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Development and physiological aspects of three species of passion fruit submitted to water stress

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ABSTRACT – The aim of this work was to evaluate the development and physiological aspects of three species of passion fruit submitted to water stress. The work was carried out at the Faculdade de Ciências Agrárias e Tecnológicas of Universidade Estadual Paulista (Unesp, Brazil). The experimental design was completely randomized (DIC), in a 3 x 3 factorial arrangement, in which three passion fruit species were used: *Passiflora gibertii*; *Passiflora foetida*, and *Passiflora edulis* interacting with three irrigation intervals, namely: 4, 8 and 12 days, with four replications. The suspension of irrigations for twelve days reduces the transpiration of passion fruit seedlings, while the suspension of irrigations every eight days reduces stomatal conductance. *Passiflora gibertii* plants have higher photosynthetic efficiency than *P. edulis* and *P. foetida. Passiflora gibertii* seedlings show greater growth than *P. foetida* and *P. edulis*, while *P. foetida* seedlings have the highest number of leaves. In plants irrigated at twelve-day intervals, fruit growth was affected.

Keywords: irrigation, *Passiflora,* stomatal conductance, transpiration.

RESUMO – **Desenvolvimento e aspectos fisiológicos de três espécies de maracujás submetidas a estresse hídrico.** O objetivo deste trabalho foi avaliar o desenvolvimento e os aspectos fisiológicos de três espécies de maracujá submetidas ao estresse hídrico. O trabalho foi realizado na Faculdade de Ciências Agrárias e Tecnológicas da Universidade Estadual Paulista (Unesp, Brasil). O delineamento experimental foi inteiramente casualizado (DIC), em esquema fatorial 3×3, no qual foram utilizadas três espécies de maracujá: *Passiflora gibertii*, *Passiflora foetida* e *Passiflora edulis* interagindo com três intervalos de irrigação, a saber: 4, 8 e 12 dias, com quatro repetições. A suspensão das irrigações por doze dias reduz a transpiração das mudas de maracujá, enquanto a suspensão das irrigações a cada oito dias reduz a condutância estomática. Plantas de *P. gibertii* têm maior eficiência fotossintética do que as de *P. edulis* e as de *P. foetida*. Mudas de *P. gibertii* apresentam maior crescimento do que as de *P. foetida* e as de *P. edulis*, enquanto mudas de *P. foetida* apresentam maior número de folhas. Plantas irrigadas em intervalos de 12 dias afetam o crescimento do fruto.

Palavras-chave: irrigação, condutância estomática, *Passiflora*, transpiração.

INTRODUCTION

Among the fruit trees grown in Brazil, passion fruit has stood out for presenting a good income option, mainly for the family farming segment. With a cultivated area of around 50 thousand hectares and a production exceeding 700 thousand tons, the country stands out as the main world producer of the fruit (Agrianual 2019), however, its productivity of 14.1 $t.ha^{-1}$ is considered low, mainly affected by factors such as water stress, nutritional deficiencies, phytosanitary problems, inadequate cultivation techniques and low use of improved cultivars (Simon & Karnatz 1983, Aguiar *et al*. 2015).

Non-irrigated crops in Brazil suffer more and more from water deficiency, which can lead to reduced growth and loss of crop productivity (Menzel *et al*. 1986, Rahmati *et al*. 2018). Water deficiency causes stomata to close, affecting various plant physiological processes, such as reducing transpiration and supplying carbon dioxide, decreasing plant growth and increasing photorespiration (Shinozaki & Yamaguchi-Shinozaki 2007). These processes can vary according to the severity, the duration of the water deficiency, the phenological phase of the plant and the genetic material (Peixoto *et al*. 2006).

Water stress is one of the environmental factors that can affect the growth and development of passion fruit (Staveley & Wolstenholme 1990). According to Menzel *et al*. (1986), in passion fruit, mild humidity stress levels can already limit the vegetative growth and the productive potential of the crop, thus recommending that the soil moisture profile should remain close to its field capacity.

