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Original Paper

Does shading mitigate water restriction in *Ormosia arborea* seedlings?

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Abstract
This study aimed to evaluate the potential of shading in reducing the stressful effect of water restriction on photosynthetic and antioxidant metabolism, favoring the growth of *Ormosia arborea* seedlings and the recovery metabolic. The shadings levels studied were: 0, 30 and 70%. Two water regimes were implemented: control, in which the plants were irrigated at 75% of soil water retention capacity and water restriction, in which the irrigation was suspended until the photosynthetic rate reached values close to zero (P0), period at which the seedlings were re-irrigated. There were two evaluation periods: P0 and recovery. We observed that water restriction reduced photosynthetic metabolism, growth and quality of *O. arborea* seedlings by increasing the activity of antioxidant enzymes. The seedlings present higher quality index, stomatal conductance, intrinsic carboxylation efficiency and transpiration when cultivated under 70% shading in recovery. The stressful effect of water restriction was mitigated by the shading of 70%, where the seedlings were able to recover, resuming their photosynthetic metabolism and quality after resumption of irrigation. In addition, 0% shading is not indicated for this species.
Key words: antioxidant enzymes, chlorophyll *a* fluorescence, ecophysiology, gas exchange, native tree.

Resumo
O objetivo deste estudo foi avaliar o potencial do sombreamento na redução do efeito estressante do déficit hídrico sobre o metabolismo fotossintético e antioxidante, favorecendo o crescimento de mudas de *Ormosia arborea* e sua recuperação metabólica. Os níveis de sombreamento estudados foram: 0, 30 e 70%. Foram implantados dois regimes hídricos: controle, em que as plantas foram irrigadas a 75% da capacidade de retenção de água no solo e restrição hídrica, em que a irrigação foi suspensa até que a taxa fotossintética atingisse valores próximos a zero (F0), período em que as mudas foram re-irrigadas. Ocorreram dois períodos de avaliação: F0 e recuperação. Observamos que a restrição hídrica reduz o metabolismo fotossintético, o crescimento e a qualidade das mudas de *O. arborea*, aumentando a atividade de enzimas antioxidantes. As mudas apresentam maior índice de qualidade, condutância estomática, eficiência de carboxilação da Rubisco e transpiração quando cultivadas sob sombreamento de 70% na recuperação. O efeito estressante da restrição hídrica foi mitigado pelo sombreamento de 70% quando as mudas foram capazes de se recuperara, retomando seu metabolismo fotossintético e qualidade após a retomada da irrigação. Além disso, 0% de sombreamento não é indicado para essa espécie.
Palavras-chave: enzimas antioxidante, fluorescência da clorofila *a*, ecofisiologia, trocas gasosas, árvore nativa.

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Introduction

Global climate change and the need to recover degraded areas has raised the attention to the in the propagation of native species (Lázaro-Nogal *et al*. 2015). The species of the family Fabaceae have good employability in human and animal nutrition, medicinal use, handcrafts, and nitrogen fixation, which has led to their exploration, requiring studies to facilitate their sustainable exploitation (Duarte *et al*. 2012).

Among the species of this Family, *Ormosia arborea* (Vell.) Harms (Fabaceae: Papilionoideae), popularly known as "olho de cabra" (Carvalho 2008), is a native and endemic species of Brazil, that occurs in the phytogeographic domains of Cerrado and Atlantic Forest (Cardoso & Meireles 2015). The species is used for ecological restoration in humid areas, being tolerant to periodic flooding (Junglos *et al*. 2016). It is found in phytophysiognomies subject to adverse climatic conditions (Ressel *et al*. 2004) and it is classified as late secondary or climax (Carvalho 2008).

Considering that species is on the verge of extinction, because of the constant devastation of its natural environment (Pick-Upau 2012), therefore actions that aim at the conservation of this genetic resource, with wide applicability and high potential for environmental, social and economic use, conservations actions are needed. In view of the difficulty of producing seedlings of *O. arborea* due to the low seedlings emergence rate, it is necessary to maintain the growth and metabolism in the few seedlings produced, requiring ecophysiological studies that may enable the cultivation and survival (Costa *et al*. 2015).

Light and water availability are abiotic factors that directly influence plant growth, once they are related to metabolic adjustments through chlorophyll *a* fluorescence and photosynthetic metabolism of each species, especially in function of their ecological group. So, since *O. arborea* plant are found in humid and shading environments (Junglos *et al.* 2016; Carvalho 2008), and that water restriction is a limiting factor for the photosynthetic metabolism of seedlings of this species (Bastos *et al.* 2022), we hypothesized that shading levels mitigates the stressful effect of water restriction, keeping gas exchanges and photochemical of photosynthesis and the growth of seedlings of *O. arborea* stable, which is made possible by the action of antioxidant enzymes, thus facilitating its recovery after the resumption of the water supply.

In this context, our aim was to evaluate the potential of shading in reducing the stressful effect of water restriction on photosynthetic and antioxidant metabolism, favoring the growth of *O. arborea* seedlings and their recovery.

Material and Methods

Plant and cultivation material

The experiment was carried out in a greenhouse at the Faculty of Agrarian Sciences of the Federal University of Grande Dourados (UFGD), in the municipality of Dourados/MS-Brazil (22º11'43.7"S, 54º56'08.5"W, 452 m). The seedlings of *O. arborea* were cultivated in 5-liter polyethylene pots containing Dystrophic Red Latosol and sand in the proportion of 1:1 (v/v), with irrigation at a 70% water retention capacity (WRC) of the substrate. When seedlings showed average height of 20 cm, were stored under three shading levels: 0% (full sun), 30% and 70%, and these levels were simulated using shade cloths, commonly known as 'sombrite®'.

The value of the photosynthetic photon flux density (PPFD) of 1,300; 700 and 400 μmol m^{-2} s^{-1} under 0%, 30% and 70% shade, respectively. The average temperatures and relative humidity, respectively, were 41.1 °C and 30% under 0% shading, 35.1 °C and 40% under 30% shading and 36.4 °C and 43% under 70% shading.

The water regimes were constituted of two conditions: control, where the plants were irrigated daily (I), *i.e.*, maintaining 75% of the soil water retention capacity according Souza *et al.* (2000) and the water restriction regime, which was composed of stressed plants (S). In the water restriction treatment, regime irrigation was suspended, until the photosynthetic rate reached to zero, followed by resumption of irrigation. During restriction we do not monitor the water status of the substrate.

The seedlings were evaluated in two periods: the first, characterized as photosynthesis zero (P0), was determined when the photosynthetic rate of the seedlings of each shading reached values close to zero. The second phase, characterized as recovery (Rec), occurred after the resumption of irrigation of the seedlings, *i.e.*, the seedlings were evaluated when the photosynthetic rate did not present statistical differences to the rates of the control seedlings, for their respective shadings.

