Research, Society and Development, v. 9, n. 12, e10891210862, 2020 (CC BY 4.0) | ISSN 2525-3409 | DOI: http://dx.doi.org/10.33448/rsd-v9i12.10862 Initial growth of clonal seedlings of *Passiflora mucronata* genotypes in response to paclobutrazol concentrations Crescimento inicial de mudas clonais de genótipos de *Passiflora mucronata* submetidos a doses de paclobutrazol El crecimiento inicial de las plántulas clonales de *Passiflora mucronata* genotipos en respuesta a las concentraciones de paclobutrazol

Received: 12/00/2020 | Reviewed: 12/00/2020 | Accept: 12/13/2020 | Published: 12/14/2020

Patrick Alves de Oliveira

ORCID: https://orcid.org/0000-0003-0077-4199 Federal University of Espírito Santo, Brazil E-mail: patrickalvesdeoliveira@hotmail.com Ariany das Graças Teixeira ORCID: https://orcid.org/0000-0001-8742-521X Federal Institute of Education, Science and Technology of Espírito Santo, Brazil E-mail: ariany.teixeira@ifes.edu.br **Guilherme Bravim Canal** ORCID: https://orcid.org/0000-0002-2691-1270 Federal University of Espírito Santo, Brazil E-mail: guilhermebravim@hotmail.com Paula Aparecida Muniz de Lima ORCID: https://orcid.org/0000-0003-1601-1786 Federal University of Espírito Santo, Brazil E-mail: aluap-lima@hotmail.com Gardênia Rosa de Lisbôa Jacomino ORCID: https://orcid.org/0000-0002-6375-629X Federal University of Espírito Santo, Brazil E-mail: gardenialisboa@gmail.com **Rodrigo Sobreira Alexandre** ORCID: https://orcid.org/0000-0002-5248-6773 Federal University of Espírito Santo, Brazil E-mail: rodrigosobreiraalexandre@gmail.com

José Carlos Lopes

ORCID: https://orcid.org/0000-0002-4880-0547 Federal University of Espírito Santo, Brazil E-mail: jcufes@bol.com.br

Abstract

Paclobutrazol (PBZ) is a plant growth regulator of the triazole group that can block the biosynthesis of gibberellic acid, resulting in reduced plant height and increased stem diameter. This study aimed to evaluate the effect of different paclobutrazol concentrations on seedling quality of Passiflora mucronata Lam. Two Passiflora mucronata genotypes were used, one resistant (G5) and one tolerant (G7) to fusariosis, prepared as cuttings treated with PBZ. The experimental design was completely randomized, in a 2 x 4 factorial arrangement corresponding to two genotypes (G5 and G7) and four PBZ concentrations (0, 5, 10, and 15 mg plant⁻¹), with three replications of four plants. Data were subjected to analysis of variance, regression analysis, and the F-test, followed by Pearson's correlation test between variables. PBZ promoted an increase in the plagiotropic shoot diameter and the leaf area index of genotypes G5 and G7, in addition to increased shoot length in genotype G5 and increased number of leaves in genotype G7. PBZ also resulted in increased photosynthesis and stomatal conductance. There was a positive correlation for genotype G5, between the plagiotropic shoot diameter and the leaf transpiration rate, and for genotype G7, between the plagiotropic shoot diameter and the number of leaves. Genotypes G5 and G7 showed different phenotypic responses when subjected to PBZ doses, highlighting the intraspecific divergence of the species.

Keywords: Passifloraceae; Passion fruit; Wild species; Triazole.

Resumo

O paclobutrazol (PBZ) é um regulador de crescimento de plantas do grupo dos triazois, capaz de bloquear a biossíntese do ácido giberélico, acarretando a redução da altura e aumento da espessura da haste da planta. Objetivou-se estudar o efeito de diferentes doses de paclobutrazol sobre a qualidade das mudas de *Passiflora mucronata* Lam. Foram utilizados dois genótipos de *Passiflora mucronata*, G5 (resistente) e G7 (tolerante) à fusariose, oriundos de estaquia tratados com PBZ. O delineamento experimental utilizado foi inteiramente casualizado, em esquema fatorial 2x4, dois genótipos (G5 e G7) e quatro concentrações de

PBZ (0; 5; 10 e 15 mg planta⁻¹) com três repetições de quatro plantas. Os dados foram submetidos a uma análise de variância geral, de regressão e teste de F, e realizado um teste de correlação linear de Pearson entre as variáveis. O PBZ promoveu aumento do diâmetro do broto plagiotrópico e do índice de área foliar nos genótipos G5 e G7; aumentou o comprimento dos brotos do genótipo G5 e o número de folhas do genótipo G7. O PBZ resultou na elevação da fotossíntese e condutância estomática. Houve correlação positiva para o genótipo G5 entre o diâmetro do broto plagiotrópico e a taxa de transpiração foliar e para o genótipo G7 entre a variável do diâmetro do broto plagiotrópico e o número de folhas. Os genótipos G5 e G7 apresentaram respostas fenotípicas diferentes quando submetidas às doses de PBZ, proporcionando divergência intraespecífica da espécie.

Palavras-chave: Passifloraceae; Maracujá; Espécie silvestre; Triazol.

