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Article

Ornamental Traits and Sensory Analysis of 'Biquinho Vermelha' Pepper Treated with Paclobutrazol

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Abstract: This work aimed to determine the effect of PBZ concentrations and application protocols on the ornamental quality and global preference of customers. PBZ was applied in five concentrations by three protocols (drench at transplanting, drench at 30 days after transplanting and immersion). At commercial maturity, the plants were evaluated for plant growth parameters related to height and canopy characteristics, fruits and leaves, in addition to sensory analysis. PBZ produced plants with darker green leaves and the drench method was more effective in modulating plant growth parameters compared to immersion. Conversely, the sensory analysis showed a greater preference among evaluators for plants treated with PBZ at 2.5 and 5 mg L^{-1} via drench at 30 DAT or through immersion (IM), even though IM was not associated with any of the ornamental quality parameters evaluated. Likewise, the application of PBZ by drench during transplanting is not recommended as the plants showed a reduced number and size of fruits, which affected the ornamental value of the plants and global preference. Therefore, PBZ applied by drench 30DAT at concentrations of 2.5 and 5 mg L^{-1} produced 'Biquinho Vermelha' pepper plants with attractive ornamental characteristics being an alternative method for producers of potted ornamental plants.

Keywords: paclobutrazol; *Capsicum chinense*; ornamental plants; growth regulator; canopy

1. Introduction

The global market of flower and ornamental plants is expanding and focused on the decoration of internal spaces [1] mainly with the use of potted plants. 'Biquinho' (*Capsicum chinense* Jacq.) is one of the most common pepper varieties in Brazil and is traditionally cultivated in conventional systems to produce fruits that are consumed fresh or processed [2]; however, this variety also has attractive characteristics for ornamental purposes. The commercial success of growing these plants as ornamentals depends on consumer attraction to their beauty, quality, vigor, color, shape and size of fruits and leaves, as well as a balanced canopy [3]. 'Biquinho' pepper fruits are cuspidal and triangular; green when immature, orange during the maturation process and red when mature. The main characteristic is the lack of pungency. This variety holds an intermediate growth, reaching an average of 60 cm in height and 1 m in canopy diameter [4]. Therefore, it may not adapt to cultivation in small pots, a necessary condition for its use to decorate indoor environments [5].

Paclobutrazol (PBZ) is an effective plant growth regulator that inhibits gibberellin synthesis by blocking the oxidation of ent-kaurene to ent-kaurenoic acid [6]. PBZ has been successfully used to reduce the height of several species of ornamental plants including roses, sunflowers, orchids, marigold, platycodon, petunia and peppers [7–13]. The ornamental quality of potted pepper plants that follows the standard of the Brazilian cooperative Veiling Holambra, located in the state of São Paulo, is measured by several quality parameters in addition to the plant height [5]. These plants must cover the top of pots, with beautiful foliage and rigid stems, exhibiting an adequate fruit number per plant and stage of maturity, besides lacking symptoms of toxicity and nutritional

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deficiency. The effectiveness of PBZ, as well as other plant growth regulators, varies with the species, cultivar, concentration, mode and time of application [14,15].

Previous studies on the effect of PBZ on 'Biquinho' pepper have already been carried out elsewhere and they are elusive. The effect of different methods of applying PBZ at a concentration of 10 μM, demonstrated a higher efficiency of the regulator when the application was via the root system immersion instead of application by foliar spray, which otherwise proved to be inefficient [16].

PBZ applied either by drenching or by immersion of the root/substrate system has already been shown to be both effective and harmful in reducing the height and improving the ornamental quality of several peppers of the species *C. annuum* and *C. chinense* [5,10,16–18], depending on concentration applied. The results of these studies show a clear genetic effect regarding the different quality parameters evaluated in response to PBZ applications.

The objective of this research was to determine the effect of PBZ concentrations and application protocols on the ornamental quality and global preference of pepper plants of the commercial variety 'Biquinho Vermelha'.

2. Materials and Methods

2.1. Plant Material and Crop Management

The plant material used in this research was the commercial variety of pepper 'Biquinho Vermelha' (*Capsicum chinense)*. The seedlings were produced from commercial seeds in a protected environment at the Federal University of São Carlos, in the city of Araras, state of São Paulo, Brazil (-22.3114976 latitude and 47.3847851 longitude, 685 m altitude). During the experiment, the mean, maximum and minimum temperatures were 21.3 °C, 27.8 °C and 16.6 °C respectively. Sowing was carried out in trays of 128 cells filled with the commercial substrate Carolina Soil® on January 23, 2023. On February 28, 2023, when the seedlings reached 2 to 3 pairs of leaves, they were transplanted into pots number 15 (1.17 L in volume; 11 cm in height, 10.45 cm in basal diameter and 14. 5 cm in upper diameter) filled with a substrate based on Pine bark (Spagnhol®). Cultivation in pots was carried out in the same place where the seedlings were produced, in a greenhouse and under natural light conditions. Irrigation was supplied to the plants intermittently with a micro-drippers system. The plants were fert irrigated three times a week manually using watering flasks. Each plant received approximately 300 mL of the nutrient solution proposed by Furlani [19].

2.2. Experimental Design

The plants were treated with the plant growth regulator paclobutrazol (PBZ) (CULTAR® 250 SC) following three application protocols: IM-Immersion, for 10 seconds, of the root-substrate of the seedlings in the PBZ solutions, immediately before the transplanting into pots; DT-Drenching of 250 mL of PBZ solutions to the cultivation substrate, applied immediately after transplanting the seedlings into pots and D30DAT-Drenching of 250 mL of PBZ solutions to the cultivation substrate, 30 days after transplanting.