Several parameters have been used to study water deficiency in plants, highlighting leaf water potential, osmotic potential, transpiration and photosynthetic activity

(Nogueira *et al*. 2001). Lopez *et al*. (1993) evaluated stomatal conductance, transpiration and photosynthetic activity for the selection of cotton genotypes with drought tolerance, while Pereira *et al*. (2003) recommend to evaluate the growth of the root system and the aerial part based on the accumulation of dry matter and the increase of the leaf area for citrus rootstocks.

The use of wild passion fruit species as rootstocks to control pathogens that cause soil diseases has been successfully adopted in recent years, becoming an alternative for cultivation in areas with a history of diseases (Cavichioli *et al*. 2011a, Nogueira Filho *et al*. 2010). Several species of native passifloras have been showing resistance to these diseases (Chaves *et al.* 2004), standing out among them the *Passiflora gibertii* N.E. Brown (Roncatto *et al*. 2004, Cavichioli *et al*. 2009). In passion fruit, there are few studies related to the effects of water limitation in the soil on growth and photosynthetic processes between species.

This work seeks to evaluate the initial growth and some physiological aspects of three species of passion fruit subjected to different water regimes.

MATERIAL AND METHODS

The experiment was carried out from May 2019 at the Universidade Estadual Paulista (Unesp), Faculdade de Ciências Agrárias e Tecnológicas, located in the municipality of Dracena, state of São Paulo, Brazil, and conducted in a greenhouse covered with plastic light-diffusing wrap of 1200 microns, with a ceiling height of 4.0 meters, with its sides closed with a Sombrite® type screen with 50 % light passage.

The experimental design was completely randomized (DIC), in a 3×3 factorial arrangement, in which three passion fruit species were used: *Passiflora gibertii, Passiflora foetida* L. and *Passiflora edulis* Sims interacting with three irrigation intervals, namely: 4, 8 and 12 days, with four replications, totaling 36 plots.

Each plot was composed of a plant; the seedlings were obtained in a commercial nursery in the region of the municipality of Adamantina, state of São Paulo and had an average size of 20 ± 3 cm; with 6 ± 1 definitive leaves and aged 60 days. The seedlings were planted in plastic pots with a volumetric capacity of 9.0 dm^3 filled with ferric Red Latosol (Embrapa 2013) and had the following chemical attributes as shown in Tab. 1.

The soil was corrected and fertilized according to the requirements of the crop (Cavichioli *et al*. 2011b) and was irrigated to determine the field capacity, where it was saturated and allowed to drain, naturally. The evapotranspiration and water volume estimates to be replaced in the irrigation intervals, were determined according to the methodology described by Casaroli & Lier (2008).

An assessment was conducted 60 days after the beginning of the experiment regarding the values of the assimilation rate of CO_2, expressed by area (A – μmol CO_2 m^{-2} s^{-1}), the Transpiration (E – mmol H_2O m^{-2} s^{-1}), the Stomatal conductance (Gs – mol H_2O m^{-2} s^{-1}), the Internal concentration of CO_2 in the substomatic chamber (Ci – μmol mol^{-1}), the Water-use efficiency (WUE – mol CO_2 mol H_2O^{-1}), determined using the formula:

$$WUE = \frac{A}{E}$$

where a portable gas exchange device Infra-Red Gas Analyzer - IRGA, brand ADC BioScientific Ltd, model LC-Pro was used, the initial conditions imposed for the measurements were 1000 μmolm^{-2} s^{-1} of photosynthetically active radiation (PAR), which was provided by LED lamps, 380 ppm of CO_2, and a chamber temperature of 28 °C between 10 am and 2 pm. It was also determined: plant height (PH) by using a millimeter ruler; number of leaves (NL) through direct counting; the stem diameter (SD) measured at 1 cm from the neck of the plant, with the aid of a 0.1 mm precision digital caliper; dry mass of aerial part (DMAP) and dry mass of root (DMR), which were set by drying the humid mass in oven with air circulation and renewal, under a steady 65 °C temperature, till they reach steady weight.