Resumen

Paclobutrazol (PBZ) es un regulador de crecimiento vegetal del grupo triazois que puede bloquear la biosíntesis del ácido giberleico, lo que resulta en una reducción en la altura y un mayor espesor del tallo de la planta. El objetivo de este estudio fue estudiar el efecto de diferentes dosis de paclobutrazol en la calidad de las plántulas Passiflora mucronata Lam. Se utilizaron dos genotipos de Passiflora mucronata, G5 (resistente) y G7 (tolerante) a fusariosis, preparado como esquejes tratados con PBZ. El diseño experimental fue completamente aleatorizado, en una disposición factorial de 2 x 4 correspondiente a dos genotipos (G5 y G7) y cuatro concentraciones de PBZ (0, 5, 10 y 15 mg planta⁻¹), con tres repeticiones de cuatro plantas. Los datos fueron sometidos a análisis de varianza, análisis de regresión y la prueba F, seguido de la prueba de correlación de Pearson entre variables. El PBZ promovió un aumento en el diámetro del brote plagiotrópico y el índice de área de la hoja de los genótipos G5 y G7; aumentó la longitud de los brotes del genotipo G5 y el número de hojas del genotipo G7. PBZ también resultó en un aumento de la fotosíntesis y la conductancia estomatal. Hubo una correlación positiva para el genotipo G5 entre el diámetro del brote plagiotrópico y la tasa de transpiración de la hoja, y para el genotipo G7 entre el diámetro del brote plagiotrópico y el número de hojas. Los genotipos G5 y G7 mostraron diferentes respuestas fenotípicas cuando se sometieron a dosis de PBZ, proporcionando la divergencia intraespecífica de la especie.

Palabras clave: Passifloraceae; Fruta de la pasión; Especies silvestres; Triazole.

1. Introduction

Brazil stands out as the leading passion fruit producer and consumer in the world (*Passiflora* spp.), with a production of 593,429 tons obtained in 41,584 hectares (Ibge, 2019), mainly cultivated by smallholder farmers in small and medium-sized properties (Furlaneto et al., 2014; Santos et al., 2016). Passion fruit yield, however, is still low (14.10 t ha⁻¹) due to the use of seeds of unknown genetic origin, coming from fruits produced in commercial orchards, in addition to the use of inappropriate crop management technologies associated with the occurrence of phytosanitary problems that have accentuated with crop expansion, especially the occurrence of soil fungi (Faleiro & Junqueira, 2016; Ibge, 2019; Flora Do Brasil, 2020).

Passiflora mucronata Lam., found in the coastal restinga vegetation of the states of Espírito Santo and Rio de Janeiro (Magnago et al., 2011; Garbin et al., 2012), is a species of high agronomic interest for investigations due to its resistance to diseases such as bacterial leaf blight, fruit and branch anthracnose, and microorganisms such as *Fusarium* (Correia, 2019). In this context, the species becomes a viable alternative as a control method for resistance detection and studies on graft compatibility with commercial species for use as rootstocks (Alexandre et al., 2013; Oliari et al., 2016; Schmildt et al., 2018). Grafting is a form of vegetative propagation that, using resistant/tolerant rootstocks, such as *P. nitida*, *P. gibertii*, *P. setacea*, and *P. alata*, allows establishing technically superior orchards compared with those formed by seeds, with greater disease and premature plant death control, enabling the multiplication of plants with better quality fruits and increased productivity, in addition to obtaining more homogeneous orchards with increased resistance to pests and diseases (Ruggiero; Corrêa, 1980), especially considering that, in areas under crop succession and irrigation, the decrease in yield caused by Fusarium wilt has been increasing (Toledo-Souza et al., 2012).

In a study conducted with mini-grafting, which consists of grafting apical segments from adult plants on young rootstocks, *P. edulis* f. *flavicarpa* was grafted on *P. mucronata* rootstocks, with an 80% success rate (Alexandre et al., 2013). In another study, the *P. edulis* f. *flavicarpa* scion grafted on *P. mucronata* resulted in more than 90% success (Morgado et al., 2015). Likewise, the mini-grafting of *P. edulis* on *P. mucronata* resulted in an 89% success rate (Oliari et al., 2016).

However, despite the viability of grafting *P. edulis* on *P. mucronata*, the stem of *P. mucronata* has a smaller diameter than other Passifloraceae commercial species (Mauri et al.,

2020), which may hinder the growth of the commercial species due to their likely greater radial expansion, making the grafting process unfeasible. However, for better use of the species for grafting, it is necessary to use plants with larger diameters, justifying the need to study alternatives to modify the structure of the stem in *P. mucronata*, such as stem thickening, among which is the use of growth regulators.

Among these growth regulators, paclobutrazol (PBZ) or β -[4(chlorophenyl) methyl-1H-1,2,4,- triazole-1-ethanol] is one of the triazoles that has been used in agricultural crops, acting as an inhibitor of gibberellin biosynthesis and effectively promoting root formation in several species, such as tomato (*Solanum lycopersicum* L.) (Silva & Faria Junior, 2011) and wheat (*Triticum aestivum* L.) (Peng et al., 2014; Wang et al., 2014; Tesfahun & Menzir, 2018). PBZ can block the biosynthesis of gibberellic acid, resulting in reduced height and increased stem thickness (Berova & Zlatev, 2000; Téllez et al., 2020).

