Australian Journal of Crop Science AJCS

Aust J Crop Sci. 19(03):220-224 (2025) | https://doi.org/10.21475/ajcs.25.19.03.p146 ISSN:1835-2707

Impacts of passion fruit woodiness disease (Cowpea Aphid-Borne Mosaic Virus) on single-leaf gas exchange of *Passiflora edulis.*

Marco Antônio da Silva Vasconcellos[1], Wallace de Paula Bernado[2], Ellen Moura do Vale[2], Anne Reis Santos[2], Guilherme Augusto Rodrigues de Souza[2], Weverton Pereira Rodrigues[3], Eliemar Campostrini*[2]

[1]Department of Plant Science, Institute of Agronomy, Federal Rural University Federal of Rio de Janeiro, BR 465, km 7, Seropédica, RJ 23890-000, Brazil
[2]Plant Physiology Laboratory, LMGV, Agricultural Science and Technology Center, State University of North Fluminense, Av. Alberto Lamego, 2000, Campos dos Goytacazes, RJ 28013-602, Brazil
[3]Center of Agricultural, Natural and Literary Sciences, State University of the Tocantina Region of Maranhão (UEMASUL), Estreito, MA, Brazil

***Corresponding author: Eliemar Campostrini**

Received:* *07/09/2024

Revised:* *08/10/2024

Accepted:* *20/12/2024

Abstract: Passion fruit woodiness disease (PWD) is primarily caused by potyviruses (Cowpea Aphid-Borne Mosaic Virus) and is considered the most economically important and a limiting factor for passion fruit production in Brazil. Understanding the effects of the infectious process on the host is one of the goals of parasitism physiology. The experiment was conducted at the commercial panting in Rio de Janeiro state, Brazil, with 10 healthy plants of yellow passion fruit (*Passiflora edulis*) [control (CTL) leaf], cv. FB200 with 7 months of age (beginning of fruit production), and 10 plants with PWD were selected. Leaves of field-grown passion fruit plants infected with PWD were selected to determine its influence on single-leaf gas exchange. Leaf stomatal conductance, transpiration and net CO2 assimilation rate were reduced by PWD infection. Negative effects on single-leaf gas exchange in the yellow passion fruit supported our initial hypothesis that PWD cause impacts on stomatal conductance, transpiration, and net CO_2 assimilation rate. These results suggest that reduced growth, yield, and fruit quality in PWD-infected passion fruit plants is caused, at least partially, by reduced single-leaf gas exchange.

Keywords: Photosynthesis, stomatal conductance, transpiration, *Passiflora edulis.*
Abbreviations: A_{net} - net CO2 assimilation rate; CTL – Control; E – Transpiration rate; g_s – Stomatal conductance; LT – Leaf temperature; PPFD – Photosynthetic photon flux density; PWD - Passion fruit woodiness disease.

Introduction

The yellow passion fruit (*Passiflora edulis*) is widely distributed in tropical and subtropical areas of the world and becomes popular because of balanced nutrition and health benefits (He et al., 2020). Currently, Brazil is the largest producer of this fruit. According to the Agricultural Census from 2021, 683,993 tons of passion fruit were harvested in Brazil on 44,827 hectares, thus estimating an average yield per hectare of 15.25 tons. The value of production reached 1,533,905 million reais (IBGE, 2024).
However, the cultivation and production of passion fruit are severely affected by various pathogens, such as viruses, bacteria, and fungi. Among them, viral disease is an extremely serious disease of passion fruit and a yield-limiting factor for the crop (Chen et al., 2021). Passion fruit woodiness disease (PWD) is considered the most economically important and a limiting factor for passion fruit production in Brazil (Cerqueira-Silva et al., 2014). The main characteristic symptoms of PWD are reduced size and deformation of the plants because of shortening of the internodes; different levels of wrinkling and mosaic formation, with the possible occurrence of bubbles or blisters on the leaf surface; formation of mosaics on leaves; and woodiness and deformation of the fruits (Oliveira et al., 2013; Cerqueira-Silva et al., 2014).
When plants are attacked by pathogens, physiological and photosynthetic properties are often impaired. Infection by pathogens may lead to a decrease in photosynthesis rates and changes in the photosynthetic apparatus (Alves et al., 2011). To trigger defense mechanisms that deter pathogens by reallocating resources at the expense of growth, a reduction in photosynthetic capacity in the remaining leaf tissues may represent a 'hidden cost' of defense (Bilgin et al., 2010).
Understanding the effects of the infectious process on the host is one of the goals of parasitism physiology. Knowledge of the way in which a pathogen mobilizes and alters the physiology and growth of the host plant can help to establish the basis for disease control or management and allow for the suppression or reduction of damage to crops.

