Initial Growth and Morpho-Anatomical Development of “Jacaranda” at Different Levels of Luminosity

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Abstract
Thus, the objective of the research was to evaluate the initial growth and development of seedlings under *Platymiscium floribundum* at different levels of luminosity. The experimental design was completely randomized in a 4 x 10 factorial scheme (four levels of shade: full sun, 50%, 65% and 80% with ten replications per treatment. The height, number of leaves was evaluated monthly. At the end of the experiment at 180 days, leaf dry matter, stems, roots, and dry matter. In addition, measurements were made of the variables related to gas exchange and plant anatomy of leaves. The results obtained were observed higher growth and developed satisfactorily at full sun), without presenting morphological changes. For the gas exchange did not differ statistically. It is concluded that the initial growth of *P. floribundum* seedlings was superior in the light level in full sun, providing favorable conditions for their development.

Keywords: Anatomy, Chlorophyll, Forest Species, Physiology, Stomatal Conductance.

1. INTRODUCTION AND OBJECTIVES

*Platymiscium floribundum* Vogel., it is a forest species belonging to the Fabaceae family, popularly known as sacambu, jacaranda and jacerandá-do-coastal, a species with characteristics of the humid forest of the Atlantic hillside (Freire et al., 2019). The species has great economic potential, due to the production of wood, being biodiesel component (Nogueira et al., 2020), disease treatment (Freire et al., 2019). Studies on the morpho-anatomical behavior of Brazilian flora consists of generating knowledge that makes it possible to determine ideal conditions for production of interest for the recovery of degraded areas.

Some environmental factors influence plant growth and development, such as light and temperature (Felsemburgh et al., 2016; Syzmanska et al., 2017). Exposure of the plant to total sunlight can lead to changes in morphological, physiological and anatomical changes, as well as in stomatal density and / or changes in the proportion of photosynthetic tissues, leading to a change in the distribution of biomass (Gomes et al., 2019). As well as, cell damage and plant death (Syzmanska et al., 2017).

Among the various components of the environment, light levels are essential for the formation of seedlings for reforestation, aiming at obtaining higher quality plants to withstand climatic storms and adequate growth at the final production site (Salles et al., 2017). Therefore, shading screens become an alternative against the harmful effects of high incidence of solar radiation and extreme temperatures, microclimate conditions proper for the growth of seedlings destined for reforestation (Zare et al., 2019).

Studying the responses of these species to variations in luminosity is of great importance for the practical knowledge of the needs of these plants and their use in reforestation, reforestation and forest formation (Gomes et al., 2019).

The present study aimed to evaluate the initial growth and development of seedlings under *P. floribundum* at different levels of luminosity for reforestation.
2. MATERIALS AND METHODS

The work was carried out in the vegetation house belonging to the Laboratório de Ecologia Vegetal Departamento de Fitotecnia e Ciências Ambientais, Universidade Federal da Paraíba (UFPB), Areia, PB.

P. floribundum seedlings were produced with seeds from matrix trees located in the city of Areia, PB. The fruits were completely healthy and showed no signs of predation at the beginning of the natural dehiscence process. After collection, the fruits were placed in paper bags and sent to the Laboratório de Ecologia Vegetal, opened manually with scissors to remove the seeds.

2.1. Germination test

The seeds were disinfested by means of 1% sodium hypochlorite solution for 3 minutes and rinsed in distilled water for 1 minute. 100 seeds were used, which were divided into four repetitions of 25. The seeds were considered photoblastic positive, germinated in the presence of light, therefore, they were placed in a regime of white light. The seeds were placed on three sheets of Germitest paper. Moistened with distilled water in the amount equivalent to 2.5 times the dry paper mass, and placed in germinating chambers of the type Biochemical Oxygen Demand (B.O.D.) adjusted to constant temperatures of 25°C (Alves et al, 2016). When germination stabilized at 14 days after sowing, normal seedlings were selected, that is, they had an average of 5 cm in length and 1 pair of cotyledons. those with structures with root and aerial part. (Brasil, 2009).

2.2. Levels of shading

40 seedlings were selected, averaging 10 cm in length and 1 pair of cotyledons. Subsequently, the seedlings placed in bags of 15 cm x 30 cm polyethylene black plastics with a capacity of 2 liters containing substrate based on vegetable soil, soil mixed with remnants of organic matter, such as leaves, stems and bark and with perforations below to drain excess water. Wooden frames approximately 1.0 m high, top and side covered with commercial black polyethylene shading screen (Sombrite®) were used to obtain the different levels of shading. 10 replicates were placed in each, totaling 40 seedlings were evaluated in each treatment. The seedlings were submitted to four levels of shading: full sun, 50%, 65%, and 80%. The irrigation sprinkler system was performed manually, when necessary, to replenish water

2.3. Growth variables

Monthly, the following growth variables were measured: plant height considering the soil level, the distance between the plant lap and the insertion of the last expanded leaf measured with a millimeter ruler. The number of leaves with counts in each plant of each treatment.