At each evaluation period, the following characteristics were evaluated in the second pair of fully expanded leaves without symptoms of nutritional deficiency or injuries:

Leaf water potential (Ψ_w)

The leaves were collected and conditioned in a refrigerated container to prevent transpiration. In laboratory, the leaf water potential was obtained from measures read immediately after leaf collection, between 7:00 and 10:00 a.m., using a Scholander-type pressure chamber (Portable Plant water status console - model 3115 - TECNAL), and the results were expressed in MPa.

Leaf area (LA), chlorophyll index and Dickson Quality Index (*DQI*)

Total leaf was measured with the aid of the Li 3100 - Area Meter (Nebraska - USA). Chlorophyll index was measured using the (SPAD 502 - MINOLTA Chlorophyll meter).

Seedling quality was determined based on the Dickson Quality Index (*DQI*) according to the height of aerial parts (H), diameter (D), total dry mass (TDM), which is given by the sum of the dry mass of aerial parts (DMAP) and roots (DMR), using the following equation DQI = TDM/[(H/D) + (DMAP/DMR)] (Dickson *et al*. 1960).

Gas exchange

Gas exchange were measured with a portable photosynthesis measurer (LCIPro- SD ADC Bio Scientific Ltd.) The following items were evaluated: photosynthetic rate - A (μmol m^{-2} s^{-1}), stomatal conductance - gs (mol m^{-2} s^{-1}), transpiration - E (mmol m-^{2} s-^{1}) and intracellular CO_2 concentration - C_i (μmol mol^{-1}), based on these data the intrinsic carboxylation efficiency (A/C_i), intrinsic water use efficiency (A/gs) and water use efficiency ($WUE = A/E$) were calculated. The determination of gas exchange was performed between 8:00 and 11:00 a.m.

Chlorophyll *a* fluorescence

Chlorophyll *a* fluorescence was obtained with the aid of portable fluorometer model OS-30p (Opti-Sciences Chlorophyll Fluorometer, Hudson, USA) evaluating the potential quantum efficiency of photosystem II (F_v/F_m), the effective efficiency of absorbed energy conversion (F_v/F_0) and the basal quantum production of non-photochemical processes in PS II (F_0/F_m). For fluorescence analysis, the leaves were submitted to a period of 30 minutes of adaptation to the dark with the aid of adaptor clips. The determination of chlorophyll *a* fluorescence was performed between 8:00 and 11:00 a.m., on the same leaves of the second pair of fully expanded leaves, considering photosynthetic photon flux of 1,500 μmol m^{-2} s^{-1}.

Activity of antioxidant enzymes

Activity of antioxidant enzymes was evaluated in leaves and roots of each treatment, using frozen tissue in liquid nitrogen, to avoiding metabolic changes. From each sample, one gram was macerated in 6 mL of solution containing 0.3 g of Polyvinylpyrrolidone (PVP) diluted in 100 mL of potassium phosphate buffer (0.2 M). Then, the samples were centrifuged at 4,000 rpm for 20 minutes at 4 °C and the supernatant was used as enzymatic extract. The activity of superoxide dismutase (SOD), catalase (CAT) and peroxidase (POD) was analyzed according to Broetto (2014).

Experimental design and statistical analysis

For each evaluation period, the experiment was carried out in a completely randomized design in a split-plot scheme. The plots were characterized by water regimes (I, E) and the subplots represented the three levels of shading (0%, 30% and 70%) with four replicates, represented by four pots with two plants in each pot. The data were subjected to homogeneity of variance and normal distribution by Shapiro-Wilk test. The data were submitted to variance analysis and once the F test ($p < 0.05$) effects were significant, the means of the plots were submitted to the Bonferroni t test and the means of the subplots submitted to the Tukey test ($p < 0.05$), with the aid of the statistical software SISVAR (Ferreira 2014).

Results

Leaf water potential (Ψ_w)

We observed a reduction in water potential (Y_w) in seedlings of *O. arborea* under water restriction in P0, regardless of shading. In Rec, the seedlings recovered the values and there was no significant difference between the irrigated and stressed seedlings irrespective shading level. However, under the 70% shading the seedlings presented Y_w higher than that of the seedlings under 0% and 30% shading, emphasizing that the higher level of shading allowed the greater recovery of the Y_w (Fig. 1a).

Chlorophyll index

The seedlings under 70% shading presented higher chlorophyll index in both water regimes and in the two evaluation periods (P0 and Rec) and the lowest indexes under 0% shading, in both water availability conditions (Fig. 1b). In Rec, only seedlings under 70% shading recovered their chlorophyll index.

Leaf area (LA) and Dickson Quality Index (*DQI*)

Leaf area (LA) decreased in P0 with water restriction, regardless of light conditions. Although in Rec only seedlings under 0% shading recovered LA, and under 30% shading LA was significantly higher when compared to other cultivation conditions, the values in all conditions under previous water restriction were still low. The LA of seedlings under 70% shading and irrigated remained higher in the two evaluation periods (Fig. 1c).

Dickson Quality Index (*DQI*) decreased in P0 and remained lower in seedlings under water restriction, even after the irrigation was resumed. However, in Rec, the seedlings regained their quality, since there were no significant differences among seedlings that were irrigated and previously stressed. However, under 70% shading the seedlings obtained higher quality (Fig. 1d).

Gas exchange

Photosynthetic rate (*A*) decreased under water restriction, presenting an average value of 0.53 μmol m^{-2} s^{-1} in order not to cause irreversible damage, which occurred 15 days after the suspension of irrigation for the seedlings under 0% shading, and at 20 days, for seedlings under 30% and 70 % shading. (Fig. 2a). The seedlings under 0% shading maintained lower *A*, even after resumption of irrigation (Fig. 2b). The recovery of *A* occurred at 18 days after the resumption of irrigation of the seedlings under 0% shading and