This study aimed to quantify the loss of photosynthetic capacity in virus-infected leaves, as viruses damage chloroplasts, to provide a disease restriction, supporting the decision-making process for uprooting (eradication) of plants exhibiting symptoms, since virus-infected plants exhibit reduced photosynthetic activity. Thus, we hypothesized that PWD cause negative impacts on single-leaf gas exchange.

Results

Micro-environmental data

The maximum photosynthetic photons flux density (PPFD) values were 1340.3 ± 66.38 and 1271 ± 72.72 μmol m^{-2} s^{-1}, CTL and PWD treatments at 9:00h, respectively, during the measures (Fig. 1a). In both treatments, high PPFD was at 11:00h, showing 1792.7 ± 80.73 and 1847.9 ± 24.91 μmol m^{-2} s^{-1} for CTL and PWD treatments, respectively. However, maximum PPFD measured in both treatments was at 14:00h, reaching approximately 1933.1 ± 14.97 and 1814.3 ± 68.94 μmol m^{-2} s^{-1}, for CTL and PWD, respectively. At 16:00h, a considerable PPFD reduction was observed, showing 1589.5 ± 67.03 and 1390.7 ± 57.47 μmol m^{-2} s^{-1}.

In both treatments, leaf temperature (LT) values increased similarly as PPFD (Figure 1b), and the maximum LT value was obtained 14:00h reaching 37.79 ± 0.15 and 37.94 ± 0.2° C in CTL and PWD treatments, respectively. In addition, highest relative humidity was observed in both treatments at 9:00h with 51.03 ± 0.6 and 47.45 ±1.18 % in CTL and PWD, respectively (Figure 1c).

Leaf gas-exchange measurements

The PWD showed significant reduction on A_{net} during the day (Figure 2a). The PWD caused a significant decrease of 41.5% in A_{net} compared to CTL at 9:00h (ca. 9.74 ± 0.49 vs ca. 16.65 ± 0.68 μmol CO_2 m^{-2} s^{-1}, respectively). At 11:00h, PWD decreased A_{net} values by 41% compared to CTL treatment (ca. 9.82 ± 0.54 vs ca. 16.39 ± 0.34 μmol CO_2 m^{-2} s^{-1}, respectively). At 14:00h, a decrease in C-assimilation in CTL occurred. However, PWD remained with significantly decreased (41%) in Anet values when compared to CTL treatment (ca. 9.53 ± 0.54 vs ca. 13.97 ± 0.60 μmol CO_2 m^{-2} s^{-1}, respectively). On the end of day (16:00h), a light increased in C-assimilation occurred in CTL and decreased in PWD treatment in C-assimilation when compared to 14:00h (ca. 9.03 ± 0.40 vs ca. 14.74 ± 0.72 μmol CO_2 m^{-2} s^{-1}, respectively).