However, the effect varies according to the developmental stage of the plant, the product concentration, species, cultivar, time, and type of application (Rademacher, 2000; Mabvongwe et al., 2016; Oliveira et al., 2020). In *Solanum lycopersicum* L., PBZ application resulted in reduced plant height, increased stem diameter and leaf number, and altered root architecture (Pal et al., 2016), while in *Solanum tuberosum* L., PBZ application increased stem diameter by 74% (Mabvongwe et al., 2016).

It is then verified that PBZ use can assist in increasing stem thickness in *P. mucronata*, allowing the grafting of other commercial species. Thus, this study aimed to evaluate the effect of different paclobutrazol concentrations on seedling quality of genotypes of *Passiflora mucronata*.

2. Material and Methods

The study was conducted from January to May 2020 in a plant nursery at the Center of Agricultural Sciences and Engineerings of the Federal University of Espírito Santo (CCAE/UFES), in Alegre, ES, Brazil. Two *P. mucronata* genotypes were used in the experiment, originated from the Cedro Farm, located in the municipality of Jaguaré, in the northern region of the state of Espírito Santo. The genotypes were grown in a vertical shoot positioning system in the experimental area of CCAE/UFES (20° 45'50" S and 41° 31' 58" W, 250 m) and in the Ponte da Braúna Farm (20° 45' S and 41° 29' W, 138 m), Rive district,

Alegre, ES. According to the Köppen international classification, the climate of the region is classified as Cwa, humid hot tropical, with cold and dry winters (Inmet, 2020).

Two genotypes were used, namely G5 (resistant) and G7 (tolerant), respectively resistant and tolerant to *Fusarium solani* and *Fusarium oxysporum* f. sp. *passiflorae* (Correia, 2019), originated from parent plants of *Passiflora mucronata* Lam. with five years of age grown in a vertical shoot positioning system. The herbaceous cuttings, each with two buds and measuring 10 cm, were prepared by removing the leaves, cutting the upper portion in a straight cut above the bud, and the lower portion in a bevel cut in the opposite position to a bud. Subsequently, the cuttings were treated with an aqueous solution in their basal portion for 10 seconds, with the application of 1,000 mg L⁻¹ of indolebutyric acid (IBA) (Alexandre et al., 2014), and planted in 50 cm³ polyethylene tubes filled with the Bioplant[®] substrate, being kept in a plant nursery provided with a mist irrigation system. After 60 days of planting, the substrate, acclimatized for seven days under a polyolefin fabric (50% shading), and selected according to the size of the shoots, which were standardized in a 20 cm length, keeping only one shoot per plant.

After 30 days, the plants were treated with different concentrations of the paclobutrazol (PBZ) plant regulator $(0, 5, 10, \text{ and } 15 \text{ mg plant}^{-1})$, and applied via soil.

The experimental design was completely randomized, in a 2 x 4 factorial arrangement corresponding to two genotypes (G5 and G7) and four paclobutrazol concentrations (0, 5, 10, and 15 mg plant⁻¹) with three replications of four plants. Based on the work carried out by Siqueira et al., (2008) and França et al., (2018).

At 45 days after PBZ application, the following variables were analyzed: plagiotropic shoot length (cm) – (CP) - measured from the base to the top of the shoot using a measuring tape; orthotropic shoot diameter (cm) – (DO) - measured at two centimeters from the base of the cutting; plagiotropic shoot diameter (cm) – (DP) - measured at two centimeters from the base of the shoot; leaf area index (cm²) – (IAF) - obtained with a millimeter ruler by measuring the largest leaf blade width (L) and determined by the model: AFE = 1.8963 L^{1.7275} (Schmildt et al., 2017); and the number of leaves per plant – (NF). The following physiological variables were also measured: net CO₂ assimilation rate (A, µmol CO₂ m⁻² s⁻¹), stomatal conductance (Gs, mol H₂O m² s⁻¹), measured with an infrared gas analyzer (IRGA Licor 6800XT). The physiological analyses with the IRGA were conducted from 9:00 a.m. to

12:00 p.m., on cloudless days, by sampling intermediate, three fully expanded leaves without any visual anomaly per plant. The photosynthetically active radiation was standardized under an artificial saturating light source of 1,000 μ mol photons m⁻² s⁻¹, and the CO₂ concentration within the chamber was established at 400 ppm.

The data were subjected to regression analysis due to the high coefficient of determination (R^2) and the significance of all regression coefficients. In all tests, p <0.01 was used as the significance value. The free *software* R was used for all statistical analyses (R Core Team, 2020).

3. Results and discussion

It was found that the length of the shoots of genotype G5 did not differ statistically between PBZ doses, showing a mean of 28.33 centimeters. However, genotype G7 showed a cubic behavior; initially increasing up to the dose of 5.0 mg plant⁻¹, when it reached the highest shoot length value. However, with the further increase of PBZ doses, shoot size decreased along with internode elongation, without affecting the number of leaves (Figure 1A). This behavior suggests that high PBZ doses are associated with a reduction in plant height, with an antagonistic and inhibitory action on the biosynthesis of gibberellins (GAs), affecting shoot elongation and corroborating the results obtained in *Eragrostis tef* (Zucc.) Trotter (Tesfahun & Menzir, 2018).

