and chlorophyll a fluorescence parameters in four species of variegated plants, *Fittonia albivenis*, *Alternanthera ficoidea*, *Tradescantia spathacea* and *Hypoestes phyllostachya*, as well as the control species, *Jasminum mesnyi*, and *Tradescantia pallida*. To determine which are most suitable for environments with high and low light intensities. The analysis of leaf pigments were carried out by spectrophotometry, and to analyze the fluorescence emission of chlorophyll-a PAM-2500 fluorometer was used, whose parameters were calculated from the JIP test. The chlorophyll a levels ranged from 27.67 to 110.88 (μg mL^{-1} gMF^{-1}), those of chlorophyll b ranged from 10.29 to 40.64 (μg mL^{-1} gMF^{-1}). Carotenoid titers ranged from 2.6275 to 26.1925 (μg mL^{-1} gMF^{-1}), and anthocyanins ranged from 0 to 2.25679 (μg mL^{-1} gMF^{-1}), with the highest titers occurring for the control species *J. mesnyi* and the variegated *F. albivenis*. The fluorescence parameters differed statistically among the variegated plant species concerning the controls. We conclude that the species *T. spathacea* presents better adaptation to environments with high levels of light radiation. In environments with more significant variation in light radiation, the species variegated *H. phyllostachya* and *A. ficoidea* will develop better. Finally, the species variegated *F. albivenis* must be maintained in environments with low levels of light radiation.
Keywords: photosynthesis, OJIP curve, energy dissipation.

Plantas características de ambientes de sub-bosque apresentam colorações variadas em suas folhas denominadas de variegações. Estas tendem a regular, por meio de suas concentrações de pigmentos foliares, seus processos fotossintéticos. O objetivo desta pesquisa foi comparar as concentrações de pigmentos foliares e de parâmetros da fluorescência da clorofila *a* em quatro espécies de plantas variegadas, *Fittonia albivenis*, *Alternanthera ficoidea*, *Tradescantia spathacea* e *Hypoestes phyllostachya*, bem como das espécies controles, *Jasminum mesnyi* e *Tradescantia pallida*, para determinar quais são mais propicias aos ambientes residenciais com altas e baixas intensidades luminosas. A análise dos pigmentos foliares foi realizada por espectrofotometria e a de emissão de fluorescência da clorofila *a* através do fluorômetro PAM-2500, cujos parâmetros foram calculados a partir do teste-JIP. Os teores de clorofila *a* variaram de 27,67 a 110,88 (μg mL^{-1} gMF^{-1}), os de clorofila *b* variaram de 10,29 a 40,64 (μg mL^{-1} gMF^{-1}). Os títulos de carotenoides variaram de 2,6275 a 26,1925 (μg mL^{-1} gMF^{-1}) e os de antocianinas variaram de 0,00 a 2,26 (μg mL^{-1} gMF^{-1}), sendo que os maiores títulos ocorreram na espécie controle *J. mesnyi* e na variegada *F. albivenis*. Os parâmetros da fluorescência diferiram estatisticamente entre as plantas variegadas em relação aos controles. Concluímos que a espécie *T. spathacea* apresenta melhor adaptação a ambientes com altos índices de radiação luminosa. Em ambientes com maior variação desta as espécies *H. phyllostachya* e *A. ficoidea* se desenvolverão melhor. Por fim, a espécie *F. albivenis* deve ser mantida em ambientes com baixos índices de radiação luminosa.
Palavras-chave: fotossíntese, curva OJIP, dissipação energética.

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1. INTRODUCTION

Floriculture, which can be defined as the set of commercial activities related to the market of plant species with ornamental purposes, is very promising in Brazilian agriculture. In 2000, the search for ornamental plants led to an annual per capita consumption of R$ 20.00, moving around three billion reais per year [1]. In 2019, average consumption of BRL 31.50 per capita was recorded, representing an increase of 63.4%, with an estimated movement of around eight billion reais by the sector of the Brazilian landscaping market [2]. According to the Brazilian Institute of Floriculture (Ibraflor), the ornamental plant sector grew around 10% in 2020, and new purchase formats, such as delivery, promoting the market to 5% in 2021 [3].

Among the most representative and widespread species of ornamental plants are the variegated plants, characterized by the development, throughout their evolutionary processes, of several multicolored patterns on their leaf surfaces [4]. Several internet advertisements indicate them as suitable plants for ornamentation in the interior of homes, where the availability of sunlight is sometimes compromised. However, as the multiplicities of colors of these plants occur as a function of the dynamic exposure to light radiation to which they are submitted in understory environments [5-7], each species reacts physiologically differently when placed in environments with different availability of light.

Plant variegations aim to regulate light capture, optimizing their photosynthetic processes [4, 8]. Different concentrations of photosynthetic pigments in palisade mesophylls lead to colors in the plant variegation [9]. These differences directly affect leaf tissue density and, consequently, light penetration through it [6]. Among the best-known leaf pigments, chlorophylls (chlorophyll a and chlorophyll b), carotenoids, and anthocyanins stand out.

