

## ARTICLE

# Use of paclobutrazol as a growth reduction strategy for ‘*Vincent’s Choice*’ ornamental sunflower

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## Abstract

Floriculture has experienced exponential growth in Brazil, generating substantial revenue in the ornamental sector through the use of decorative flowers. Specific morphological traits are required to meet market standards for potted ornamental plants, such as compact architecture and restricted growth. Although ‘*Vincent’s Choice*’ sunflower is widely accepted by growers, its large size limits its commercialization in pots. This study aimed to evaluate different doses of paclobutrazol (PBZ), a plant growth regulator that inhibits the gibberellin biosynthesis pathway, the primary hormonal route for shoot elongation. PBZ was applied at 0, 100, 200, 300, 400, and 500 mg L<sup>-1</sup>, with five replicates per treatment. The variables analyzed included plant height, number of leaves, stem diameter, leaf area, fresh and dry biomass, and postharvest traits such as inflorescence diameter, colorimetric parameters (L\*, a\*, and b\*), visual quality (graded scale), and anatomical traits (transverse leaf blade thickness). The concentration of 500 mg L<sup>-1</sup> applied to the substrate demonstrated the most effective growth regulation without inducing phytotoxicity. This dose significantly reduced plant height and maintained postharvest quality, resulting in compact plants suitable for the producer and consumer markets of potted ornamental sunflower.

**Keywords:** floriculture, *Helianthus annuus* L., ornamental plants, plant growth regulator, plant height.

## Resumo

No Brasil, a floricultura tem crescido exponencialmente com um grande faturamento no setor de decoração, utilizando flores ornamentais. Para atender ao padrão do mercado de plantas ornamentais envasadas, é necessário que estas apresentem características morfológicas específicas, como uma arquitetura compacta e crescimento limitado. Embora o girassol cultivar ‘*Vincent’s Choice*’ seja bem aceito entre os produtores, seu grande porte impede sua comercialização em vaso. Objetivou-se estudar diferentes doses de Paclobutrazol (PBZ), um redutor de crescimento que atua na rota da biossíntese de giberelinas, principal hormônio de crescimento das plantas. Foram utilizadas concentrações de 0, 100, 200, 300, 400 e 500 mg L<sup>-1</sup> de paclobutrazol, utilizando cinco repetições para cada tratamento. Foram analisadas as variáveis altura da planta, número de folhas, diâmetro da haste, área foliar, pesos frescos e secos, além de análises pós-produção, como diâmetro da abertura da inflorescência, parâmetros L\*, a\* e b\* do colorímetro, qualidade visual através de uma escala de notas, e da análise anatômica (largura transversal das folhas). A concentração de 500 mg L<sup>-1</sup> aplicada ao substrato demonstrou ser a mais eficaz na regulação do crescimento, sem induzir fitotoxicidade. Essa dose reduziu significativamente a altura das plantas e manteve a qualidade pós-colheita, tornando as plantas compactas e adequadas para o mercado produtor e consumidor de plantas ornamentais envasadas.

**Palavras-chave:** altura da planta, floricultura, *Helianthus annuus* L., plantas ornamentais, regulador de crescimento de plantas.

## Introduction

The floriculture production chain has expanded exponentially in Brazil in recent years. Among its diverse segments, ornamental flower decoration stands out as one of the most profitable. In 2024, the sector recorded R\$ 21,3 billion in sales (Ibraflor, 2025). In 2024, the floriculture and ornamental plant industry employed 264.875 people, according to a study by the Center for Advanced Studies in Applied Economics (CEPEA) at Esalq/USP, in partnership with the Brazilian Institute of Floriculture (Ibraflor).

Among ornamental plants, sunflower (*Helianthus annuus* L.) has generated significant interest among consumers. It has a short growth cycle, is easy to manage and propagate, and can be used both for cut flowers and potted plants, making it versatile for arrangements and various decorative purposes (Nascimento et al., 2019; Ezz et al., 2021). In addition to its many uses, several sunflower cultivars are available, including ‘*Vincent’s Choice*’, considered the best hybrid in Brazil (Alves et al., 2025). This cultivar key advantage is its pollen-free nature, which increases its longevity for ornamental use. It can also be sown year-round, providing flexibility for growers. Its cycle ranges from 55 to 60 days, whereas Sunbright Kids, a pot cultivar, has a much longer cycle of 140 to 150 days (Sakata, 2023).