The g_s values was always lower in PWD than CTL leaf in all diurnal period (Figure 2b). At 9:00h period, PWD treatment showed significantly lower g_s than CTL (± 36%) (ca. 0.16 ± 1.01 vs ca. 0.25 ± 0.01 mol H_2O m^{-2} s^{-1}). AT 11:00h and 14:00h, g_s in PWD was 0.16 ± 0.01 and 0.17 ± 0.01, while in CTL treatment was 0.21 ± 0.01 and 0.21 ± 0.01 mol H_2O m^{-2} s^{-1}. PWD treatment maintained somewhat lower values of g_s at 16:00h (ca. 0.18 ± 0.01 mol H_2O m^{-2} s^{-1}), while CTL maintained somewhat higher in g_s (± 22%) (ca 0.23 ± 0.01 mol H_2O m^{-2} s^{-1}) when compared to PWD treatment.

The PWD showed significant lower *E* than CTL treatment in all diurnal period (Figure 2b). PWD decreased E values by 30.5% compared to CTL treatment at 9:00h (ca. 4.2 ± 0.00023 vs. ca.6.0 ± 0.23 mmol H_2O m^{-2} s^{-1}, respectively). At 11:00h, *E* in PWD treatment was 6.2 ± 0.25 while CTL was ca. 6.9 ± 0.23 mmol H_2O m^{-2} s^{-1}.

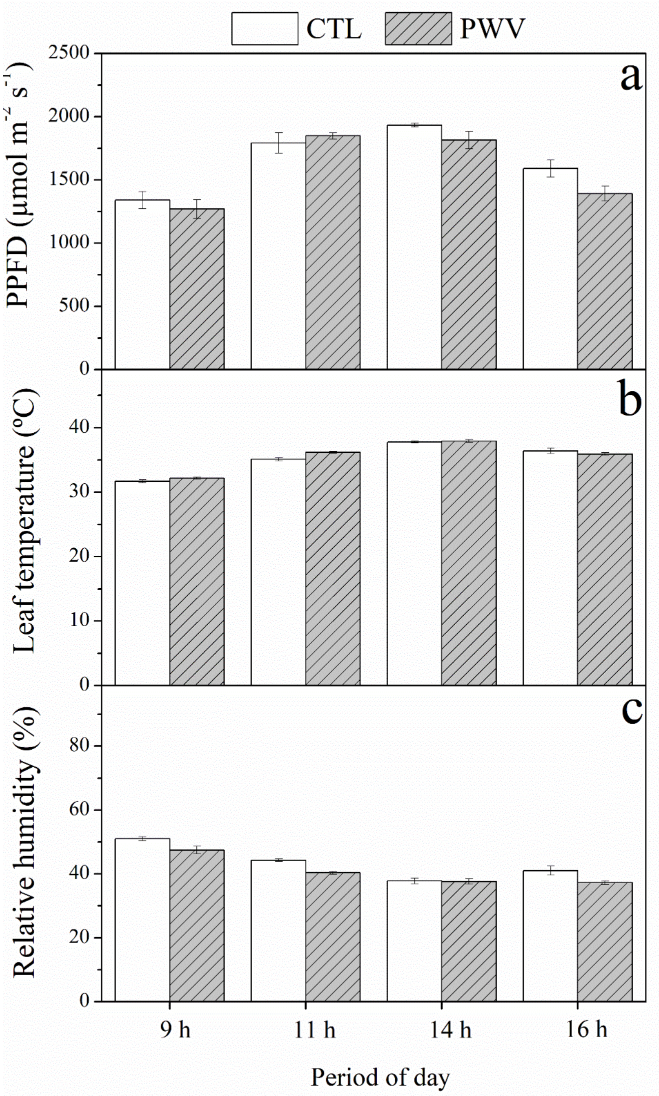

Fig 1. Add a general legend. a) PPFD (μmol m^{-2} s^{-1}) on the leaf. b) Leaf temperate (°C). c) Relative humidity (%) inside of LI-6200 chamber. Results during the measures. CTL (healthy leaf) and PWD (leaf-associated vírus).