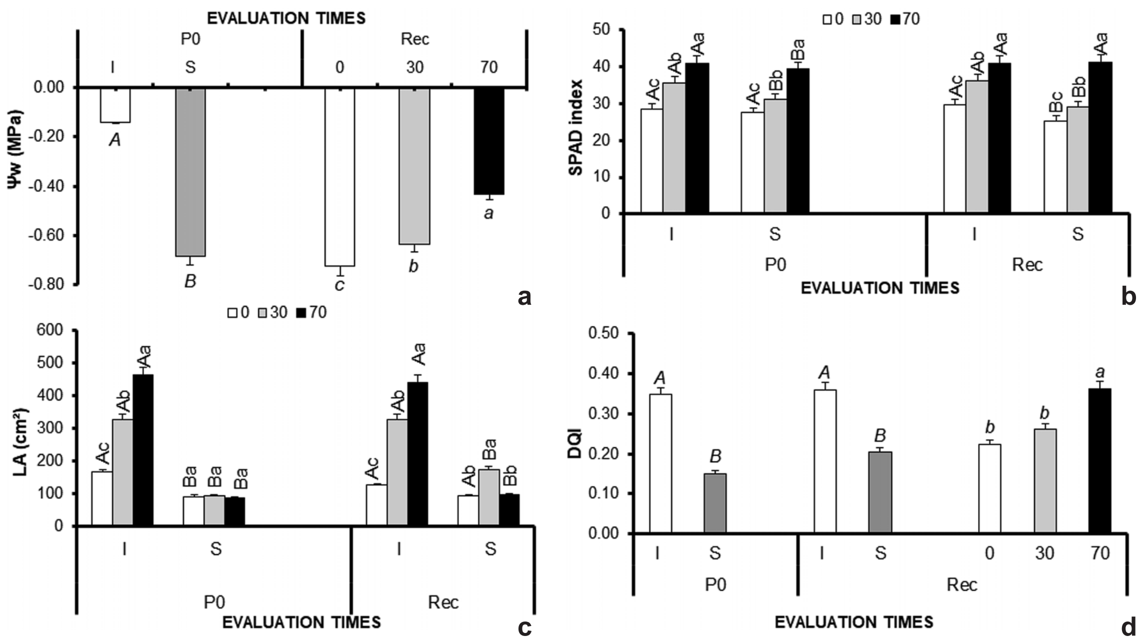

Figure 1 – a-d. Seedlings of *Ormosia arborea* under different water regimes: irrigated (I) and stressed (S) and in different shadings levels (shading): 0%, 30% and 70% – a. leaf water potential - Ψ_w; b. chlorophyll index - SPAD; c. leaf area - LA; d. Dickson Quality Index - *DQI*. Capital letters in italics compare the different water regimes. Lowercase letters in italics compare the different shadings (a, d). Uppercase letters compare the different water regimes in the same shading and lowercase letters compare the different shadings in the same water regime (b, c). The means followed by different letters indicate significant differences by Bonferroni *t* test for water regimes, and by Tukey test for shadings ($p < 0.05$) (n= 4).

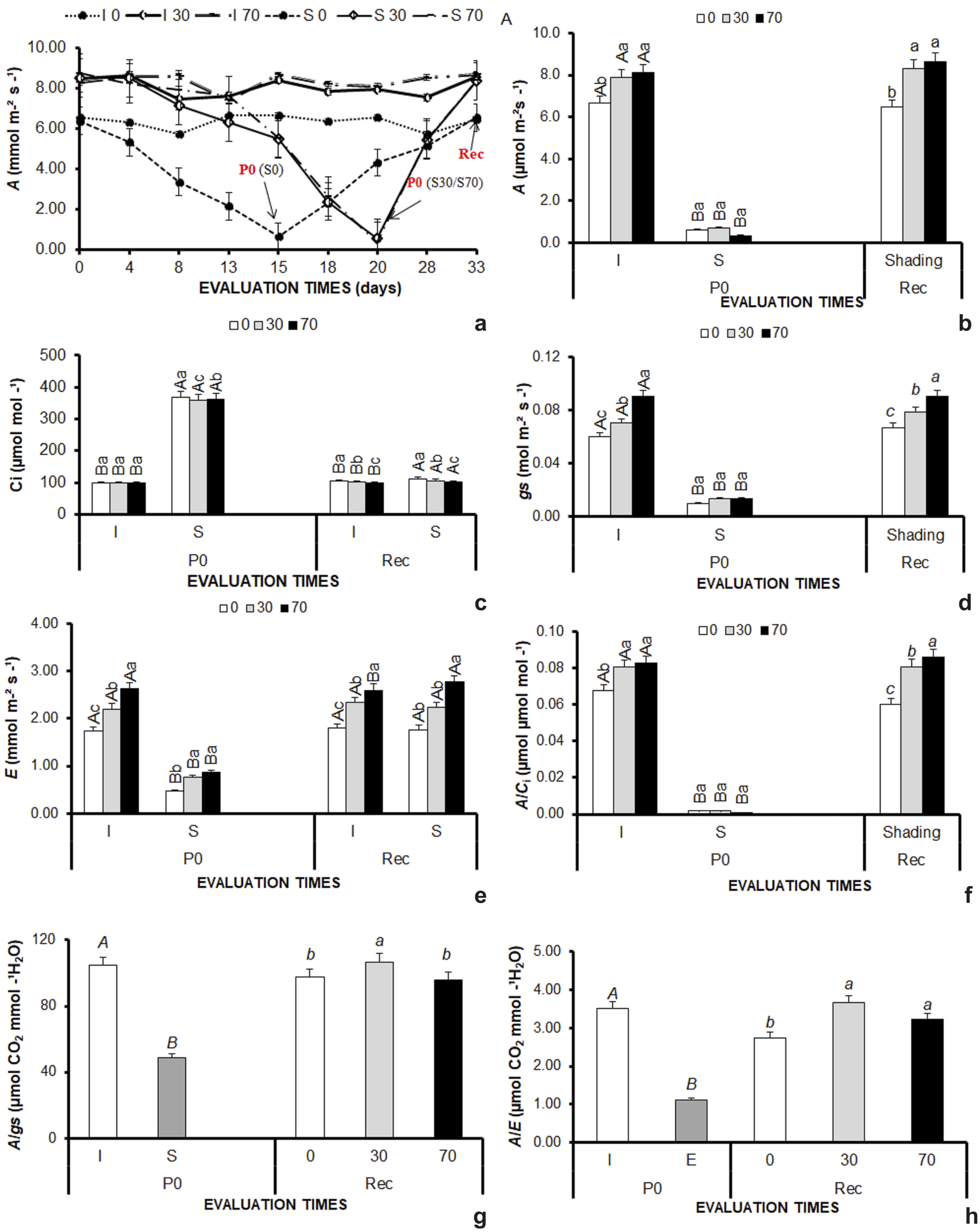

Figure 2 – a-h. *Ormosia arborea* seedlings under different water regimes: irrigated (I) and stressed (S) in different shadings levels (shading): 0%, 30% and 70% – a,b. photosynthetic rate - *A*; c. internal CO_2 concentration - C_i; d. stomatal conductance - *gs*; e. transpiration rate - *E*; f. instantaneous efficiency of CO_2 carboxylation - A/C_i; g. intrinsic efficiency of water use - *A*/*gs*; h. instantaneous efficiency of water use - *A*/*E*. Capital letters in italics compare the different water regimes. Lowercase letters in italics compare the different shadings. Uppercase letters compare the different water regimes in the same shading and lowercase letters compare the different shadings in the same water regime. Means followed by different letters indicate significant differences by Bonferroni *t* test for water regimes and Tukey test for shadings ($p < 0.05$) (n= 4).

at 13 days for the seedlings under 30 and 70% shading.