According to Almeida et al. (2024), meeting market standards for potted ornamentals requires specific morphological traits such as compact

architecture and controlled growth. Additionally, not only does the commercial success of a potted plant depend on its visual appeal, but also on postharvest longevity. Therefore, appropriate postharvest management tailored to each species and environmental condition is essential (Siqueira et al., 2024). Due to increased demand for high-quality ornamental plants and growing restrictions on chemical residues, the use of conventional postharvest preservatives has become limited (Almeida et al., 2020). In this context, growth regulators offer an alternative by delaying flower senescence (Siqueira et al., 2024).

Despite the high potential and popularity of ‘*Vincent’s Choice*’, which is regarded as one of Brazil’s best cut flower hybrids (Alves et al., 2025), its large size hinders its use as a potted ornamental and limits its marketability in that form. Therefore, strategies are necessary to control plant and inflorescence size while enhancing durability to meet the market demand for compact ornamental sunflowers.

The application of plant growth regulators to ornamental sunflowers has thus become a focus in recent research. Suzuki et al. (2018) demonstrated that the use of growth regulators such as mepiquat chloride and paclobutrazol can significantly reduce the height of the Florenza cultivar (*Helianthus annuus* cv. ‘Florenza’) without causing phytotoxicity, making the cultivar more compact and suitable for the potted ornamental plant market. Elisheba and Sudhagar (2021) evaluated paclobutrazol, cycocel, and Alar on the ‘Ring of Fire’ cultivar. Tütüncü and Çelikel

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(2022) tested AVG, Promalin, TDZ, and Etephon on the 'Sunsation F1' cultivar, highlighting the effectiveness of AVG and TDZ in improving stem quality and extending flower longevity. Mendes et al. (2025) studied paclobutrazol, trinexapac-ethyl, and chlormequat chloride on TERA 868 HO and HELIO 251, reporting reduced plant height and increased stem diameter. In a broader agricultural context, de Oliveira et al. (2023) demonstrated that paclobutrazol, mepiquat chloride, and prohexadione calcium effectively reduced stem elongation and lodging risk in common bean (*Phaseolus vulgaris*), with dose-dependent effects on seed yield and plant architecture.

Among these regulators, paclobutrazol (PBZ) stands out for its compacting effect and enhancement in ornamental quality. As a triazole compound, PBZ inhibits gibberellin (GA) biosynthesis, which involves hormones responsible for stem elongation, flower development, and senescence, without negatively affecting overall plant development or causing phytotoxicity (Desta and Amare, 2021). In addition to growth regulation, PBZ exhibits fungicidal and antimicrobial properties (NCBI, 2023). It can be applied to leaves, stems, branches, and propagative organs, but is most effective when applied to the soil or substrate, where it is passively absorbed by the roots and transported via xylem throughout the plant (Desta and Amare, 2021).

Paclobutrazol has been applied to several ornamental species (such as sunflower, lily, chrysanthemum, basil, ornamental pepper and ornamental pineapple) to produce compact and visually attractive plants (Ribeiro et al., 2019; Torres-Pio et al., 2021; Guimarães et al., 2021; Lailaty and Nugroho, 2022; Santos Filho et al., 2022; Sarkar, 2023; Tellez et al., 2024). Its effectiveness depends on cultivar dosage and application method. In sunflowers, paclobutrazol at 300–600 mg L<sup>-1</sup> reduced plant height by up to 50% in cv. Multissol without affecting biomass (Brito et al., 2016), and 300 mg L<sup>-1</sup> improved the chlorophyll index and capitulum diameter under salt stress in 'Sol Noturno' (Guimarães et al., 2021). Likewise, paclobutrazol at 90 ppm reduced height and internode length while increasing branching in 'Ring of Fire' (Elisheba and Sudhagar, 2021). When combined with supplemental lighting, paclobutrazol enhanced compactness and dry mass in petunia, geranium, pansy, and dianthus seedlings, with species-specific responses (Collado et al., 2022). In ornamental sunflower 'Golden', doses up to 6 mg a.i./pot also improved visual harmony, particularly in substrates with rice husk and sugarcane biochar (Barbosa et al., 2008).

