

## Growth and gas exchanges of red pitaya under different shading conditions

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**ABSTRACT:** Plants with yellow cladodes are commonly present in commercial red pitaya orchards, in Ceará, mainly during the warmer seasons of the year. Therefore, this study aimed to evaluate the effect of shading on the growth and gas exchanges of red pitaya. We tested five shading levels on stock plants (full sunlight, 35, 50; 65 and 80% shading) via randomized block design (RBD), with five replications and two plants per plot. After 180 days, the following characteristics were evaluated: sum of the length of the axillary shoots, number of axillary shoots, diameter, thickness of secondary shoots, photosynthetic rate, stomatal conductance of water vapor, transpiration, internal carbon dioxide concentration, ratio of internal to external CO<sub>2</sub> concentration, instantaneous and intrinsic water use efficiency, and instantaneous carboxylation efficiency. The plants presents large and thin cladodes, apparently blanching under 80% shading; therefore, we concluded that cultivation in full sunlight caused a negative effect on gas exchanges and growth of red pitaya.. The use of 35% shading at 180 days increased the photosynthetic rate, water use efficiency and carboxylation, with increase vegetative growth.

**Key words:** Cactaceae; ecophysiology; exotic fruit; *Hylocereus* sp.; light intensity

## Crescimento e trocas gasosas de pitaia vermelha sob diferentes condições de sombreamento

**RESUMO:** Em pomares comerciais de pitaia vermelha, no Ceará, é comum a presença de plantas com cladódios amarelados, principalmente nas épocas mais quentes do ano. Diante disso, objetivou-se avaliar o efeito do sombreamento sobre o crescimento e trocas gasosas de pitaia vermelha. O delineamento experimental foi em blocos casualizados (DBC), com cinco tratamentos, cinco repetições e duas plantas por parcela. Os tratamentos consistiram de cinco níveis de luminosidade (pleno sol, 35, 50; 65 e 80% de sombreamento). Aos 180 dias, realizou-se análise de caracteres vegetativos (somatório do comprimento de emissões laterais, número de emissões laterais, diâmetro e espessura de emissões secundárias) e trocas gasosas (assimilação líquida do carbono, condutância estomática ao vapor de água, transpiração, concentração interna de CO<sub>2</sub>, razão entre as concentrações interna e externa de CO<sub>2</sub>, eficiências intrínseca e instantânea de uso da água, e eficiência instantânea de carboxilação). Concluiu-se que o cultivo a pleno sol ocasiona efeito negativo nas trocas gasosas e crescimento de pitaia vermelha, ao passo que sob 80% de sombra, as plantas apresentam cladódios grandes e finos, aparentemente estiolados. O uso de 35% de sombreamento aumenta a assimilação líquida de carbono e eficiências de uso da água e carboxilação, com incremento no crescimento vegetativo regular, até os 180 dias de cultivo.

**Palavras-chave:** Cactaceae; ecofisiologia; frutíferas exóticas; *Hylocereus* sp.; intensidade luminosa

## Introduction

The red pitaya (*Hylocereus* spp.) is a cactaceous fruit from a semi-epiphytic plant, with star-shaped modified stems, called cladodes, which develops numerous adventitious roots. The flower is hermaphrodite, white, large and with nocturnal anthesis. The fruit is attractive to consumers due to its reddish pulp and pleasant slightly sweet taste (Marques et al., 2011; Ortiz-Hernández & Carrillo-Salazar, 2012).

The red pitaya is distributed mainly in Central America, in countries such as Costa Rica, El Salvador, Guatemala and Mexico (Donadio, 2009). In Ceará, the production is mainly concentrated in Limoeiro do Norte and Quixeré. Cultivation occurs throughout the year, with little decrease in the wettest months, which are usually from January to April (Almeida et al., 2016a; Nunes et al., 2014).

The average productivity is around 10 to 30 ton ha<sup>-1</sup>, depending on soil and climate conditions, cultivation techniques and age of orchard (Le Bellec et al., 2006). Light is one of the environmental factors that regulate processes such as photosynthesis, pigments biosynthesis, nitrogen assimilation and anatomy (Sesma et al., 2009).

Under natural conditions, the red pitaya is found in the shaded rainforest understories of America, which suggests that commercial cultivation requires a system of protection against exposure to direct sunlight. Within this context, preliminary studies performed in Israel (Mizrahi & Nerd, 1999; Raveh et al., 1996; Raveh et al., 1998) confirmed the need of shading, depending on local conditions. Almeida et al. (2016b) concluded that the production of high quality plants, it is necessary to collect cuttings from stock plants grown in full sunlight or 80% shade. Cavalcante et al. (2011) reported that plants grown in full sunlight, in the soils and climate conditions of Bom Jesus (PI), presented shorter development, with loss of some plots, due to the plants death from overexposure to sunlight.