All variables were subjected to normality tests, where the Shapiro-Wilk tests were used, after meeting the test precepts, the analysis of variance was performed by the F test ($p < 0.05$) and their means compared by the Tukey Test at 5 % probability, Pearson's correlation was also made between the variables (Banzatto & Kronka 2013) and the R statistical program (RStudio 2015) was used.

RESULTS AND DISCUSSION

It was found that there was an interaction between the factors for the CO_2 assimilation rate (A) as shown in Tab. 2. The species *P. gibertii* irrigated at four-day intervals showed the highest rates of assimilation of CO_2, showing that the species behave in different ways and *P. gibertii* may be more susceptible to water stress. These results differ from those of Gomes *et al*. (2018), when they found that six days after the suspension of irrigation, there was a decrease in

Table 1. Chemical attributes of the soil used in the implementation of the experiment.

pH	MO	P	K	Ca	Mg	SO_4^{-2}	H+Al	Al	SB	CTC	V	m
	mg dm^{-3}	--- (mmolc dm^{-3}) ---				mg dm^{-3}	------ (mmolc dm^{-3}) ------				--- (%) ---	
4.5	4.5	6.0	5.6	10.0	4.0	7.0	18.0	1.0	18.6	36.6	46.4	6.0

MO - Organic Matter, SB - Sum of bases, CTC - Cation exchange capacity, V - Saturation by bases and m - Saturation by aluminum.

Table 2. Average values of CO_2 Assimilation rate (A), Transpiration (E), Stomatal conductance (GS), and Internal concentration of CO_2 in the substomatic chamber (Ci) and Water-use efficiency (WUE) of species of passion fruit when grown at different irrigation intervals.

		A – μmol CO_2 m^{-2} s^{-1}		
	4 days	8 days	12 days	Average (ssp = S)
P. gibertii	13.28 aA	2.79 aB	5.59 aB	7.22 a
P. foetida	6.79 bA	7.85 aA	2.45 aA	5.70 a
P. edulis	6.43 bA	6.94 aA	3.00 aA	5.46 a
Inter. average (I)	8.83 a	5.86 ab	3.68 b	
	SMD: 5.60	CV(%): 52.20	GA: 6.12	
	F (S): 1.07ns	F (I): 7.84¨**	F (SxI): 4.33**	
		E – mmol H_2O m^{-2} s^{-1}		
	4 days	8 days	12 days	Average (ssp = S)
P. gibertii	4.17	1.62	2.76	2.85
P. foetida	3.13	3.79	1.50	2.81
P. edulis	3.83	3.20	1.87	2.97
Inter. average (I)	3.71 a	2.87 ab	2.04 b	
	SMD: 1.32	CV(%): 45.42	GA: 2.87	
	F (S): 0.05ns	F (I): 4.88*	F (SxI): 2.27ns	
		GS – mol H_2O m^{-2} s^{-1}		
	4 days	8 days	12 days	Average (ssp = S)
P. gibertii	0.25 aA	0.05 aB	0.09 aB	0.13 a
P. foetida	0.07 bA	0.09 aA	0.02 aA	0.06 b
P. edulis	0.11 bA	0.08 aA	0.04 aA	0.08 ab
Average (I)	0.14 a	0.08 b	0.05 b	
	SMD: 0.10	CV(%): 62.53	GA:0.09	
	F (S): 4.31*	F (I): 8.08**	F (SxI): 4.00*	
		Ci – μmol mol^{-1}		
	4 days	8 days	12 days	Average (ssp = S)
P. gibertii	271.16	300.83	317.08	296.36 a
P. foetida	228.08	241.33	251.33	240.25 b
P. edulis	289.41	246.00	261.66	265.69 ab
Average (I)	262.88	262.72	276.69	
	SMD: 51.86	CV(%): 19.18	GA: 267.43	
	F (S): 3.60*	F (I): 0.29ns	F (SxI): 0.74ns	
		WUE		
	4 days	8 days	12 days	Average (ssp = S)
P. gibertii	3.54	1.59	1.37	2.16
P. foetida	2.03	1.92	1.48	1.81
P. edulis	1.67	2.22	1.68	1.86
Average (I)	2.41	1.91	1.51	
	SMD: 1.00	CV(%): 50.74	GA: 1.949	
	F (S): 0.45ns	F (I): 2.52ns	F (SxI): 2.02ns	

