

# Arbuscular mycorrhizal fungi and water stress on the physiology and quality of *Parkia platycephalic* benth

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# SILVICULTURE

#### **ABSTRACT**

**Background:** The water deficit characteristic of semi-arid regions can cause negative changes in plant physiology. Arbuscular mycorrhizal fungi can meet water needs by associating with plant roots, improving seedling quality. The physiological parameters and the quality of the seedlings of Parkia platycephala Benth. were evaluated as a result of the association of isolated strains of Gigaspora rosea, *Gigaspora margarita*, *Acaulospora koskei*, *Acaulospora morrowiae*, intercalated strains (a mixture of the four strains mentioned above) and a control (without inoculation) under two conditions of water regime (with and without stress).

**Results:** Artificial inoculation of AMF is a promising management technique in the production of *P. platycephala* Benth seedlings for improving the physiological state, increasing seedling quality, and drought resilience. Under restriction water restriction conditions, the seedlings showed lower values of net CO<sub>2</sub> assimilation, leaf transpiration, stomatal conductance, and higher values of internal CO2 concentration. Under these conditions, the *G. rosea* strain stood out with the lowest rate of net CO<sub>2</sub> assimilation (A), leaf transpiration (E), and stomatal conductance (gs) with increases of 34, 49, and 70 %, respectively, compared to the control.

**Conclusion:** Stomatal closure is a strategy used by *Parkia platycephala* Benth. seedlings to prevent excessive water loss, and the Gigaspora rosea strain is responsible for mediating the best physiological conditions and better quality seedlings.

Keywords: Gas exchange; quality index; seedling production; water deficit.

#### **HIGHLIGHTS**

Arbuscular mycorrhizal fungi mitigate water stress damage in Parkia platycephala Benth. The *Gigaspora rosea* strain was excellent at improving gas exchange.

The Gigaspora rósea strain favors greater water use efficiency in plants under a water deficit The Gigaspora rosea and Gigaspora margarita strains promote a higher Dickson quality index.

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#### INTRODUCTION

The genus *Parkia* (Leguminosae) has a pantropical distribution and consists of 34 species. Its occurrence in the paleotropics has been recorded in Africa, Asia, Madagascar, and the Indo-Pacific region. In the Neotropical region, this genus occurs from Honduras to southeastern Brazil, with emphasis on the Amazon rainforest given the remarkable morphological and taxonomic diversity, encompassing 17 species with large, globose, biglobose, clavate, or subspherical inflorescences adapted for pollination, which is performed by bats. In addition, the flowers of these species have an imbricated prefloration (Hopkins, 1986), and these plants have significant socioeconomic value as they are used in food and herbal medicines (Octasari and Widyastani 2021).

Parkia platycephala Benth is one of the species that occurs widely outside the Amazon rainforest (Hopkins, 1986). In Northeast Brazil, it occurs in transition areas between the Cerrado (Brazilian savanna) or the Atlantic Forest and the Caatinga, standing out for its timber and landscape potential (Oliveira et al. 2021). Additionally, it has a significant potential for use in agricultural, forestry, and pasture systems due to its plasticity, rapid growth, and use as forage for ruminant feeding (Alves et al., 2007). This characteristic is especially relevant in regions where herds suffer from food scarcity at certain times of the year (Santos et al., 2017).

Approximately 60% of northeastern Brazil is characterized as semi-arid due to its climatic conditions represented by intense insolation, relatively high temperatures, and a rainfall regime marked by rainfall scarcity, irregularity, and concentration in a short period, mainly during three months of the year (Moura et al., 2007). The effects of climatic conditions on the survival of plant species cultivated in semi-arid regions are extremely important. Therefore, it is necessary to know the factors that limit the initial development of the species, especially abiotic factors such as water deficit, which affects the initial plant development. Furthermore, plant responses under artificial stress conditions are essential for ecophysiology and allow assessing the limits and the survival and adaptation of these species to natural stress conditions (Peguero-Pina et al., 2020; Lacerda and Mapeli, 2021; Leite et al., 2022).