Chlorophyll a (Chla) is the main pigment responsible for photosynthetic processes. Its light energy absorption peak from 400 to 450 nm is in the blue region and from 630 to 700 nm in the red region [4]. Chlorophyll b (Chlb) has its absorption peak from 450 to 480 nm and Chla, leading to the green coloration on the leaves [10]. In addition to acting as accessory pigments in photosynthesis, carotenoids absorb light at wavelengths from 470 to 570 nm. Carotenoids reflect their yellow and orange color and play a crucial role in the photooxidative protection of the plant [8]. Anthocyanins, reddish, blue, and violet pigments, in turn, are mainly present on the adaxial surface of the leaves in order to protect the plant against low temperatures, loss of water, and excess ultraviolet radiation [4]. The other pigments help absorb light at other wavelengths and transfer radiant energy to the reaction centers (RC) of photosystems I (FSI) and II (FSII), thus being called accessory pigments [8].

Linked to the analysis of leaf pigments, one of the best and most promising scientific techniques to assess the photosynthetic processes of plants is based on the fluorescence of chlorophyll a (Fchla). It is characterized by a fraction of energy dissipated by the plant, not in heat or resonance, but in the form of light [11]. The FChla yield reveals information about the levels of energetic excitations in photosynthetic systems [11]. The FChla yield providing subsidies to estimate the inhibition or damage in the electron transfer processes of photosystem II (FSII). Thus, this technique can reveal which plants are more or less susceptible to environments with greater or lesser incidences of light.

The chlorophyll a fluorescence emission profile and its various parameters can be calculated and analyzed from the JIP test. The JIP test, proposed by Strasser and Strasser (1995) [12], provides basic measurements of FChla transients in phenomenological and biophysical expressions. They quantify the efficiency of the photosynthetic process of plants in the most varied environments. Differences in chlorophyll a fluorescence transients can be assessed in plants in vivo and in situ. These fluorescence transients can be used as markers to indicate changes in processes related to the electron transport chain (ETC) and its acceptors [11, 13].

Given the above, this study aimed to evaluate the contents of chlorophyll pigments (Chla and Chlb), carotenoids and anthocyanins, and the chlorophyll a fluorescence, in four species of variegated plants, *Tradescantia spathacea* Sw., *Fittonia albivenis* (Lindl. ex Veitch.) Brummitt, *Hypoestes phyllostachya* Baker and *Alternanthera ficoidea* (L.) P.Beauv. This study also allowed us to identify which variegated species are more suitable for environments with higher levels of light radiation, which are more suitable for places with lower solar radiation levels. Our hypothesis was based on the following proposition: the variegated plant species with the highest levels of leaf pigments and with the highest energy dissipation by fluorescence are the most suitable to be allocated to environments with lower light radiation levels. Furthermore, it was possible to characterize the photosynthetic performance of each of the species evaluated.

2. MATERIAL AND METHODS

2.1 Acquisition and maintenance of evaluated species

The plant species analyzed were obtained from local commerce and kept in their respective vases in the Vegetal Physiology laboratory of the Universidade Estadual do Centro-Oeste (Unicentro), Guarapuava, Paraná, Brazil, under natural environmental conditions (Figure 1). For the experiment, two species of non-variegated ornamental plants were selected to serve as controls for the proposed analyses: 1) *Jasminum mesnyi* Hance, completely green, 2) *Tradescantia pallida* (Rose), completely purple. The species were identified through consultations with the Virtual Herbarium of Flora and Fungi (INCT). The vouchers for their exsiccates were deposited in the Unicentro herbarium (ARAUCA) under the following codes: 1087 (*Jasminum mesnyi* Hance); 1088 (*Fittonia albivenis* (Lindl. ex Veitch) Brummitt); 1085 (*Tradescantia spathacea* Sw.); 1152 (*Tradescantia pallida* (Rose) D.R. Hunt); 1086 (*Hypoestes phyllostachya* Baker.); 1089 (*Alternanthera ficoidea* (L.) P.Beauv.).

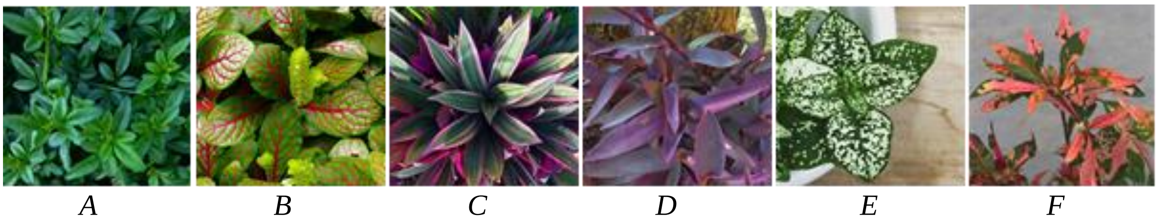

Figure 1: Plant species analyzed (A) Jasminum mesnyi; (B) Fittonia albivenis; (C) Tradescantia spathacea; (D) Tradescantia pallid; (E) Hypoestes phyllostachya; (F) Alternanthera ficoidea.

2.2 Extraction and quantification of photosynthetic pigments

In the analyzed plant species, completely expanded leaves (N=5) were selected. These were sectioned with 1.6 cm in length and 0.7 cm in width, avoiding the main vains. The samples were homogenized in a mortar and pistil with 2 mL of acetone/Tris HCl buffer solution (80:20 V/V, pH 7.8). After this step, the total volume of extracts was completed with the same solution up to 8 mL. The extracts were centrifuged at 4000 rpm for 15 minutes and, after 24 hours, the supernatant was transferred to quartz cuvettes and analyzed by spectrophotometry at wavelengths 537 nm, 647 nm and 663 nm [14]. The contents of chlorophyll a (Chla), chlorophyll b (Chlb), carotenoids and anthocyanins were calculated, and their contents were expressed in μgmL^{-1} per gMF^{-1} (MF = Fresh Mass).