PBZ-treated plants also exhibit increased levels of abscisic acid (ABA) and phytochrome, a chlorophyll component. Triazole compounds such as PBZ are known to improve tolerance to abiotic stress, including low temperature, drought, heat, pollutants, and heavy metals. These effects are likely due to increased cytokinin and ABA levels and reduced ethylene production (Desta and Amare, 2021).

Despite advances in research involving growth regulators applied to ornamental sunflowers, the *Vincent's Choice* cultivar, a modern hybrid widely used as a cut flower, had not yet been evaluated for its response to paclobutrazol for ornamental purposes. This lack of specific studies gives this research a novel and applied character, especially in light of market demands for compact, visually attractive plants with good postharvest longevity. Therefore, the objective of this study was to evaluate the effects of soil-applied paclobutrazol at different concentrations on the growth and inflorescence traits of the *Vincent's Choice* potted ornamental sunflower. The goal was to promote compact growth without phytotoxic effects, thereby meeting the commercial demands of the potted plant market.

## Materials and methods

The study was conducted from January 15 to March 23, 2024, in a greenhouse located at an altitude of 889 m, at 21°06' S and 44°15' W. Maximum and minimum temperatures, along with relative humidity, were recorded daily using a digital thermo-hygrometer. The average maximum and minimum temperatures were 47.42 °C and 20.46 °C, respectively. Relative humidity values averaged 92.67% (maximum) and 27.35% (minimum).

A randomized block design was adopted, with six treatments (paclobutrazol at 0, 100, 200, 300, 400, and 500 mg L<sup>-1</sup>) and five replications, totaling thirty experimental plots. Each plot consisted of eight

pots, each containing one plant. '*Vincent's Choice*' ornamental sunflower seeds were obtained from Sakata Seed Sudamerica® and sown in 1-L pots filled with Maxfertil® commercial substrate consisting of 85% composted pine bark, 10% vermiculite, and 5% carbonized rice hulls, supplemented with NPK fertilizer.

The product Cultar 250 SC®, containing paclobutrazol as the active ingredient, was used. At 16 days after sowing, when plants had reached an average height of 1.43 cm (measured from cotyledonary leaves), a single dose of the paclobutrazol solution was applied to the substrate in each pot at 100 mL per plant, according to treatment concentrations (Mateus et al., 2009). Throughout the experiment, standard cultivation practices were implemented to ensure healthy plant development, including daily irrigation, removal of lateral shoots, and appropriate pest and disease control measures. Additionally, substrate fertilization was carried out biweekly, following Malavolta's recommendation adapted by Torqueti et al. (2016) for potted plants.

### Plant Growth Analyses

The first growth assessment was performed 14 days after sowing, prior to the application of paclobutrazol. Subsequent measurements were taken at 15, 30, and 45 days after treatment. Parameters evaluated included plant height (measured with a ruler), number of leaves, stem diameter (measured with a caliper), leaf area, and fresh biomass. For dry biomass determination, leaves, stems, roots, and inflorescences (capitula) were separated, labeled, and dried in a forced-air oven at 65 °C until constant weight was achieved.

### Postharvest Quality Analyses

In the laboratory, inflorescence quality during flowering was evaluated every other day by visually assessing floral appearance. Twelve plants were used per treatment. Upon reaching the ideal market stage (R5), the capitulum diameter was measured using a digital caliper, and visual quality was graded using a visual scale. Ligule color was assessed with a Solab NR60CP® colorimeter based on L\*, a\*, and b\* values (CIELAB color system).

Visual quality was rated on a four-point scale, developed based on commercial criteria and aesthetic attributes relevant to the ornamental plant market (Tomiozzo et al., 2024, with adaptations). A score of 4 indicated excellent conditions and optimal marketability; score 3 represented good conditions, acceptable for sale; score 2 referred to fair conditions, suitable for display only; and score 1 indicated poor conditions, at which the plant was considered unsellable.