The use of microbiological technologies in seedling production, especially those involving functional groups, e.g., arbuscular mycorrhizal fungi (AMFs), could be a viable environmental management tool (Costa et al., 2019). Combined with mycorrhizal roots, these fungi have one of the oldest and most widespread symbiotic relations known (Carneiro et al., 1998; Shi et al., 2023), with AMFs colonizing the roots of host plants to obtain sugars. In turn, the fungi increase the plant's nutrient absorption, tolerance to root pathogens, and drought resistance through a thin network of extra-root mycelia that spread from colonized roots to the soil (Smith and Read, 2010).

Previous studies have observed that AMFs significantly increased the drought tolerance of host plants through various mechanisms, e.g., direct water absorption by extraradicular mycorrhizal hyphae at a rate of 375-760 nL water/h, contributing up to 20% of the total water content

water absorbed by roots (Ruth et al., 2011) and improving the osmotic adjustment (Wu et al., 2017; Tisarum et al., 2022).

From this perspective, under the hypothesis that mycorrhizal symbiosis improves the growth and photosynthetic metabolism of *P. platycephala* Benth, contributing to its adaptation to semi-arid regions, this study aimed to study the effect of AMF inoculation on the physiology and quality of *P. platycephala* Benth subjected to water stress. This was achieved by measuring the gas exchange and quality index of seedlings developed aiming to help them adapt to regions with water restrictions.

# **MATERIAL AND METHODS**

# Study area and experiment

The experiment was conducted in a plant nursery at the Federal University of Piauí/ Campus Professora Cinobelina Elvas (UFPI/CPCE), Piauí, Brazil, at the following geographical coordinates: 09° 04′ S, 44° 21′ W, and at a mean elevation of 277 m. The study area is located in the Brazilian semi-arid region, with a climate defined as tropical hot and humid and classified as *Aw* according to Köppen, with two well-defined seasons: a dry season between May and October, and a rainy season from November to April (Alvares et al., 2013). The experiment was conducted between October 2017 and February 2018.

The experimental design was in randomized blocks and set up in a 6 x 2 factorial arrangement corresponding to six inoculation treatments and two water regimes (with and without stress) with three replicates. The treatments consisted of the isolated strains *Gigaspora rosea* SPL 101A, *Gigaspora margarita* SCT 077A, *Acaulospora koskei* SCT 042A, and *Acaulospora morrowiae* SCT 048B, intercropped strains (mix of the four strains previously mentioned), and one control (without inoculation).

Planting was carried out manually using five seeds per pot. Inoculation was performed with 5 g of AMF of the four individual strains and 1.25 g of AMF of each strain for intercropping (Mix). After full emergence, the plants were thinned to two seedlings per experimental unit.

The arbuscular mycorrhizal fungal strains (AMFs) were provided by the International Glomeromycota Culture Collection (CICG) of the Regional University of Blumenau (FURB), Blumenau, Santa Catarina, Brazil.

The soil used in the experiment was classified as Typical Ortic Quartzarenic Neosol according to the Brazilian Soil Classification System (Santos et al., 2013). For physicochemical characterization, samples were collected from the 0-20 cm depth layer, and the analyses were performed according to the methodology described by Teixeira et al. (2017). The mean values of the soil attributes are shown in Table 1.

After being air-dried, ground, and sieved through a 2-mm sieve, the soil samples were packed in fiber bags to be autoclaved at 121 °C and 1 atm for 30 minutes. Black PVC pots with a volume of 3 L and perforated at the bottom were used for planting. Each pot received 400 g of rock fragments simulating the water flow through the soil and was then covered with tissue, after which 3 kg of autoclaved soil was

added. Dolomitic limestone was incorporated to increase the base saturation of the soil to 50% thirty days before sowing, according to the recommendations of Sousa and Lobato (2004b). No chemical fertilization was performed.

The seeds of *Parkia platycephala* Benth used in the experiment were donated by the Laboratory of Ecophysiology of UFPI/CPCE. A total of 360 seeds free of pathogens and injuries were selected, subjected to mechanical scarification to overcome dormancy, and treated with 5% sodium hypochlorite, according to procedures described by Silva et al. (2017).