2.3 Fluorescence analysis of chlorophyll a

The FChla analyses were performed on the same plants in fully expanded leaves (N=5), corresponding to pigment analysis. Fchla analyzes were performed three times on the same day, at 7:00 am, 12:00 pm, and 5:00 pm. Measurements were performed using a Fluorometer probe model PAM-2500 (Heinz Wlaz, GmB) after dark adaptation of the leaves for 15 minutes with a leaf clip. Subsequently, the JIP test was performed [12] to obtain the fast kinetic curve's induction and calculate its parameters. The chlorophyll a fluorescence parameters were calculated from the induction of the fluorescence curve and were based on estimates of energy fluxes per reaction center (RC) or per area or cross-section (CS) of the photosynthetic apparatus present in the thylakoids of the sampled leaves (Table 1).

Table 1: Chlorophyll a fluorescence parameters were obtained from the curve induction by the JIP test. Adapted from Goltsev et al. (2016) [11].

Parameters	**Formula**	**Definition**
$F_0 = F_{50\mu s}$ (O)		Fluorescence at 50 µs.
$F_J = F_{2ms}$ (J)		Fluorescence at 2 ms.
$F_I = F_{30ms}$ (I)		Fluorescence at 30 ms.
F_M (P)		Maximum fluorescence
$F_{300\mu s}$		Fluorescence at 300 µs.
Calculated parameters		
F_V	$= (F_M - F_{50\mu s})$	Variable fluorescence
V_I	$= (F_{30ms} - F_{50\mu s}) / (F_m - F_{50\mu s})$	Relative Variable fluorescence at 30 ms.
V_J	$= (F_{2ms} - F_{50\mu s}) / (Fm - {}_{F50\mu s})$	Relative Variable fluorescence at 2 ms.
M_0	$= [4. (F_{300\mu s} - F_{50\mu s}) / (F_m - F_{50\mu s})]$	Net rate of closed FSII.
Specific parameters (RC)		
ABS/RC	$= [(TR_0/RC) / (TR_0/ABS)]$	Effective size of antenna complex per active RC.
ET_0/RC	$= [(TR_0/RC) (ET_0/ TR_0)]$	Electron transport rate per RC.
DI_0/RC	$= [(ABS/RC) - (TR_0/RC)]$	Active RC dissipation.
TR_0/RC	$= (M_0/VJ)$	Maximum capture by RC.
Quantum Yields		
δ_{Ro} (RE_0/ET_0)	$= (F_M - F_I) / (F_M - F_J)$	Probability of the electron being transported from the PQ to the photosystem I acceptor.
Ψ_{Eo} (ET_0/TR_0)	$= 1 - V_J$	Probability of the excitation energy captured by the RC II to move an electron after QA.
φ_{Po} (TR_0/ABS)	$= F_v/F_m = 1 - (F_{50\mu s} / F_M)$	Maximum quantum efficiency of photosystem II.
φ_{Do} (DI_0/ABS)	$= DI_0/ABS = 1 - \varphi Po = (F_{50\mu s}/F_M)$	Maximum quantum efficiency of non-photochemical excitation.
φ_{Eo} (ET_0/ABS)	$= \varphi Po.\ \Psi_0 = [1 - (F_{50\mu s}/F_M)] (1 - V_J)$	Maximum quantum efficiency of electron transport.
φ_{Ro} (RE_0/ABS)	$= [1 - (F_0/F_M)] (1 - V_I) = 1 - F_I/F_M$	Maximum quantum efficiency of the electrons that reach the receiver.
Phenomenological Flows (CS)		
ABS/CS	Proportional to F0	Number of photons absorbed by CS.
TR_0/CS	$= (ABS/CS) (TR_0/ABS)$	Energy captured by CS.
DI_0/CS	$= (ABS/CS) - (TR_0/CS)$	Energy dissipated by CS.
ET_0/CS	$= (ET_0/RC) (RC/CS)$	Electron transport by CS.
Vitality indices		
PI_{ABS}	$(RC/ABS) [\varphi Po/(1 - \varphi Po)][\Psi 0/(1 - \Psi_0)$	Performance index (potential) for the conservation of photon energy absorbed by the FSII for the reduction of electron acceptors.
PI_{CS}	$(RC.\varphi Po.\Psi_0) /(CS.1 - \varphi Po.1 - \Psi_0)$	Performance index, an indicator of the functional activity of the FSII expressed per unit of cross-section of the illuminated area.
$PI_{totalABS}$	$PI_{ABS}.\ (\delta Ro/1 - \delta Ro)$	Total performance index, indicating the integral functional activity of PSII, PSI and intersystem electron transport chain.
$PI_{totalCS}$	$PI_{CS}.\ (\delta Ro/1 - \delta Ro)$	Total performance index, indicating the integral functional activity of PSII, PSI and intersystem electron transport chain expressed per unit cross-section of the illuminated area

2.4 Statistical analysis

The JIP test parameters and photosynthetic pigment concentrations were analyzed by Generalized Linear Models (GLM) with gamma distribution. Significant differences between pigment contents were analyzed using pairwise comparison and Bonferroni significance, while the JIP test parameters were evaluated using the effect model test. The analyzes were carried out in Environment R [15]. The Principal Component Analysis (PCA) was performed in the software ORIGIN 9.6.

