Rodriguésia 72: e00662020. 2021
http://rodriguesia.jbrj.gov.br
DOI: http://dx.doi.org/10.1590/2175-7860202172066

Original Paper

Ecophysiological response of *Astronium fraxinifolium* (Anacardiaceae) in degraded and non-degraded brazilian Cerrado

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Abstract

Plants native from Cerrado generally have peculiar characteristics that allow tolerating water and nutritional stress. *Astronium fraxinifolium* is a Anacardiaceae tree of from Brazilian Cerrado. The aim of this research was to characterize *A. fraxinifolium* leaves morphophysiologically, in order to recognize characteristics related to acclimatization of the species in different soil conditions. Two populations of *A. fraxinifolium* were sampled in different study areas, A1 (Degraded Soil) and A2 ("Undegraded Soil"). Nitrogen compounds, total carbohydrates, chlorophyll, nutritional content, stomatal density and gas exchanges were quantified, comparing the areas. A high number of stomata was observed on the abaxial surface of *A. fraxinifolium* leaves, with a higher density occurring in A1 individuals. The values of chlorophyll and boron content were significantly higher in A2 plants. It's possible that the lowest concentration of boron in A1 plants is related to chlorophyll production. Regardinf the other analysis, there weren't significant differences between the areas. The results show that this species undergoes changes in production of chlorophyll, but liquid photosynthesis isn't impaired, considering the low chlorophyll content in A1 being compensated by the higher stomatal density. Thus, these changes may be the result of acclimating this species to different environmental conditions to which it's exposed.

Key words: chlorophyll content, nutritional deficiency, physiological characterization, stomatal density.

Resumo

Plantas nativas do Cerrado geralmente apresentam características peculiares que permitem tolerar o estresse hídrico e nutricional. *Astronium fraxinifolium*, é uma das espécies arbóreas de Anacardiaceae do Cerrado brasileiro. O objetivo desta pesquisa foi caracterizar morfofisiologicamente as folhas de *A. fraxinifolium*, com o objetivo de reconhecer características relacionadas a aclimatação da espécie em diferentes condições de solo. Para tanto, duas populações de *A. fraxinifolium* foram amostradas em diferentes áreas de estudo, A1 (Solo degradado) e A2 ("Solo não degradado"). A partir dessas amostras foram quantificados compostos nitrogenados, carboidratos totais, amido, clorofila, conteúdo nutricional, densidade estomática, trocas gasosas, alocação de carbono e altura da parte aérea, comparando as áreas de estudo. Um número elevado de estômatos foi observado na superfície abaxial das folhas de *A. fraxinifolium*, com uma densidade estomática mais elevada ocorrendo em indivíduos de A1. Já os valores de clorofila a, b e total foram significativamente maiores em A2. O conteúdo nutricional não diferiu entre as áreas, exceto o boro, que apresenta maior concentração nas plantas de A2. É possível que a menor concentração de boro nas plantas A1 esteja relacionado a produção de clorofila. Em relação às demais análises, não houve diferenças significativas entre as áreas. Os resultados mostram que esta espécie sofre alterações na produção de clorofila, mas a fotossíntese líquida não é perturbada, sendo o baixo teor de clorofila em A1 compensado pela maior densidade estomática. Assim, essas mudanças podem ser o resultado da aclimatação dessa espécie às diferentes condições ambientais às quais está exposta.

Palavras-chave: teor de clorofila, deficiência nutricional, caracterização fisiológica, densidade estomática.

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Introduction

Due to human activities, there has been a progressive increase in the environmental degradation events (De Long *et al.* 2015). Soil degradation directly affects local vegetation once soil conditions is one of the determinant factors to the development of plants, and so it may directly compromise the infiltration capacity (Deon *et al.* 2018), soil water storage and root growth (Dinis *et al.* 2015; Cambi *et al.* 2017).