During 14:00h, PDW maintained reduced E values compared to CTL treatments (ca 7.5. ± 0.53 vs. ca. 8.9 ± 0.39 mmol H_2O m^{-2} s^{-1}, respectively). At 16:00h, PDW caused a reduction of 17% in E values compared to CTL treatments, with values of 6.9 ± 0.30 and 8.3 ± 0.28 mmol H_2O m^{-2} s^{-1}, respectively.

Discussion

Passion fruit woodiness virus (PWD) is one of the most important phytosanitary problems which affects the passion fruit crop, causing large production losses world-wide (Santos-Jiménez et al., 2022). When infected, plants have a decreased in leaf area and fruit weight, with a decreased number of fruits, quality, and your commercial values (Nascimento et al., 2006). In the leaves, the major symptoms of disease include severe foliar mosaic, chlorotic spots, roughness, leaf blister, distortion, deformation, and irregular thickness (Peruch et al., 2009; Colariccio et al., 2018). In addition, plants infected with PWD can cause sever yield losses and reduction on plant life by 50% (Fischer and Rezende, 2008).

In this study, physiological responses in the yellow passion fruit (*Passiflora edulis*) supported our initial hypothesis that PWD causes impacts on single-leaf gas exchange. The PWD negatively effects on ± 41% on A_{net} during the all-day periods (Figure 2a). Leaf infected with PWD showing anatomical changes in the cell wall structure and/or the conformation and distribution of organelles, altering the biochemical processes and synthesis and productivity (Grove and Marsh, 2011; El-Banna et al., 2014; Xiao et al., 2016). Consequently, a reduction in CO2 assimilation was observed, due to dependence to stomatal conductance, and green leaf area, because photosynthesis use the sunlight energy intercepted for leaves for convert CO_2 in carbohydrates, being responsible for 90% of the biomass and yield of crops (Zelitch, 1982; Simkin et al., 2019). In fact, PWD shows a bubbles or blisters occurrence on the leaf surface, which can affect the net CO_2 assimilation rate (Rezende, 2006).
Stomatal conductance ($g_{s)}$ is quantified as the capacity for exchanging CO_2 and water vapor, typically normalized by leaf area (Xiong and Flexas, 2020). Our study showed that CTL plants had higher g_s than PWD infected plants (Figure 2b), which supports their higher A_{net} values in CTL plants (Figure 2a). The stomata can differ in density and size, in addition, their distribution can generate high g_s impact (de Boer et al., 2016; Muir, 2018; Drake et al., 2019). We suggest that PWD leaves showed the greatest morphological and anatomical changes, with an important impact on leaf expansion, stomatal distribution, and consequently metabolism and production of photoassimilates (Fernández-Calvino e al., 2014; Murray et al., 2016). Thus, gs is the main key physiological parameter that can affect the productivity under both optimum growth and abiotic conditions (Roche et al., 2015; Rahnama et al., 2010). In addition, any change in leaf morphological traits, mainly stomatal density and distribution, and epidermal parameters can significantly affect single-leaf gas exchange and their relationships with an environmental factor (Woodward, 1987; Nilson and Assmann, 2007).
The morphological and anatomical leaves changes of PWD cause a stronger reduction on g_s, consequently a significantly reduction in net CO_2 assimilation rate and transpiration per unit leaf area (*E*) (Figure 2c), since the balancing between carbon assimilation and transpiration rate depends on the fine-tuning of the stomatal aperture (Hasanuzzaman et al., 2023). Finally, our results suggest that PDW can reduce mineral nutrient uptake, biomass production, as well as passion fruit yield.