The concentrations used in each of the application protocols were 0; 2.5; 5.0; 7.5 and 10 mg L-1 of the commercial product which are equivalent to 0; 0.63; 1.25; 1.88 and 2.50 mg of the active ingredient, respectively. In control plants, application protocols were followed replacing the PBZ solution with tap water. Thus, 15 treatments were applied (3 application protocols x 5 concentrations of PBZ).

When 50% of the plants in each treatment reached commercial maturity (at least 30% of ripe fruits) the plants were evaluated for the ornamental quality characteristics described below. In plants where the application was made by immersion and drenching at 30DAT, the evaluations were made on June 8, 2023 (101DAT) and in those where PBZ was applied by drenching at transplanting on June 22, 2023 (115DAT).

2.3. Ornamental Quality Traits

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The height of the plants was measured from the base of the stem to the apex of the canopy (considering the last leaf completely expanded) using a ruler and expressed in cm. The canopy's longitudinal and transverse diameters were measured using a tape measure, observing the plant from above and at an angle of 90° to the stem. The canopy compactness was determined by the transverse and longitudinal diameter ratio, where values closer to one (1) indicate a more circular canopy [10].

The SPAD chlorophyll index was determined with the SPAD-502 chlorophyll meter (Spectrum Technologies Inc., Plainfield, IL), using measurements from nine leaves, three from the basal part, three from the middle part and three from the upper part of each plant. The leaf color parameters were obtained in the same position using the Minolta® Chroma Meter - CM-25d spectrophotometer. Positive values of parameters a* and b* indicate red and yellow coloration and negative values of a* and b* indicate green and blue coloration, respectively. The L* parameter indicates luminosity (0 = black and 100 = white) and the hue angle (h°) shows where the color is located in the diagram, where 0° and 360° correspond to red, 90° to yellow, 180° the green color and 270° the blue color. The intermediate angles correspond to a combination of these colors. Chroma (C*) indicates chromaticity, the vivacity of the color, represented by the distance from the center to the end of the diagram, which varies from 0 to 60 [20].

The total number of leaves and fruits of each plant were counted. The length of the leaves was measured from the insertion of the petiole to the apex of the limbus in a longitudinal direction, with a ruler and expressed in cm. The width of the leaf was measured at the longest transverse length of the limb, also using a ruler and expressed in cm. The leaves used for length and width measurements were the same leaves on which the colorimetric and SPAD chlorophyll index measurements were carried out.

The length and diameter of the fruits were determined with a caliper using the average of measurements of nine fully ripe fruits per plant when available and chosen randomly from the canopy or the maximum number of fruits on each plant, when smaller than nine.

The aerial part of the plant was detached from the pot at the substrate level with pruning shears and its fresh weight was obtained on a scale. Plant fullness was determined by the relationship between the shoot fresh weight and the height of the plant. Throughout the cultivation of pepper plants, daily assessments were carried out on each plant to identify the day of the opening of the first flower, with the result expressed several days after transplanting for anthesis.

2.4. Sensory Analysis

The project was approved by the Research Ethics Committee, CAAE 74062923.0.0000.5504. The sensory analysis of the plants was carried out at the Sensory Analysis Laboratory of the Federal University of São Carlos. For this, one plant from each treatment was used, totaling 15 potted plants coded with three digits and presented to the evaluators all at once. All plants presented were at commercial maturity (at least 30% of ripe fruits). 125 participants responded to the global preference test, showing interest and availability using a seven-point structured hedonic scale (7-liked extremely; 6-liked a lot; 5-liked moderately; 4-neither liked/nor disliked; 3-disliked slightly; 2-disliked it very much; 1-disliked it extremely).

2.5. Statistical Analyses

A completely randomized design with five plants per treatment was used. Treatments were arranged in factorial scheme 3 x 5 (3 application protocols x 5 concentrations of PBZ). Data related to agronomic and sensorial variables were subjected to analysis of variance (ANOVA) and means were separated using Tukey's and Scott-Knott´s tests, both at $p \leq 0.05$ for agronomic and sensorial variables respectively. A principal component analysis (PCA) was performed to assess the major vector variation among agronomic and sensorial traits associated with paclobutrazol application protocols and concentrations. All assessments were accomplished with the R software (v. 4.3.2) [21]. The outputs were plotted using the R package 'ellipse' [22] considering the Euclidean distances at 95%

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confidence ($p<0.05$). Traits loading contribution >0.4 was set up as a minimum threshold for variable selection.

3. Results

3.1. Ornamental Quality Traits

The analysis of variance presented in Table 1 showed a significant interaction between the concentrations and application protocols of paclobutrazol for plant height, height of first bifurcation, longitudinal and transverse diameters of the canopy, SPAD chlorophyll index, plant fullness, leaves length and width and for the C*, a* and b* parameters of the colorimetric analysis. The canopy compactness, fruit number and length, fresh weight of the aerial part and the days to anthesis were significantly affected individually by the concentrations and application protocols of PBZ. The L* and h° parameters were significantly affected only by PBZ concentrations and the number of leaves and fruit diameter only by the application protocol.

Table 1. Analysis of variance for the ornamental quality traits of 'Biquinho Vermelha' pepper treated with solutions of PBZ at concentrations of 0; 2.5; 5.0; 7.5 and 10 mg L^{-1} applied in three different application protocols (AP): 1-Immersion, for 10 seconds, of the root-substrate of the seedlings in the PBZ solutions, immediately before the transplanting into pots; 2-Drenching of 250 mL of PBZ solutions to the cultivation substrate, applied immediately after transplanting the seedlings into pots and 3-Drenching of 250 mL of PBZ solutions to the cultivation substrate, 30 days after transplanting. Assessments were made at the commercial maturity of plants (when at least 50% of the plants in each treatment had 30% of the fruits ripe).