Chlorophyll *a* and *b* concentrations were also measured using the ChlorophyllLog CFL1030 (Falker®) electronic chlorophyll meter, following the methodology described by Santos Filho et al. (2022). This device calculates the Falker chlorophyll index by considering the total contents of chlorophyll *a* and *b*, rather than relying solely on light wavelength absorbance. Measurements were taken from the third fully expanded leaf (from top to bottom) of each plant during the reproductive stage.

### Anatomical Analyses

Anatomical analyses were performed on fully expanded leaves, which were collected and fixed in 70% GL alcohol. Freehand transverse sections were prepared by making horizontal cuts deep enough to observe mesophyll thickness.

The sections were obtained from 0.5-cm fragments taken from the median region of the leaf blade. These fragments were clarified in a 50% sodium hypochlorite solution, rinsed in distilled water, stained with safranin and astra blue, and mounted in 50% glycerin to measure the thickness of the adaxial and abaxial epidermis (Kraus and Arduin, 1997). Leaf blade cross-sections were obtained from three leaves per treatment, collected from the upper third of the plants. Three fields of view were analyzed per replicate, totaling fifteen observations per treatment. Slides were examined under an Olympus CX23® optical microscope and photographed using an Olympus EP50® camera. Measurements were taken using the ImageJ software.

### Statistical Analysis

To analyze plant growth parameters (plant height, number of leaves, stem diameter, leaf area, fresh and dry weight of leaves, stems, roots, and capitula), postharvest quality traits (capitulum diameter, *L*, *a*, and *b* colorimetric parameters, and visual grading scale), and anatomical traits (leaf blade thickness), analysis of variance (ANOVA) was performed using the F-test ( $p < 0.05$ ), followed by regression analysis. For postharvest quality and anatomical data, a completely randomized design (CRD) was adopted. Statistical analysis was conducted using the Sisvar software version 5.6 (Ferreira, 2019) for growth data and SPSS version 2.1 for postharvest and anatomical analyses.

### Results

#### Plant Growth Analyses

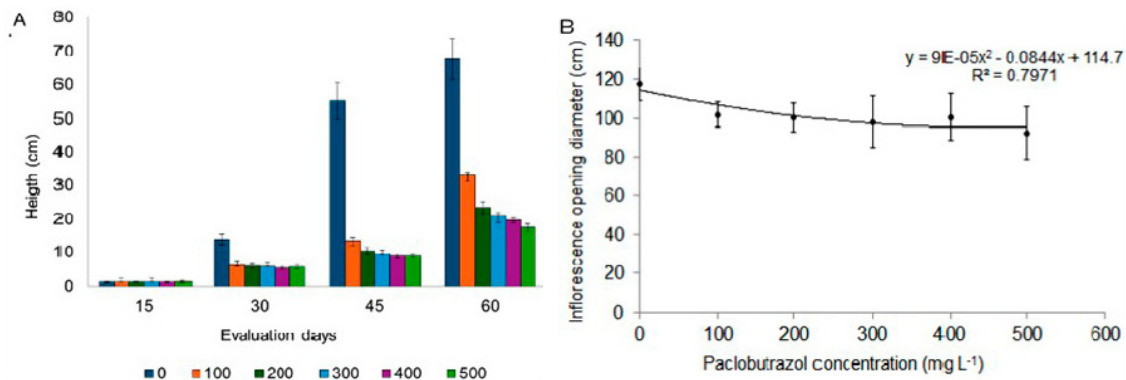
The application of the growth regulator paclobutrazol resulted in a linear reduction in plant height of ‘*Vincent’s Choice*’ ornamental sunflower (Fig. 1). This decrease was more pronounced at 45 and 60 days after planting, with the control treatment ( $0 \text{ mg L}^{-1}$ ) showing the highest average heights (1.395 cm at 15 days and 67.6 cm at 60 days), while the highest concentration ( $500 \text{ mg L}^{-1}$ ) resulted in the lowest values (1.52 cm at 15 days and 17.475 cm at 60 days). The average plant heights observed at 60 days were 67.6, 32.935, 23.515, 20.935, 19.66, and 17.475 cm for the concentrations of 0, 100, 200, 300, 400, and  $500 \text{ mg L}^{-1}$  of paclobutrazol, respectively (Fig. 2A).