Materials and methods

Experimental site and species description

The experiment was conducted in a commercial panting of yellow passion fruit (*Passiflora edulis* FB200 with 7 months of age, at the beginning of fruit production), in Vera Cruz (22º13'11" S and 49º49'10" W, at 628 m de altitude), Rio de Janeiro state, Brazil. On April 1999, 10 healthy plants of yellow passion fruit [control (CTL) leaf], and 10 plants with passion fruit woodiness virus (PWD) were selected after showing the typical visual symptoms of of mosaic virus. Plants were set at a spacing of 3.5 m x 3.0 m in topography with mild slopes and sandy-textured soil classified as a Dark Red Latosol (Oxisol) prepared, fertilized and irrigated (≈ 6 and 10 L H_2O $plant^{-1}$ day^{-1})

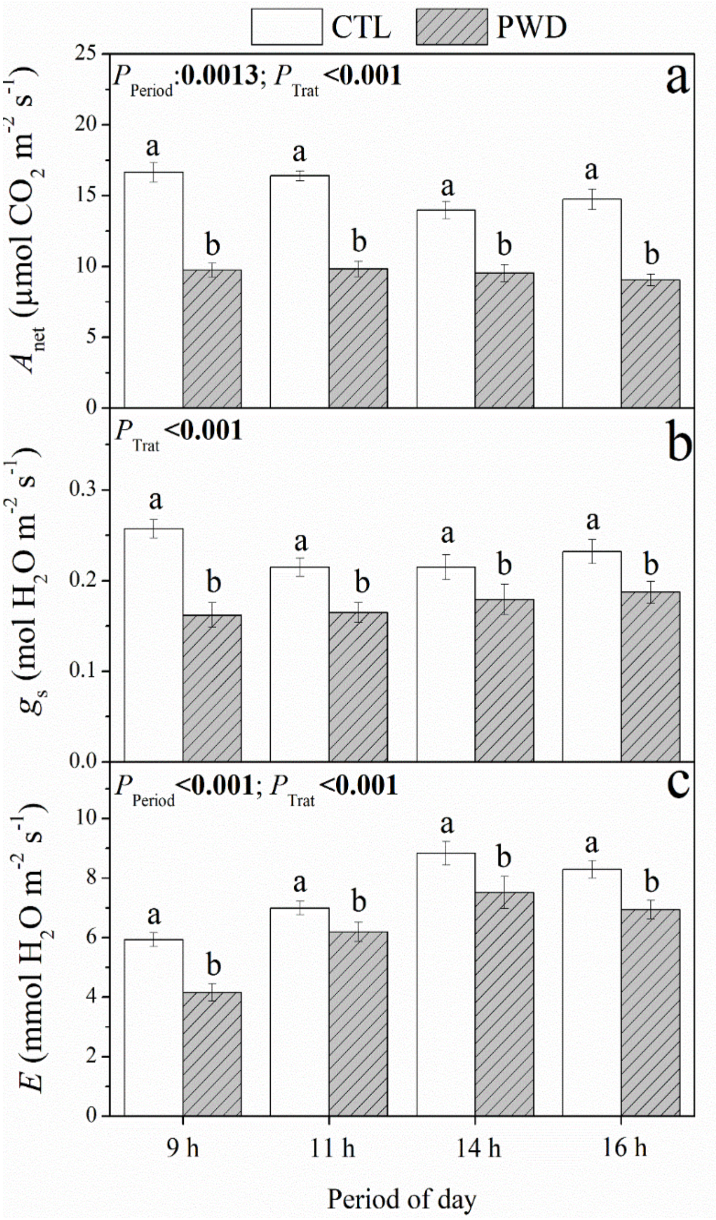

Fig 2. a) Net CO_2 assimilation rate (A_{net}). b) Stomatal conductance (g_s). c) Transpiration (*E*). (n=10). CTL (healthy leaf) and PWD (leaf-associated virus) treatments. The ANOVA P-vales are shown (n=10). P < 0,05 was considered significant. Vertical bars represent estimated S.E.

According to (Piza Júnior et al., 1997). The remaining cultural practices were carried out in accordance with Rizzi et al. 1998.