3. RESULTS

Chla was the leaf pigment that presented the highest concentrations in leaf tissues of all analyzed species, followed by Chlb, carotenoids and then anthocyanins (Figure 2). Regarding Chla, the variegated species *F. albivenis* and *H. phyllostachya* did not differ statistically from *J. mesnyi*, the green control. On the other hand, they differed significantly from the entirely purple control, *T. pallida*, which did not differ from the variegated species *A. ficoidea* and *T. spathacea*. Concerning Chlb concentrations, *F. albivenis* was the only species that differed statistically from the purple control, T. pallida. The variegated species *F. albivenis* did not present levels of anthocyanins statistically different from the green control, *J. mesnyi*. All analyzed species presented significantly different levels of this pigment compared to the purple control, *T. pallida*. In addition, the variegated plants showed levels of carotenoids statistically different from the two control species ($p < 0.05$) (Figure 2).

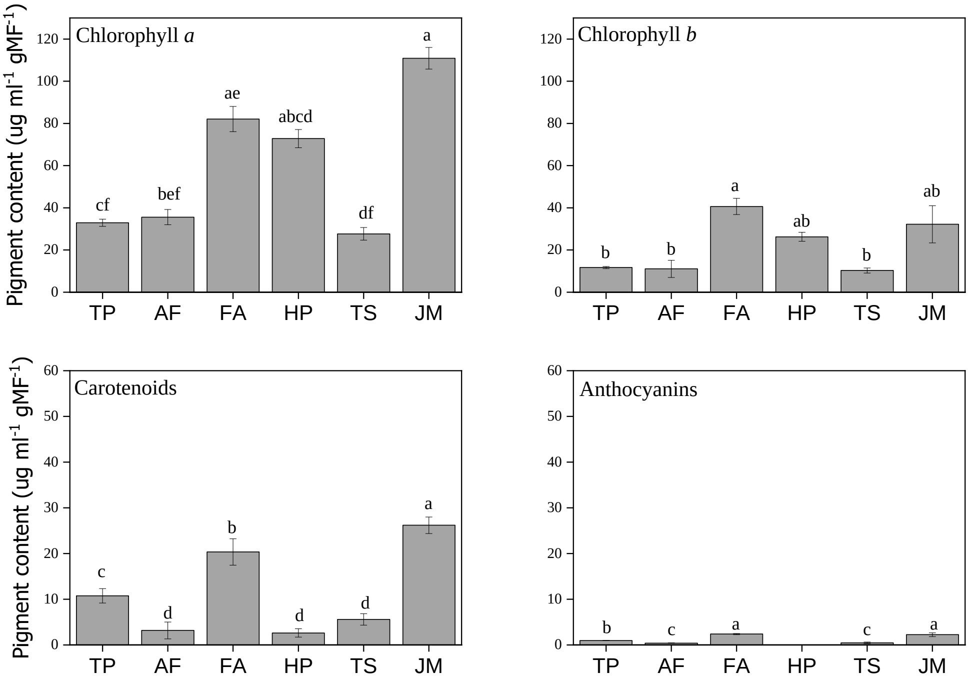

Figure 2: Contents of chlorophyll a, chlorophyll b, carotenoids, and anthocyanins in leaves of the analyzed species. TP – Tradescantia pallida*; AF –* Alternanthera ficoidea*; FA –* Fittonia albivenis*; HP –* Hypoestes phyllostachya*; TS –* Tradescantia spathacea*; JM –* Jasminum mesnyi*. Means followed by the same lowercase letter do not show significant differences (p < 0.05) between species for each type of pigment analyzed by MLG.*

Principal component analysis (PCA) was performed on the same data set, encompassing all response variables simultaneously (Figure 3). Principal components 1 and 2 explain 96.89% of the data variance. The main component 1 highlighted the plant species that presented the highest (*J. mesnyi* and *F. albivenis*) and lowest values (*T. pallida, H. phyllostachya, A. ficoidea, T.*

spathacea) for the leaf pigment contents evaluated in this work (Figure 3). The main results of PCA showed that *J. mesnyi*, completely green, had the highest contents of Chla and carotenoids (2nd quadrant). In contrast, the variegated species, *F. albivenis*, had the highest contents of Chlb and anthocyanins (3rd quadrant). The purple control, *T. pallida*, and the variegated *T. spathacea* were grouped together as they presented the lowest contents of Chla and carotenoids (4th quadrant). Likewise, the variegated *H. phyllostachya* and *A. ficoidea* were grouped as they presented the lowest levels for Chlb and anthocyanins (1st quadrant) (Figure 3).