Ornamental quality traits	PBZ	AP	PBZ X AP
Plant height	*[1]	*	*
First bifurcation height	*	*	*
Canopy longitudinal diameter	*	*	*
Canopy transverse diameter	NS	*	*
Canopy compactness	*	*	NS
Fruit number	*	*	NS
Leave number	NS	*	NS
Fruit diameter	NS	*	NS
Fruit length	*	*	NS
Shoot fresh weight	*	*	NS
SPAD Index	*	*	*
Plant fullness	*	*	*
Leaf length	*	*	*
Leaf width	*	*	*
Days to anthesis	*	*	NS
Lightness (L*)	*	NS	NS
Red/green coordinate (a*)	*	*	*
Yellow/blue coordinate (b*)	*	*	*
Chroma (C*)	*	*	*
Hue angle (h°)	*	NS	NS

[1] NS, *, non-significant and significant at 5% probability respectively.

All PBZ application protocols caused a reduction in plant height. This reduction was significant at all concentrations used as compared to the control when PBZ was applied by drench at transplanting (DT). However, when PBZ was applied via drench at 30DAT (D30DAT) a significant effect on height reduction was observed only from a concentration of 5 mg L^{-1} compared to the control. When applied as immersion (IM), PBZ was effective in significantly reducing plant height (24.4%) compared to control plants only at the highest concentration (10 mg L^{-1}) (Table 2).

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Table 2. Effect of PBZ concentrations and application protocols on the ornamental quality traits of 'Biquinho Vermelha' pepper. The data was collected between June 8 and 22, 2023 and represents the average of 5 plants.

Ornamental quality traits	AP[A]	PBZ concentrations (mg L^{-1})[B]					CV (%)
		0	2,5	5,0	7,5	10,0	
Plant height (cm)	IM	34,8 Ba[C]	32,8 Aab	29,9 Aab	30,7 Aab	26,3 Ab	16,2
	DT	44,4 Aa	20,6 Bbc	25,5 ABb	26,5 ABb	14,3 Bc	
	D30DAT	31,4 Ba	25,1 Bab	19,0 Bb	20,3 Bb	17,7 Bb	
First bifurcation height (cm)	IM	8,1 Aa	7,8 ABa	6,8 ABa	7,6 ABa	6,1 Ba	19,2
	DT	9,2 Aa	6,4 Bb	5,2 Bb	5,7 Bb	4,1 Bb	
	D30DAT	8,5 Aa	8,9 Aa	8,0 Aa	8,0 Aa	8,4 Aa	
SPAD chlorophyll Index	IM	52,6 Aa	56,6 Ba	57,2 Aa	55,6 Ba	58,8 Ba	6,9
	DT	51,3 Ac	67,0 Aab	62,8 Ab	65,8 Aab	72,1 Aa	
	D30DAT	53,6 Ab	53,9 Bab	60,0 Aab	56,9 Bab	60,9 Ba	
Plant fullness ($g.cm^{-1}$)	IM	3,3 Aa	3,5 Ba	3,6 ABa	3,7 ABa	3,7 Ba	20,4
	DT	2,2 Ab	5,0 Aa	3,0 Bb	3,1 Bb	6,0 Aa	
	D30DAT	2,9 Ab	4,0 ABab	4,7 Aa	4,8 Aa	4,5 Ba	
Canopy longitudinal diameter	IM	43,6 Aa	38,6 Aa	39,0 Aa	43,1 Aa	36,2 Aa	14,7
	DT	45,3 Aa	30,2 Bb	30,5 Bb	30,4 Bb	22,6 Bb	
	D30DAT	34,10 Ba	29,3 Bab	25,4 Bb	25,2 Bb	24,4 Bb	
Canopy transverse diameter	IM	24,9 Ba	30,3 Aa	29,4 Aa	35,0 Aa	29,1 Aa	19,3
	DT	34,4 Aa	22,0 Bab	26,4 Ab	25,3 Bb	20,8 Bb	
	D30DAT	28,4 ABa	23,7 ABa	22,7 Aa	22,4 Ba	21,7 ABa	
Leaf lenght (mm)	IM	59,0 Aa	57,2 Aab	53,1 ABab	55,4 ABab	50,7 Bb	6,9
	DT	56,0 Aa	48,6 Bb	48,2 Bb	50,4 Bab	47,2 Bb	
	D30DAT	55,6 Aa	61,6 Aa	57,9 Aa	56,5 Aa	57,4 Aa	
Leaf width (mm)	IM	39,0 Aa	35,5 Bab	33,3 Bb	36,3 Aab	32,1 Bb	7,9
	DT	36,1 Aa	31,0 Cb	30,4 Bb	31,6 Bab	29,1 Bb	
	D30DAT	38,3 Aa	41,1 Aa	39,1 Aa	39,1 Aa	39,7 Aa	
Chroma (C*)	IM	19,4 ABa	19,3 Aa	17,2 Aa	17,8 Aa	16,4 Aa	11,0
	DT	21,5 Aa	13,0 Bbc	15,4 ABb	15,4 Ab	11,6 Bc	
	D30DAT	18,4 Ba	16,6 Bab	14,0 Bb	15,8 Aab	13,6 Bb	

[A]AP-Application protocol: IM-Immersion, for 10 seconds, of the root-substrate of the seedlings in the PBZ solutions, immediately before the transplanting into pots; DT-Drenching of 250 mL of PBZ solutions to the cultivation substrate, applied immediately after transplanting the seedlings into pots and D30DAT-Drenching of 250 mL of PBZ solutions to the cultivation substrate, 30 days after transplanting. [B]Concentrations of the commercial product (CULTAR® 250 SC). [C]Means followed by the same lowercase letter in the rows and uppercase letters in the columns do not differ from each other using the Tukey test at 5% probability.