Thus, environmental factor such as soil conditions, water, light, temperature, etc., influence the morphophysiological characteristics of the plants and so, environmental changes can cause structural changes (Costa *et al.* 2012; Alameda & Villar 2012; Aref *et al.* 2013; Cambi *et al.* 2016) and physiological (Warren *et al.* 2011; Alameda & Villar 2012; Lu *et al.* 2012; Cambi *et al.* 2016; Wang *et al*. 2018).

In the literature there are several reports of natural regeneration in sites with degraded soils with growth of several species representing several taxa (Ferreira *et al.* 2010; Araújo *et al.* 2006; Higuchi *et al.* 2006; Gama *et al.* 2002; Campos & Landgraf 2001). These observations indicate the high capacity of these plants to acclimatise to environmental changes. For example, xerophytic species exhibit adaptive characteristics that allow them to survive in rocky soils with low nutrient availability and undergoing large periods of drought (Fahn & Cutler 1992).

A species that occurs in high frequency in recovering degraded areas in the Brazilian Cerrado is *Astronium fraxinifolium* Schott (Venturoli *et al.* 2011; Marangon *et al*. 2008; Neri *et al.* 2005). It is commonly known as "Gonçalo-Alves", and it is a pioneer, deciduous, heliophyte, native xerophytic tree plant from the Cerrado (Lorenzi 2002).

Considering that environmental changes, such as degradation, can cause modifications in plant morphophysiology the present study aimed to evaluates morphology and physiology of the leaves of *Astronium fraxinifolium*, in order to verify the occurrence of morphophysiological changes in two populations species occurring in non-degraded and degraded areas and too evaluate the soil physical-quality by the construction of the dam of the Ilha Solteira hydroelectric power plant, Brazil. Therefore, we evaluated the stomatal density, specific leaf area, gas exchange, nutritional status, chlorophyll content, quantification of nitrogen compounds, total carbohydrate and starch.

Material and Methods

The botanical material was sampled at FEPE (Fazenda de Ensino, Pesquisa e Extensão, city of Selvíria, MS, Brazil) belonging to UNESP/FEIS. The sampling area has an average annual rainfall of 1,300 mm with an average temperature of 23.5 °C, climate type is AW, according to Köeppen. The soil is classified as dystrophic red Latosol, sandy clay loam texture (Embrapa 2013).

The area where the farm is located had a great impact in the 1970s, and a large area had about 8.6 m deth of soil removed to be used in the construction of the dam of the Ilha Solteira Hydroelectric Power Plant and, with this, the soil horizons A and B were lost, as well as great part of the nutrients (Modesto *et al.* 2009).

In this way, samples were collected in two different areas. The first area, which had a direct impact due to soil removal for the construction of the dam (A1), shows *Astronium fraxinifolium* specimens that had spontaneous sprout after soil removal. The second area, which was not directly impacted by the construction of the dam (A2), is a place where the species was cultivated, obtaining all the necessary conditions for its initial growth and development.

This natural regeneration in the area occurred at the same time that the planting of the seedlings of this species was carried out in the non-degraded area, so that both populations are close to 30 years old. It should be emphasized that only the impact caused by the removal of the surface layers of the soil for the construction of the hydroelectric dam is being considered, and not impacts coming from other sources.

In all, 20 trees from each studied area were choosen for sampling.

Soil analysis

For the physical analysis, 10 undeformed samples were sampled from each study area, 5 of them collected at depths of 0–20 cm and 5 of 20–40 cm, using the volumetric ring method (Teixera *et al.* 2017). This analysis was performed at the Soil Physics Laboratory of Unesp Ilha Solteira.

The chemical analysis of the soil was performed at the Soil Fertility Laboratory of Unesp Ilha Solteira. For this analysis, 10 composite samples collected from 20–40 cm deep

were collected, using the method described by Raij *et al.* (2001).