For the orthotropic shoot diameter (DO), there was no significant interaction between genotypes G5 and G7 and between PBZ concentrations within each genotype, with a mean of 0.34 and 0.35 cm, respectively.

The plagiotropic shoot diameter (DP) of genotypes G5 and G7 was significantly affected by the PBZ doses, with quadratic growth. There was an increase in diameter with PBZ application, with the maximum efficiency achieved at the concentrations of 12.75 and 14.75 mg plant⁻¹, determining diameters of 0.1284 and 0.1519, with an increase of 33.88 and 40.12%, respectively (Figure 1B). This growth can be explained by the plant growth regulating properties of PBZ, which acts by altering the levels of plant hormones such as gibberellins, abscisic acid (ABA), and cytokinins (Fletcher & Hofstra, 1990). PBZ acts as a gibberellin antagonist, which, besides reducing cell elongation, also increases shoot diameter and lignin accumulation (Pal et al., 2016; Rademacher, 2018). Thicker shoots favor maintenance and the production phase of the plant and act against the possible rupturing

caused by abiotic factors such as wind and rain and plant handling during crop management practices, constituting an essential characteristic that facilitates grafting.



Figure 1. A. Plagiotropic shoot length (cm); B. Plagiotropic shoot diameter (cm);

**(p≤0.001); ^{ns}not significant; Source: Elaborated by authors (2020).

The leaf area indexes (IAF) between genotypes G5 and G7 did not differ significantly in response to the PBZ doses. A quadratic adjustment was observed for the genotypes, with the maximum response at the PBZ dose of 7.89 mg plant⁻¹, resulting in an IAF of 14.03 cm², 20.29% higher than the control (Figure 2A). These results disagree with those found by Benett et al. (2014), who observed a reduction in the leaf area index (IAF) of plants treated with PBZ. However, with the increase in the PBZ doses, the leaf area index decreased. The leaf area index provides an indication of the photosynthetic surface, allowing to obtain an essential indicator for the compensation of plant responses to environmental factors (Lopes et al., 2004).





**(p≤0.001); ^{ns}not significant; Source: Elaborated by authors (2020).

The number of leaves (NF) of genotype G5 showed no statistical difference between the PBZ doses applied, with a mean of 8.5 leaves plant⁻¹ (Figure 2B). These results suggest that PBZ, applied at low concentrations, does not change the number of leaves. However, leaf emission is reduced at higher doses (Braun & Garth, 1986; Jiao et al., 1986; Sankhla et al., 1986; Vu & Yelenosky, 1992; Siqueira et al., 2008), an opposite behavior to that of genotype G7, which showed an increasing linear growth of this variable with PBZ application (Figure 2B). Different phenotypic responses were observed in the studied genotypes (G5 and G7), suggesting the existence of intraspecific divergence between populations of *P. mucronata*, corroborating the results observed by França et al. (2018).

Photosynthesis (*A*) showed a cubic response in the plants of genotype G7, while the plants of genotype G5 showed quadratic growth, increasing up to the maximum efficiency dose of 7.97 plant⁻¹, with maximum values of 3.99 μ mol CO₂ m⁻² s⁻¹, 23.91% higher than the control. However, as the dose increased, photosynthesis reduced (Figure 3A). This may be

related to the effect of PBZ in increasing the levels of endogenous cytokinins, consequently resulting in increased chloroplast differentiation, increased biosynthesis and lower degradation of chlorophyll in the leaves, and delayed degradation of the ribulose 1.5-bisphosphate carboxylase-oxygenase (rubisco) enzyme, optimizing the carboxylation capacity of the plant (Skowron et al., 2016; Moura et al., 2017).

Figure 3. A. Net CO₂ assimilation rate (A, µmol CO₂ m⁻² s⁻¹); **B.** Stomatal conductance (g_s , mol H₂O m⁻² s⁻¹); **C.** Transpiration rate (E, mmol H₂O m⁻² s⁻¹).





**(p≤0.001); ^{ns}not significant; Source: Elaborated by authors (2020).

The stomatal conductance (g_s) of genotypes G5 and G7 showed opposite cubic responses.

The leaf transpiration rate (*E*) of genotype G5 showed a similar quadratic response to that of g_s with growing PBZ doses up to 14.35mg plant⁻¹, with a transpiration rate (*E*) of 3.75 mmol H₂O m⁻² s⁻¹, 27.55% higher than the control (Figure 3C). This result may reflect a better soil-plant-atmosphere continuum (Paul et al., 2017), which directly influences the nutrient absorption capacity and, as mentioned, may have influenced the increase in the diameter and length of plagiotropic shoots. However, when applying the PBZ doses, genotype G7 showed a cubic response (Figure 3C). Nevertheless, although G7 showed lower rates of leaf transpiration, stomatal conductance, and photosynthesis, it showed higher values for the number of leaves and plagiotropic shoot length than genotype G5, which may have resulted in the larger plagiotropic shoot diameter of genotype G7.

Regarding the physiological variable of substomatal CO2 concentration (*Ci*) between genotypes G5 and G7, there were no interactions or isolated effects of the PBZ concentrations within each genotype. This may be associated with the little variation in stomatal conductance values, promoting similar CO2 entry into the substomatal cavity of the plants treated with PBZ.