The roots were washed in running water to remove all soil residues, dried on absorbent paper and packed in kraft paper bags, taken to the forced air circulation oven at 65 °C until reaching constant weight. Then the dry matter of the leaves (LDR), the stems (SDM), the roots (RDM) were weighed. With the dry matter data, the total dry mass was calculated.

2.4. Photosynthetic Variables

At 130 days after sowing the evaluation of the gas exchange was performed using an IRGA LC-Pro Sd Portable, ADC BioScientific, UK. Measurements were performed on the leaves of the median third of each plant in the period from 10 to 11 h in the morning. The variables were rates of liquid photosynthesis (A), stomatal conductance (Gs), transpiration (E), internal carbon (Ci).

The relative chlorophyll content of the leaves was determined using a portable device, ClorofiLOG®, model CPL 1030, by means of six readings per plant in the medium part of the leaf.

2.5. Anatomical variables

The leaves submitted to gas exchange evaluations were collected, sections of the middle third of the second expanded leaf (in the apex-base direction), and fixed in 70% alcohol (formaldehyde, acetic acid and ethyl alcohol) (Johansen, 1940) and conducted to the Laboratório de Ecologia e Reprodução de Plantas, where the preparation of the slides was carried out for the purpose of anatomical studies.

In order to obtain the cuts, it was made by clarification in sodium hypochlorite, followed by triple washing in distilled water, 1% methylene blue staining (transversal and paradérmico respectively) (Kraus & Arduin, 1997) and mounting in glycerinated water. For the parasideric cuts, the stomatal density, expressed in mm², in the micrometric and objective 40 x following the technique of Labouriau et al. (1961). The leaf thickness was evaluated by means of transverse sections of semipermanent leaves with three leaves from each treatment, with 10 replicates. Measurements made by the Model microscope equipped with a Micrometer Ocular. The measurements were performed in three fields.
per leaf, making a total of 30 measurements for each leaf tissue per treatment.

Semipermanent slides were obtained from cross sections of fresh material washed in distilled water, diaphanized in commercial sodium hypochlorite for 15 min and washed again three times in distilled water for 5 min each. After the material was stained in 1% methylene blue and 50% ethanol for 30 sec and washed in distilled water for 1 min, the sections were blanket-capped with 50% glycerin and sealed with colorless enamel (Kraus & Arduin, 1997).

The following variables were determined: stomatal density (number of stomata per mm$^2$); polar diameter of stomata and equatorial diameter of stomata. The polar and equatorial diameter of stomata was calculated based on the average of their length and width (adapted from Johansen, 1940). In the cross section of the leaf blades, leaf thickness, total cross-sectional area of the adaxial epidermis and abaxial epidermis were evaluated and the stomatal pore (length and transversal) was also evaluated.

2.6. Statistical Analysis

The experiment was conducted in a completely randomized design. The data of height and number of leaves were submitted to a polynomial regression analysis. For the other variables, analysis of variance was performed, and the averages were compared using the Tukey test, at a level of 5% probability using the SAS® statistical program.

3. RESULTS AND DISCUSSION

A higher height was observed in the plants in an environment with 80% shading, 10.55 cm high, while the 50% shading treatment had an average of 9.5 cm (Figure 1A). The number of leaves presented by the seedlings at after sowing was significantly affected by shading. The number of leaves was higher in plants grown under 80% shading, followed by treatment full sun, while 50% of shaded plants emitted few leaves during the experimental period (Figure 1B).

The investment in height presented by plants in the shade occurs in response to the faster growth promoted by the greater investment in cell stretching (Mota et al., 2013). This can be an adaptation mechanism to escape the light deficit, since they are not able to tolerate low light intensities, adjusting their metabolic rates (Morais Neto et al., 2000).

Regarding the height of the Jatobá plants (Hymenaea courbaril var. Stilbocarpa), there was a significant difference with a 50% and 80% shade screen, because higher shading triggers a superior adaptation mechanism to escape the light deficit (PagliarinI et al., 2017). Brazilian woody seedlings (Caesalpinia echinata Lam.) Showed similar behavior in relation to height, in the shade treatments of 20 and 40% (Aguiar et al., 2011). Dalmolin et al. (2015) studying sambaiba plants (Curatella americana L.) in full sun showed a higher number of leaves when compared to plants grown in the shade, a typical characteristic of deciduous plants.

The plants under shading had a shoot length, however, they did not increase their mass in the same proportion, which leads us to consider that the shading condition is limiting for the Tabebuia aurea, indicating typical behavior of escape to the shade. Because the plants presented this elongation in the stem due to the low light intensity that they were submitted (Souza Pinto et al., 2017).