**Fig. 1.** Effects of different paclobutrazol concentrations ( $\text{mg L}^{-1}$ ) on the size of ornamental sunflower plants grown in pots, evaluated 60 days after seedling transplant. Scale bar: 12 cm.

Capitulum diameter also showed a decreasing trend with increasing paclobutrazol concentrations 60 days after planting. The control treatment ( $0 \text{ mg L}^{-1}$ ) recorded the highest average value (118.05 cm), while the lowest average was observed at  $300 \text{ mg L}^{-1}$  (80.00 cm). The average

diameters were 118.05, 102.07, 105.71, 80.00, 101.77, and 92.72 cm for the treatments with 0, 100, 200, 300, 400, and  $500 \text{ mg L}^{-1}$  of paclobutrazol, respectively (Fig. 2B).



**Fig. 2.** Changes in plant height over time (A) and inflorescence diameter (B) of ‘*Vincent’s Choice*’ ornamental sunflower in response to paclobutrazol concentrations ( $\text{mg L}^{-1}$ ).

A linear decrease in leaf area was observed in response to increasing paclobutrazol concentrations (Fig. 3A), and the number of leaves began to decrease from  $300 \text{ mg L}^{-1}$  onward (Fig. 3B). It was not possible to fit a consistent model for stem diameter over time.

Fresh leaf biomass followed a quadratic regression pattern, with a decreasing trend beginning at  $400 \text{ mg L}^{-1}$  (Fig. 3C). No statistically significant differences were observed for dry leaf biomass.

Fresh stem biomass decreased as paclobutrazol doses increased, with a slight increase at the highest concentration (Fig. 3D). Dry stem biomass, however, was reduced starting at  $300 \text{ mg L}^{-1}$  (Fig. 3E).

Regarding root biomass, both fresh and dry weight increased compared to the control, with peak values at  $300 \text{ mg L}^{-1}$ , followed by reductions at  $400 \text{ mg L}^{-1}$  and above (Fig. 3E, F).

Finally, paclobutrazol also influenced chlorophyll *a* and *b* concentrations in leaves, resulting in increased pigment levels compared to the control (Fig. 4A–B).

#### Postharvest Quality Analyses

The diameter of inflorescence opening in ‘*Vincent’s Choice*’ ornamental sunflower showed a statistically significant difference, with more pronounced reductions at higher paclobutrazol concentrations, without compromising overall inflorescence quality (Fig. 5).