The intracellular CO_2 concentration (C_i) increased significantly for stressed seedlings (E) in P0, and this increase was 72.9% when compared to irrigated seedlings. After resumption of irrigation, the values decreased, becoming similar to the values of plants under irrigation daily (I) (Fig. 2c).

The stomatal conductance (*gs*), transpiration (*E*) and carboxylation efficiency (A/C_i) decreased with water restriction regardless of shading, except for *E*, which presented reduction in both shadings (Fig. 2d-f). Once the irrigation was resumed, the seedlings recovered the values of these characteristics, and *gs* and A/C_i were higher under 70% shading.

For *A*/*gs* and *A*/*E* in P0, the stressed seedlings presented lower ratios, recovering after re-irrigation, and were higher under 30% shading in Rec (Fig. 2g-h).

Chlorophyll *a* fluorescence

The quantum efficiency of photosystem II (F_v/F_m) decreased in stressed seedlings regardless of shading levels, and after resumption of irrigation there was no significant difference between treatments (Fig. 3a). The basal quantum production of the non-photochemical processes of photosystem II (F_0/F_m) was lower in P0 under 30% shading, however, in Rec it was higher, followed by seedlings under 0% shading (Fig. 3b). The effective efficiency of the photosystem in the conversion of absorbed energy (F_v/F_0) also decreased in P0 for seedlings under water restriction, after resumption of irrigation, the seedlings recovered their values, but F_v/F_0 was higher under 30% shading (Fig. 3c).

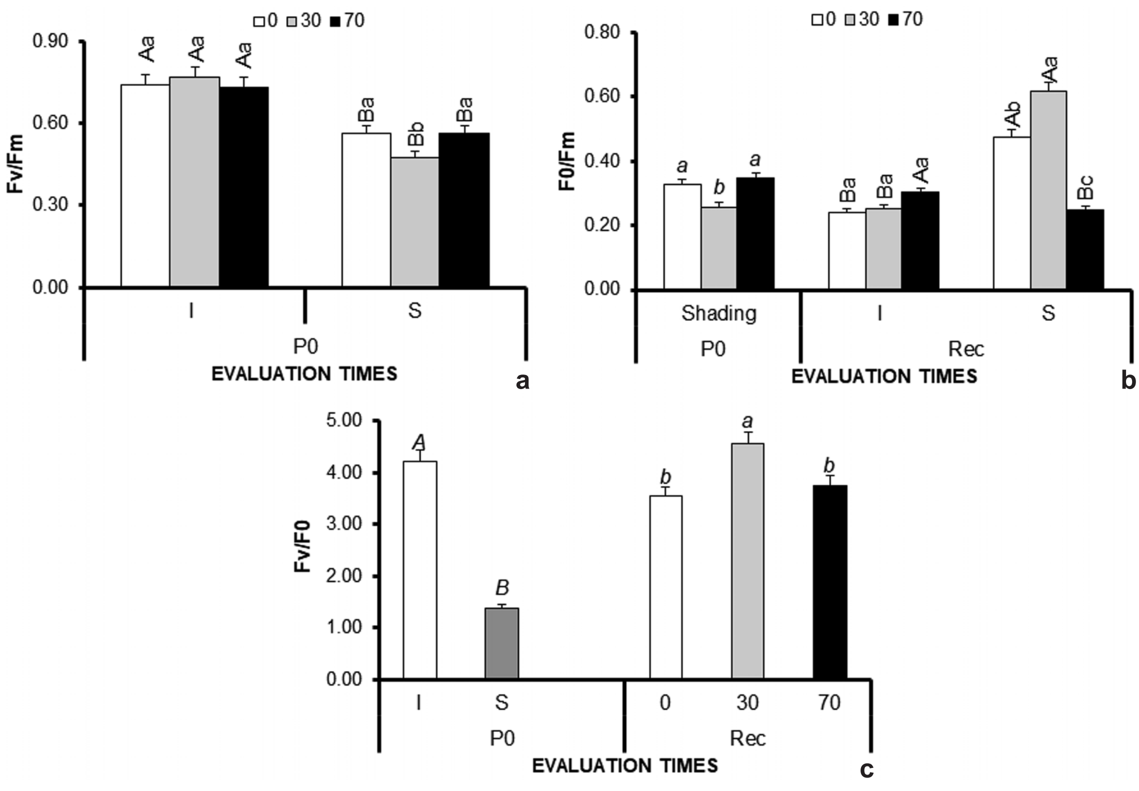

Figure 3 – a-c. *Ormosia arborea* seedlings under different water regimes: irrigated (I) and stressed (S) in different shadings levels (shading): 0%, 30% and 70% – a. photochemical efficiency in photosystem II - F_v/F_m; b. basal quantum production of non-photochemical processes of photosystem II - F_0/F_m; c. effective efficiency of the photosystem in the conversion of absorbed energy - F_v/F_0. Uppercase letters in italics compare the different water regimes. Lowercase letters in italics compare the different shadings. Uppercase letters compare the different water regimes in the same shading and lowercase letters compare the different shadings in the same water regime. Means followed by different letters indicate significant differences by Bonferroni *t* test for water regimes and Tukey test for shadings ($p < 0.05$) (n = 4).

Activity of antioxidant enzymes

The SOD activity in the leaf as well as in the root in Rec presented similar values between treatments, regardless of the water condition, although the seedlings previously stressed and under shading still maintained higher values (Fig. 4a,b).

The CAT in the leaf during Rec decreased in previously stressed seedlings, however under 0% shading it remained higher (Fig. 4c). At the root, after reirrigation, even though all seedlings under previous restriction water had their enzymatic activity decreased, the values remained higher than the control treatments, in the seedlings under 0% shading the value was higher than the others shading levels (Fig. 4d).

The activity of POX in leaf after rehydration of the seedlings under 70% and 30% shading was lower in the Rec (Fig. 4e). In the root, the behavior was similar to that of CAT, and in seedlings under 0% shading, *i.e.*, present higher enzymatic activity, regardless of water regime and evaluation time, with higher values in stressed seedlings (Fig. 4f).