Table 1. Orthotropic shoot diameter, plagiotropic shoot diameter, shoot length, leaf area index, number of leaves, photosynthesis, stomatal conductance, leaf transpiration rate, and substomatal CO_2 concentration of clonal seedlings of *Passiflora mucronata* Lam. genotypes subjected to paclobutrazol doses.

	Orthotropic shoot diameter (cm)				Plagiotropic shoot diameter (cm)			
Genotypes	PBZ (mg Plant)				PBZ (mg Plant)			
-	0.0	5	10	15	0.0	5	10	15
G5	0.31a	0.40a	0.32a	0.32a	0.1008b	0.1225b	0.1341b	0.1316b
G7	0.33a	0.38a	0.40a	0.33a	0.1183a	0.1300a	0.1683a	0.1541a
CV (%)	6.80				2.16			
Genotypes	Shoot length (cm)				Leaf area index (cm ²)			
G5	19.83b	30.91a	20.50b	25.33b	11.9a	15.9a	11.7a	13.2a
G7	28.41a	27.25b	26.66a	31.00a	10.8a	13.6a	13.9a	11.7a
CV (%)	6.56				9.34			
Genotypes	Number of leaves				Photosynthesis			
G5	8.0b	9.91b	7.25b	8.91b	3.27a	3.76a	4.06a	3.35a
G7	10.75a	12.16a	12.83a	13.66a	2.33b	3.23b	2.51b	2.70b
CV (%)	13.33				7.54			
Genotypes	Stomatal conductance			Leaf transpiration rate				
G5	0.09a	0.073b	0.116a	0.086a	3.00a	3.25a	3.83a	3.70a
G7	0.08a	0.106a	0.066b	0.086a	2.61b	3.40a	1.94b	2.90b
CV (%)	14.98			5.28				
Genotypes		Substomatal CO ₂ concentration						
G5		292.8a		303.3a		297.5a	308.3a	
G7		321.4a		312.7a		302.8a	322.4a	
CV (%)		3.37						

¹Means followed by the same lowercase letter in the column do not differ by the Tukey test ($p \le 0.005$); Source: Elaborated by authors (2020).

When comparing the genotypes G5 and the genotype G7 within each dose, the diameter of the orthotropic branch did not differ, however the diameter of the plagiotropic branch of the genotype G7 was greater than the genotype G5 in all doses tested, which can improve the grafting process, facilitating the growth of commercial species due to its greater radial expansion (Table 1).

The shoot length of the G7 genotype was greater than the genotype G5 at doses of 0; 10 and 15 mg Plant⁻¹. Consequently, the highest number of leaves presented by the genotype G7 is observed in all doses, which may be correlated with the greater length of the shoots. The leaf area index did not differ between genotypes (Table 1).

For the physiological variables, the genotype G5 has a higher photosynthetic rate than the genotype G7 at all doses. However, the substomatic concentration of CO_2 did not differ between the genotypes. The rate of leaf transpiration of the genotype G5 was higher at doses 0; 10 and 15 mg planta⁻¹ (Table 1).

Thus, the dissimilar behavior between *P. mucronata* genotypes submitted to doses of paclobutrazol, corroborates the results observed by França et al. (2018), suggesting that there is intraspecific divergence between populations.

Considering the better interpretation of agronomic traits obtained, comparisons were made between the variables. Pearson's linear correlation (r) was used to verify the associations between variables for genotypes G5 (Figure 4) and G7 (Figure 5).

The plagiotropic shoot diameter (DP) and the leaf transpiration rate (*E*) showed a positive linear correlation with the PBZ concentrations (C), resulting in 0.85 and 0.83, respectively (Figure 4), suggesting that the PBZ dose applied linearly interferes with the plagiotropic shoot diameter and the leaf transpiration rate in genotype G5. Similarly, it is worth noting that DP showed a strong correlation with *E* (0.87). These responses obtained may be related to the effects of PBZ on the hormonal changes induced in plants, characterized by the inhibition of the synthesis of gibberellins, increasing the content of abscisic acid and gibberellin in the cells (Jaleel et al., 2007), corroborating the results of increased diameter observed in young coffee plants treated with PBZ up to the dose of 600 mg L⁻¹ (D'arêde et al., 2017).



Figure 4 – Pearson's correlations for genotype G5.

C- PBZ doses; DP- plagiotropic shoot diameter; NF- number of leaves; CB- plagiotropic shoot length; AF- total leaf area index; DO- orthotropic shoot diameter; A- photosynthesis; g_s -stomatal conductance; *Ci*- substomatal CO₂ concentration; *E*- and leaf transpiration rate; Source: Elaborated by authors (2020).

The number of leaves (NF) showed a positive linear correlation with shoot length (CP), resulting in 0.84. Likewise, CP showed a positive linear correlation with the total leaf

area index (AF), resulting in 0.78 (Figure 4), suggesting that shoot growth in length increases the number of internodes, consequently emitting a higher number of leaves. However, NF and CP showed a negative linear correlation with stomatal conductance (g_s), corresponding to -0.78 and -0.65, respectively (Figure 4), suggesting that while one trait increases, the other decreases, and vice-versa. This behavior shows that the plant can still control stomatal conductance even with greater length and number of leaves. PBZ can promote the activation of systems that increase resistance to abiotic stresses, such as the ability to control the total or partial closure of stomata, changing the plant source-sink relationship; that is, despite the greater length and number of leaves, the plant still has mechanisms to control stomatal conductance (Mohan et al., 2015; Srivastava et al., 2016)

In genotype G7, the plagiotropic shoot diameter and the number of leaves showed a positive linear correlation with the PBZ concentrations applied, resulting in 0.8 and 0.86, respectively; that is, the higher the PBZ dose, the higher the DP and NF values. There was also a positive linear correlation between DP and NF, resulting in 0.73 (Figure 5).