Stomatal density

To determine stomatal density, it was performed cuttings in the medial portion of the third leaflet of *Astronium fraxinifolium* leaves of each study area, and such cuts (rectangles of approximately 1 × 2 cm) were fixed on the glass slides with fast drying glue, ester of cyanoacrylate (Segatto *et al.* 2004). After total drying of the glue, the plant material was removed from the blade leaving only the impression of the abaxial leaf face. Afterwards, photographs were taken of the blades with the abaxial face impressions of each sample with a 40X magnification. The total number of stomata was counted in each image. Considering the scale bar (µm) expressed in each photograph, the total area of the photo was calculated in μm^2, from which it was transformed into mm^2, making it possible to estimate the number of stomata per mm^2.

Specific leaf area

The analysis of the specific leaf area was performed with digitization of 10 leaflets of the trees collected by the leaf area scanner belonging to Analytical Development Company Limited. Afterwards, they were oven dried at 65 ºC (Gobbi *et al.* 2011) for 72 hours. The specific leaf area was calculated, which consists of the ratio between the leaf area (cm^2) and the weight of the leaf dry mass (g).

Gas exchange analysis and carbono allocation

The liquid photosynthesis, stomatal conductance and the internal CO_2 concentration in the sub-stomatic chamber were evaluated using a portable gas exchange device, Infra Red Gas Analyzer (IRGA) (Richardson *et al.* 2017; Paixão *et al.* 2017); brand ADC BioScientific Ltd, model LC-Pro, it was set 1,000 µmol m^{-2} s^{-1} of photosynthetically active radiation (PAR), provided by LED lamps and 380 ppm CO_2 and chamber temperature at 28 °C. A branch was separated from the plant and immediately afterwards, gas exchange and carbon allocation of three random leaflets were measured. It should be noted that the measurements were made on a sunny day at dawn in both areas, avoiding climatic interference in the data.

Total chlorophyll determination

Fresh leaf samples were collected from both study areas. The dosage of chlorophylls *a* and *b* was performed according to Hiscox & Israeltam (1979). The leaves were cut into thin strips (1 mm) in 50 mg samples. Three replicates of the assay were made for each sample. The plant material was placed in a test tube with 7 mL of DMSO (dimethyl sulfoxide), covered with glass beads and kept in water bath at 65 ºC for 30 minutes in the dark. After cooling at room temperature, the extract was read in spectrophotometer at 645 and 663 nm (Arnon 1949). The content of the pigments was obtained from the equations proposed by Arnon, as follow below:

$$C_a\ \mu g.ml^{-1} = (12.70 \times A_{663}) - (2.69 \times A_{645})$$

$$C_b\ \mu g.ml^{-1} = (22.90 \times A_{645}) - (4{,}68 \times A_{663})$$

$$C_a + C_b\ \mu g.ml^{-1} = (20.20 \times A_{645}) + (8.02 \times A_{663})$$

* Data expressed as µg/g FW (micrograms per gram of fresh weight).

Extraction and quantification of nitrogen compounds, total carbohydrate and starch

To extract nitrogen compounds, total carbohydrate and starch it was used the method described by Bieleski & Turner (1966). For each 1g of fresh material it was added 10 ml of MCW solution (60% methanol, 25% chloroform, 15% ml H_2O). The material was well crushed, homogenized and then centrifuged. Then, 1 ml Chloroform + 1.5 ml H_2O was added for each 4 ml supernatant recoverd. The solution recovered was kept for 24-hour in fridge for phase separation and the water-soluble phase was used for the analysis of amino acids and total soluble carbohydrates.

After the first extraction, it was added 0.1 N NaOH (at a ratio of 1:10, w/v) to the remaining pellet, which was resuspended and centrifuged, and then the supernatant was recovered and used for total soluble proteins quantification.

After protein extraction, it was added 30% perchloric acid (at a ratio of 1:10, w/v) to the remaining pellet, which was resuspended and centrifuged, and then the supernatant was recovered and used for starch quantification.

Total soluble amino acids were quantified according to Yemm & Cocking (1955); total soluble protein was quantified according to Bradford

(1976); total soluble carbohydrate and starch was quantified according to Umbreit *et al.* (1957).