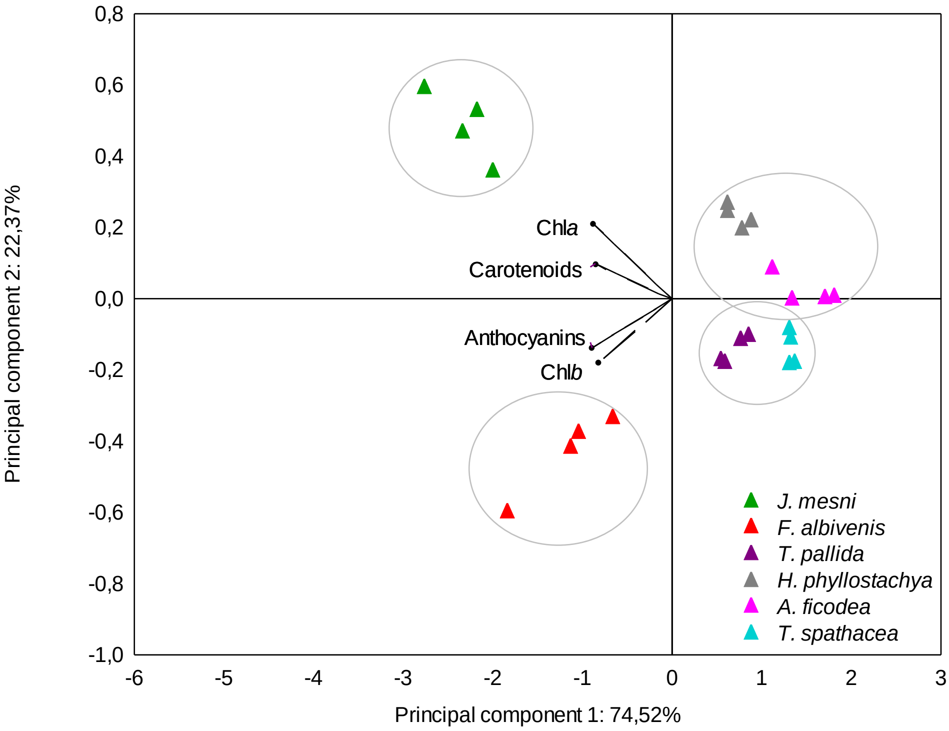

Figure 3: Projection of variables and six plant species in the factor plane (PC1 and PC2).

All plant species analyzed presented distinct polyphasic chlorophyll a fluorescence emission curves (Figure 4A). Therefore, there were statistically significant differences ($p < 0.05$) between the 28 chlorophyll a fluorescence parameters estimated by the JIP test in the evaluated plants (Table 2). The green control, *J. mesnyi*, and the variegated species, *F. albivenis*, presented the highest values for the basic parameters of chlorophyll a fluorescence, namely: F_{50us}, F_{300us}, F_{2ms}, F_{30ms}, not significantly different from each other, but differ from the other variegated species and the purple control (Figure 4B). The highest value for F_M was also observed for the green control (Figure 4B), without statistical differences for *F. albivenis* and *T. spathacea* (Table 2).

From these basic parameters, other parameters about FChla were calculated. The green control, *J. mesnyi*, presented the highest values of F_V and V_I. On the other hand, it was *F. albivenis* who differed statistically (Table 2) from all other species, showing the lowest values for these parameters (Figure 4B). The variegated plant species also differed from each other and the controls regarding the VJ parameter. The purple control, *T. pallida*, and the variegated one, *T. spathacea*, showed the lowest values for Mo (Figure 4B), differing statistically from the other species, which did not differ from each other (Table 2). *F. albivenis* showed the highest values for the specific parameters (ABS/RC, ET_0/RC, DI_0/RC and TR_0/RC) and the activities in the reaction centers (RC) of photosystem II (Figure 4B), differing significantly of all other plant species analyzed (Table 2).

The species *F. albivenis* had the highest φRo and φDo (Figure 4B), differing statistically from the other species (Table 2). The two plants of the *Tradescantia* genus had the highest φPo, φEo, and Ψo, demonstrating significant differences from the other species (Table 2). In *T. spathacea* the highest index for δRo was recorded (Figure 4B). The two control species did not differ statistically from each other in this parameter (Table 2). The parameters referring to the phenomenological fluxes per leaf cross-sectional area (CS) were also evaluated in this work. The values observed for the green control species, *J. mesnyi*, were the highest for ABS/CS and TR_0/CS (Figure 4B), differing significantly from the other species analyzed (Table 2).

Table 2: Statistical significance of the OJIP curve parameters in the control and variegated plants analyzed. Different letters represent significant differences. AF - Alternanthera ficoidea*; HP -* Hypoestes phyllostachya*; TS -* Tradescantia spathacea*; JM -* Jasminum mesnyi*; FA -* Fittonia albivenis*; TP -* Tradescantia pallida.