Planting was performed manually in October 2017 using five seeds per pot. During sowing, 5 g of the AMFs of the four individual strains and 1.25 g of each strain were inoculated into the intercrop (mix). After complete emergence, 30 days after sowing (DAS), the seedlings were thinned to two plants per experimental unit.

Soil moisture was maintained at approximately 60% of pot capacity (PC). The PC was determined by the water content removed from the soil after saturation and by capillarity until drainage ceased. The pots were irrigated in a controlled manner by periodically weighing the containers using a precision balance with a capacity of 10 kg, followed by the replacement of the water loss by evapotranspiration in the period, maintaining the pots close to PC (60%).

The water stress treatments underwent three periods of irrigation suspension beginning 35, 70, and 105 days after sowing (DAS). Irrigation was interrupted until leaf curling, i.e., the irrigation suspension periods occurred at different intervals, with a twelve-day interval at 35 DAS, a nine-day interval at 70 DAS, and a seven-day interval at 105 DAS. After some time, the water supply to non-irrigated plants was resumed until reaching 60% PC (rehydration).

# **Gas exchange measurements**

The gas exchange evaluations in *P. platycephala* Benth were performed at 120 DAS. One plant (the most developed) was chosen in each pot to determine the data on the net  $CO_2$  assimilation rate (A), leaf transpiration (E), stomatal conductance (gs), internal  $CO_2$  concentration (Ci), water-use efficiency (WUE = A/E), intrinsic water-use efficiency (WUEi = A/gs), instantaneous carboxylation efficiency (ICE = A/Ci), the ratio of the internal to external  $CO_2$  concentration (Ci/Ca), and leaf temperature (Tleaf).

An infrared gas analyzer (Infrared Gas Analyzer-IRGA, model GFS-3000, Heinz Walz, Germany) was used in the experiment. The measurements were made between

9:51 a.m. and 11:09 a.m. using the third youngest leaflet of each plant. The parameters of air temperature (Tair), relative air humidity (RHair), and photosynthetically active radiation (PAR) inside the greenhouse were also determined during the evaluations. The vapor pressure deficit (VPD) was calculated based on the values of Tair and RHair for the same times, according to Tetens (1930).

# **Seedling quality index**

The seedling quality index was determined according to Dickson et al. (1960) by considering the shoot, root, and total dry mass indicators. The height and stem diameter of the seedlings (Porto et al., 2020) were determined using the following formula: DQI = TDM/(H/ND)+(SDM/ RDM), where DQI = Dickson quality index; TDM = total dry mass (g); SDM = shoot dry mass (g); RDM = root dry mass (g); H = height of the shoot part (cm); ND = neck diameter (mm).

# Statistical analysis

The data were subjected to the F-test by analysis of variance (ANOVA). Whenever the means were significant, they were subjected to the Scott-Knott test at 5% of probability. Simultaneously, with the statistical analyses, the basic assumptions of the ANOVA, normality of errors, and homogeneity of variances were tested for all variables analyzed. Next, Pearson's correlation analysis ( $p \le 0.05$ ) was performed between the physiological parameters and the DQI under different water conditions. All procedures were performed using the R software (R Core Team, 2016).

#### **RESULTS**

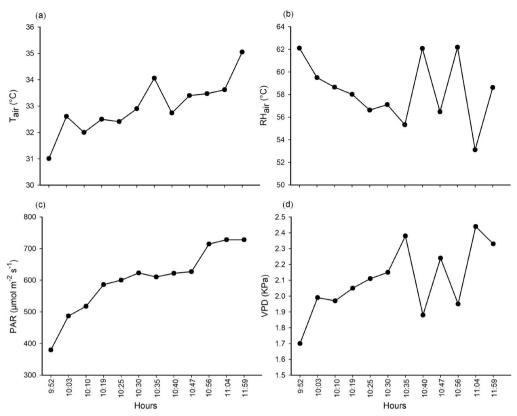
# Microclimate in the plant nursery during the gas exchange evaluations

During the evaluations of the physiological parameters in the plant nursery, the mean air temperature ranged from 31.01 to 35.05 °C, with the highest value occurring at 11:09 a.m., close to the times of greatest solar radiation (Figure 1a). The mean relative air humidity ranged from 53.11 to 62.19 % (Figure 1b), and the photosynthetically active radiation ranged from 379.50 to 728.00  $\mu$ mol m  $^{-2}$  s  $^{-1}$ , with the highest mean values recorded at 11:04 a.m. and 11:09 a.m. (Figure 1c) , respectively. The vapor.