Nutritional analysis

The sampled leaves were washed with distilled water containing a little detergent and then rinsed with distilled water in successive portions to remove any impurity and detergents. Then the leaves were placed on absorbent paper.

The leaves were placed in perforated paper bags and then dried in a forced air circulation oven, with a temperature ranging from 65 to 70 ºC. After dried, they were milled using a stainless steel mill to avoid contamination of the sample mainly by iron, zinc and copper. The grinded samples were subjected through a 1 mm sieve. This sieved material was used to determine N, P, K, Ca, Mg, S, Cu, Fe, Mn and Zn contents according to Malavolta *et al.* (1997).

Statistical analysis

To quantify nitrogen compounds, total carbohydrates, starch, nutritional content, gas exchange and carbon allocation, the analyzes consisted of three random leaflets from each individual, among the 20 field trees we randomly sampled 6 to carry out the analysis aforementioned. For the analysis of chlorophyll, stomatal density and specific leaf area, 20 trees from each area were sampled and evaluated.

All data were submitted to descriptive statistics, using R Core Team software (2018), in order to evaluate the possible morphophysiological differences in plants of the different areas studied.

Results

Soil characteristics

The physical analysis of the soil showed that A1 and A2 present very different characteristics for the apparent density, macroporosity, microporosity and total porosity attributes (Tab. 1).

The chemical characteristics of the soil that presented differences between the areas were: sum of bases (SB); aluminum saturation (m%); cation exchange capacity (C.E.C); organic matter content (MO). SB was 9.5 $mmol_c\ dm^{-3}$ for $A1^{3+}$ and 12.1 $mmol_c\ dm^{-3}$ for A2. For m%, an average of 43% was observed for A1 and 52% for A2. Soil C.E.C was 34.5 and 76.1 $mmol_c\ dm^{-3}$ for A1 and A2 respectively, with MO of 13 g dm^{-3} in A1 and 23 g dm^{-3} in A2. The soil in both areas is acidic, with values of 4.4 for A1 and 4.1 for A2.

Table 1 – Bulk density (BD), macroporosity (MA), microporosity (MI) and total porosity (PT) of soil A1 and A2, at depths of 0–20 cm and 20–40 cm.

	BD (kg dm^{-3})		MA ($m^3\ m^{-3}$)		MI ($m^3\ m^{-3}$)		TP ($m^3\ m^{-3}$)	
	0–20 cm	**20–40 cm**	**0–20 cm**	**20–40 cm**	**0–20 cm**	**20–40 cm**	**0–20 cm**	**20–40 cm**
A1	1.70 a	1.73 a	0.087 a	0.090 a	0.258 a	0.236 a	0.345 a	0.325 a
A2	1.40 b	1.33 b	0.132 b	0.155 b	0.338 b	0.330 b	0.470 b	0.485 b

* different letters indicate statistical difference between the two areas ($p < 0.05$; $n = 10$). Student's t-test was applied to all data.

Morphophysiology of plant leaves

The epidermis of the *Astronium fraxinifolium* leaf presents cells with curved walls and anomocytic stomata, which are observed on both sides of the leaf, which is therefore called amphistomatic (Muir 2015). In the abaxial epidermis there are stomata in great numbers throughout the leaf surface, which is almost completely composed of stomata and guard cells. On the adaxial side, stomata and guard cells are present only in regions closer to the rib. A total of 1,801.5 and 1,619.5 stomata/mm^2 on the abaxial surface were observed for leaves coming from the areas A1 and A2 ($p = 0.035$; $n = 20$), respectively, with A1 being statistically different from A2.