Parameter	**p-value**	**AF**	**HP**	**TS**	**JM**	**FA**	**TP**
$F_{50\mu s}$	0.000	0.35953e	0.42507c	0.38823d	0.55383a	1.0000a	0.37171e
F_{2ms}	0.000	0.67683b	0.69975a	0.80766c	0.99162a	1.0000a	0.72286c
F_{30ms}	0.000	0.68224d	0.68553c	0.85013c	1.0000a	0.86383a	0.79899d
F_M	0.000	0.66895c	0.66917c	0.87325a	1.0000a	0.87007a	0.80038b
$F_{300\mu s}$	0.000	0.59883b	0.67345a	0.68062c	0.91871a	1.0000a	0.61469c
F_V	0.000	0.66895d	0.62553d	0.94970b	1.0000b	0.45485a	0.85773c
V_I	0.000	1.00000c	0.98651b	0.93278d	0.95699c	0.80361a	0.96361d
V_J	0.003	1.00000a	0.99706a	0.88908c	0.94317b	0.88387c	0.85484d
M_0	0.000	0.86690a	0.99991a	0.71461b	0.88256a	1.0000a	0.69242b
ABS/RC	0.000	0.39349d	0.49467b	0.33747e	0.43140c	1.0000a	0.34613e
ET_0/RC	0.000	0.62878d	0.73533b	0.70227e	0.76431c	1.0000a	0.74369e
DI_0/RC	0.000	0.1863d	0.27606c	0.12992b	0.2079b	1.0000a	0.14056c
TR_0/RC	0.000	0.76467e	0.88633c	0.70932d	0.83179b	1.0000a	0.71444c
δRo	0.000	0.40431c	0.45818b	0.57111a	0.52404c	1.0000b	0.44292c
Ψo	0.001	0.79029e	0.79453d	0.95053a	0.87238c	0.65806b	1.0000c
φPo	0.000	0.92597d	0.85650c	1.0000a	0.91956b	0.47832a	0.98296c
φDo	0.000	0.47280d	0.55461d	0.38562b	0.48035a	1.0000e	0.40568c
φEo	0.000	0.74350c	0.68904d	0.96248a	0.81242b	0.46304e	1.0000a
φRo	0.000	0.54637d	0.57944d	1.0000a	0.78204b	0.84584e	0.80038f
ABS/CS	0.000	0.66287c	0.66917d	0.87325b	1.0000a	0.37007e	0.80038b
TR_0/CS	0.000	0.66895d	0.62553c	0.94970d	1.0000a	0.45485b	0.85773d
DI_0/CS	0.000	0.35953c	0.42507b	0.38823c	0.55383c	1.0000a	0.37171c
ET_0/CS	0.001	0.59072b	0.55279c	1.0000a	0.96889a	0.48062d	0.96249a
RC/CS	0.001	0.65580d	0.53113e	1.0000b	0.91427a	0.34094f	0.89820c
PI_{ABS}	0.000	0.57194c	0.49604d	0.96191a	0.68136b	0.06051e	1.0000a
PI_{CS}	0.000	0.47980c	0.44187d	1.0000a	0.57540b	0.06341e	0.97788a
$PI_{TotalABS}$	0.000	0.39228d	0.33204d	0.9000a	0.73787c	0.19202d	0.9000b
$PI_{TotalCS}$	0.002	0.37287d	0.35499d	0.8000a	0.73090c	0.49647d	0.8500b

Fittonia albivenis presented the highest value for DI_0/CS, also differing statistically from the other species. For this parameter, both control species presented intermediate values (Figure 4B). *Tradescantia spathacea, T. pallida* and *J. mesnyi* did not show significant differences regarding the ET_0/CS parameter (Table 2).

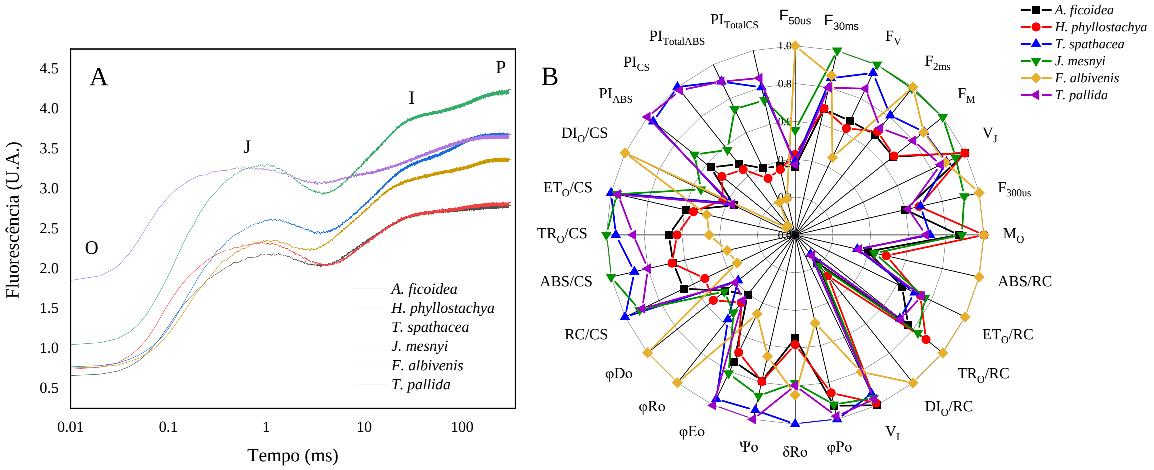

Figure 4: (A) Kinetic curve of chlorophyll a fluorescence emission of the plant species analyzed: O (F_{50us}); J (F_{2ms}); I (F_{30ms}); P (F_M). (B) Radar chart with the parameters of the JIP test about the emission of FChla by the analyzed plant species. T. pallida - Tradescantia pallida*; A. ficoidea* – Alternanthera ficoidea*; F. albivenis* – Fittonia albivenis*; H. phyllostachya* – Hypoestes phyllostachya*; T. spathacea* – Tradescantia spathacea*; J. mesnyi* – Jasminum mesnyi.