However, most of Ceará's commercial orchards are grown in full sunlight, evidencing the presence of plants with yellow cladodes, mainly during the warmer times of the year. To date, the effects of full exposure to solar radiation on the gas exchanges of red pitaya remain unknown, in the Brazilian climates conditions. Based on the above, this study was conducted in order to assess the effect of the use of shading on red pitaya by evaluating their process of growth and gas exchanges.

## Material and Methods

The experiment was conducted from August 2013 to February 2014, in a greenhouse located in the horticulture sector of the Department of Plant Science, Agricultural Science Center of the Federal University of Ceará in Fortaleza (CE) (3°43'02" S latitude and 38°32'35" WGR longitude, elevation 19.6 m) (IPECE, 2016), which Köppen (1918) classifies as Aw. This region belongs to the tropical rainy climate group, with an average annual temperature of 26.5 °C.

The experiment was set up as a randomized block design, with five replications and two plants per plot. Plants were placed in five shading levels (0, 35, 50, 65 e 80%) using screen mesh.

The cuttings of red pitaya (*Hylocereus* sp.) were collected from stock plants 120 days old grown in the horticulture sector. The propagation material used for cuttings was selected based on health and vigor, with an average length of 25 cm (Pontes Filho et al., 2014). Cuttings were transplanted "with clod" to plastic pots of 11 L<sup>-1</sup>, filled with sand soil, clay and organic material (1:1:1). The substrate had the following chemical characteristics: M. O. = 28.86 g kg<sup>-1</sup>; pH = 7.0; P = 30 mg kg<sup>-1</sup>; K<sup>+</sup> = 1.82 cmol dm<sup>-3</sup>; Ca<sup>2+</sup> = 5.8 cmol dm<sup>-3</sup>; Mg<sup>2+</sup> = 2.2 cmol dm<sup>-3</sup>; Na<sup>+</sup> = 1.72 cmol dm<sup>-3</sup>; Al<sup>3+</sup> = 0.25 cmol dm<sup>-3</sup>; H + Al = 1.16; C/N = 10; V = 91%.

After rooting and emergence of the first bud, the cuttings were pruned, leaving only one bud. Afterwards the pots were transferred to field conditions, spaced 0.70 m in rows and 2.00 m between rows, and positioned on a brick base about 0.10 m high. The plant staking started after 60 days, and was carried out by attaching the axillary shoots to a wooden stake (positioned in the center of the vase) about 1.20 m high.

Weeds were controlled by hand hoeing. Water was supplied via located irrigation, applying 850 ml of water per plot, twice a day, for 4 minutes, resulting in 4.76 L per week. This information was based on results obtained by Mizhari (2014) for the cultivation of *Hylocereus* sp.

Mineral fertilization was performed manually, dissolved in water, at thirty-day intervals. The doses were according to Almeida et al. (2014) and Corrêa et al., (2014), at the following concentrations: 4.10 g vase<sup>-1</sup> of KCl, 10.49 g vase<sup>-1</sup> of triple superphosphate, 19.67 g vase<sup>-1</sup> of ammonium sulphate, and 0.60 g vase<sup>-1</sup> of FTE BR-12, for the supply of potassium, phosphorus, nitrogen, sulfur and micronutrients (1.8% boron, 1.8%, 0.8% copper, 2.0% manganese, 0.1% molybdenum, 9.0% zinc), respectively. Apart from the triple superphosphate and FTE BR-12, which entire amounts were applied during transplantation, the remaining nutrient supplies were divided in six equal installments (October and December 2013; February, April and June 2014). For each period of application, we prepared a nutrient solution with the total amount of each fertilizer. After this, we manually applied the nutrient solution using plastic cups (100 ml).

The following morphological features were analyzed after 180 days, during the stage of vegetative development: diameter of secondary shoots (DSS) – Determined by measuring the medium portion of the secondary shoots (measured in the transverse direction of each costilla, with a digital caliper); thickness of secondary shoots (TSS) – determined by measuring the medium portion of the upper costillas (measured in the transverse direction, with a digital caliper); number of axillary shoots (NAS) – measured by counting the axillary cladodes; the sum of the length of the axillary shoots (SLAS) – determined by the sum of the length of the axillary shoots, which were measured with a millimeter tape in the longitudinal direction of each cladode.