The RDM and SDM variables did not differ statistically between shade levels, however for LDM the treatment full sun verified higher contents for dry mass, (0.431g) it was superior to other treatments (Figure 2).
Figure 2. Dry matter of roots, dry matter of stems and dry matter of leaves of Platymiscium floribundum plants at different levels of shading.

In contrast, Dantas et al. (2011) working with shade levels in true catingueira (Caesalpinia pyramidalis) seedlings, which also belong to the Fabaceae family, obtained fresh mass of shoot and root smaller in the treatment of 50% compared to full sun. Plants that grow under greater luminous intensity have a greater accumulation of dry mass in the root allowing a greater absorption of water and nutrients, a strategy that would guarantee the plant the capacity to withstand higher rates of photosynthesis and transpiration in more enlightened environments (Silva et al., 2007). The seedlings of P. floribundum presented a slight increase in the production of RDM under higher luminosity intensities, noting the good adaptive capacity of the species at different light incidences.

3.1. Gas exchange

On the parameters of gas exchange photosynthesis (A), transpiration (E) and stomatal conductance (Gs), there was no significant difference when the plants were submitted to conditions of shading levels. At the moment of measurement, the internal carbon (CO₂) there was a significant difference between the treatments, the increase in the internal concentration of CO₂ verified in the treatment of 50% shading (Table 1).

Table 1. Mean values of photosynthesis (A), transpiration (E), stomatal conductance (Gs) and internal carbon of Platymiscium floribundum plants at different levels of shading.

<table>
<thead>
<tr>
<th>Shading</th>
<th>Photosynthesis (µmol m⁻² s⁻¹)</th>
<th>Transpiration (mol m⁻² s⁻¹)</th>
<th>Stomatal conductance (mmol m⁻² s⁻¹)</th>
<th>Internal carbon (µmol mol⁻¹)</th>
</tr>
</thead>
<tbody>
<tr>
<td>full sun</td>
<td>3.40a</td>
<td>1.40a</td>
<td>0.046a</td>
<td>244.00b</td>
</tr>
<tr>
<td>50%</td>
<td>3.60a</td>
<td>2.60a</td>
<td>0.136a</td>
<td>290.40ab</td>
</tr>
<tr>
<td>65%</td>
<td>3.20a</td>
<td>2.60a</td>
<td>0.143a</td>
<td>319.20a</td>
</tr>
<tr>
<td>80%</td>
<td>5.80a</td>
<td>2.80a</td>
<td>0.088a</td>
<td>264.80ab</td>
</tr>
<tr>
<td>CV%</td>
<td>23.09</td>
<td>31.12</td>
<td>18.20</td>
<td>24.16</td>
</tr>
</tbody>
</table>

*Means followed by the same letter in the column are statistically equal to each other by the Tukey test at 5% significance.

Thus, it appears that gas exchange at levels of shading is an intrinsic characteristic of each species. Given this information, it is necessary to report the importance of light for its development. The main factor that influences this process is the quantity and quality of light energy (Barros et al., 2019), as well as the need for photosynthesis to interact with chlorophyll (Oliveira; Gualtieri, 2017). The high luminosity over a certain time can harm the plants, in order to cause greater absorption of photons and less assimilation, thus being able to cause the photoinhibition or death of the plant (Taiz et al., 2017). According to the same author, the low light provides inadequate amounts of energy for the plants, also limiting the photosynthetic performance during the photochemical phase.

The leaf is one of the main organs involved in performing photosynthesis, so it is considered as the one that most responds promptly to environmental variations, as they aim to understand the changes that plants make to leaf morpho-anatomy to facilitate their survival (Lemos, et al., 2020).

It can be observed that the species P. floribundum revealed to be a species of leaf like hypoestomatic, that is to say, they present restricted stomata in the abaxial face, as for the type were found stomata of the paracítico type.

In the environments where plants were exposed to radiation intensities, all morpho-anatomical attributes verified statistically significant differences between stomatal density, leaf thickness, except for the size of the guard cells (length), data set forth in Table 2. The stomatal density (number of stomata/mm²) in the treatment under 0% (full sun) and 50% shading showed a reduction as a function of the increase of luminosity. For the stomatal (polar) dimensions were significantly affected (P <0.05) by shading, while the length did not differ statistically. The equatorial diameter were higher in plants developed 65% shading (15.33 μm) indicating the effect of the availability of solar radiation on the stomatal dimensions.

Thus, the reduction of the size of the stomata of plants grown in full sunlight are probably adaptations of the plant to minimize the loss of water, because the smaller the stomata, the faster the stomatal pore opening and closing process (Schmidt et al. 2017).