Discussion

Ormosia arborea seedlings kept under 30 and 70% of shading presented superior physiological performance than seedlings under 0% shading, especially when submitted to water restriction, which is explained by their late secondary classification (Carvalho 2008). Based on the results of higher *Yw*, chlorophyll index, seedling quality and photosynthetic metabolism after resumption of irrigation, during the recovery period, we believe that shading mitigated the stressful effects of water restriction by favoring seedling recovery, corroborating our hypothesis, although shading did not prevent seedlings from showing signs of stress during water restriction (P0).

The water potential is directly related to the physiological behavior of plants, and in conditions of low soil water supply, photosynthesis is inhibited, which in most species is due to stomatal closure, which is the first plant protection mechanism to prevent water loss and turgor maintenance (Hu *et al*. 2010). This relationship corroborates the results found in this research with stressed seedlings of *O. arborea*, which, during water restriction in P0, presented lower values of Ψ_w, *A*, *gs*, *E*, A/C_i, *DQI* and chlorophyll index. However, shading mitigated the stressful effect of water restriction and favored the recovery of these characteristics after the resumption of irrigation. The seedlings under 0% shading remained with lower Ψ_w even with the resumption of irrigation, since high light incidence probably impaired the maintenance of the integrity of the water transport system, triggering metabolic disorders, causing embolism in metaxylem, however, this characteristic was not evaluated in our study.

The higher quality of seedlings and their ability to perform photosynthetic metabolism under shading can guarantee the plant greater competitive advantage when occupying clear recovering environments of degraded areas. The excess of light on the photosynthetic apparatus of seedlings not adapted to such environments causes photoinhibition, leading to partial or total inactivation of the photosystem (Araújo & Deminicis 2009). The oxidative damage to the photosynthetic apparatus that led to degradation of chlorophyll molecules or deficiency in their synthesis, as seen in *Eugenia uniflora* L. plants (Martinazzo *et al*. 2007), which presented lower mean chlorophyll values in 0% shading and higher under 50% shading, making it possible to ensure greater use of solar radiation. We observed in the seedlings of *O. arborea* in this work the maintenance of lower levels of chlorophyll under 0% shading, although we should consider that under shading there is a tendency to increase this characteristic as a resource to optimize light absorption.

We emphasize that the beneficial effect of shading in the mitigation of water stress observed in the biochemistry of photosynthesis was not expressive on photochemistry, evaluated in this research by the chlorophyll *a* fluorescence characteristics, which are quantified to evaluate stressful cultivation conditions and possible damage to the photochemical apparatus. However, the activity of antioxidant enzymes increased with stress and, in general, shading favored the recovery of activity levels, where the reduction of activity was more significant.

Under water stress conditions, leaf area and chlorophyll contents in leaves can be reduced and thus reduce photosynthesis (Reis *et al*. 2020). The reduction in leaf area as a function of water restriction was more pronounced in seedlings of *O. arborea* than the chlorophyll index (Fig. 1b-c), and this reduction in leaf areas is a strategy that species presents to minimizes leaf transpiration and therefore maintains turgidity in the rest of the plant (Bento *et al*. 2016).

Similarly, reduction of leaf area in seedlings of *Alibertia edulis* Rich. (Jeromine *et al*. 2019), *Hymeneae courbaril* L. (Silva *et al*. 2016), *Euterpe oleracea* Mart. (Mar *et al*. 2013) was observed when these plants were cultivated under water restriction. The reduction of chlorophyll index and leaf area in *Calophyllum brasiliense* Cambess. under water restriction was pointed as an indicator of impairment to plant growth in these water conditions (Reis *et al*. 2020).

Plants under water restriction in P0 presented a *DQI* value below the one indicated in literature, which is 0.20 (Hunt 1990). In Rec, the plants irrigated and 70% shading presented *DQI* values well above the suggested minimum, evidencing better performance in biomass gain for the plants under these conditions, when compared to seedlings under 0% shading.

The photosynthetic rate was negatively influenced by water restriction in seedlings