Characteristics of photosynthetic rate ( $A$ ), stomatal conductance of water vapor ( $g_s$ ), transpiration ( $E$ ), internal carbon dioxide concentration ( $C_i$ ) and ratio of internal to external  $\text{CO}_2$  concentration ( $C_i C_a^{-1}$ ) were determined at the 180th day after planting, with an infrared portable gas analyzer – IRGA (ADC, Bioscientific Ltd. Hoddesdon, UK). Using the photosynthetic rate, transpiration, stomatal conductance of water vapor and internal carbon dioxide concentration values. The intrinsic water use efficiency was calculated as  $A g_s^{-1}$ , the instantaneous water use efficiency, was calculated as  $A E^{-1}$ , and the instantaneous carboxylation efficiency as  $A C_i^{-1}$ .

The measurements for gas exchanges were obtained during the early morning hours (02:00 - 04:00 AM), due to the pitaya's CAM metabolism. This schedule was set up as the period of maximum  $\text{CO}_2$  fixation in the plant, in previous monitoring trials carried out by the research group. These evaluations were performed in healthy and vigorous cladodes after adapting the tweezers of the IRGA.

Data were submitted to an analysis of variance (F-test with significance level  $p < 0.05$ ). The comparison of means was performed using the Tukey test for the gas exchanges characteristics and regression to the morphological characteristics. The gas exchanges results were presented in graphic form composed by means and standards errors of each treatment. Statistical analyses were conducted using the statistical program *Statistical Analysis System* (SAS) version 9.1 (SAS Institute, 2003).

Estimates of phenotypic correlations of Pearson were obtained for most traits evaluated, considering all of them were important for the study.

## Results and Discussion

Treatments caused a significant effect ( $p < 0.05$ ) on morphological characteristics analyzed. The coefficients of variation oscillated from 3.00 to 8.73%, which indicates good experimental precision (Table 1).

The sum of the length of axillary shoots (SLAS) values increased concurrently with an increase of shading levels. We estimated a range of 56.87 cm, between SLAS of plants grown under 80% shading and full sunlight, which represents the highest and lowest mean, respectively (Figure 1A). These results corroborate those of Andrade et al. (2006) and Cavalcante et al. (2011), who noted an increase in length of lateral cladodes in pitayas grown under shading.

Plants grown under shading, followed similar number of shoots (NAS) as in full sunlight, however under shaded conditions they presented cladodes with a higher length (SLAS). It is likely that blanching of cladodes, mainly in plants grown in 65 and 80% shading, had positively influenced the increase of SLAS.

Regarding DSS and TSS, the highest values were obtained in plants grown under full sunlight (Figures 1C and 1D), similar to the results obtained by Cavalcante et al. (2011), in cultivation of red pitaya in Bom Jesus (PI), under different luminosity conditions. According Gonçalves et al. (2010) the increase of thickness in photosynthetic organs, in full sunlight, aims to protect the photosynthetic apparatus, to regulate the internal levels of light and  $\text{CO}_2$  and to allow the cells to be protected from dehydration and dryness. Therefore, it is possible that the increase of TSS, in full sunlight, worked as photo-protective mechanism in red pitaya, which is a plant hemi-epiphyte.

The reduction in diameter of secondary shoots (DSS) and thickness of secondary shoots (TSS), and the increase of SLAS in shaded plants, especially under highest shading levels (65 to 80%), resulted in blanching, which corroborates the hypothesis by Merten (2003) for etiolated cactaceous. This author found that etiolated plants have strait and long cladodes, similar to our results.

Regarding the gas exchanges, treatments caused a significant effect ( $p < 0.05$ ) on gas exchanges of red pitaya, represented by  $A$ ,  $g_s$ ,  $E$ ,  $A g_s^{-1}$ ,  $A E^{-1}$ ,  $C_i C_a^{-1}$ ,  $A C_i^{-1}$ . The coefficients of variation oscillated from 6.18 to 21.59%, which indicates good experimental precision (Tables 2 and 3).