Figure 5 – Pearson's correlations for genotype G7.

C- PBZ doses; DP- plagiotropic shoot diameter; NF- number of leaves; CB- plagiotropic shoot length; AF- total leaf area index; DO- orthotropic shoot diameter; A- photosynthesis; g_s -stomatal conductance; *Ci*- substomatal CO₂ concentration; *E*- and leaf transpiration rate; Source: Elaborated by authors (2020).

The physiological variables related to stomatal conductance (C_s) and the leaf transpiration rate (E) showed a linear correlation with photosynthesis (A), resulting in 0.74 and 0.63, respectively (Figure 5). Likewise, g_s and E showed a linear correlation of 0.73; that is, when one variable increases, the other also increases, and vice versa.

4. Conclusions

The studied genotypes (G5 and G7) showed different phenotypic responses when subjected to PBZ doses, highlighting the intraspecific divergence of the species.

The plagiotropic shoots of the seedlings of genotypes G5 and G7 of *Passiflora mucronate* show larger diameters with the application of 10 mg plant⁻¹ of paclobutrazol.

The application of the 10 and 5 mg plant⁻¹ PBZ doses increases photosynthesis in the seedlings of genotypes G5 and G7 of *Passiflora mucronata*.

For further studies, it is recommended to apply paclobutrazol doses in other genotypes of *P mucronata* and as they can be applied over time and prolong the evaluation time until frui harvesting.

Acknowledgments

The authors thank the Federal University of Espírito Santo for providing facilities and equipment available for research; the Coordination for the Improvement of Higher Education Personnel (CAPES) and the National Council for Scientific and Technological Development, for financial support and doctoral and PIBIC scholarships to the fourth and fifth authors and productivity in research to the sixth and seventh authors, respectively; to the Foundation for Research and Innovation Support of Espírito Santo (FAPES), for the granting of a master's scholarship to the first author and research rate to the seventh author (FAPES Notice No. 19/2018 - Research Rate - FAPES Process N°. 82195510).

References

Alexandre, R. S., Costa, P. R., Chagas, K., Mayrinck, L. G., Detoni, J. L., Schmildt, E. R. (2014) Enraizamento adventício de estacas do maracujazeiro silvestre *Passiflora mucronata* Lam.: forma de veiculação e concentrações do ácido indol-3-butírico. *Revista Ceres*, v. 61(1), 567-571.

Alexandre, R. S., Lopes, J. C., Tiradentes, A. T., Bruckner, C. H., Otoni, W. C. (2013) Metodologia de minienxertia em maracujazeiros. *Revista Brasileira de Fruticultura*, 35(1), 329-332.

Alvares, C. A., Stape, J. L., Sentelhas, P. C., Gonçalves, J. L. M., Sparovek, G. (2013) Köppen's climate classification map for Brazil. *Meteorologische Zeitschrift*, 22(1), 711-728.

Babu, M., Reddy, A. S., Basha, P. A., Naik, S. T. (2014) Transpiration and photosynthesis as affected by triazoles in mulberry (*Morus alba* L.). *Indian Journal of Advances in Chemical Science*, 2(4), 271-274.

Benett, K. S. S., Junior, M. J. D. A. F., Benett, C. G. S., Seleguini, A., Lemos, O. L. (2014) Utilização de paclobutrazol na produção de mudas de tomateiro. Comunicata Scientiae, 5(2), 164-169.

Bernardes, P. M., Nicoli, C., Alexandre, R., Guilhen, J., Fontes, M. P., Ferreira, A., Ferreira, M. (2020) Vegetative and reproductive performance of species of the genus *Passiflora*. *Scientia Horticulturae*, 256(1), 109193.

Braun, J. W. & Garth, J. K. L. (1986) Growth and fruiting of 'HeritGAe' primocane fruiting red raspberry in response to Paclobutrazol. *HortScience*, 21(1), 437-439.

Correia, A. O. (2019). Propagação e avaliação de passifloráceas submetidas a estresses bióticos e abióticos. Tese de Doutorado na produção vegetal. Espirito Santo: Alegre.

D'arêde, L. O., Matsumoto, S. N., Santos, J. L., Viana, A. E. S., Silva, P. A. R. (2017) Morfofisiologia do crescimento vegetativo inicial de cafeeiros arabica submetidos a aplicação via foliar de paclobutrazol. *Coffee Science*, 12(3), 451-462.

Do Brasil, F. (2020). em construção. 2017. Jardim Botânico do Rio de Janeiro.

Garbin, M. L., Corrijo, T. T., Sansevero, J. B. B., Sánchez-Tapia, A., Scarano, F. R. (2012) Subordinate, not dominant, woody species promote the diversity of climbingplants. *Perspectives in Plant Ecology, Evolution and Systematics*, 14(1), 257-265.