The application of PBZ by D30DAT produced significantly shorter plants when compared to IM at all concentrations of PBZ used, except for control plants. The moment of application of PBZ by drenching whether at transplanting or 30DAT did not significantly influence plant height (Table 2).

For the first bifurcation height, significant difference was only observed in all concentrations relative to the control when PBZ was applied by DT. These plants showed a first bifurcation height, on average, 35.8% lower compared to D30DAT in all concentrations used, except for the control.

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However, IM produced plants with a first bifurcation height significantly lower than D30DAT only at the highest concentration, 10 mg L-1 of PBZ (Table 2).

An increase in the SPAD chlorophyll index of leaves and plant fullness of plants treated with PBZ was observed. When PBZ was applied by DT, the leaves were greener, with a higher SPAD chlorophyll index compared to the leaves of control plants, regardless of the concentration used (Table 2). However, when applied by drenching 30 days later, this was only observed when the highest concentration was used (10 mg L-1 of PBZ). Leaves of plants treated with PBZ by DT were greener than when applied by IM and D30DAT in all concentrations except the control and the concentration of 5 mg L-1 of PBZ.

Plants treated with PBZ applied by DT showed improved plant fullness only at the lowest and highest concentrations compared to the control, not showing a direct relationship between the concentrations used and this characteristic (Table 2). However, when applied by D30DAT, better plant fullness was observed in all concentrations from 5 mg L-1. When the regulator was applied by IM, the increase in SPAD chlorophyll index and plant fullness was not significant (Table 2).

PBZ reduced the canopy longitudinal and transverse diameter of the canopy when applied by drenching, either at transplanting or at 30DAT. This reduction was significant when PBZ was applied by DT in all concentrations and from a concentration of 5 mg L-1 as compared to the control for the longitudinal diameter and transverse diameter of the canopy, respectively. D30DAT only significantly affected canopy longitudinal diameter from a concentration of 5 mg L-1 compared to untreated plants. However, the application of PBZ by IM did not have a significant effect on both diameters (Table 2).

Comparing the application protocols there was no significant difference in the longitudinal diameter of the plants in which PBZ was applied by drenching at transplanting or 30DAT, except the control. However, these two protocols produced plants with a longitudinal diameter significantly smaller than those in which PBZ was applied by IM (Table 2). The transverse diameter was significantly smaller than the control at concentration of 5 mg L-1 when PBZ was applied by DT. In the other two protocols, no significant difference was observed between concentrations (Table 2).

Except for control plants, PBZ applied by DT from a concentration of 2.5 mg L-1 produced plants with smaller leaves in both length and width when compared to the D30DAT application protocol. Only at the highest concentration (10 mg L-1) was there a significant difference in leaf length between IM and D30DAT. At this concentration, IM produced plants with smaller leaves in length. For leaf width, comparing these last two protocols, there was only the exception of the control and the concentration of 7.5 mg L-1. In the other concentrations, when applied by IM, the plants presented leaves smaller in width than those to which it was applied D30DAT (Table 2).

In the instrumental color analysis, the leaves of all treatments were yellowish green, since all a* values were negative indicating green color and all b* values were positive indicating yellow color (data not shown). The C* values were significantly lower in all concentrations used than the control when PBZ was applied by DT application protocol. The same was observed at concentrations of 5 and 10 mg L-1 when PBZ was applied by D30DAT and no effect was observed on the chromaticity of the leaves when PBZ was applied by IM. Comparing the application protocols, except for control plants, there was no significant difference in the chromaticity of the leaves between drenching at transplanting and at 30DAT, regardless of the concentration of PBZ used (Table 2).

A reduction in L* and an increase in h° values was observed with the application of PBZ. Except for the concentration of 7.5 mg L-1, all PBZ concentrations used were significantly lower than the control for these two parameters (Table 3). From the L*, C* and h° data, obtained under D65 illuminant and 10° observer, images were obtained with the color of the leaves of each PBZ concentration using the EASYRGB color calculator [23]. Checking the images, it was possible to see a darker green tone of the leaves treated with PBZ at different concentrations compared to the control, with this difference being more subtle at the concentration of 7.5 mg L-1 (Figure 1).

PBZ concentrations (mg L^{-1})				
0	2,5	5,0	7,5	10,0

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Figure 1. Colors obtained from the EASYRGB color calculator in 'Biquinho Vermelha' pepper leaves treated with different concentrations of paclobutrazol.

The plant canopy became significantly more circular with increasing concentrations from the concentration of 7.5 mg L^{-1}. Furthermore, increasing PBZ concentrations caused a significant delay in flowering from a concentration of 5.0 mg L^{-1}. This delay varied from 3 to 4 days at concentrations of 5, 7.5 and 10.0 mg L^{-1} compared to control plants (Table 3). However, although there was a delay in the beginning of flowering, this did not affect fruiting. The plants had on average 61.9 fruits. No significant difference was observed in the length of these fruits and the fresh mass of the aerial part when compared to the control in any of the PBZ-used concentrations (Table 3).