** - significant at the 1 % probability level ($p<0.01$); * - significant at the 5 % probability level ($0.01 =< p < 0.05$); ns - not significant ($p >= 0.05$); SMD: significant minimum difference; F: f value calculated in the variance analysis; the means followed by the same letter do not differ statistically. CV: coefficient of variation; GA: general average; Small letters compare averages in the column. Capital letters compare averages in the line. Letters that are the same do not differ among themselves by Tukey's test at 5 % probability within the same factor.

the availability of water in the soil, causing a reduction in the water potential of the soil, in the water potential of the leaves and in the photosynthetic rate of passion fruit plants propagated by seeds and grafted. Cavalcante *et al*. (2001) studying physiological responses in yellow passion fruit seedlings (*P. edulis*) inoculated with arbuscular mycorrhizal fungi and subjected to water stress, found that plant watering suspension for seven days induced significant changes in diffusive resistance and leaf temperature in uninoculated passion fruit seedlings.

A statistical difference was observed only in the irrigation intervals factor, where plants irrigated after 12 days had less transpiration (E) than plants irrigated every four days, showing that the availability of water in the soil-plant-atmosphere system is crucial for the development of the plant, given that this deleterious effect is corroborated with the stomatal conductance (GS) which was also significant, where an interaction between the factors was observed, thus the correlation between the variables was significant and positive as shown in Fig. 1 and in Tab. 3. Cavalcante *et al*. (2001) found that passion fruit seedlings subjected to seven-day water stress had lower rates of transpiration compared to plants that were subjected to water stress. This water restriction can cause a significant loss in the rate of photosynthesis, as in this process water is the electron donor molecule for the formation of NADPH+ that will be used in the chemical phase of photosynthesis (Taiz & Zeiger 2006).

The species of *P. gibertii* when irrigated in the interval of 4 days showed higher stomatal conductance (GS) in relation to *P. foetida*, not differing from *P. edulis*. This greater conductance shows that the stomata present in the leaves were open, and that it allows a higher rate of CO_2 assimilation (Ci), thus guaranteeing a greater carbon fixation in the dry mass and relegating to a greater growth or accumulation of dry mass as shown in Tab. 4. The reduction in stomatal conductance can limit the CO_2 retention rate and, consequently, the concentration of CO_2 is reduced in intercellular spaces due to the consumption of CO_2 by photosynthetic activity (Daley *et al*. 1989). Paiva *et al*. (2005), studying the bean culture, observed the reduction of stomatal conductance in response to low values of soil water potential and high values of vapor pressure deficit. In this work it was verified that in both species the reduction of stomatal conductance for passion fruit seedlings started after eight days of irrigation interval, showing that in the initial phase of the crop the producer must be aware of the water restriction in the interval from 4 to 8 days, and with a requirement of approximately 954.98 mm per year and a Kc ranging from 0.42 to 1.12 (Silva & Klar 2002). These differences between plant sizes can be seen in Fig. 2.

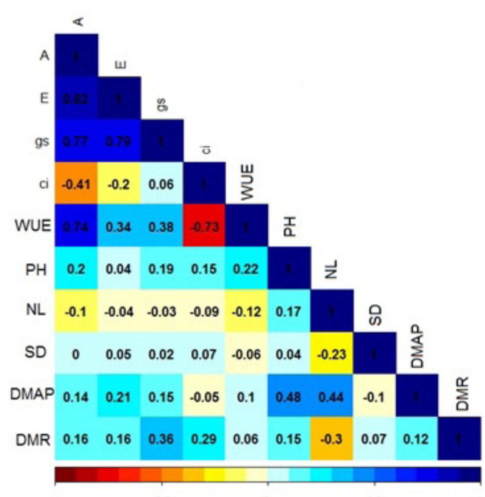

Figure 1. *r* values of Pearson correlation between the variables analyzed in the passion fruit species after the irrigation intervals.