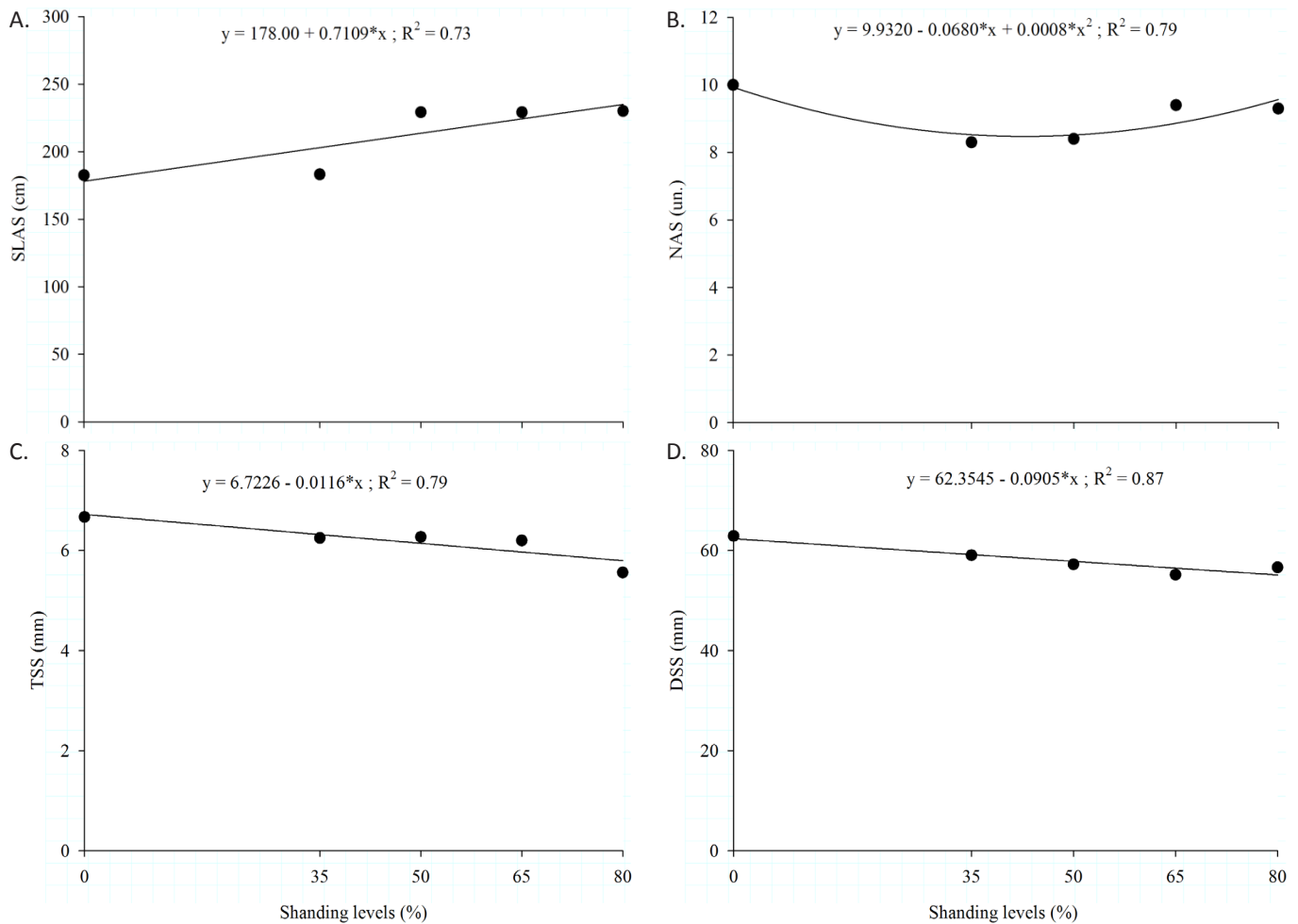
The use of screens with 35, 50 and 65% shading increased photosynthetic taxes. The greater mean was obtained under 35% shading, followed by 50 and 65% shading, which did not differ among them. Plants grown in full sunlight and 80% shading resulted in lower photosynthetic taxes (Figure 2A). Therefore, it is possible that the excess of radiation in full sunlight had induced in damages to the photosynthetic apparatus or stomatal limitation ( $A$  and  $g_s$  decreased concurrently with an increase of  $C_i$ ). More over, the shading of plants at 80% led to the blanching of cladodes (increase of SLAS and decrease of TSS), with consequent reduction of the photoassimilated productive capacity.

According to Baliza et al. (2012) and Coelho et al. (2010), plants grown in high shading level can present anatomical changes that limit photosynthesis, such as, reduction in thickness of

**Table 1.** Summary of the analysis of variance for the following characteristics at 180 days: sum of the length of axillary shoots (SLAS), number of axillary shoots (NAS), diameter of secondary shoots (DSS) and thickness of secondary shoots (TSS).

Source of variation	Degree of freedom	Average squares			
		Sum of the length of axillary shoots	Number of axillary shoots	Diameter of secondary shoots	Thickness of secondary shoots
Shading	4	3256.6089**	2.5850*	44.4766**	0.7968**
Blocks	4	64.0846 <sup>ns</sup>	0.7714 <sup>ns</sup>	2.3782 <sup>ns</sup>	0.0118 <sup>ns</sup>
Residue	16	176.9158	0.6288	3.0387	0.0488
C.V. (%)		6.31	8.73	3.00	3.57

C.V. - Coefficient of variation; - \*Significant at a level of 5% by the F test; \*\* - Significant at a level of 1% by the F test; <sup>ns</sup> - Not significant by the test F.