Table 3. Effects of paclobutrazol concentrations on canopy compactness (CC), number of fruits (NFr), fruit length (LFr), leaf lightness (L*), leaf hue angle (h°), shoot fresh weight (FrW) and days after transplantation for anthesis (A) in 'Biquinho Vermelha' pepper. The data was collected between June 8 and 22, 2023 and represents the average of 5 plants.

PBZ (mg L^{-1})[A]	L*	h°	CC	FrW	A	NFr	LFr
0	38,8 a[B]	114,8 b	0,7 b	101,2 ab	38,1 c	67,9 a	22,5 ab
2,5	37,1 b	115,9 a	0,8 ab	103,3 a	39,9 bc	64,0 a	23,5 a
5,0	37,0 b	116,1 a	0,8 ab	89,6 ab	41,1 ab	56,3 a	21,8 ab
7,5	37,7 ab	115,5 ab	0,9 a	95,1 ab	42,4 a	64,7 a	21,7 ab
10,0	36,7 b	116,3 a	0,9 a	86,1 b	41,5 ab	56,5 a	21,0 b
CV (%)	3,2	0,9	14,0	15,8	5,6	19,7	9,7

[A]Concentrations of the commercial product (CULTAR® 250 SC); [B]Means followed by the same letter in the lines do not differ from each other using the Tukey test at 5% probability.

Drenching PBZ, either at transplanting or at 30DAT, produced plants with more circular canopy compared to plants in which PBZ was applied by IM (Table 4). The anthesis of plants in which PBZ was applied by DT was delayed by approximately 2 days compared to IM (Table 4). Plants in which PBZ was applied by DT had significantly smaller number of fruits and these fruits were smaller in length than those of plants in which the other two protocols were applied. However, the D30DAT application protocol produced fruits significantly smaller in diameter than the fruits of plants to which PBZ was applied by IM and DT (Table 4).

Table 4. Effects of paclobutrazol application protocols on canopy compactness (CC), number of fruits (NFr), number of leaves (NLe), fruit diameter (DFr), fruit length (LFr) and days after transplanting for anthesis (A) in 'Biquinho Vermelha' pepper. The data was collected between June 8 and 22, 2023 and represents the average of 5 plants.

AP[A]	CC	A	NFr	LFr (mm)	DFr (mm)	NLe	FrW
IM	0,7 b[B]	39,6 b	69,7 a	22,7 a	12,5 a	118,5 ab	108,1 a
DT	0,8 a	41,7 a	53,7 b	20,9 b	13,0 a	128,2 a	85,4 b
D30DAT	0,9 a	41,0 ab	62,2 a	22,7 a	11,2 b	101,3 b	91,6 b
CV (%)	14,0	5,6	19,7	9,7	10,1	23,0	15,8

[A]AP-Application protocol: IM-Immersion, for 10 seconds, of the root-substrate of the seedlings in the PBZ solutions, immediately before the transplanting into pots; DT-Drenching of 250 mL of PBZ solutions to the cultivation substrate, applied immediately after transplanting the seedlings into pots and D30DAT-Drenching of 250 mL of PBZ solutions to the cultivation substrate, 30 days after transplanting. [B]Means followed by the same letter in the lines do not differ from each other using the Tukey test at 5% probability.

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The number of leaves was significantly lower in plants treated with PBZ applied by D30DAT compared to DT (Table 4). The fresh weight of the aerial part of the plants was significantly reduced when the application was made by drenching regardless of when this application was made (Table 4).

3.2. Sensory Analysis

Global preference test showed that participants had a greater preference for plants to which PBZ was applied using the IM and D30DAT application protocol in the two lowest concentrations, 2.5 and 5.0 mg L^{-1}, as well as the control plant of D30DAT where PBZ was not applied (Table 5). The ratings obtained by the 125 interviewees in all these treatments were presented as "moderately liked". Furthermore, when the highest concentration (10 mg L^{-1}) was applied by DT and D30DAT, the plants showed the lowest scores in terms of global preference together with untreated and treated with 5 mg L^{-1} by DT application protocol. All these plants are in the description 'neither like nor dislike'. The plants to which concentrations of 2.5 and 7.5 mg L^{-1} were applied using the same method had intermediate grades (Table 5).

Table 5. Results of overall preference test of 'Biquinho Vermelha' pepper plants treated with PBZ.

Application protocol	PBZ (mg L^{-1})[A]	Overall preference[B]
Immersion	0	4,82 b[C]
	2,5	5,18 a
	5,0	5,54 a
	7,5	4,89 b
	10,0	4,70 c
Drenching at transplanting	0	4,02 d
	2,5	4,85 b
	5,0	4,14 d
	7,5	4,63 c
	10,0	4,42 c
Drenching at 30DAT	0	5,42 a
	2,5	5,33 a
	5,0	5,03 b
	7,5	4,78 b
	10,0	4,36 c
CV (%)		27,4

[A]Concentration of the commercial product (CULTAR® 250 SC).

3.3. Principal Components Analysis (PCA)

From 24 inputs, 21 variables presented loading contributions higher than 0.4, which were delineated through two major components (PCA1 and PCA2). Together, such variables explained over 48%, 65%, and 56% of the total variation in IM, DT, and D30DAT, respectively (Fig. S1 A, B, and C). Based on the PCA biplot, the clustering was better evidenced in both DT and D30DAT methods (Fig. S1 B and C). On the other hand, no marked discrepancy is observed in IM (Fig. S1 A). In DT, the biplot scaled two main clusters based on the vector of similarities. PCA1 positively scaled singularity for leaf color parameters (a*, SPAD, h°) and sensory analysis (GP) and negatively scaled with growth parameters (Height, LDC, TDC, LW, 1stBH). Such response was closely associated with PBZ application or absence, respectively (Fig. S1 B). Likewise, in D30DAT, PCA1 positively scaled with leaf color (a*, SPAD, h°) contrasting growth parameters (LDC, Height and TDC). However, it showed otherwise a singularity between growth traits and sensorial analysis parameters, which also varied over the range of PBZ concentrations. This was evidenced by three to four clusters displayed in the biplot (Figure 2).