A statistical difference was found only between the passion fruit species for the variable internal concentration of CO_2 in the substomatic chamber (Ci) where *P. foetida* had the lowest average values (Tab. 2). This result is contrary to other studies that demonstrate that this species is tolerant to water stress, however in this work it was observed that this species is sensitive to water stress since it had the lowest averages in the other variables, so it is important for the producer to pay attention for its choice as a rootstock for other species, as it demands greater attention in irrigation during its cycle.

It is worth mentioning that the internal concentration of CO_2 is important because the productivity of a plant can be analyzed as the product of intercepted solar energy and the CO_2 set over a period (Taiz & Zeiger 2006, Andrade *et al*. 2019). In general, when plants are under some type of stress, they reduce stomatal conductance and transpiration and increase water use efficiency. Under these conditions, the rate of photosynthesis is also reduced (Ferraz *et al*. 2012).

Table 3. Matrix of significant linear regressions of Pearson significant interactions of the variables analyzed in the passion fruit species after the irrigation intervals.

	Y = βo + βi.. + βnN	p value	R²
A	A = -0.6113453 + 2.34144776 E	<0.0001	0.6771
	A = 2.34140361 + 40.0860624 GS	<0.0001	0.5978
	A = 14.8202040 – 0.03249543 Ci	0.0061	0.1648
	A = 0.36360213 + 2.95748360 WUE	<0.0001	0.5516
E	E = 1.52673535 + 14.3092014 GS	<0.0001	0.6168
WUE	WUE = 1.25518227 + 0.24122932 E	0.0288	0.1140
	WUE = 1.48738171 + 4.89185180 GS	0.0180	0.1412
DMAP	DMAP = 9.16908127 + 0.03026795 PH	0.0042	0.2269
	DMAP = 10.3226178 + 0.16806260 NL	0.0078	0.1924

Table 4. Average Plant Height (PH); Number of Leaves (NL); Stem Diameter (SD); dry mass of aerial part (DMAP) and dry mass of root (DMR) of passion fruit species when cultivated in different irrigation intervals.

		Plant Height (AP)		
	4 days	8 days	12 days	Average(ssp=S)
P. gibertii	125.50	123.25	94.00	114.25 a
P. foetida	92.75	80.75	66.62	80.04 b
P. edulis	81.50	52.25	49.75	61.16 b
Average (I)	99.91 a	85.41 ab	70.12 b	
	SMD: 18.95	CV(%): 22.02	GA: 85.15	
	F (S): 24.71**	F (I): 7.57**	F (SxI): 0.71ns	
		Number of Leaves (NF)		
	4 days	8 days	12 days	Average(ssp=S)
P. gibertii	9.00	7.00	5.50	7.16 b
P. foetida	13.50	14.25	12.50	13.41 a
P. edulis	5.75	4.50	4.25	4.83 b
Average (I)	9.41	8.58	7.41	
	SMD: 3.91	CV(%): 45.67	GA: 8.47	
	F (S): 15.79**	F (I): 0.81ns	F (SxI): 0.20ns	
		Stem Diameter (SD)		
	4 days	8 days	12 days	Average(ssp=S)
P. gibertii	2.74	2.86	2.95	2.85
P. foetida	2.91	2.13	3.11	2.72
P. edulis	2.97	2.58	2.93	2.83
Average (I)	2.87	2.52	3.00	
	SMD: 0.61	CV(%): 21.67	GA: 2.80	
	F (S): 0.16ns	F (I): 1.95ns	F (SxI): 0.78ns	
		Dry Mass of Aerial Part (DMAP)		
	4 days	8 days	12 days	Average(ssp=S)
P. gibertii	12.78	13.09	10.11	11.99
P. foetida	11.78	12.62	11.35	11.90
P. edulis	12.73	11.11	10.12	11.32
Average (I)	12.43 a	12.27 ab	10.52 b	
	SMD: 1.86	CV(%): 15.71	GA: 11.74	
	F (S): 0.48ns	F (I): 3.94*	F (SxI): 0.88ns	
		Dry Mass of Root (DMR)		
	4 days	8 days	12 days	Average(ssp=S)
P. gibertii	5.27	4.88	4.96	5.04 a
P. foetida	3.77	3.92	3.17	3.62 b
P. edulis	5.34	3.72	4.42	4.49 ab
Average (I)	4.79	4.17	4.19	
	SMD: 1.09	CV(%): 24.74	GA: 4.38	
	F (S): 5.22*	F (I): 1.29ns	F (SxI): 0.82ns	