Table 1: Physicochemical characterization of the typical Ortic Quartzarenic Neosol used in the experiment.

рН	H+AI	Al	Ca	Mg	Ca + Mg	K	SB	CEC	Р
H <sub>2</sub> O				cmc	ol <sub>c</sub> dm <sup>-3</sup> ———				mg dm <sup>-3</sup>
4.6	3.47	0.2	0.53	0.13	0.66	0.1	0.77	4.23	1.75
Cu	Fe	Mn	Zn	V	m	SOM	Clay	Silt	Sand
_	mg (	dm-3			%	_	g l	kg <sup>-1</sup>	
0.97	85.37	7.13	1.75	18.1	20.7	6	69	11	920

SB = sum of bases; CEC = cation-exchange capacity; V = saturation of bases; m = saturation of aluminum; SOM = soil organic matter.



**Figure 1:** Growing conditions inside the greenhouse during gas exchange evaluations in Parkia platycephala Benth. seedlings inoculated with different arbuscular mycorrhizal fungi in management with and without stress. Air temperature - T<sub>air</sub> (a), relative air humidity - RH<sub>air</sub> (b), photosynthetically active radiation – PAR (c) and vapor pressure deficit - VPD (d).

Pressure deficit between leaf and air ranged from 1.70 kPa to 2.44 kPa (Figure 1d). The highest value recorded for VPD culminates with the lowest RH value and the highest PAR. The plants generally showed greater transpiration when approaching 11 a.m., when the air temperatures and solar radiation were higher and the relative humidity was lower.

#### **Gas exchange and Quality index**

The analysis of variance of the data obtained during the stress period showed a significant effect of the single factors (except for the Ci of AMF and Tleaf under stress) and the interaction between the water regime and arbuscular mycorrhizal fungi on the physiological quality of *Parkia platycephala* Benth. (Table 2).

Under water restriction conditions, the seedlings showed lower values of net  $\mathrm{CO}_2$  assimilation (A), leaf transpiration (E), and stomatal conductance (gs) and higher values of internal  $\mathrm{CO}_2$  concentration (Ci) (Figure 2). Furthermore, under water stress conditions, the *G. rosea* strain stood out with the lowest rate of net  $\mathrm{CO}_2$  assimilation (A), leaf transpiration (E), and stomatal conductance (gs) with increases of 34, 49, and 70 %, respectively, compared to the control.

Better values of net  ${\rm CO_2}$  assimilation (A), leaf transpiration (E), and stomatal conductance (gs) were obtained when using the *A. morrowiae* strain without

stress conditions. High gs and E values suggest higher water consumption by the plant when this strain is used. Except for the treatment using *A. koskei*, the internal CO<sub>2</sub> concentration (Ci) in plants under water deficit was higher than in irrigated plants. The irrigated treatments with *A. koskei* and *G. rosea* differed from the control, showing the highest means. There was no variation between treatments in this parameter under water stress.

For the water-use efficiency (WUE), the highest means were observed in the control treatment for both conditions (Figure 3a). In the non-stressed condition, the highest mean of this parameter was observed when using the fungus *G. rosea*. The plants under stress did not differ from the control.

With regard to the intrinsic water-use efficiency (WUEi) the treatments using *A. morrowiae*, *G. margarita*, and the mix of strains showed differences in relation to the water conditions. The control treatment had the highest mean when subjected to water stress (Figure 3b). In contrast, without water stress, the plants showed no differences. The treatments using the fungi *A. koskei*, *G. rosea*, and the strain mix did not differ in relation to the water conditions of the present study.