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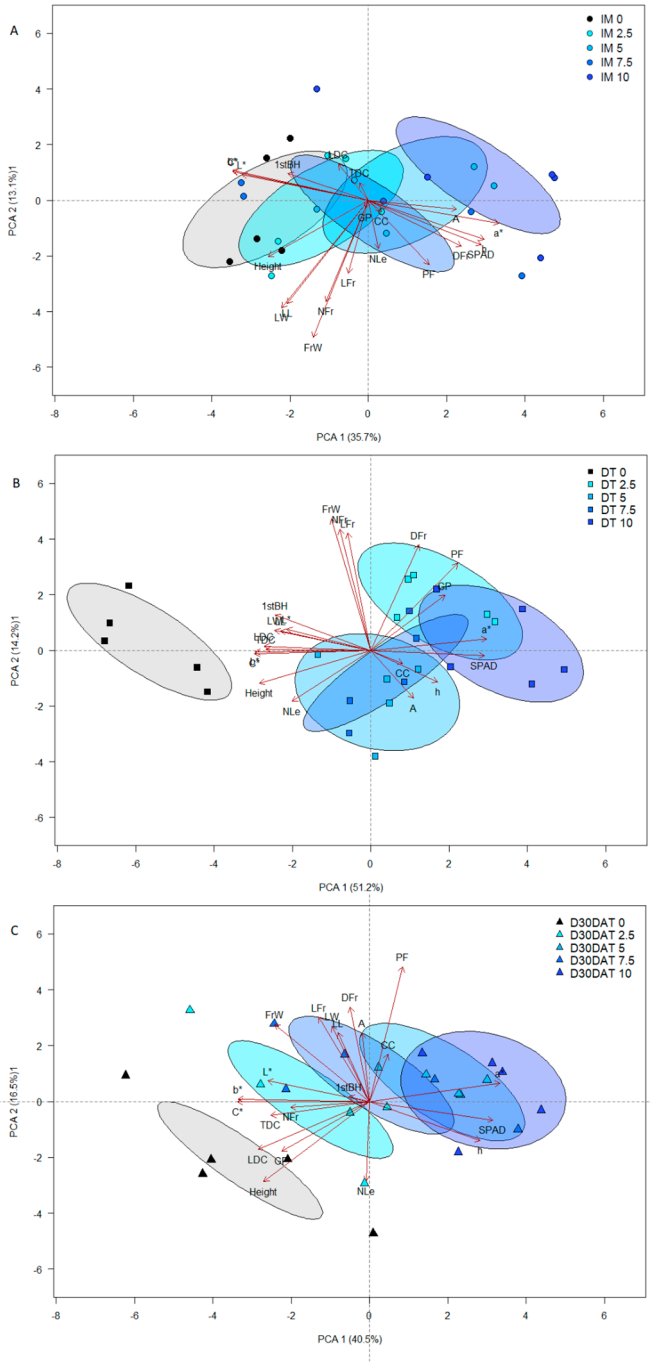

Figure 2. Principal component analysis biplot presenting major axes of variation for parameters related to ornamental traits and global preference (GP) in 'Biquinho Vermelha' pepper in three PBZ application protocols (A, Immersion; B, Drenching at transplant; and C, Drenching at 30 DAT). The arrows are vectors of correlation among variables and ellipses represent delineated groups based on normally distributed data for concentration of PBZ (0, 2.5, 5, 7.5 and 10 mg/L) in each application method. Symbols are mean values with n = 5 per group. Captions: Height (plant height); 1stBH (first bifurcation height); LDC (canopy longitudinal diameter); TDC (canopy transverse diameter); CC (canopy compactness); NFr (fruits number); NLe (leaves number); DFr (fruits diameter); LFr (fruits length); Frw (shoot fresh weight); SPAD (SPAD index); PF (plant fullness); LL (leaves length); LW (leaves width); A (days after transplant to anthesis); L* (lightness); a* (the red/green coordinate); b* (the yellow/blue coordinate); C* (chroma) e h (hue angle); GP (global preference).

4. Discussion

4.1. Ornamental Growth Traits

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In studies previously reported, plants treated with paclobutrazol by drenching showed a greater effect in reducing plant height regardless of the moment of application, at transplantation or 30DAT compared to the immersion root-substrate system in the solutions. The application of PBZ by drenching also proved to be efficient in controlling the growth of pepper plants of the varieties Pitanga, Bode Amarela and several accessions from Germplasm Banks of both species *C. annuum* and C. *chinense* [5,16–18], as well as several other ornamental potted plants [7,9,11,13,24–28].

Unlike plant height, there was a difference between the first bifurcation height when comparing the application by drenching at both moments, transplanting and 30DAT (Table 2). Applied PBZ during transplantation, the first bifurcation height was significantly lower. The branches of *Capsicum* plants follow a dichotomy model, when the plant reaches 15 to 20 cm in height, a young branch ends with one or several flowers, forming the first bifurcation (two new vegetative branches), which emerge from the axil leaves and will continue to grow until new flowers are formed. This vegetative process is repeated throughout the growth period conditioned by apical dominance and hormonal dependence [29]. Thus, when applied 30 days after transplanting, the plant remains longer free from the action of PBZ in inhibiting gibberellin synthesis, and consequently, the reduction in the length of the internode, allowing this first branch to reach a greater height before occurs the first bifurcation.