Virus detection

The presence of virus in yellow passion fruit plants was confirmed according by Inoue et al. (1995). For this, four isolates of PWD were collected from yellow passion fruit and characterized. Filamentous virus particles, typical of potyviruses, were seen in leaf dip preparations from infected samples. In many leaf parenchyma cells of infected plants some chloroplasts appeared with different degrees of disorganization. Serological reactions using antiserum against a Brazilian isolate of PWD gave positive results.

Table 1. Average temperature (°C), Minimum temperature (°C), Maximum temperature (°C), Rain (mm), Humidity (%), Rainy days (d); Photoperiod (h). Date obtained in 1998 and 1999.

	January	February	March	April	May	June	July	August	September	October	November	December
Average temperature °C)	26.4	26.6	26.7	26	25.1	24.2	23.5	23.4	24.1	25	25.7	26.3
Minimum temperature (°C)	24.8	25	25.1	24.5	23.8	23	22.3	22.2	22.7	23.6	24.2	24.7
Maximum temperature (°C)	28.3	28.4	28.5	27.5	26.4	25.5	24.9	24.9	25.7	26.9	27.8	28.4
Rain (mm)	69	72	94	162	189	158	129	95	75	71	70	51
Umidity (%)	78%	78%	80%	82%	82%	81%	79%	78%	77%	78%	78%	78%
Rainy days (d)	12	13	15	17	17	17	17	16	13	11	10	9
Photoperiod (h)	9.6	9.3	8.8	8.1	7.6	7.6	7.7	7.9	8.1	8.5	9	9.5

Indirect ELISA demonstrated that all isolates were serologically related to each other and also to *Cowpea aphid-borne mosaic virus* (CABMV). Leaf samples were collected and subjected of mechanical inoculation in other individuals of yellow passion fruit and cowpea (*Vigna unguiculata*), Virus infection was confirmed by serology and inoculation of indicator plants. The four viral isolates obtained from passionfruit were capable of infecting several plant species, although a difference in the intensity of symptoms induced by each isolate was observed in some hosts. Indirect ELISA method showed that the passion fruit isolates were serologically related to each other, and also to the potyvirus (CABMV).

Leaf gas-exchange measurements

Leaf gas exchange was determined at 9 h, 11 h, 14 h, and 16 h, in a mature leaf (6^{th} leaf from apex) from each plant, using an infrared gas analyzer (LI-6200, Li-Cor Inc., Lincoln, NE). The CO_2 concentration was ≈370 µmol mol^{-1} inside the leaf chamber. The leaf area was 0,00068 m² (volume chamber was 125 mL). Incident photosynthetic photons flux density (PPFD) on the chamber, leaf temperature and relative humidity inside the chamber were recorded. Net CO_2 assimilation rate (*A*net, µmol CO_2 m^{-2} s^{-1}), stomatal conductance (g_s, mol H_2O m^{-2} s^{-1}), and transpiration rate (*E*, mmol H_2O m^{-2} s^{-1}) were measured.

Experimental design and statistical analysis

The experiment was carried out using a completely randomized design, in a 2 x 4 double factorial scheme for the evaluations carried with the leaf gas exchange measurements, considering 2 treatments (CTL, PWD), four periods (9 h, 11 h, 14 h and 16 h). Ten repetitions were performed for each treatment, totaling 20 plants, with measurements taken from one leaf per plant. The data were submitted to analysis of variance (ANOVA). Least-squares means and respective statistical errors (S.E.) were estimated from the fitted GLMs, the S.E. derived from GLM ANOVA being a residual mean square error for each response variables. Statistical analyses were performed using a R version 4.3.2, package 'ExpDes' (R Core Team, 2021). Graphs were generated using a OriginPro 2017 software.