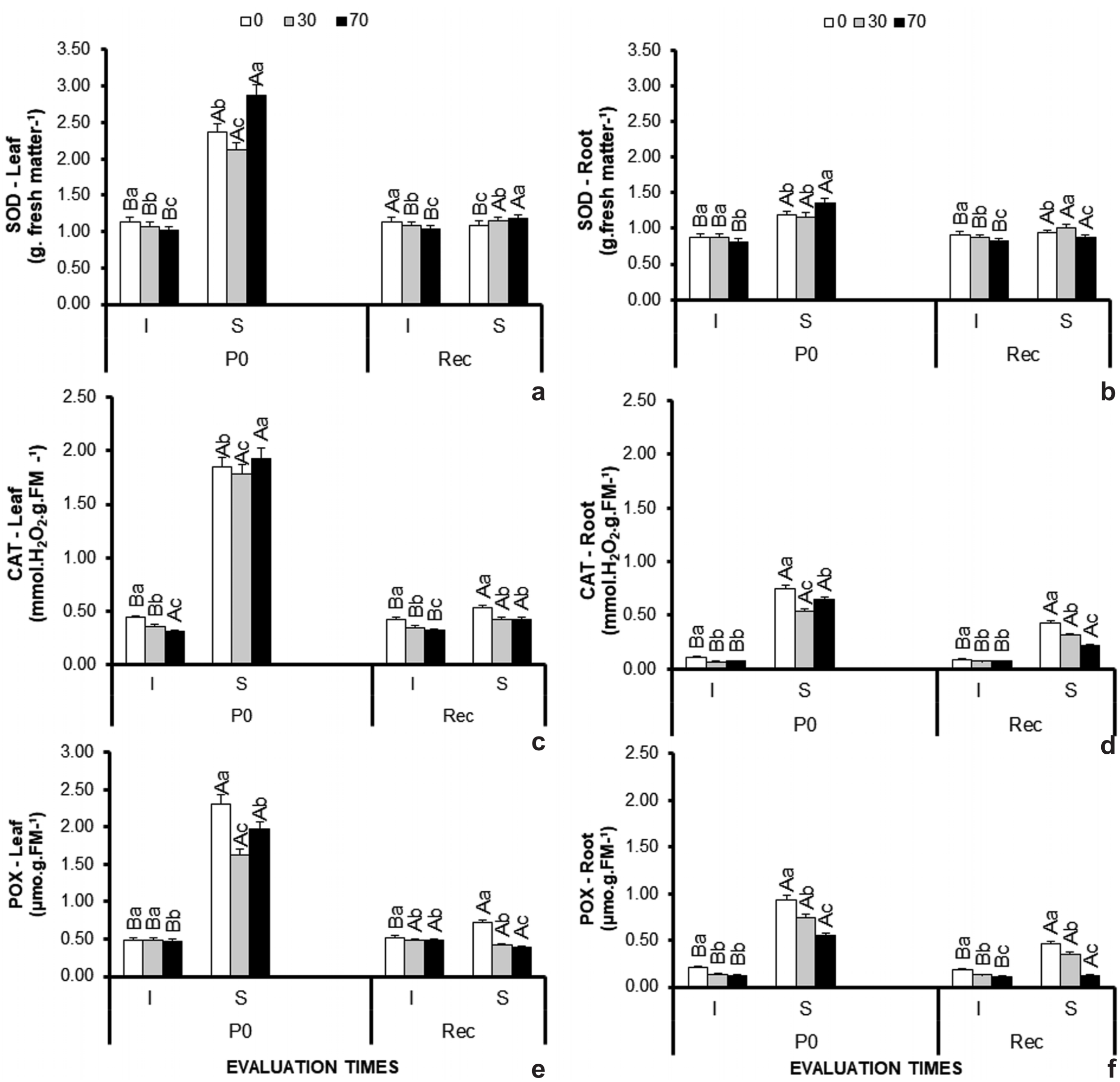

Figure 4 – a-f. *Ormosia arborea* seedlings under different water regimes: irrigated (I) and stressed (S) in different shadings levels (shading): 0%, 30% and 70% – a,b. mean values of CAT catalase enzyme activity – a. in leaf; b. in root; c,d. peroxidase enzyme POX – c. in leaf; d. in root; e,f. superoxide dismutase enzyme SOD – e. in leaf; f. in root. Uppercase letters compare the different water regimes in the same shading and lowercase letters compare the different shadings in the same water regime (Fig. b,c). Means followed by different letters indicate significant differences by Bonferroni *t* test for water regimes and Tukey test for shadings ($p < 0.05$) (n = 4).

under 0% shading, reaching values close to zero in less time (15 days) and also, the seedlings showed the largest number of days to recover (18 days), when compared to shaded seedlings, which showed 13 days. Variations in the recovery time of photosynthetic metabolism to previous water restriction were observed in the literature for different species such as *Campomanesia adamantium* (Cambess.) O. Berg. (Junglos *et al*. 2016) and *Hancornia speciosa* Gomes (Scalon *et al*. 2015) in which recovery was observed in approximately 4 days, and for *Khaya ivorensis* A. Chev. (Albuquerque *et al.* 2013) and *Campomanesia xanthocarpa* (Mart) O. Berg. (Bento *et al*. 2016), that needed 7 days to recover. However, we suggest that the potential for recovery varying between species and edaphoclimatic conditions.

The intercellular CO_2 concentration in plants under water restriction in P0 increased significantly, and in seedlings under 0% shading it was 3.7 times higher when compared to the other cultivation conditions, however, the values significantly reduced in Rec, which favored the increase of A/C_i values especially in seedlings under 70% shading. The increase of C_i on period of P0 in seedlings under water restriction is a consequence of the lower efficiency of carboxylation A/C_i and *gs*, as observed for other tree species such as *C. brasiliense* (Reis *et al.* 2018) *H. courbaril* (Freitas *et al*. 2018) *Schinus terebinthifolius* Raddi (Nunes *et al*. 2017).

Intrinsic and instantaneous efficiency of water use (*A*/*gs* and *A*/*E*, respectively) reduced when seedlings were stressed in P0, but recovered in the Rec, being higher in seedlings cultivated under 30% shading. The maintenance of *A* rate associated with lower values of *gs* and *E* are characteristics of plants tolerant to lower water availability in the soil, which leads to higher values of *A*/*gs* and *A*/*E* ratios (Ma *et al*. 2004).

We observed in literature that the behavior of these characteristics varies between species belonging to similar successional groups, suggesting that each species has a characteristic physiological plasticity that confers potential adjustment to different environmental conditions. Similarly, to that observed for *O. arborea*, seedlings of *Campomanesia xanthocarpa* Cambess. showed reduction of *A*, *gs* and A/C_i, when submitted to water restriction, however, shading minimized this reduction (Bartieres *et al.* 2020), evidencing the mitigating effect of shading on the stress caused by water restriction. We emphasize that when comparing transpiration and stomatal conductance in *Pterogyne nitens* Tul., *Myroxylon peruiferum* L. f. and *Aspidosperma polyneuron* Müll.Arg. under natural light conditions and different levels of irrigation, Tonello & Teixeira Filho (2012) observed that the greater increase in *E* and *gs* occurs with the increase of solar radiation, different from that observed for *O. arborea* in our study, but it decreases under reduction of water availability, similar to behavior of *O. arborea* seedlings.

The potential quantum efficiency of photosystem II (F_v/F_m) and the effective efficiency of the photosystem in the conversion of absorbed energy (F_v/F_0) decreased under conditions of low water availability, leading to an increase in photoinhibition processes (Lage-Pinto *et al*. 2012). However, the seedlings recovered F_v/F_m with the resumption of irrigation regardless of shading, with minimum values higher than 0.75. Values below these indicate a decline in PS II efficiency (Bolhàr-Nordenkampf *et al*. 1989; Baker & Rosenqvist 2004), expressing stressful condition. We emphasize that the values of F_v/F_0 were lower than 4.0, which is the limit for the cultivation condition to also be considered stressful (Zanandrea *et al*. 2006; Reis & Campostrini 2011), indicating possible damage to the electron transport chain (Azevedo *et al.* 2011). In Rec, higher values of F_v/F_0 were observed in the seedlings under 70% shading showing the recovery potential of *O. arborea* seedlings in this condition.

The maximum efficiency of non-photochemical process in photosystem II (F_0/F_m) increased in seedlings stressed by water restriction, however, in rec under 70% shading there was a reduction in values, which got closer to the maximum limit of 0.20, found in literature for plants in good cultivation conditions (Rohácek 2002). The other conditions suggest that the seedlings were still showing signs of stress and damage in their photochemical apparatus with significantly higher values.

The sensitivity of *O. arborea* seedlings to water restriction was confirmed by the increased activity of SOD, CAT and POX enzymes in P0 in both leaves and roots. However, the effect of shading on antioxidant activity varied according to each enzyme. During water restriction, the highest activity of SOD and CAT in leaves was observed under 70% shading, and for POX, the highest activity was under 0% shading. With reirrigation, the activity of enzymes evaluated in the Rec reduced in both organs and was statistically lower in seedlings under shading levels, especially under 70% shading, excluding SOD activity in leaves, which was lower under 0% shading.