**Figure 1.** Effects of shading on the growth of pitaya, after 180 days. 1A: length of axillary shoots (SLAS), 1B: number of axillary shoots (NAS), 1C: diameter of secondary shoots (DSS) and 1D: thickness of secondary shoots (TSS).

**Table 2.** Summary of the analysis of variance for the following characteristics at 180 days: photosynthetic rate (*A*), stomatal conductance of water vapor (*g<sub>s</sub>*), transpiration (*E*) and intrinsic water use efficiency (*A g<sub>s</sub><sup>-1</sup>*).

SV	DF	Average squares			
		<i>A</i>	<i>g<sub>s</sub></i>	<i>E</i>	<i>A g<sub>s</sub><sup>-1</sup></i>
Shading (S)	4	68.0364**	0.0006*	0.0213**	14.5765**
Blocks	4	2.5088 <sup>ns</sup>	0.0002 <sup>ns</sup>	0.0023 <sup>ns</sup>	493.8693 <sup>ns</sup>
Residue	16	1.9242	0.0002	0.0016	252.2636
C.V. (%)		12.73	21.59	6.18	8.99

SV - Source of variation; DF - Degree of freedom; C.V. - Coefficient of variation; \* - Significant at a level of 5% by the F test; \*\* - Significant at a level of 1% by the F test; <sup>ns</sup> - Not significant by the test F.

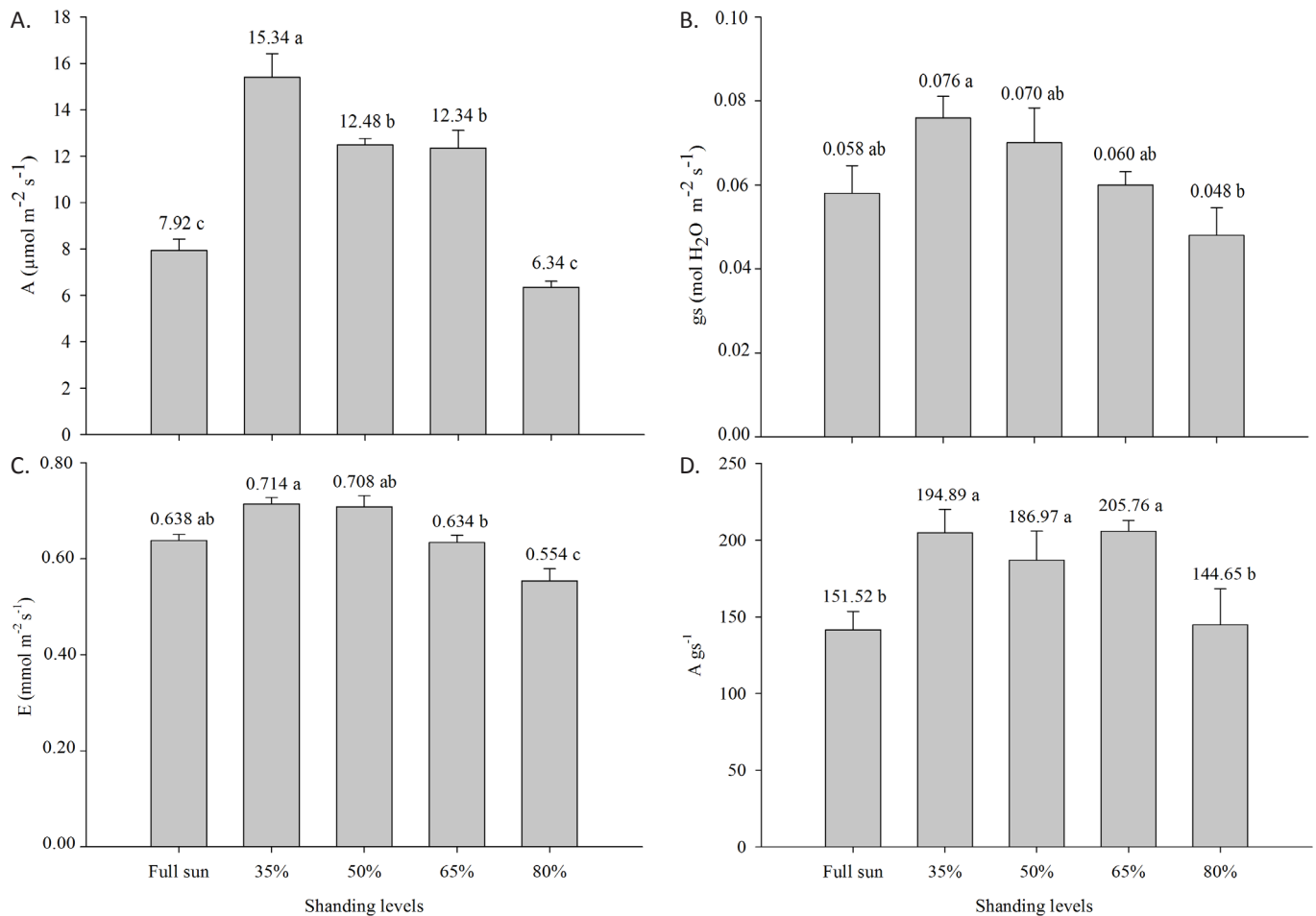
**Table 3.** Summary of the analysis of variance for the following characteristics at 180 days: instantaneous water use efficiency (*A E<sup>-1</sup>*), internal carbon dioxide concentration (*C<sub>i</sub>*), ratio of internal to atmospheric CO<sub>2</sub> concentration (*C<sub>i</sub> C<sub>a</sub><sup>-1</sup>*) and ratio photosynthetic rate into internal to atmospheric CO<sub>2</sub> concentration.