Conclusion

Negative effects on single-leaf gas exchange in yellow passion fruit supported our initial hypothesis that PWD cause impacts morphological and anatomical on the leaves. In addition, reduction in stomatal conductance, transpiration, and net CO2 assimilation rate were also observed. These results suggest that reduced growth, yield, and fruit quality common in PWD-infected passion fruit plants is caused, at least partially, by reduced single-leaf gas exchange.

Acknowledgments

This work was supported by CNPq Researcher fellowships to: EC (303166/2019-3); Fundação Carlos Chagas de Apoio à Pesquisa do Estado do Rio de Janeiro (FAPERJ) grants (E-26/202.759/2018, E-26/210.309/2018, and E-26/210.037/2020, EC)

References

Alves AA, Guimarães LM da S, Chaves AR de M, DaMatta FM, Alfenas AC (2011) Leaf gas exchange and chlorophyll a fluorescence of *Eucalyptus urophylla* in response to *Puccinia psidii* infection. Acta Physiologiae Plantarum. 33:1831–1839.

Bilgin DD, Zavala JA, Zhu JIN, Clough SJ, Ort DR, DeLUCIA EH (2010) Biotic stress globally downregulates photosynthesis genes. Plant Cell Environ. 33:1597–1613.

Cerqueira-Silva CBM, Conceição L, Souza AP, Corrêa RX (2014) A history of passion fruit woodiness disease with emphasis on the current situation in Brazil and prospects for Brazilian passion fruit cultivation. Eur J Plant Pathol. 139:261–270.

Chen L, Sun D, Zhang X, Shao D, Lu Y, An Y (2021) Transcriptome analysis of yellow passion fruit in response to cucumber mosaic virus infection. PLoS One. 16:e0247127.

Colariccio A, Garcez RM, Rodrigues LK, Eiras M, Peruch LAM, Chaves ALR (2018) Doenças causadas por vírus na cultura do maracujazeiro (*Passiflora edulis*). In: Peruch LAM, Schroeder AL (ed) Maracujazeiro-azedo: polinização, pragas e doenças, 1st edn. Epagri, Florianópolis. 1.

de Boer HJ, Price CA, Wagner-Cremer F, Dekker SC, Franks PJ, Veneklaas EJ (2016) Optimal allocation of leaf epidermal area for gas exchange. New Phytol. 210:1219–1228.

Drake PL, de Boer HJ, Schymanski SJ, Veneklaas EJ (2019) Two sides to every leaf: water and CO_2 transport in hypostomatous and amphistomatous leaves. New Phytol. 222:1179–1187.

El-Banna OHM, Awad MA, Abbas MS, Waziri HM, Darwish HS (2014) Anatomical and ultrastructural changes in tomato and grapevine leaf tissues infected with *Tomato ringspot virus*. Egypt J Virol. 11:102–111.

Fernández-Calvino L, Osorio S, Hernández ML, Hamada IB, del Toro FJ, Donaire L, Yu A, Bustos R, Fernie AR, Martínez-Rivas JM, Llave C (2014) Virus-induced alterations in primary metabolism modulate susceptibility to *Tobacco rattle virus* in Arabidopsis. Plant Physiol. 166:1821–1838.

Fischer IH, Rezende JAM (2008) Diseases of passion flower (*Passiflora* spp.). Pest Technol. 2:1–19.

Grove J, Marsh M (2011) The cell biology of receptor-mediated virus entry. J Cell Biol. 195:1071–1082.

Hasanuzzaman M, Zhou M, Shabala S (2023) How does stomatal density and residual transpiration contribute to osmotic stress tolerance? Plants. 12:494.

He X, Luan F, Yang Y, Wang Z, Zhao Z, Fang J, Wang M, Zuo M, Li Y (2020) *Passiflora edulis*: An insight into current researches on phytochemistry and pharmacology. Front Pharmacol. 11:617.

IBGE (2024) Produção Agropecuária. Available in: https://www.ibge.gov.br/explica/producao-agropecuaria/maracuja/br. Accessed 24 October 2022.