Regarding the specific leaf area, no significant difference was observed between the populations, presenting a mean of 82.87 cm^2/g for A2 and 77.66 cm^2/g for A1 ($p = 0.17$; $n = 20$). Considering gas exchanges and carbon allocation, there was no significant difference between the areas for all variables: stomatal conductance, internal CO_2 concentration in the sub-stomatic chamber, liquid photosynthesis and transpiration (Tab. 2). For chlorophyll analysis, significant differences were

Table 2 – Net photosynthesis (A - $\mu mol\ CO_2\ m^{-2}\ s^{-1}$), transpiration (E - $mmol\ H_2O\ m^{-2}\ s^{-1}$), stomatal conductance (g_S - $mol\ H_2O\ m^{-2}\ s^{-1}$) e internal concentration of CO_2 (C_i - $\mu mol\ CO_2\ mol\ m^{-2}\ s^{-1}$) in leaves of *Astronium fraxinifolium* area A1 and A2.

	A1	A2	p- value
A	13.91 ± 2.08 a	9.24 ± 1.28 a	0.074
E	1.33 ± 0.12 a	1.52 ± 0.21 a	0.670
g_s	0.08 ± 0.01 a	0,11 ± 0,02 a	0.830
C_i	203.28 ± 25.18 a	216.42 ± 32.20 a	0.730

* different letters indicate statistical difference between the two areas ($p < 0.05$; $n = 36$). For A, E e g_s the Wilcoxon test was used, considering that such data were non-parametric, for Ci, the Student's t-test was applied.

found for the three variables. Chlorophyll *a*, *b* and total chlorophylls were significantly higher in A2 (Tab. 3).

The mineral content of *Astronium fraxinifolium* leaves showed that there was no significant difference between the two populations for all macro and micronutrients, except for boron (B), which was significantly higher in A2, with an average of 27 mg/kg A2 and 17 mg/kg in A1 (Tab. 4). Concerning the data of nitrogen compounds, total soluble proteins, total carbohydrate and starch, also there were no significant differences (Tab. 5).

Discussion

Soil characteristics

There is a close relationship between total porosity and bulk density, when density increases, total porosity decreases (Chen *et al.* 2014), with high density being indicative of soil compaction (Rosa-Filho *et al.* 2009). Total porosity is composed of microporosity (soil water volume) and macroporosity (soil aeration). With the reduction of pores in the soil, in addition to making aeration difficult, it hinders root penetration and water infiltration into the soil (Calonego *et al.* 2017).

Bowen (1981) states that a soil with a density greater than or equal to 1.55 kg dm^{-3} already has compaction that can affect root growth. For agriculture, the ideal is that the soil has microporosity between 0.250 and 0.330 $m^3\ m^{-3}$ and macroporosity 0.170 and 0.250 $m^3\ m^{-3}$, with the ideal total porosity of 0.500 $m^3 m^{-3}$ (Kiehl 1979; Lima *et al.* 2007).

Studies conducted in preserved Cerrado areas show that soils in this biome have values close to or above those indicated for agriculture. Sena *et al.* (2017), for example, observed 0.470 $m^3\ m^{-3}$ of total porosity in a soil of a preserved Cerrado area. Oliveira *et al.* (2013) and Araújo *et al.* (2007), who also evaluated preserved Cerrado soil, observed values of 0.580 and 0.650 $m^3\ m^{-3}$ respectively, for total porosity.

As observed in this research, the A1 presents lower values than those presented in conserved Cerrado soil and recommended for agriculture, presenting compacted soil.

The m% is high in both areas, considering that soils with m% greater than 50% are considered alic, containing huge amount of exchangeable aluminum (Ronquim 2010), low CEC and acidic pH, characteristics that are common in Cerrado soils (Haridasan 2008; Skorupa *et al.* 2012).

Table 3 – Concentration of chlorophylls *a* and *b* and total chlorophylls (µg/gMF) in leaves of *Astronium fraxinifolium* from areas A1 and A2.

	A1	A2	p- value
Chlorophyll *a*	1972.76 ± 49.46 b	2171.11 ± 62.07 a	0.013
Chlorophyll *b*	582.46 ± 22.62 b	661.12 ± 20.13 a	0.010
Total Chlorophyll	2554.63 ± 67.93 b	2831.56 ± 76,77 a	0.007

* different letters indicate statistical difference between the two areas ($p < 0.05$; $n = 40$). For Chlorophyll b the Wilcoxon test was used, considering that such data were non-parametric, for the others, Student's t-test was applied.