SV	DF	Average squares			
		<i>A E<sup>-1</sup></i>	<i>C<sub>i</sub></i>	<i>C<sub>i</sub> C<sub>a</sub><sup>-1</sup></i>	<i>A C<sub>i</sub><sup>-1</sup></i>
Shading (S)	4	95.1653**	1036.7464**	0.0067**	0.0014**
Blocks	4	3.2905 <sup>ns</sup>	15.1264 <sup>ns</sup>	0.00009 <sup>ns</sup>	0.00004 <sup>ns</sup>
Residue	16	4.0666	46.1164	0.0003	0.00004
C.V. (%)		12.20	2.99	2.93	12.64

SV - Source of variation; DF - Degree of freedom; C.V. - Coefficient of variation; \*\* - Significant at a level of 1% by the F test; <sup>ns</sup> - Not significant by the test F.

photosynthetic organ, which culminates in lower radiation use efficiency. Therefore, the shorter TSS in plants grown under 80% shading may have caused low light utilization, interfering in photoassimilated reserves.

The highest transpiration means were obtained in full sunlight, 35 and 50% shading, which did not differed each other (Figure 2C). Souza et al. (2011) can explain this result. According to the authors, highest transpiration



**Figure 2.** Effects of shading on the gas exchanges of red pitaya, after 180 days. 2A: photosynthetic rate ( $A$ ) ( $\text{dms} = 2.69$ ); 2B: stomatal conductance of water vapor ( $g_s$ ) ( $\text{dms} = 0.026$ ); 2C: transpiration ( $E$ ) ( $\text{dms} = 0.078$ ); 2D: intrinsic water use efficiency ( $A/g_s$ ) ( $\text{dms} = 30.83$ ). Means followed by the same letter did not differ between each other by the Tukey test.

values in shaded plants are due to lower variation of relative humidity, usually common to environments kept in these luminosity conditions. In general, the increment of stomatal conductance was instrumental in increasing the transpiration. The highest  $g_s$  were obtained in full sunlight, 35 and 50% shading, which did not differ each other. The lower value was in 80% shading, probably due to the low productive capacity (Figure 2B).

Although there has been an increase of  $g_s$  and  $E$ , in a linear ratio, under 35, 50 and 65% shading, plants presented high water use efficiencies (Figures 2D and 3A). According Oliveira et al. (2011), estimates of water use efficiency relate how much water is lost via transpiration so that the plant can integrate a molecule of  $\text{CO}_2$  in some organic compound. Zapp & Taylor (2017) substantiates that, physiologically, a specific type of metabolism, called CAM metabolism, characterizes the cactaceous. This system presented high water use efficiency when compared to other metabolisms (C3 e C4).

Graham & Nobel (2005) reported that transpiration and water movement in stem of red pitaya could influence the diameter growth, mainly under stress conditions. Therefore, it is likely that the larger DSS obtained in plants grown under full sunlight be related to a mechanism of water reserve and subsequent acclimatization to adverse conditions.