Faleiro, F. G. & Junqueira, N. T. V. (2016). Maracujá: o produtor pergunta, a Embrapa responde. Embrapa Cerrados-Livro técnico (INFOTECA-E).

Fletcher, R. A. & Hofstra, G. (1990) Improvement of uniconazole-induced protection in wheat seedlings. *Journal of Plant Growth Regulation*, 9(207), 1-4.

França, J. M., Venial, L. R., Costa, E. B., Schmildt, E. R., Schmildt, O., Bernardes, P. M., Tatagiba, S. D., Lopes, J. C., Ferreira, M. F. S., Alexandre, R. S. (2018) Morphophysiology, phenotypic and molecular diversity of auxin-induced *Passiflora mucronata* Lam. (Passifloraceae). *Anais da Academia Brasileira de Ciências*, 1(1), 1799-1814.

Furlaneto, F. P. B., Esperancini, M. S. T., Martins, A. N., Okamoto, F., Vidal, A. A., Bueno,O. C. (2014) Análise energética do novo sistema de produção de maracujá amarelo na região de Marília-SP. *Ciência Rural*, 44(2), 235-240.

Hartmann, H. T., Kester, D. E., Davies Jr, F. T., Geneve, R. L. (2011) *Plant propagation*: principles and practices. 8th ed. Boston: Prentice Hall.

Ibge - Instituto Brasileiro de Geografia e Estatística. (2019). Available: http://www.cnpmf.embrapa.br/Base_de_Dados/index_pdf/dados/brasil/maracuja/b1_maracuja .pdf.

Inmet - Instituto Nacional de Meteorologia, 2020. Available: https://portal.inmet.gov.br/ ?r=estacoes/estacoesAutomaticas.

Jaleel, C. A., Kishorekumar, A., Manivannan, P., Sankar, B., Gomathinayagam, M., Gopi, R., Panneerselvam, R. (2007) Alterations in carbohydrate metabolism and enhancement in tuber production in white yam (*Dioscorea rotundata* Poir.) under triadimefon and hexaconazole applications. *Journal of Plant Growth Regulation*, 53(1), 7-16.

Jiao, J., Tsujita, J. J., Murr, D. P. (1986) Effects of paclobutrazol and A-Rest on growth, flowering, leaf carbohydrate, and leaf senescence in 'Nellie White' Easter lily (*Lilium longiflorum* Thunb.). *Scientia Horticulturae*, 30(1-2), 135-141.

Junqueira, N. T. V., Braga, M. F., Faleiro, F. G., Peixoto, J. R., Bernacci, L. C. (2005) Potencial de espécies silvestres de maracujazeiro como fonte de resistência a doenças. In: Faleiro, F. G., Junqueira, N. T. V., Braga, M. F. *Maracujá*: germoplasma e melhoramento genético. Planaltina: Embrapa Cerrados, 81-106.

Lopes, C. M., Andrade, I., Pedroso, V., Martins, S. (2004) Modelos empíricos para estimativas da área foliar da videira na casta jaen. *Ciência e Técnica Vitivinícola*, v. 19(1), 61-75.

Mabvongwe, O., Manenji, B. T., Gwazane, M., Chandiposha, M. (2016) The effect of paclobutrazol application time and variety on growth, yield, and quality of potato (*Solanum tuberosum* L.). *Advances in Agriculture*, 2016(1), 1-5.

Magnago, L. F. S., Martins, S. V., Pereira, O. J. (2011) Heterogeneidade florística das fitocenoses de restingas nos estados do Rio de Janeiro e Espírito Santo. *Revista Árvore*, 35(1), 245-254.

Mohan, R., Vyas, D., Bhat, H. A., Kaur, T. D., Dhar, A. (2015) Exploring possibilities of induction of water stress tolerance in mulberry in rainfed condition by application of paclobutrazol. *Journal of Global Biosciences*, 4(9), 3301-3310.

Morgado, M. A. D., Bruckner, C. H., Rosado, L. D. S., Santos, C. E. M. (2015) Desenvolvimento de mudas de maracujazeiro-azedo enxertadas em espécies silvestres de Passiflora. *Revista Brasileira de Fruticultura*, 37(1), 471-479.

Moura, F. B., Vieira, M. R. D. S., Simoes, A. D. N., Silva, S. L., Medeiros, D. C., Paes, R. D. A., Júnior, W. S. (2017) Participation of cytokinin on gas exchange and antioxidant enzymes activities. *Indian Journal of Plant Physiology*, 22(1), 16-29.

Oliari, L., Giles, J., Mayrinck, L., Oliveira, J. P., Lopes, J., Otoni, W., Schmildt, E., Aoyama, E., Alexandre, R. S. (2016) Mini-grafting of adult *Passiflora edulis* Sims f. *flavicarpa* Deg.

scions onto vegetatively propagated adult rootstocks of *P. mucronata* Lam. Australian Journal of Crop Science, 10(1), 490-496.

Oliveira, G. P. (2020). Uso do paclobutrazol na produção de manga. *Research, Society and Development*, 9(7), 1-16.

Pal, S., Zhao, J., Khan, A., Yadav, N. S., Batushansky, A., Barak, S., Rachmilevitch,
S. (2016) O paclobutrazol induz tolerância no tomate ao déficit de irrigação através de efeitos diversificados na morfologia, fisiologia e metabolismo das plantas. *Scientific Reports*, 6(1), 39321.