The greater plant fullness presented by plants treated with PBZ by D30DAT from a concentration of 5 mg L-1 prevents the substrate from being visible when the plants are viewed from above [10]. Despite the reduction in vegetative growth observed, the shoot fresh weight of plants treated with PBZ did not differ from control counterparts regardless of the concentrations used. In Petunia, on the contrary, a reduction in shoot fresh weight plants treated with the regulator was observed [13], and according to these authors, plant fullness is a characteristic measuring the general quality of plants since the density of their growth is an important attribute of appearance.

Plant fullness is given by the relationship between the fresh weight of the aerial part and the height of the plant, so the lack of PBZ concentration effect for this trait in IM treated plants is due also to the absence of significative effect of PBZ concentrations in height of plants treated with this protocol, even with the greater shoot fresh weight observed on plants treated this way compared to the others two protocols. The greater shoot fresh weight of IM treated plants is explained by the lower effectiveness of this protocol in reducing vegetative growth compared to the application of PBZ by drenching either at transplanting or at 30DAT.

The smaller canopy longitudinal diameter observed in plants to which PBZ was applied by DT at all concentrations and from a concentration of 5 mg L-1 when applied by D30DAT compared to untreated plants corroborates what was observed in Pitanga, ENAS-5007 and ENAS-5032 peppers [17,18] and in sunflower, zinia, basil and physalis grown in pots [24,27,30]. Since canopy compactness is obtained by the relationship between transverse and longitudinal diameters the smaller longitudinal diameter of drenching PBZ treated plants either at transplanting or at 30DAT, made these plants more circular and compact than those to which the PBZ was applied by immersion.

Higher values of canopy compactness were only observed at higher concentrations of PBZ, above 7.5 mg L-1. However, previously applied in higher concentrations (20 to 60 mg L-1) in 'Biquinho Vermelha' pepper by drench at 15DAT and in concentrations of 25 to 75 mg L-1 applied by immersion in 8 accessions of pepper plants, paclobutrazol showed no effect on canopy compactness [5,10].

The shorter length and width of the leaves of the plants to which PBZ was applied by DT is expected because when the application was made 30 days later the plants had an additional month of vegetative growth before undergoing the action of the growth regulator in inhibition of gibberellin synthesis. Reduction in the length and width of leaves of plants treated with PBZ at concentrations ranging from 25 to 75 mg L-1 compared to control plants was observed in 3 and 4 of the 8 pepper accessions evaluated, for leaf length and width, respectively [10]. In ornamental pineapple, plants treated with PBZ had a 64% reduction in leaf length [28].

The reduction in the number of leaves of plants treated with PBZ by D30DAT compared to application by DT was visually proportional to the reduction in height, without affecting the ornamental appearance of the plants. Corroborating the results found in this study, studying only

 doi:10.20944/preprints202409.2343.v1

the effect of PBZ application methods, without variation in its concentration, also did not observe a significant difference between the number of 'Biquinho' pepper leaves when PBZ was applied by drenching at transplant related to those in which the growth regulator was applied by immersion. However, for the 'Bode Amarela' variety contrary to what was observed in this work, the immersion method proved to be more effective and reduced the number of leaves by 68% compared to application by drenching at transplant [16].

Depending on the plant species, PBZ can delay or promote flowering [31]. Delays in flowering of plants treated with PBZ above 5 mg L-1 as compared to the control and between plants treated by DT as compared to IM were observed in this work. As in this research, the effect of PBZ concentration on delay in anthesis was also observed in potted zinnia and sunflower plants [30]. In platycodon, there was no significant effect of anthesis as compared to the concentrations of PBZ applied to the four varieties studied: 'Blue', 'Lavander', 'Pink' and 'White'. In mango, the effect is usually the opposite, with reports of induction and/or advancement of flowering being common [32–36].

Despite the delay in the beginning of flowering observed from the concentration of 5 mg L-1 this effect did not affect fruiting. However, reported that at higher doses, paclobutrazol caused a drastic reduction in the number of fruits when applied to 'Biquinho Vermelha' pepper by drenching at 15DAT [5]. On the contrary, in the same study, these authors did not observe a significant effect on the number of fruits in 'Bode Amarela' pepper and in the 2345PB and 2334PB accessions, clearly showing that there is a genetic and concentration effect concerning this characteristic. These variations in fruit set responses were also clearly observed in eight pepper accessions where PBZ was applied by immersion at concentrations ranging from 25 to 75 mg L-1 [10]. The fruits of treated plants, regardless of the concentration used, also had no change in length compared to untreated plants, a desirable characteristic for ornamentation. However, the choice of application method is important, since PBZ applied by DT reduced the number of fruits and the fruits produced by these plants were smaller in length. Ornamental pepper plants have their fruits as the main decorative element [10] and, therefore, the number and characteristics of the fruits produced are relevant factors in evaluating the ornamental quality of these plants.

4.2. Ornamental Color Traits

Greener leaves with a higher SPAD chlorophyll index were also reported in sunflower plants, peppers Biquinho Vermelha, Bode Amarela and Pitanga [5,8,17]. However, this effect is dependent on the concentration used in each mode of application, being significant when PBZ was applied earlier, at the time of transplantation. Applied 30 days later, only the highest concentration was able to significantly increase the chlorophyll index of the leaves. PBZ is capable of increasing cytokinin levels and consequently increasing chlorophyll synthesis in addition to preventing its degradation [37]. The contrast between the color of leaves and fruits is a visual attraction for consumers of ornamental peppers [38]. Thus, the increase in chlorophyll content after the application of PBZ intensifying the green can make the plants more attractive to the consumer, contributing to their commercialization.