The instantaneous carboxylation efficiency (ICE) was lower in seedlings that underwent water restriction (Figure 3c). Under water stress using *G. rosea, G. margarita*, and the mix, the plants obtained better results than the other treatments. Except for the *A. koskei* strain, the seedlings

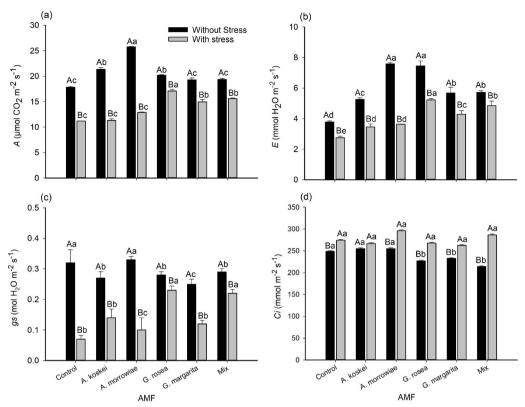
under stress showed a higher ratio of internal to external  ${\rm CO}_2$  concentration (Ci/Ca) in relation to well-watered treatments (Figure 3d), and stress conditions resulted in no differences between treatments. The leaf temperature (Tleaf) varied in the water conditions studied and in the different treatments for each condition (Figure 3e). Without water restriction, the treatment with the mix of strains showed a lower value for this parameter. Under stress conditions, the treatments with *A. koskei, A. morrowiae*, and *G. rosea* had the lowest temperatures.

Except for the control, the Dickson quality index (DQI) was lower with water restriction (Figure 3f). However, even with no significant differences from the control, there was a higher mean of this parameter for the no-stress condition. The treatments in the no-stress condition showed different variations, with the highest and lowest averages being found in the *A. morrowiae* and control treatments, respectively. When under stress, the seedlings using *G. rosea* and *G. margarita* obtained better results.

**Table 2:** Mean square and significance of the analysis of variance of physiological parameters and seedling quality in *Parkia platycephala* Benth. submitted to inoculation of different arbuscular mycorrhizal fungi (AMF), in the managements without water stress and with water stress.

		Mean	square		
SV	А	Е	gs	Ci	WUE
AMF	17.64***	6.801***	0.005620***	728.5ns	1.34428***
Stress	415.41***	31.659***	0.184900***	12197.0***	0.16538*
AMF x Stress	22.82***	1.928***	0.010740***	629.9*	0.38687***
CV AMF (%)	5.60	2.32	7.21	7.51	6.10
CV Stress (%)	5.60	2.97	9.06	5.05	5.87
		Mean	stress		
Without stress	20.63	5.91	0.29	238.76	3.61
With stress	13.83	4.03	0.15	275.57	3.48
		Mean	s AMF		
Control	14.51	3.26	0.19	261.51	4.39
A. koskei	16.32	4.35	0.21	260.93	3.67
A. morrowiae	19.32	5.61	0.22	275.49	3.48
G. rósea	18.64	6.34	0.26	247.22	2.99
G. margarita	17.11	4.98	0.19	247.60	3.44
Mix	17.48	5.29	0.25	250.24	3.30
SV	WUEi	ICE	Ci/Ca	Tleaf	DQI
AMF	1936.1***	0.000278**	0.005291	1.6609***	0.000764***
Stress	11263.2***	0.011378***	0.07111***	0.0006 <sup>ns</sup>	0.008711***
AMF x Stress	2949.2***	0.000238**	0.005084**	5.9714***	0.000798***
CV AMF (%)	10.97	9.86	7.38	0.73	5.71
CV Stress (%)	12.48	7.65	4.55	1.23	5.29
		Mean	stress		
Without stress	71.57	0.09	0.60	33.73	0.09
With stress	106.95	0.05	0.69	33.74	0.06
		Mean	s AMF		
Control	113.70	0.06	0.67	34.38	0.06
A. koskei	78.86	0.06	0.64	33.70	0.07
A. morrowiae	100.19	0.07	0.68	33.37	0.09
G. rósea	72.79	0.08	0.61	33.29	0.08
G. margarita	100.51	0.07	0.61	34.40	0.08
Mix	69.50	0.07	0.63	33.30	0.07