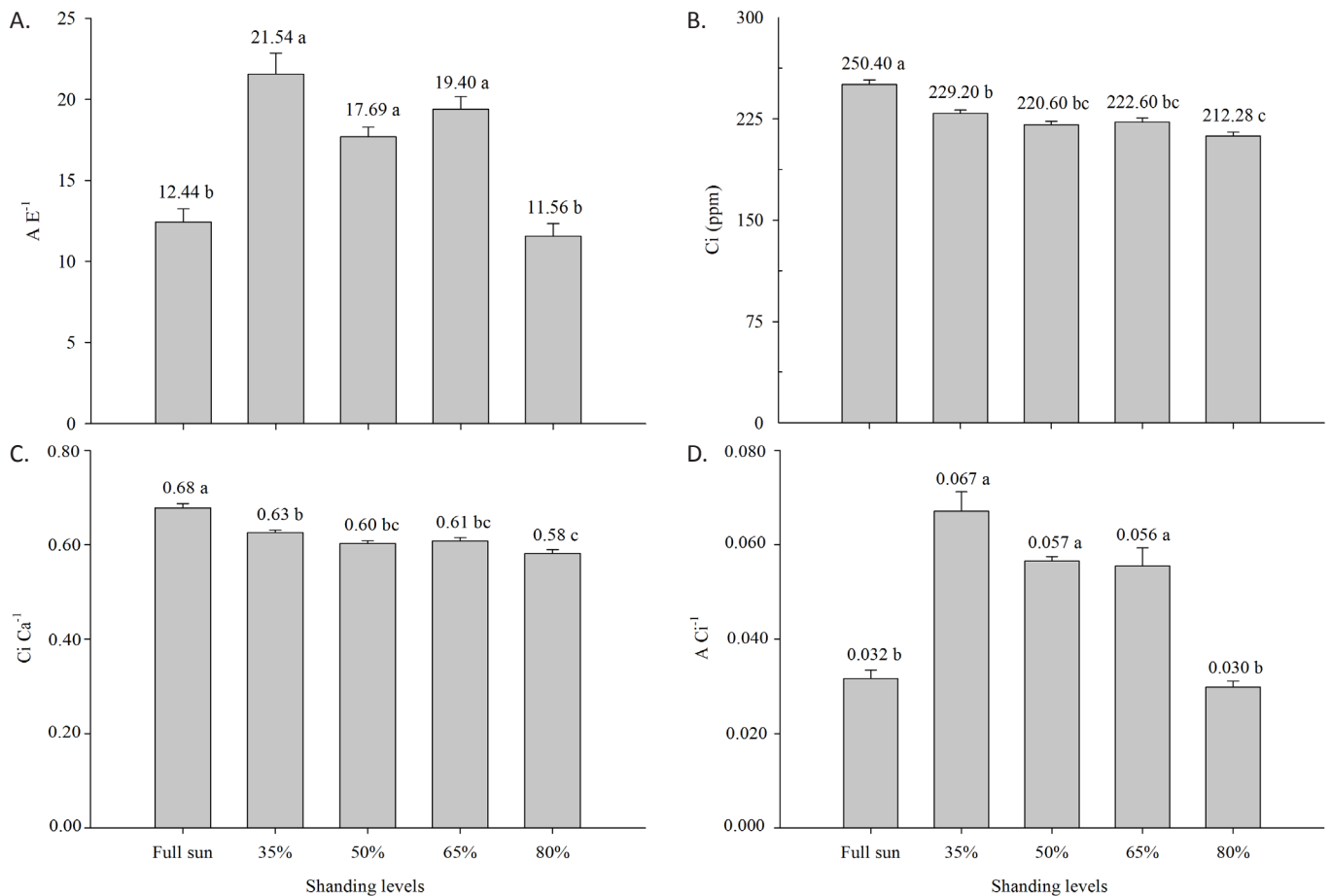
Regarding  $C_i$ , the highest mean was obtained in sunlight. The increase of  $C_i$  in this condition, may be related to the fall in the photosynthetic activity. That is, a high  $C_i$  value associated to low  $g_s$  values would indicate a decrease in carboxylation efficiency, with subsequent reduction of  $A$ .

In relation to carboxylation efficiency ( $A/C_i$ ), the lowest values were achieved under full sunlight condition and 80% shading (Figure 3D). In full sunlight, the result can be justified by the increase of  $C_i$ , which caused reduction in stomatal conductance ( $g_s$ ) and photosynthetic rate ( $A$ ). Whereas, the low carboxylation efficiency encountered at 80% shading, can be explained by the reduced productive capacity ( $A$ ), probably associated to a low radiation.

For the correlations between vegetative characters and gas exchanges, we observed that the photosynthetic rate ( $A$ ) was positively correlated with water use efficiencies and number of axillary shoots, in full sunlight conditions (Table 4).