Table 4 – Concentration of macro and micronutrientes in leaves of *Astronium fraxinifolium* in areas A1 and A2.

	A1	A2	p- value
N g/Kg	21.18 ± 1.03 a	22.53 ± 0.85 a	0.380
P g/Kg	1.45 ± 0.12 a	1.45 ± 0.08 a	1
K g/Kg	8.03 ± 0.55 a	903 ± 0.45 a	0.230
Ca g/Kg	7.85 ± 1.29 a	5.80 ± 0.46 a	0.195
Mg g/Kg	3.7 ± 0.31 a	2.78 ± 0.30 a	0.085
S g/Kg	1.43 ± 0.07 a	1.90 ± 0.18 a	0.057
B mg/Kg	17.16 ± 1.67 b	27.33 ± 1.66 a	0.002
Cu mg/Kg	8 ± 0.84 a	12.16 ± 2.06 a	0.120
Fe mg/Kg	167.5 ± 29.40 a	215 ± 54.92 a	0.630
Mn mg/Kg	52.16 ± 16.25 a	41.83 ± 10.04 a	0.632
Zn mg/Kg	11 ± 0.74 a	11 ± 1.31 a	0.806

* different letters indicate statistical difference between the two areas ($p < 0.05$; $n = 6$). For N, Ca, Cu, Fe the Wilcoxon test was used, considering that such data were non-parametric, for the other nutrientes the Student's t-test was applied.

Morphophysiology of plant leaves

Chlorophylls play a key role in photosynthesis and are components of the antenna complex, light absorption, excitation of the Photosynthesis Reaction Center and water photoxidation (Taiz & Zeiger 2009). In the studies of Lima *et al.* (2004) with *Oryza sativa*, Neves *et al.* (2009) with *Eugenia uniflora*, Massacci *et al.* (2008) with cotton (*Gossypium* sp.) and Jaleel *et al.* (2008) with *Catharanthus roseus* who showed a decrease in chlorophyll when presented with different types of plant stress, a result similar to that observed for *A. fraxinifolium*.

It has been reported in several studies that photosynthetic rate changes in plants subjected to environmental stresses (Rashid *et al.* 2018; Karimi & Tavallali 2017; Silva *et al.* 2017; Silva & Arrebaça 2004), such observations could indicate that the photosynthetic capacity of the plant was affected by stress and that, possibly, the individuals of A1 would present low photosynthetic rate. However, there was no change in net photosynthesis between the two populations.

In the literature, there are species that under environmental impact present an increase in leaf area (Gobbi *et al.* 2011; Rad *et al.* 2011), and in other species there is a decrease (Scalon *et al.* 2011; Machado *et al.* 2010; Maranho *et al.* 2006; Alves *et al.* 2001), in addition it should be expected increase in leaf area in leaves of *Astronium fraxinifolium* as a way to compensate the low amount of chlorophyll.

However, such landings did not occur. Despite this, the species in general has a very high stomatal density, as observed in *Miconia pycnoneura* (Howard 1969). The change in stomatal density is observed in studies with other species, and may be caused by several types of environmental changes

Table 5 – Concentration of soluble amino acids, soluble proteins, total carbohydrate and starch (μmol/gMF) in leaves of *Astronium fraxinifolium* in areas A2 and A1.