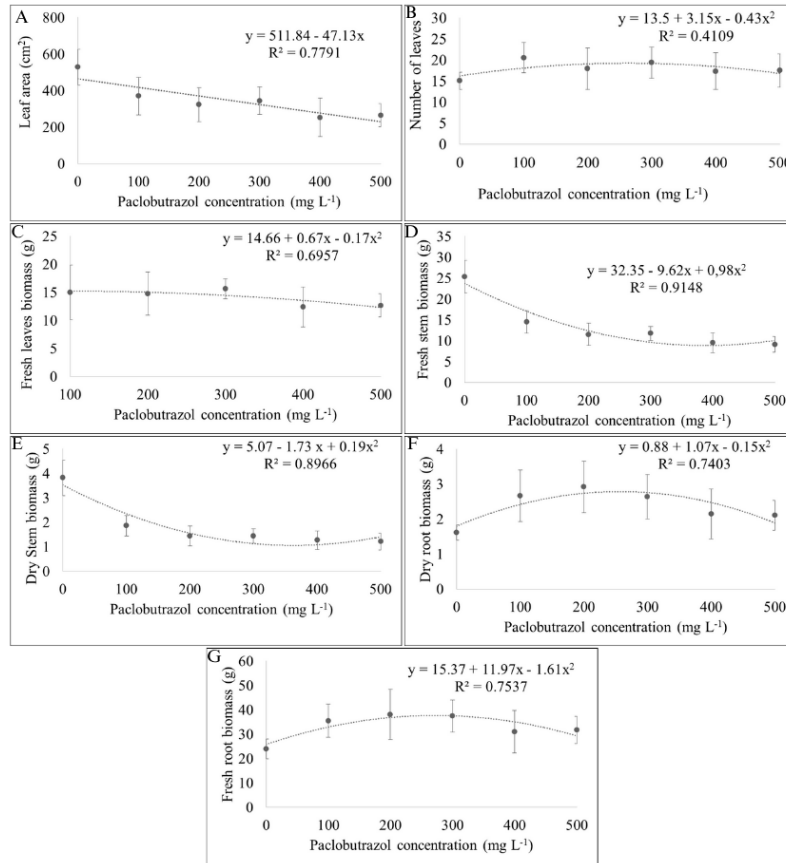


Fig. 3. (A) Leaf area, (B) number of leaves, (C) fresh leaf biomass, (D) fresh stem biomass, (E) dry stem biomass, (F) dry root biomass, and (G) fresh root biomass of 'Vincent's Choice' ornamental sunflower as a function of paclobutrazol concentrations (mg L<sup>-1</sup>).

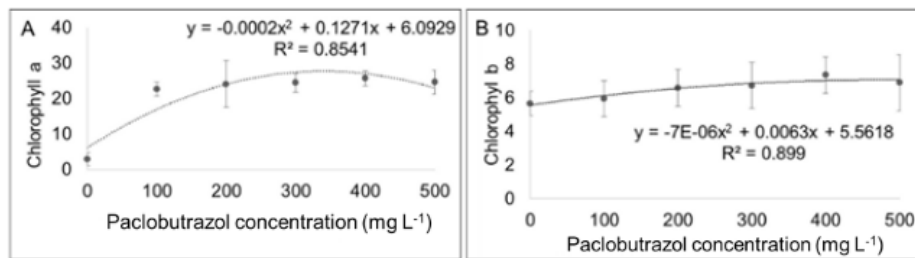


Fig. 4. Chlorophyll a and b content in 'Vincent's Choice' ornamental sunflower in response to paclobutrazol concentrations (mg L<sup>-1</sup>).

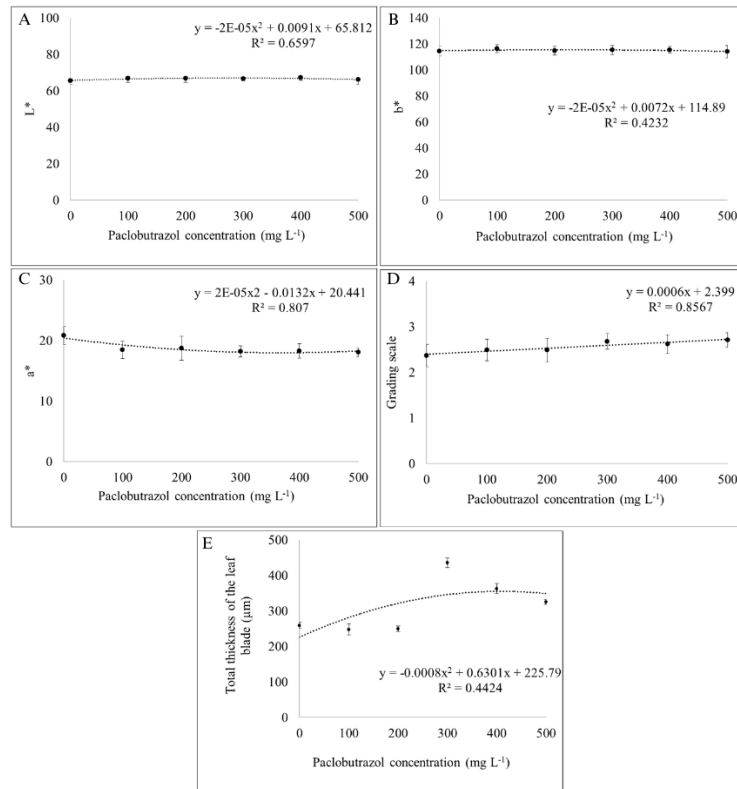


Fig. 5. Visual comparison of inflorescence quality in 'Vincent's Choice' ornamental sunflower during the postharvest period at different paclobutrazol concentrations (mg L<sup>-1</sup>). (A-F) Excellent quality. (A.1-F.1) Final stage of the plant cycle.