Rate of net  $CO_2$  assimilation (A,  $\mu$ mol de  $CO_2$   $m^2$   $s^{-1}$ ), leaf transpiration (*E*, mmol de  $H_2O$   $m^2$   $s^{-1}$ ), stomatal conductance (*gs*, mol de  $H_2O$   $m^2$   $s^{-1}$ ), internal  $CO_2$  concentration (Ci,  $\mu$ mol  $m^2$   $s^{-1}$ ), water use efficiency (WUE,  $\mu$ mol de  $CO_2$   $m^2$   $s^{-1}$ / mmol de  $H_2O$   $m^2$   $s^{-1}$ ), intrinsic water use efficiency (WUEi,  $\mu$ mol de  $CO_2$   $m^2$   $s^{-1}$ / mol de  $H_2O$   $m^2$   $s^{-1}$ ), instantaneous carboxylation efficiency (ICE,  $\mu$ mol  $m^2$   $s^{-1}$ ), ratio between internal and external  $CO_2$  concentrations (Ci/Ca,  $\mu$ mol  $m^2$   $s^{-1}$ /  $\mu$ mol  $m^2$   $s^{-1}$ ), leaf temperature (Tleaf,  $^{\circ}$ C) and Dickson quality index (DQI).  $^{*}$ ,  $^{**}$ ,  $^{***}$  and ns: significant at the 5, 1, 0.1 % and not significant, respectively.



**Figure 2:** Evaluation of the physiological parameters of *Parkia platycephala* Benth. seedlings inoculated with different arbuscular mycorrhizal fungi in management with and without stress. Net CO<sub>2</sub> assimilation rate – A (a), leaf transpiration – E (b), stomatal conductance - gs (c), and internal CO<sub>2</sub> concentration – Ci (d). Different lowercase letters indicate a significant difference between the AMF in each water regime and different capital letters indicate a significant difference between the water regimes, within each AMF, by the Scott-Knott test at 5% significance.

# Correlation between the gas exchange parameters and the Dickson quality index

Pearson's correlations for the variables representing gas exchange and DQI of *Parkia platycephalic* Benth plants grown under different water conditions are presented in the correlograms in Figure 4, and the correlations ranged from moderate to very strong for both conditions. In the non-stressed condition (4a), parameter A showed positive correlations with E (p < 0.01), WUEi (p < 0.05), and ICE (p < 0.01).

In contrast, in the stress condition, the same parameter was positively correlated with E (p < 0.01), gs (p < 0.01), ICE (p < 0.01), and Tleaf (p < 0.05) (Figure 4b). In both conditions, parameter E correlated WUE and WUEi, with a positive correlation without stress (p < 0.01) and a negative correlation with stress (p < 0.01), beyond ICE (p < 0.01). However, only in the stress condition did E have a very strong correlation with gs. When under stress, the gs obtained a strong correlation with WUE, WUEi and ICE, all presenting a p < 0.01. Without stress, there was only a moderate and negative correlation with WUEi. Such results demonstrate the variation of the effects caused on the physiology of plants cultivated under different water conditions, mainly influencing plant growth.