Inoue AK, Mello RN, Nagata T, Kitajima EW (1995) Characterization of passion fruit woodiness virus isolates from Brasília and surrounding region, Brazil. Fitopatol Bras. 20:479–487.

Lin P-A, Chen Y, Ponce G, Acevedo FE, Lynch JP, Anderson CT, Ali JG, Felton GW (2022) Stomata-mediated interactions between plants, herbivores, and the environment. Trends Plant Sci. 27:287–300.

Muir CD (2018) Light and growth form interact to shape stomatal ratio among British angiosperms. New Phytol. 218:242–252.

Murray RR, Emblow MS, Hetherington AM, Foster GD (2016) Plant virus infections control stomatal development. Sci Rep. 6:1–7.

Nascimento AVS, Santana EN, Braz ASK, Alfenas PF, Pio-Ribeiro G, Andrade GP, De Carvalho MG, Murilo Zerbini F (2006) Cowpea aphid-borne mosaic virus (CABMV) is widespread in passionfruit in Brazil and causes passionfruit woodiness disease. Arch Virol. 151:1797–1809.

Nilson SE, Assmann SM (2007) The control of transpiration. Insights from Arabidopsis. Plant Physiol. 143:19–27.

Oliveira EJ de, Soares TL, Barbosa C de J, Santos-Filho HP, Jesus ON de (2013) Disease severity from passion fruit to identify sources of resistance in field conditions. Rev Bras Frutic. 35:485–492.

Peruch LAM, Schroeder AL, Colariccio A, Guimarães L, Chagas CM (2009) Doenças do maracujazeiro amarelo. Epagri, Florianópolis. 145.

Piza Júnior CT, Quaggio JA, Meletti LMM, Silva JR, São José AR, Karati R (1997) Maracujá. In: Raij B, Cantarella H, Quaggio JA, Furlani AMC (ed) Recomendação de adubação e calagem para o Estado de São Paulo, 2nd edn. Instituto Agronômico/Fundação IAC, Campinas. 100.

R Core Team (2020) R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna.

Rahnama A, James RA, Poustini K, Munns R (2010) Stomatal conductance as a screen for osmotic stress tolerance in durum wheat growing in saline soil. Funct Plant Biol. 37:255–263.

Rezende JAM (2006) Práticas culturais para prevenção e convivência com as viroses do maracujazeiro. In: Sampaio AC, Fumis TF, Rossi AD, Almeida AM, Garcia MJM (ed) Manejo no controle do vírus do endurecimento dos frutos (PWV) do maracujazeiro, 1st edn. Multipress, Jaboticabal. 1.

Rizzi LC, Rabello LR, Morozini Filho W, Savazaki ET, Kavati R (1998) Cultura do maracujá azedo. CATI, Campinas.

Roche D (2015) Stomatal conductance is essential for higher yield potential of C_3 crops. Crit Rev Plant Sci. 34:429–453.

Santos-Jiménez JL, de Barros MC, Vidal AH, Ribeiro SG, Bergter EB, Vaslin MFS (2022) A fungal glycoprotein mitigates passion fruit woodiness disease caused by *Cowpea aphid-borne mosaic virus* (CABMV) in *Passiflora edulis*. Biocontrol. 67:75–87.

Simkin AJ, López-Calcagno PE, Raines CA (2019) Feeding the world: improving photosynthetic efficiency for sustainable crop production. J Exp Bot. 70:1119–1140.

Woodward FI (1987) Stomatal numbers are sensitive to increases in CO_2 from pre-industrial levels. Nature. 327:617–618.

Xiao Y, Tholen D, Zhu XG (2016) The influence of leaf anatomy on the internal light environment and photosynthetic electron transport rate: exploration with a new leaf ray tracing model. J Exp Bot. 67:6021–6035.

Zelitch I (1982) The close relationship between net photosynthesis and crop yield. Bioscience. 32:796–802.