Peng, D., Chen, X., Yin, Y., Lu, K., Yang, W., Tang, Y., Wang, Z. (2014) Lodging resistance of winter wheat (*Triticum aestivum* L.): Lignin accumulation and its related enzymes activities due to the application of paclobutrazol or gibberellin acid. *Field Crops Research*, 157(1), 1-7.

Rademacher, W. (2018) Chemical regulators of gibberellin status and their application in plant production. *Annual Plant Reviews online*, 49(1), 359-403.

Rademacher, W. (2000) Growth retardants: effects on gibberellin biosynthesis and other metabolic pathways. *Annual Review of Plant Physiology and Molecular Biology*, 51(1), 501-531.

Roseli, A. N. M., Ying, T. F., Ramlan, N. F. (2012) Morphological and physiological response to *Syzygium myrtifolium* Walp. to paclobutrazol. *Sains Malays*, v. 41(10), 1187-1192.

Sankhla, N., Davis, T. D., Jolley, V. D. (1986) Upadhyaya, A. Effect of Paclobutrazol on the development of iron chlorosis in soybeans. *Journal of Plant Nutrition*, 9(7), 923-934.

Santos, C. H. B., Cruz Neto, A. J., Junghans, T. G., Jesus, O. N., Girardi, E. A. (2016) Estádio de maturação de frutos e influência de ácido giberélico na emergência e crescimento de *Passiflora* spp. *Revista Ciência Agronômica*, v. 47(3), 481-490.

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

Schmildt, E. R., Oliari, L. S., Alexandre, R. S., Silva, F. O. Dos R. Da., Schmildt, O. (2018) Histological aspects of mini-grafting of *Passiflora edulis* Sims. and *Passiflora mucronata* Lam. *Revista Brasileira de Fruticultura*, 40(2), 351-357.

Schmildt, E. R., Oliari, L. S., Schmildt, O., Alexandre, R. S., Pires, F. R. (2017) Determinação da área foliar de *Passiflora mucronata* a partir de dimensões lineares do limbo foliar. *Revista Agro@mbiente On-line*, 10(4), 351-357.

Silva, K. S. & Faria Junior, M. J. A. (2011) Uso de paclobutrazol como estratégia para redução do porte e da brotação lateral de plantas de tomateiro. *Ciência e Agrotecnologia*, v. 35(3), 539-546.

Siqueira, D. L. De, Cecon, P. R., Salomão, L. C. C. (2008) Desenvolvimento do limoeiro 'Volkameriano' (*Citrus volkameriana* Pasq.) submetido a doses de paclobutrazol e ácido giberélico. *Revista Brasileira de Fruticultura*, v. 30(3), 764-768.

Skowron, E., Trojak, M., Sobala, T. Molecular marker of delayed senescence in transgenic tobacco with enhanced cytokinin level. *World Scientific News*, 51(1), 13-25.

Srivastava, A. K., Pasala, R., Minhas, P. S., Suprasanna, P. (2016) Plant bioregulators for sustainable agriculture: integrating redox signaling as a possible unifying mechanism. *Advances in Agronomy*, 137(1), 237-278.

Téllez, H. O., Bomfim, G. V., Carvalho, A. C. P. P., Azevedo, B. M., Lozano, C. H. G. (2020). Paclobutrazol no desenvolvimento de mudas de plantas matrizes de abacaxizeiro ornamental. *Research, Society and Development*, 9(10), 1-16.

Tesfahun, W. & Menzir, A. (2018) Effect of rates and time of paclobutrazol application on growth, lodging, and yield and yield components of tef [*Eragrostis tef* (Zucc.) Trotter] in

Adadistrict, East Shewa, Ethiopia. *Journal of Biology, Agriculture and Healthcare*, 8(3), 104-117.

Toledo-Souza, E. D., Silveira, P. M., Café-Filho, A. C., Lobo-Júnior, M. (2012) Fusarium wilt incidence and common bean yield according to the preceding crop and the soil tillage system. *Pesquisa Agropecuária Brasileira*, 47(1), 1031-1037.

Vu, J. C. V. & Yelenosky, G. (1992) Growth and photosynthesis of sweet orange plants treated with Paclobutrazol. *Journal of Plant Growth Regulation*, 11(1), 85-89.

Wang, C., Ruan, R., Yuan, X., Hu, D., Yang, H., Li, Y., Yi, Z. (2014) Relationship between lignin metabolism and lodging resistance of culm in buckwheat. *Journal of Agricultural Science*, 6(9), 29-36.

Zhang, M., Duan, L., Tian, X., He, Z., Li, J., Wang, B., Li, Z. (2007) Uniconazole-induced tolerance of soybean to water deficit stress in relation to changes in photosynthesis, hormones and antioxidant system. *Journal of Plant Physiology*, 164(6), 709-717.

Percentage of contribution of each author in the manuscript

Patrick Alves de Oliveira – 30% Ariany das Graças Teixeira – 15% Guilherme Bravim Canal – 15% Paula Aparecida Muniz de Lima – 10% Gardênia Rosa de Lisbôa Jacomino – 10% Rodrigo Sobreira Alexandre – 10% José Carlos Lopes – 10%