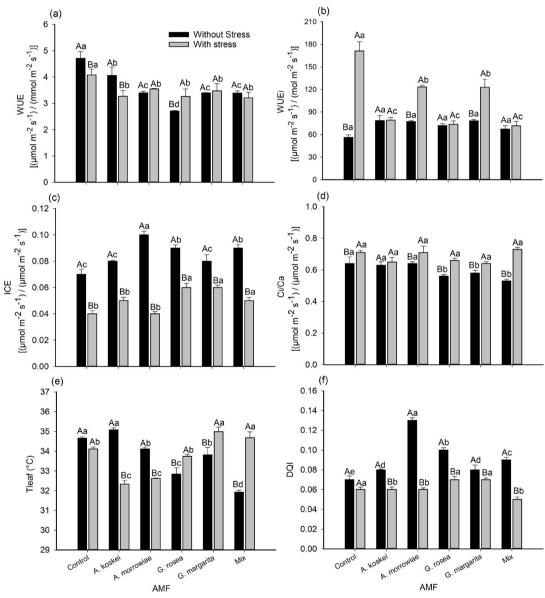
### DISCUSSION

It is well-known that water stress (drought) is one of the more critical aspects of plant cultivation in the semiarid region. However, strategies to mitigate deleterious effects are being developed, and the use of AMFs stands out as a promising option. In the present study, this strategy was adopted in young plants of *P. platycephala* Benth with positive effects, resulting in the acceptance of the hypothesis initially proposed. Due to the atmospheric conditions of the plant nursery, the increase in transpiration (Figure 2) could be related to the higher DPV. This behavior agrees with the literature as various authors stress that environments with high atmospheric evaporation demand have a higher plant water loss tendency through transpiration (Hsiao et al., 2019; Lintunen et al., 2021; Chen et al., 2023).

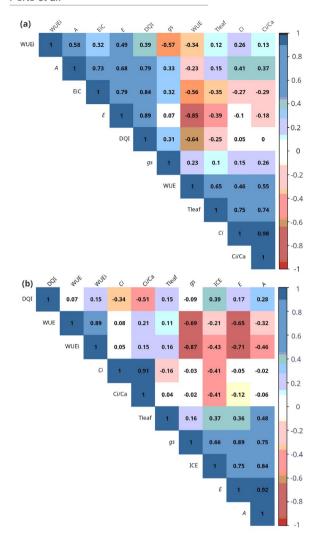
The region of Bom Jesus has shown changing trends in its climatology, moving from a dry sub-humid climate to a semi-arid climate, implying reductions in local water availability (Fernandes et al., 2020) and higher evapotranspiration rates. This sudden change in the climatic conditions can favor the development of adaptive mechanisms in the regional flora and make other species more susceptible to extinction. As expected, under drought conditions, the seedlings of *P. platycephala* Benth show

lower values of net photosynthesis, stomatal conductance, and transpiration as a mechanism to reduce water loss, highlighted by the inversely proportional relationship between transpiration and the intrinsic and instantaneous water-use efficiency, i.e., the WUE-WUEi values tend to be naturally higher when the *E* decreases (Figure 3).

It has been proposed that these responses may be closely related to an increase in stomatal density, which may also reduce stomatal size, thereby improving the hydraulic capacity of the leaf (Hasanuzzaman et al., 2023). However, Engineer et al. (2016) stressed that although the decreased *gs* is beneficial to limiting leaf water loss by *E*, closer stomata also reduce the cooling capacity of leaves, which, in turn, can increase their water stress. Nevertheless, although the stomatal conductance decreased as a function of water stress (Figure 2), the presence of AMFs of the species *A. koskei* and *A.morrowiae* contributes to maintaining the leaf temperature in seedlings of *P. platycephala* Benth under drought conditions (Figure 3). The treatment with the mixed inoculum (MIX), in turn, was more effective for this variable when water availability was not restricted.



**Figure 3:** Evaluation of the physiological parameters and growth of Parkia platycephala Benth. seedlings inoculated with different arbuscular mycorrhizal fungi in management with and without stress. Water use efficiency - WUEi (a), intrinsic water use efficiency (b), instantaneous carboxylation efficiency (c), ratio between internal and external CO<sub>2</sub> concentrations (d), leaf temperature (e) and Dickson quality index (f). Different lowercase letters indicate a significant difference between the AMF in each water regime and different capital letters indicate a significant difference between the water regimes, within each AMF, by the Scott-Knott test at 5% significance.



**Figure 4:** Correlogram of Pearson between physiological traits and the Dickson quality index (DQI) analyzed in *Parkia platycephala* Benth. subjected to inoculation of different arbuscular mycorrhizal fungi in management without water stress (a) and with water stress (b). Rate of net  $\mathrm{CO}_2$  assimilation (A), leaf transpiration (E), stomatal conductance (gs), internal  $\mathrm{CO}_2$  concentration (Ci), water use efficiency (WUE), intrinsic water use efficiency (WUE), instantaneous carboxylation efficiency (ICE), the ratio between internal and external  $\mathrm{CO}_2$  concentrations (Ci/Ca), leaf temperature (Tleaf) and Dickson quality index (DQI).