It is likely that the plants grown under this condition had restricted stomatal conductance and photosynthesis, due to the lower water use efficiency. In other words, plants possibly increased their mechanisms of water reserve and photoassimilation, with consequent reduction of productive capacity. Within this context, the vegetative growth in full





**Figure 3.** Effects of shading on the gas exchanges of red pitaya, after 180 days. 3A: instantaneous water use efficiency ( $A E^{-1}$ ) (dms =3.91); 3B: internal carbon dioxide concentration ( $C_i$ ) (dms = 13.18); 3C: ratio of  $C_i C_o^{-1}$  (dms = 0.04); 3D: carboxylation efficiency ( $A C_i^{-1}$ ) (dms =0.012). Means followed by the same letter did not differ between each other by the Tukey test.

**Table 4.** Pearson correlation coefficients ( $r_f$ ) among gas exchange characteristics [internal carbon dioxide concentration ( $C_i$ ), transpiration ( $E$ ), stomatal conductance of water vapor ( $g_s$ ), photosynthetic rate ( $A$ ), carboxylation efficiency ( $A C_i^{-1}$ ), instantaneous water use efficiency ( $A E^{-1}$ ), intrinsic water use efficiency ( $A/g_s$ )] and vegetative characteristics [sum of the length of the axillary shoots (SLAS); number of axillary shoots (NAS); diameter (DSS) and thickness of secondary shoots (TSS)], under full sunlight conditions (top diagonal) and 80% shading (bottom diagonal), after 180 days.

Character	$C_i$	$E$	$g_s$	$A$	$A C_i^{-1}$	$A E^{-1}$	$A g_s^{-1}$	SLAS	NAS	DSS	TSS
$C_i$		0.54	0.94*	0.41	-0.10	0.23	-0.83	0.52	0.02	0.37	-0.07
$E$	-0.68		0.53	0.01	0.24	-0.31	-0.63	-0.32	0.00	0.80	0.71
$g_s$	-0.15	0.83		0.40	0.08	0.22	-0.89*	0.29	0.00	0.47	0.13
$A$	0.07	-0.48	-0.68		0.62	0.95*	0.06	0.45	0.88*	-0.48	-0.29
$A C_i^{-1}$	0.00	0.00	0.00	0.00		0.50	0.22	-0.42	0.79	-0.10	0.45
$A E^{-1}$	0.40	-0.85	-0.88*	0.87*	0.00		0.25	0.54	0.83	-0.71	-0.51
$A g_s^{-1}$	-0.10	-0.66	-0.97**	0.69	0.00	0.81		-0.10	0.42	-0.76	-0.30
SLAS	-0.45	0.77	0.62	0.08	0.00	-0.40	-0.49		0.14	-0.49	-0.88
NAS	0.65	0.08	0.62	-0.44	0.00	-0.33	-0.77	0.06		-0.55	-0.11
DSS	-0.10	0.40	0.35	0.35	0.00	-0.03	-0.32	0.88*	0.08		0.77
TSS	-0.18	0.15	0.18	-0.82	0.00	-0.56	-0.17	-0.51	0.04	0.79	

\*\* and \* Significant at a level of 1% and 5% by the F test, respectively. The rest were not significant.

sunlight occurred through multiple and small buds with strong correlations among NEL,  $A$ ,  $A E^{-1}$  and  $A g_s^{-1}$ .

As for diameter and thickness of axillary cladodes in plants grown under full sunlight, we observed strong negative correlations between these variables and the water use efficiency. This allows us to affirm that the thickening of cladodes can be related to water accumulation and

photoassimilated reserves, as well as to photo-protection (Table 4).

It is important to highlight the strong positive correlations between vegetative growth (SLAS and NAS) and increase of  $g_s$  and  $E$  in cultivation under 80% shading. In this case, the increase of green mass culminated in reduction of photosynthetic rate and water use efficiency ( $A E^{-1}$  e  $A g_s^{-1}$ ),

probably due to the blanching of cladodes (thin, elongated and with low production capacity) (Table 4).

In summary, after 180 days of evaluation, the best incomes were obtained under 35% shading. This result is in the range of 50 to 60% shading, recommended by Nobel & De La Barrera (2004), and Mizrahi & Nerd (1999), for pitaya commercial cultivation. This demonstrates that the red pitaya presents shading demands in the initial growth phase.

Therefore, the use of screens aiming to reduce the plants exposure to solar radiation was suitable in this study, since we observed positive influence in growth and gas exchanges of red pitaya under 35% shading.

## Conclusions

Red pitaya performed better under shading conditions, mainly with plants shaded at 35%. In these conditions, the plants showed higher photosynthetic activity, water use efficiency, carboxylation and vegetative growth, at 180 days of evaluation. This provides strong foundation for the production of pitaya propagules in greenhouse.

## Acknowledgments

We thank CAPES for the financial support (Project 23038.006862/2010-02 - Notice 029/2010) and to CNPQ for the Doctoral Scholarship.

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