** - significant at the 1 % probability level ($p < 0.01$); * - significant at the 5 % probability level ($0.01 =< p < 0.05$); ns - not significant ($p >= 0.05$); SMD: significant minimum difference; F: f value calculated in the variance analysis; the means followed by the same letter do not differ statistically. CV: coefficient of variation; GA: general average; Small letters compare averages in the column. Capital letters compare averages in the line. Letters that are the same do not differ among themselves by Tukey's test at 5 % probability within the same factor.

This was the case in this study, when the assimilation rate of CO_2 and the transpiration were reduced with intervals of twelve days of irrigation and stomatal conductance with eight days of interval. The different species of passion fruit and the different irrigation intervals did not influence the efficiency of water use (Tab. 2).

For Plant Height (PA) a significant effect was observed individually between the factors, the species *P. gibertii* presented higher average value, and the plants had a better development with an irrigation interval of 4 days. It shows again that the factor with water restriction is the one that most influences the development of the vegetable, since the

Figure 2. Passion fruit species under different irrigation intervals. **A.** *Passiflora gibertii* with a 4-day interval; **B.** *P. gibertii* with a 8-day interval; **C.** *P. gibertii* with a 12-day interval; **D.** *P. foetida* with a 4-day interval; **E.** *P. foetida* with a 8-day interval; **F.** *P. foetida* with a 12-day interval; **G.** *P. edulis* with a 4-day interval; **H.** *P. edulis* with a 8-day interval; **I.** *P. edulis* with a 12-day interval. Bar = 30 cm.

need for water in the soil-plant-atmosphere system needs to be in line with the requirements of each species of passion fruit. With this restriction there is a lower efficiency in the photosynthetic rate and consequently a lower dry mass deposition in the vegetable.

In *P. foetida* the highest number of leaves was found, 13.41 leaves/plant, 63.98 % higher than the species *P. edulis* (Tab. 4), which had the lowest number of leaves (4.83 leaves/plant). There were no differences between the number of leaves of *P. gibertii* and *P. edulis*, which agrees with the results obtained by Nogueira Filho *et al*. (2011). The irrigation interval did not affect the number of leaves, which was not expected, as the availability of water is a limiting factor for the development of plants (Govêa *et al*. 2018). A positive correlation was found between the number of leaves and the dry mass aerial part as shown in Fig. 1 and Tab. 3. It was expected that, with the increase in the number of leaves, a greater production of dry mass of the aerial part would occur in the species *P. foetida*, however, this was not the case, as these species did not differ from each other, only the irrigation interval where plants were irrigated with an interval of 4 days showed the highest dry mass aerial part.

There were no differences for stem diameter between the species studied and neither the interference of the irrigation interval as shown in Tab. 4, corroborating with Cavichioli *et al*. (2009) who also did not observe differences for stem diameter between *P. gibertii* and *P. edulis* when they studied these species for rootstock. However, a positive interaction was found between stem diameter and dry mass aerial part (Fig. 1 and Tab. 3).

The largest dry root dry mass was found in *P. gibertii* which did not differ from *P. edulis*. The different irrigation intervals did not influence the dry root dry mass (Tab. 4). It was expected that the roots of the plants would be compromised with the restriction of water in the soil, because this organ is the first to suffer for being in contact with the soil, these damages can be observed as a reduction in its diameter or in opposite situations, that is, in flooding, there may be an expansion in its diameter due to the appearance of aerenchyma (Govêa *et al*. 2018).

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