4. DISCUSSION

The plant species analyzed in our study, variegated and controls, presented different concentrations of leaf pigments and distinct patterns of chlorophyll a fluorescence emission. The light irradiation changes in understory environments result in different excitations of photosystems in plants [16]. Therefore, plants, especially variegated ones, originating from these places. These differences through acclimatization responses result in different concentrations of photosynthetic pigments.

The analyzed species had higher Chla contents than any other photosynthetic pigment (Figure 2). This fact is widely discussed in several studies [5, 17-21] since Chla operates as the central molecule responsible for capturing light energy. This way, it can boost the electron transport reactions in the first stage of photosynthesis [19]. In this sense, we can conclude that variegated plants and even completely purple plants, such as the control *T. pallida*, have the highest levels of Chla, compared to other pigments, as this is the constituent molecule of CR, therefore, the protagonist in capturing light energy.

The *J. mesnyi, F. albivenis* and *H. phyllostachya* species presented the highest concentrations of photosynthetic pigments (Figure 2). These same species showed a predominance of green leaf areas. Studies corroborate the results observed in this study by explaining that the greenest areas of variegated plants have the highest contents of Chla and Chlb compared to yellowish, pink, or reddish areas [4, 18, 22]. However, the predominance of greenish areas on the leaf surfaces of the analyzed plants was not directly related to the contents of carotenoids and anthocyanins. This fact is because the variability in leaf color, the variegations, is due to the different contents of photosynthetic pigments in plant tissues, especially carotenoids and anthocyanins, responsible for colors differ from green [6, 22].

In the present study, it was expected that the control species *T. pallida*, being completely purple, had the highest levels of anthocyanins. However, this species expressed lower values of anthocyanin than even our completely green control, *J. mesnyi*. This fact may have occurred because *J. mesnyi* had the highest concentrations of Chla that tend to mask the color of anthocyanins, even though they are abundant in plant tissue [6]. Thus, a purple plant may have lower values of colored pigments when compared to a completely green plant.

The relationship between the levels of Chla/carotenoids and Chlb/anthocyanins observed in this study served as a marker to distinguish the variegated species from each other and the control (Figure 3). The protein association between the constituents of the light-harvesting complex and Chla molecules with the carotenoids is confirmed by the species *J. mesnyi*, *H. phyllostachya* and

A. ficoidea quadrants 1 and 2 of the PCA (Figure 3). These shreds of evidence reinforce the inherent biochemical dependence of photosynthesis with Chla molecules and carotenoids [8, 23, 24]. The association of Chla and carotenoids, in this way, increases the absorption of wavelengths in which light can promote photosynthesis.

The species with the highest content of photosynthetic pigments presented the highest values for the basic parameters of FChla ($F_{50\mu s}$ = O, $F_{300\mu s}$, F_{2ms} = J, F_{30ms} = I, F_M = P). This fact is supported by studies that reveal a positive correlation between the concentration of pigments in the membranes of thylakoids, especially Chla, and the net rate of photosynthesis [6, 18, 24]. Another vital relationship we noticed was by the FChla analysis because of the more significant the concentration of pigments in the FSII, the greater the number of plastoquinone molecules QA and QB (F_{2ms} (J), F_{30ms} (I)). Thus, the FSII reaction centers end up being closed due to the reduction of the entire electron transport chain (the result of their saturation in the FSI acceptor), increasing the emission of $F_{50\mu s}$ (O) to F_M [13, 25, 26]. Therefore, a higher potential activity rate of FSII was observed in the control species *J. mesnyi*, completely green, compared to the variegated plant species and the purple control, *T. pallida*.

Variegated plants differed statistically from each other, and controls for specific parameters of activities in the RC: ABS/RC, ET_0/RC, DI_0/RC, and TR_0/RC serving as markers for distinction among such species (Table 2). The differences in the values of these parameters are essential to keep the PQ pool low. This plasticity reflects the photosynthetic acclimatization process of plants to environmental conditions [16, 26, 27]. It is demonstrated that the variations in TR_0/CS are mainly due to the decrease in the density of the RC/CS reaction centers in the species and reveals that the greater regulation is carried out by the inactivation of a fraction of the reaction centers [28].Thus, it is observed that plants improve the reception of their primary source of energy, and this helps to determine their adaptation to environmental conditions, stimulating the development of acclimation and plasticity mechanisms.

Variegated plants and the control showed different statistically of the parameter electron transport beyond the QA (ΨO). This difference showed that the amount of light energy captured by the antenna complexes and directed to the reaction centers of each species is also different [11]. The lower the value of this parameter, as shown by *F. albivenis*, the lower the probability of final production of NADPH for the carboxylation reactions of plants [29]. The variegated species *F. albivenis, H. phyllostachya* and *A. ficoidea* presented the lowest photosynthetic efficiencies of FSII (φPo) and electron transport (φEo). These species presented an insufficient capacity to reduce QA compared to the control species and the variegated *T. spathacea*. Thus, these species tend to have higher maximum quantum efficiencies of non-photochemical excitation (φDo) [11]. In our study, we found no correlation between these parameters and the concentration of photosynthetic pigments. It suggests that each species independently adapts its biophysical and metabolic conditions to optimize its photosynthetic processes.