Stomatal morphological parameters, including size, density and location, varied between environments, but luminosity is one of the environmental factors that most influence this variation (Liu et al., 2016), thus showing that plants have the capacity to adjust these characteristics in response to changing environment.
Table 2. Stomatal density (stomata per mm²), diameter of stomata (polar and equatorial), leaf thickness (μm) and stomatal pore length and transversal of *Platymiscium floribundum* plants at different levels of shading.

<table>
<thead>
<tr>
<th>Shading</th>
<th>Stomatal density (mm²)</th>
<th>Diameter (μm)</th>
<th>Leaf thickness (μm)</th>
<th>Stomatal pore (μm)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Polar</td>
<td>Equatorial</td>
<td>Length</td>
</tr>
<tr>
<td>full sun</td>
<td>40.66a</td>
<td>33.66a</td>
<td>14.83ab</td>
<td>5.20b</td>
</tr>
<tr>
<td>50%</td>
<td>36.66a</td>
<td>41.00a</td>
<td>10.83c</td>
<td>9.55a</td>
</tr>
<tr>
<td>65%</td>
<td>27.16b</td>
<td>37.00a</td>
<td>15.33a</td>
<td>8.75a</td>
</tr>
<tr>
<td>80%</td>
<td>25.50b</td>
<td>35.66a</td>
<td>11.88c</td>
<td>9.00a</td>
</tr>
<tr>
<td>CV%:</td>
<td>10.27</td>
<td>12.71</td>
<td>18.45</td>
<td>6.01</td>
</tr>
</tbody>
</table>

*Means followed by the same letter in the column are statistically equal to each other by the Tukey test at 5% significance.*

The leaf thickness was influenced by shading levels: greater thickness when grown at 50.65 and 80% shading (Table 2).

The stomatal pore length did not show significant differences in the different levels of shading, however, the transversal pore length was higher in the condition of 80% shading and 50% shading (Table 2).

The chlorophyll/a content was significantly higher in the 65% shade-grown plants, the chlorophyll/b concentration was affected at 0% (full sun), so in the present study it was observed that shade leaves had a higher concentration of chlorophyll when compared to the sun. We grouped the data to calculate the total chlorophyll ratio, which did not show a significant difference (Table 3).

Table 3. Chlorophyll a and b contents, chlorophyll a / b ratio of *Platymiscium floribundum* plants at different levels of shading.

<table>
<thead>
<tr>
<th>Shading</th>
<th>Chlorophyll /a μg cm²</th>
<th>Chlorophyll /b μg cm²</th>
<th>Chlorophyll Relation a/b μg cm²</th>
</tr>
</thead>
<tbody>
<tr>
<td>full sun</td>
<td>27.50c</td>
<td>6.00c</td>
<td>20.16a</td>
</tr>
<tr>
<td>50%</td>
<td>32.40b</td>
<td>9.80a</td>
<td>23.00a</td>
</tr>
<tr>
<td>65%</td>
<td>38.83a</td>
<td>9.21ab</td>
<td>4.83a</td>
</tr>
<tr>
<td>80%</td>
<td>32.85b</td>
<td>8.85bc</td>
<td>22.50a</td>
</tr>
<tr>
<td>CV%:</td>
<td>7.82</td>
<td>23.80</td>
<td>15.97</td>
</tr>
</tbody>
</table>

*Means followed by the same letter in the column are statistically equal to each other by the Tukey test at 5% significance.*

Plants under shading conditions tend to increase chlorophyll concentration to maximize light capture (Azevedo, 2014), for those cultivated under low light intensities present higher levels of chlorophyll. In addition, under appropriate lighting conditions, chlorophyll molecules are synthesized and degraded in the same proportion and under intense radiation conditions, degradation is more pronounced (Ferreira et al., 2012).

The perspective of the results obtained in the present study goes through concepts related to the evidenced or behavior similar to that found for most species of tropical forests, that is, limitation in the conditions of luminosity, but with capacity to maintain, in these conditions, the positive growth rate.

The adaptive capacity under different lighting conditions can even be used in reforestation projects or in consortium with other more demanding species in relation to the amount of light.

However, the species responded to different ambient light patterns through morphological and anatomical adjustments with efficiency. This facilitates its occurrence in the different environments and demonstrates its capacity of phenotypic plasticity and adaptation to the variations of luminosity.

4. CONCLUSION

The initial growth of *P. floribundum* seedlings was superior in the light level in full sun, providing favorable conditions for their development.

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Erratum: Initial Growth and Morpho-Anatomical Development of “Jacaranda” at Different Levels of Luminosity

In the article “Initial Growth and Morpho-Anatomical Development of “Jacaranda” at Different Levels of Luminosity”, with the DOI code number: https://doi.org/10.1590/2179-8087-FLORAM-2020-0091, published at 28(4): e20200091, where it was written:

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