The efficiency of the antioxidant system of the plant increases according to its tolerance to the effects caused by reactive oxygen species (ROS) (Giannakoula *et al.* 2010). In stressful situations, plants can modulate responses according to the increase in protein expression level and activity of antioxidant enzymes such as CAT, POX and SOD (Lima *et al*. 2002). These enzymes have antioxidant properties, acting on reduced organic substrates and protecting the cell from the harmful effect of ROS in plants subjected to stress (Sofo *et al.* 2015).

The increase in SOD activity in seedlings under 70% shading may justify the better physiological performance of these seedlings, since SOD is considered the first line of defense against the toxic effects of ROS (Gill &Tuteja 2010; Amador *et al.* 2012). After dismutation of superoxide into hydrogen peroxide, intracellular levels of H_2O_2 formed by SOD need to be eliminated, activity responsible by POX and CAT because they present high affinity with H_2O_2 (Locato *et al*. 2010; Barbosa *et al.* 2014). This relationship is proven by the increase of CAT and POX enzymes in *O. arborea* seedlings submitted to water restriction. However, we emphasize that oxygen reactive species and the products of the action of these enzymes were not quantified in this research, suggesting that further studies should be conducted to better know this type of antioxidant defense in *O. arborea*.

Similarly, the activity of antioxidant enzymes increased both in the aerial parts of the plants and their roots, in response to water restriction in *Schinus terebinthifolius* Raddi seedlings (Nunes *et al*. 2017). They were also observed in seedlings of *C. brasiliense* seedlings (Reis *et al*. 2018), *Copaifera langsdorffii* Desf. (Rosa *et al.* 2017).

The results observed in this research allow us to corroborate information from the literature regarding the sciophile classification of *O. arborea* and its sensitivity to water restriction, in addition, they also suggest that it is a species that can be used in projects of restoration of degraded areas. The species can undergo some periods of water restriction, but need some degree of shading, found in mature and secondary forests, agroforestry systems, or plantations for restoration. Thus, the planting of *O. arborea* in these different silvicultural systems would be more sustainable compared to open fields especially to alleviate the effects of drought stress during the initial establishment of plants.

Water restriction reduced photosynthetic metabolism, growth and quality of *O. arborea* seedlings by increasing the activity of antioxidant enzymes. The seedlings present higher quality (DQI) and gas exchange (*gs*, A/C_i and *E*) when cultivated under 70% shading in Rec. The stressful effect of water restriction was mitigated by the shading of 70%, where the seedlings were able to recover, resuming their photosynthetic metabolism and quality after resumption of irrigation. In addition, 0% shading is not indicated for this species.

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References

Albuquerque MPF, Moraes FKC, Santos RIN, Castro GLS, Ramos EMLS & Pinheiro HÁ (2013) Ecofisiologia de plantas jovens de mogno-africano submetidas a déficit hídrico e reidratação. Pesquisa Agropecuária Brasileira 48: 9- 16.

Amador ML, Sancho S, Bielsa B, Gomes-Aparisi J & Rubiocubetas MJ (2012) Physiological and biochemical parameters controlling waterlogging stress tolerance in Prunus before and after drainage. Physiolgiae Plantarum 144: 357-368.

Araújo SAC & Deminicis BB (2009) Fotoinibição da fotossíntese. Revista Brasileira de Biociencia 7: 463-472.

Azevedo NAD, Pereira PPA, Costa DP & Santos ACC (2011) Fluorescência da clorofila como uma ferramenta possível para seleção de tolerância à salinidade em girassol. Revista Ciência Agronômica 42: 893-897.

Baker NR & Rosenqvist E (2004) Applications of chlorophyll fluorescence can improve crop production strategies: an examination of future possibilities. Journal of Experimental Botany 55: 1607-1621.

Barbosa MR, Silva MMA, Willadino L, Ulisses C & Camara TR (2014) Geração e desintoxicação enzimática de espécies reativas de oxigênio em plantas. Ciência Rural 44: 379-390.

Bartieres EMM, Scalon SDPQ, Dresch DM, Cardoso EAS, Jesus MV & Pereira ZV (2020) Shading as a means of mitigating water deficit in seedlings of *Campomanesia xanthocarpa* (Mart.) O. Berg. Notulae Botanicae Horticultural Agrobotanici Cluj-Napoca 48: 234-244.

Bastos SS, Scalon SPQ, Souza APR, Dresch DM, Junglos FS, Scalon LQ & Mussury RM (2022) Photosynthetic metabolism and antioxidant in *Ormosia arborea* are modulated by abscisic acid under water deficit? Brazilian Journal of Biology 82: e244331.

Bento LF, Scalon SPQ, Dresch DM & Pereira ZV (2016) Potential of recovery of *Campomanesia xanthocarpa* Mart. *ex* O. Berg seedlings from water deficit. African Journal of Agricultural Research 11: 2775-2785.

Bolhàr-Nordenkampf HR, Long SP & Baker NR (1989) Chlorophyll fluorescence as probe of the photosynthetic competence of leaves in the field: a review of current instrument. Functional Ecology 3: 497-514.

Broetto F (2014) Métodos de trabalho em bioquímica vegetal e tecnologia de enzimas. Cultura Acadêmica, Botucatu. 92p.

Cardoso DBOS & Meireles JE (2015) *Ormosia*. *In* List of Species of Flora of Brazil. Botanical Garden of Rio de Janeiro. Available at <http://floradobrasil2015.jbrj.gov.br/FB29802>.

Carvalho PER (2008) Espécies Arbóreas Brasileiras. Embrapa Informação Tecnológica, Brasília. Pp. 211-215.

Costa AS, Freire ALO, Bakke IA & Pereira FHF (2015) Respostas fisiológicas e bioquímicas de plantas de aroeira (*Myracroduon urundeuva* Allemão) ao déficit hídrico e posterior recuperação. Irriga 20: 705-717.

Dickson A, Leaf AL & Hosner JF (1960) Quality appraisal of white spruce and white pine seedling stock in nurseries. Forest Chronicle 36: 10-13.

Duarte MR, Dranka ERK & Yano M (2012) Leaf Microscopic Characters of *Ormosia arborea* (Vell.) Harms, Fabaceae. Latin American Journal of Pharmacy 31: 526-530.

Ferreira DF (2014) Sisvar: a guide for its Bootstrap procedures in multiple comparisons. Ciência e Agrotecnologia 8: 109-112.

Freitas VMB, Scalon SPQ, Dresch DM, Bastos SS & Souza APR (2018) Influence of exogenous application of abscisic acid on gas exchanges in *Hymenaea courbaril* L. (Fabaceae) seedlings subjected to water deficit. Floresta 48: 163-172.

Giannakoula A, Moustakas M, Syros T & Yupsanis T (2010) Aluminum stress induces up-regulation of an efficient antioxidant system in the Al-tolerant maize line but not in the Al sensitive line. Environmental Experimental Botany 67: 487-494.