	A1	A2	p- value
Aminoacids	2.62 ± 0.74 a	4.08 ± 2.03 a	0.818
Soluble proteins	21.71 ± 4.04 a	12.40 ± 3.68 a	0.431
Total carbohydrate	0.60 ± 0.07 a	0.52 ± 0.05 a	0.416
Starch	0.51 ± 0.09 a	0.94 ± 0.18 a	0.088

* different letters indicate statistical difference between the two areas ($p < 0.05$; $n = 6$). For amino acids and total carbohydrate the Wilcoxon test was used, considering that such data were non-parametric, for the other metabolites the Student's t-test was applied.

such as: water stress (Nemerkéri *et al.* 2018; Khan *et al*. 2010; Batista *et al.* 2010; Grisi *et al.* 2008), salt stress (Barbiere *et al*. 2019; Shabala *et al.* 2013; Orsini *et al.* 2012), shading (Gobbi *et al.* 2011), pollution (Alves *et al.* 2008; Maranho *et al.* 2006) and even the rise in altitude (Kučerová *et al.* 2018). Thus, it is believed that such characteristics are adaptations that allow the survival of the species under conditions of environmental stress, so that there is an increase in the number of stomata as a way to compensate the low production of photosynthetic pigments.

Thus, the fact that the plant does not need to invest heavily in chlorophyll production indicates that under such stress would be more likely to establish itself in the environment, using the few nutrients it can capture more effectively, since it is an expensive molecule to be produced by being composed of large amounts of carbon and nitrogen (Porra *et al.* 1994; Taiz & Zeiger 2009). This observation can be confirmed by the fact that there is no significant difference in the levels of nitrogen compounds, proteins, total carbohydrates and starch, so that this reflects the efficiency of nutrients utilization by the species.

Another change observed among populations was the concentration of boron in leaves, a factor that may be related to the characteristics of the soil. Boron, although not fully understood, is an essential nutrient for plants (Matoh & Kobayashi 1998; Taiz & Zeiger 2009), and it is known that it has in important role in keeping the integrity of the cell wall, together with calcium (Cakmak & Römheld 1997; Matoh & Kobayashi 1998; Blevins & Lukaszewski 1998; Goldbach & Wimmer 2007).

Thus, deficiency of this nutrient can cause problems in the integrity of the cell wall (Cakmak & Römheld 1997) impairing root elongation (Dell & Huang 1997), as well as cause anatomical changes in vascular tissues (Hajiboland *et al.* 2012). Also, boron deficiency may lead to decreased levels of chlorophylls and carotenoids (Moustafa-Farag *et al.* 2015).

All these morphophysiological characteristics presented by the species allow it to be tolerant to environmental disturbance that it is exposed, so that it can survive and develop in a degraded soil (A1). This is corroborated by the fact that the species occurs by natural regeneration in A1 and does not present altered photosynthetic rate. Although there is a change in the chlorophyll content, we can infer that this factor is offset by an increase in stomatal abundance.

Observing all features, it is possible to emphasize the importance of the use of *A. fraxinifolium* for reforestation and recovery of degraded areas, since it is a pioneer plant species that survives under stressful environmental conditions for most species. It could not only reforest areas along with other species, but facilitating the establishment of early secondary species in the ecological succession process (Ferreira *et al.* 2010; Walker & Wardle 2014).

It can be inferred that the environmental stress, selects species with ability to adjust to the environment, so that these survive in different conditions, which can be observed in the species *A. fraxinifolium*.

We conclude that the decrease in boron content, the lower levels of chlorophylls and the increase in stomatal density can be an acclimation to the environmental changes to which the species has been exposed. These data confirm that *A. fraxinifoluim* presents great plasticity adapting the conditions of environmental stress. This fact is of extreme importance for studies of the recovery of degraded areas since it is a pioneer species and could be very used in the process of ecological succession of these areas.

Acknowledgements

The authors thanks the grant conceived by 'Conselho Nacional de Desenvolvimento Científico e Tecnológico' (CNPq), CAPES (Coordenação de Aperfeiçoamento de Pessoal de Nível Superior), the scholarship conceived to the second author, the support the publication of this paper (CAPES/AUXPE 88881.593505/2020-01) and the 'Fundação de Amparo à Pesquisa do Estado de São Paulo' for the scholarship for scientific initiation of the first author (n. 2017/00277-3)..

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Area Editor: Dr. Marcelo Mielke
Received in April 27, 2020. Accepted in August 05, 2020.