The parameters ABS/CS, TR_0/CS, and DI_0/CS, referring to the phenomenological fluxes of plants by leaf cross-sectional area (CS), also proved to be good markers for distinguishing variegated plants from controls. The same did not happen with the electron flow per leaf area, ETO/CS, since the green control, *J. mesnyi*, did not differ from the control *T. pallida* and the variegated *T. spathacea*. This similarity may be associated with an apparent and similar ability to reduce QA [25]. Studies show that the energy absorption per unit of cross-section (ABS/CS) is directly related to the concentrations of photosynthetic pigments by plants, as they represent the amount of photon energy absorbed by the antenna complexes associated with the active and inactive RCs of the FSII [11, 26, 28]. Our work corroborates this research since the species that presented the highest values for ABS/CS were the green control, *J. mesnyi*, and the variegated *F. albivenis*. However, this species (*F. Albivenis*) presented the lowest energy flux trapped by the RCs of the FSII (TR_0/CS), while *J. mesnyi* presented the highest index. This parameter represents the fraction of the photon flux absorbed and used for the separation and stabilization of the primary charge of FSII as $P680^+QA^-$.

Therefore, the number of electrons transferred to QA in the electron transport chain is equal to the number of electrons captured by the FSII reaction centers [26]. This characteristic proves that even though they do not differ statistically regarding the concentration of Chla, Chlb, and anthocyanins, both species differ significantly regarding the number of active (open) and inactive (closed) reaction centers. This plasticity guarantees that each species has its mechanisms of photosynthetic activities.

The high values of the energy dissipation parameter per reaction center (DI_0/RC) and, consequently, per cross-section (DI_0/CS) of *F. albivenis* can be explained by the fact that this species showed low densities of active RCs per cross-section (RC/CS). *F. albivenis* present greater susceptibility to high irradiances than the other species in this study, including the control species. The accommodation was caused by damage caused to the FSII, resulting in their low photochemical efficiency (φPo) [30]. The high density of inactive reaction centers in this species may have overloaded the activity of its active reaction centers, as indicated by its high ABS/RC and TR_0/RC values.

The plants analyzed that presented the highest contents of anthocyanins were the ones that attested the greatest thermal energy dissipation in the FSII per unit of cross section (DI_0/CS). These pigments, mostly present in the spongy parenchyma of plants, absorb light at wavelengths ranging from 400 to 600 nm, coinciding with the spectrum of light absorption by chlorophyll b [14]. Studies indicate a photoprotective role of these molecules by acting as "filters" of light energy in the epidermal layers of leaves [4, 14, 18]. In addition to this primordial function in variegated plants, these pigments help the processes of transpiration, nutrient absorption, metabolism and their growth rates, directly linked to the photosynthetic processes of plants [18]. Therefore, plants with high levels of anthocyanins such as *F. albivenis* and *J. mesnyi* can physiologically adapt better in environments with high incidences of sunlight.

The two species of the *Tradescantia* genus, one of them being the purple control, showed the highest performance indices, followed by the green control, *J. mesnyi*. As a result, high functional activities of photosystems per unit cross-section (PI_{CS}) have been observed for these species. This activity implies a high level of photon energy conservation performance by the FSII to reduce QA and QB (PI_{ABS}) in these species [25, 26]. These values are in line with the values of φPo, however, with greater sensitivity in evaluating the efficiency of FSII as they are related to the absorption, capture, and transfer of excitation energy, providing a broader view of the fluorescence of chlorophyll [26, 30]

Together with their respective controls, the variegated plants differed much more in their $PI_{TotalABS}$ and $PI_{TotalCS}$ indices than concerning PI_{ABS} and PI_{CS}. These differences imply that these last two parameters, when multiplied by the efficiency values of the electron transport flux to the FSI acceptors and the phenomenological fluxes, can be used as markers for species distinction regarding the energy conservation of absorbed photons antenna complexes [31]. *Fittonia albivenis* showed the lowest values for these parameters despite having high levels of photosynthetic pigments, demonstrating that its low density of active reaction centers and high energy dissipation in the form of heat are the leading causes for the reduction of their vitality indices.

5. CONCLUSION

Tradescantia pallida, one of the control species since it is entirely purple and the variegated *T. spathacea* presented the best strategies for photon absorption, energy trapping, electron transport, and final PSII acceptors in photosynthetic complexes. The green control species, *J. mesnyi*, and the variegated *H. phyllostachya, A. Ficoidea*, showed intermediate performances concerning these mentioned parameters. On the other hand, the species *F. albivenis*, showed the lowest photosynthetic performance. Its great susceptibility to the inactivation of its reaction centers and high energy dissipation rates in the form of heat, despite high contents of Chlb and anthocyanins. Thus, *F. albivenis* demonstrated less efficient mechanisms to regulate and use light energy for carbohydrate synthesis.

Given the above, in domestic environments with high levels of light radiation during the day, in places such as windows and balconies, for example, preference should be given to the maintenance and cultivation of the variegated plant species *T. spathacea*. The variegated *H. phyllostachya* and *A. ficoidea* can be exposed in residential environments with more significant variations in light radiation, such as kitchens and living rooms. Finally, the variegated *F. albivenis* is the best option to be kept in environments with low light radiation levels, such as washrooms. Thus, we accept the hypothesis that motivated our study. The variegated species with the highest levels of photosynthetic pigments and the highest energy dissipation by fluorescence would be the most suitable for environments with lower light radiation levels.

6. BIBLIOGRAPHIC REFERENCES

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