Gill SS & Tuteja N (2010) Reactive oxygen species and antioxidant machinery in abiotic stress tolerance in crop plants. Plant Physiol Biochem 48: 909-930.

Hu L, Wang Z & Huang B (2010) Diffusion limitations and metabolic factors associated with inhibition and recovery of photosynthesis from drought stress in a C perennial grass species. Physiologiae Plantarum 139: 93-106.

Hunt GA (1990) Effect of styroblock design and cooper treatment on morphology of conifer seedlings. *In*: Rose R, Campbell SJ & Landis TD (1990) Target seedling symposium, meeting of the western forest nursery associations, general technichal report RM-200. Roseburg: Proceedings. United States Departament of Agriculture, Forest Service, Collins. Pp. 218-222.

Jeromine TS, Mota LHS, Scalon SPQ, Dresch DM & SCalon L (2019) Efeitos do substrato e da disponibilidade de água no crescimento inicial de *Alibertia edulis* Rich. Floresta 49: 89-98.

Junglos FS, Junglos MS, Dresch DM, Pereira NS, Kodama FM & Scalon SPQ (2016) Recovery of the photosynthetic capacity of *Campomanesia adamantium* (Myrtaceae) after water deficit. Brazilian Journal of Botany 39: 541-546.

Lage-Pinto F, Bernini E, Oliveira JGD & Vitoria AP (2012) Photosynthetic analyses of two native Atlantic Forest species in regenerative understory of eucalyptus plantation. Brazilian Journal of Plant Physiology 24: 95-106.

Lázaro-Nogal A, Matesanz S, Godoy A, Pérez-Trautman F, Gianoli E & Valladares F (2015) Environmental heterogeneity leads to higher plasticity in dry-edge populations of a semiarid Chilean shrub: insights into climate change responses. Journal of Ecology 103: 338-350.

Lima ALS, Damatta FM, Pinheiro HA, Totola MR & Loureiro ME (2002) Photochemical responses and oxidative stress in two clones of *Coffea canephora* under water deficit conditions. Environmental Experimental of Botany 47: 239-247.

Locato V, Pinto MC, Paradiso A & Gara L (2010) Reactive oxygen species and ascorbateglutathione interplay in signaling and stress responses. *In*: Gupta SD (ed.) Reactive oxygen species and antioxidants in higher plants. Science Publishers, Enfield. Pp. 45-64.

Ma CC, Gao YB, Guo HY & Wang JL (2004) Photosynthesis, transpiration, and water use efficiency of *Caragana microphylla*, *C. intermedia*, and *C. korshinskii*. Photosynthetica 42: 65-70.

Mar CC, Conceição HEO, Santos ABR, Viégas IJM & Silva FSN (2013) Produção de assa seca e área foliar de açaizeiro sob déficit hídrico. Revista Agroecologia 5: 14-23.

Martinazzo EG, Anese S, Wandscheer ACD & Pastorini LH (2007) Efeito do sombreamento no crescimento inicial e teor de clorofila foliar de *Eugenia uniflora*

Linn (Pitanga) - Família Myrtaceae. Brazilian Journal Biological Science 5: 162-164.

Nunes DP, Scalon SPQ, Dresch DM & Gordin CRB (2017) Photosynthetic and enzymatic metabolism of *Schinus terebinthifolius* Raddi seedlings under water déficit. Ciência e Agrotecnologia 41: 676-682.

Pick-Upau (2012) Avaliação da germinação e superação de dormência de sementes de olho-de-cabra (*Ormosia arborea* (Vell.) Harms) em condições de viveiro aliado à cultura indígena Guarani. Darwin Society Magazine 2: 1-27.

Reis FO & Campostrini E (2011) Microaspersão de água sobre a copa: um estudo relacionado às trocas gasosas e à eficiência fotoquímica em plantas de mamoeiro. Revista Brasileira de Agrociência 17: 284-295.

Reis LC, Foresti AC, Scalon SDPQ, Dresch DM & Pereira ZV (2018) Effect of water deficit and abscisic acid on photosynthetic and antioxidant metabolism in seedlings of *Calophyllum brasiliense* (Cambess.). Cerne 24: 387-396.

Reis LC, Scalon, SPQ, Dresch DM, Foresti AC, Santos CC & Pereira ZV (2020) Chlorophyll a fluorescence as an indicator of water stress in *Calophyllum brasiliense*. Notulae Botanicae Horticultural Agrobotanici Cluj-Napoca 48: 210-220.

Ressel K, Guilherme FAG, Schiavini I & Oliveira PE (2004) Ecologia morfofuncional de plântulas de espécies arbóreas da Estação Ecológica do Panga, Uberlândia, Minas Gerais. Brazilian Journal of Botany 27: 311-323.

Rohácek K (2002) Chlorophyll fluorescence parameters: the definitions, photosynthetic meaning and mutual relationships. Photosynthetica 40: 13-29.

Rosa DB, Scalon SDPQ, Cremon T, Ceccon F & Dresch DM (2017) Gas exchange and antioxidant activity in seedlings of *Copaifera langsdorffii* Desf. under different water conditions. Anais da Academia Brasileira de Ciências 89: 3039-3050.

Scalon SPQ, Kodama FM, Dresch DM, Mussury RM & Pereira ZV (2015) Gas exchange and photosynthetic activity in *Hancornia speciosa* Gomes seedlings under water deficit conditions and during rehydration. Bioscience 31: 1124-1132.

Silva CA, Dourado Neto D, Silva CJ & Freitas CA (2016) Desenvolvimento de mudas de *Hymenaea courbaril* em função de recipientes e lâminas de irrigação. Revista Arvore 40: 487-498.

Sofo A, Scopa A, Nuzzaci M & Vitti A (2015) Ascorbate peroxidase and catalase activities and their genetic regulation in plants subjected to drought and salinity stresses. International Journal Molecular Science 16: 13561-13578.

Souza CC, Oliveira FA, Silva IF & Amorim Neto MS (2000) Avaliação de métodos de determinação de água disponível e manejo da irrigação em terra roxa sob cultivo de algodoeiro herbáceo. Revista Brasileira de Engenharia Agrícola e Ambiental 4: 338-342.

Tonello KC & Teixeira Filho J (2012) Ecofisiologia de três espécies arbóreas nativas da Mata Atlântica do Brasil em diferentes regimes de água. Irriga 17: 78-85.

Zanandrea I, Nassi FDL, Turchetto AC, Braga EJB, Peters JA & Bacarin MA (2006) Efeito da salinidade sob parâmetros de fluorescência em *Phaseolus vulgaris*. Revista Brasileira de Agrociência 12: 157-161.

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