The C* parameter or color saturation indicates the purity of the color, being used to determine the degree of difference of a tone compared to a gray color with the same luminosity, in which the higher the chroma values, the greater the color intensity of the samples [39]. In general, plants treated with PBZ by drenching based only on chromaticity values show less intense green, except for those treated at 30DAT at concentrations of 2.5 and 7.5 mg L-1. However, lower brightness (L*) and higher hue angle (h°) values indicated a darker shade of green and higher chlorophyll content in the leaves, respectively, compared to untreated plants. These changes were more subtle at the concentration of 7.5 mg L^{-1}, which meant that the difference was not significant only at this concentration. The h° value can be correlated with the chlorophyll content in leaves, where values close to 180° indicate leaves with a high chlorophyll content, and values close to 90° indicate chlorotic leaves, with a low chlorophyll content [40].

4.3. Sensory Analysis

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Global preference test showed that despite the insignificant effect on plant growth parameters, the IM application in low concentrations of PBZ (2.5 and 5 mg L^{-1}) produced plants with characteristics that attract the public as well as plants treated with PBZ by D30DAT in the same concentrations. The control at D30DAT is also among the most preferred plants (score 5.42). It is probably associated with the lower average height of the plants (31.4 cm) compared to the plants control of other application protocols. The control plants from IM protocol had an average height of 34.8 cm and an intermediate score (4.82) and the control plants from DT had an average height of 44.4 cm and the worst score of participants preference among all plants (4.02). The growth and development of all control plants were associated with the limitation of root growth by the volume of the pot used. However, as the volume of the pots was the same for all treatments and replications (1.17L), this difference in height between control plants may be due to environmental factors, such as light, which may have benefited the production of photo assimilates and the consequent growth and development of some plants to the detriment of others, despite the randomization carried out in the greenhouse.

The ornamental quality analysis shows that the effects of paclobutrazol were generally more significant on the characteristics of plants DT treated. However, two of these plants had intermediate scores (4.85 and 4.63 for concentrations of 2.5 and 7.5 mg L^{-1} respectively) and three of them are among the lowest scores in terms of overall preference among respondents (0, 5 and 10 mg L^{-1}). In a study carried out on the acceptance and preference of the consuming public of ornamental peppers grown in pots, when the interviewees were asked about the most relevant factors considered at the time of purchase, the color of the peppers followed by the general appearance of the plant, were the aspects most cited. The format of the plant was mentioned by only 4% of respondents and 'plant size' by only 2% [38]. According to these authors, the simultaneous occurrence of fruits of different colors on the same plant, due to different stages of maturation, contributes to the ornamental appearance. This color variability can be observed in 'Biquinho Vermelha' pepper plants at commercial maturity, when green (immature), yellow-orange (in the transition phase of the maturation process) and red (ripe) fruits are found, being a species of plant attractive for ornamental purposes.

Paclobutrazol is classified as a low-toxic product (category 4) and has been used in commercial greenhouses for more than 20 years [30], however, it is important to follow the recommendations printed on its labels for safe use in humans and the environment.

4.4. Interplay between Ornamental Traits and Sensory Analysis

The contrast between the color of leaves and fruits has been indicated as a visual attraction for consumers of ornamental peppers [38]. In this study, untreated plants and those that received the lowest concentration of PBZ by D30DAT, obtained the highest values of the parameters correlated to leaf color, especially SPAD and h°, which indicate leaves with a more intense/darker green, with a higher content of chlorophyll. However, the negative correlation between these parameters and the sensory analysis did not influence the interviewees' preference. Furthermore, these taller and less compact plants had greater preference over shorter and more compact plants. These observations indicate that the plant's general appearance may be more important, the plant height and leaf color being of little relevance when analyzed in isolation [38].

Plants treated with PBZ by DT had the interviewees' preference correlated positively with the more intense/darker green color of the leaves and negatively with the growth parameters. Plants smaller in total height and of the first bifurcation, in canopy diameter and with smaller leaf width and greener leaves had greater general preference within the group of plants treated by DT. However, although in this case the reduction in plant size and the greener color of the leaves were positively correlated with the interviewee's preference, application by DT produced plants with the lowest preference scores when all protocols were compared. This response was associated with a more drastic reduction in plant growth parameters. Taken together, these data suggest that DT protocol application of PBZ is not indicated for this pepper variety.

5. Conclusions

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PBZ treatment resulted in plants with darker green leaves and a striking contrast between fruits and foliage. Applying PBZ to 'Biquinho Vermelha' pepper plants via drench, either at transplanting or 30 days after transplanting (DAT), proved more effective in regulating plant growth and reducing canopy diameter. While applying PBZ by immersion had minimal impact on growth characteristics, sensory analysis revealed a higher preference for these plants compared to those treated with a low-concentration PBZ drench at 30 DAT. However, PBZ immersion is not recommended for enhancing ornamental quality, as it has little effect on key growth variables. Similarly, drench application at transplanting is discouraged because it leads to fewer and smaller fruits, diminishing the ornamental value of the plants and resulting in lower preference among participants. In contrast, drenching at 30 DAT with PBZ concentrations of 2.5 and 5 mg L^{-1} produced Biquinho Vermelha pepper plants with appealing ornamental traits, making it a viable option for ornamental plant producers.

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