The higher internal CO<sub>2</sub> concentration in the leaves of *P. platycephala* Benth in the water restriction treatment (Figure 2) could be related to the reduced CO<sub>2</sub> assimilation rate in response to the fall in the production of adenosine triphosphate (Lawson et al. 2008) and the reduced enzymatic efficiency of the Calvin cycle, including limited regeneration of ribulose-1,5 bisphosphate (Kaur et al., 2021; GAO et al., 2018). The association with AMFs played a crucial role in improving the photosynthetic apparatus and the seedling quality of *P. platycephala* Benth, thus validating the tested hypothesis. Plants mycorrhized with *G. rosea* showed

better results under water deficit conditions, whereas plants without water restriction showed better performance when inoculated with *A. morrowiae*. Therefore, these results highlight the importance of demonstrating the divergences between the effects manifested by the variables when plants are under different water conditions.

Some environmental factors influence the colonization and functionality of AMFs, e.g., the host's specificity, seasonality, temperature, initial P content in the soil, soil pH, and light availability (Jamiołkowska et al., 2018; Han et al., 2023; Rodrigues et al., 2023). Likewise, soil water availability can also favor the colonization of some fungal species. Soil water excess and shortage are harmful to mycorrhizal colonization, but minimal water conditions favor the fungus-plant symbiosis (Augé, 2004; AL-Karaki and Williams, 2021). This scenario highlights a possible optimal range of soil moisture availability for each species of AMFs that colonize *P. platycephala* Benth.

These results also highlighted the importance of AMFs in improving the physiological conditions of seedlings of *P. platycephala* Benth, resulting in plants with higher values of the Dickson morphological quality index. In forestry sciences, inoculation with AMFs is used to maximize seedling growth in plant nurseries, reducing costs and increasing plant survival in the field (Oliveira Júnior et al., 2017). Similar results to this study are described in the literature, with reports of Forest species being highly responsive to artificial inoculation with AMFs, e.g., *Apuleia leiocarpa* (Oliveira Júnior et al., 2017), *Schizolobium parahyba* var. *amazonicum* (Brito et al., 2017), *Schinopsis brasiliensis* (Oliveira et al., 2015), *Ziziphus joazeiro*, and *Pseudobombax simplicifolium* (Oliveira et al., 2017).

The success of the initial establishment of tree seedlings is also related to the capture and use of primary resources such as water, light, and nutrients (Porto et al., 2020). The selection of pioneer and heliophilous tree species such as P. platycephala Benth (Carvalho, 2014), with a high carbon assimilation potential and efficient use of light associated with specific fungal strains for each Forest species and each edaphoclimatic condition, can favor the timely recovery of degraded areas, especially in locations with intense solar radiation and low water availability. These results suggest that water restriction associated with the artificial inoculation of AMFs could be a useful strategy to produce seedlings of *P. platycephala* Benth with greater rusticity about the use-efficiency of water, light, and nutrients. AMFs can be important facilitators for the growth and establishment of seedlings of *P. platycephala* Benth in stressful environments such as the semi-arid region of northeastern Brazil.

## **CONCLUSIONS**

As expected under water stress conditions, to decrease water loss, seedlings of *Parkia platycephala* Benth. promote partial stomatal closure by decreasing stomatal conductance and decreasing the rate of net  $\mathrm{CO}_2$  assimilation and leaf transpiration. On the other hand, inoculation with arbuscular mycorrhizal fungi promoted better physiological

conditions in plants under water restriction conditions. Under water stress, the plants that used the *Gigaspora rosea* strain had better physiological conditions and better quality seedlings.

#### **AUTHORSHIP CONTRIBUTION**

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Processing: DLP; RIO; JGC
Analysis: DLP; RIO; JGC; DVC

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