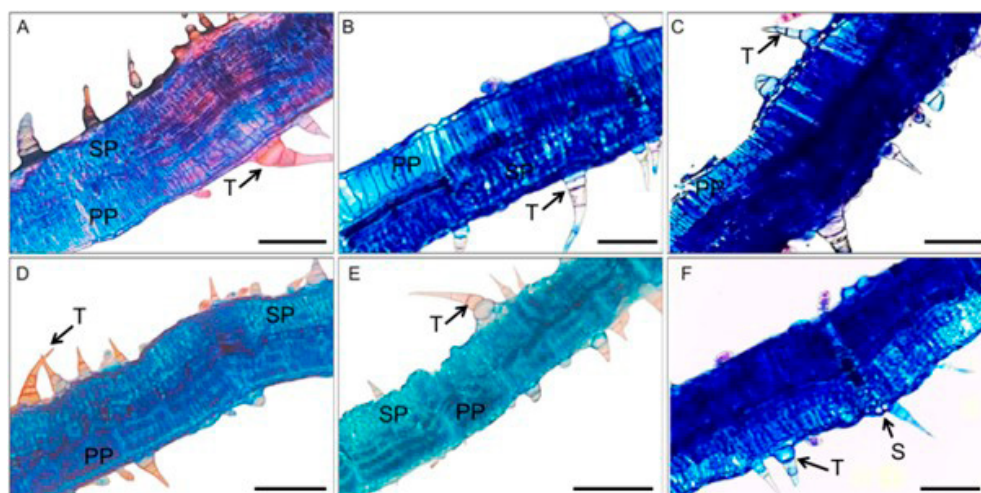
The  $L^*$  and  $b^*$  values from the CIELAB color system did not differ significantly among treatments, indicating that paclobutrazol application did not affect luminosity (L) or the blue–yellow color dimension (B) of the ligules (Fig. 6A–B). However,  $a^*$  exhibited a significant difference, with a reduction in values compared to the control, indicating a lower intensity of reddish tones in plants treated with paclobutrazol (Fig. 6C).

Regarding the visual quality of the inflorescences, a significant difference was observed between the control and the treated groups. Scores obtained from paclobutrazol-treated plants approached level 3 on the rating scale, indicating a good aesthetic standard for commercial purposes (Fig. 6D).

The analysis of total leaf blade thickness demonstrated that plants treated with paclobutrazol exhibited a statistically significant increase in thickness compared to the control (Figs. 6E and 7A–F).



**Fig. 6.** Colorimetric parameters, visual quality scale, and total leaf blade thickness of ‘*Vincent’s Choice*’ ornamental sunflower (*Helianthus annuus*) as affected by paclobutrazol concentrations ( $\text{mg L}^{-1}$ ). (A)  $L^*$ , (B)  $b^*$ , (C)  $a^*$ , (D) Visual quality rating, and (E) Total leaf blade thickness.



**Fig. 7.** Anatomical changes in the leaves of ‘*Vincent’s Choice*’ ornamental sunflower (*Helianthus annuus*) in response to different paclobutrazol concentrations ( $\text{mg L}^{-1}$ ). PP = palisade parenchyma; PS = spongy parenchyma; T = trichome; S = stomata. A: control; B:  $100 \text{ mg L}^{-1}$ ; C:  $200 \text{ mg L}^{-1}$ ; D:  $300 \text{ mg L}^{-1}$ ; E:  $400 \text{ mg L}^{-1}$ ; F:  $500 \text{ mg L}^{-1}$ . Scale bars: a, d, and E =  $150 \mu\text{m}$ ; B, C, and F =  $100 \mu\text{m}$ .

## Discussion

### Plant Growth Analyses

The reduction in plant height observed in this study supports the findings of Brito et al. (2016), Wanderley et al. (2014), and Mateus et al. (2009), who reported similar effects in different sunflower cultivars treated with paclobutrazol. This behavior can be assigned to the inhibition of gibberellin biosynthesis, caused by the regulator action on the cyclization of GGPP into ent-kaurene, as described by Taiz and Zeiger (2024) and demonstrated by Zhang et al. (2022) and Suzuki et al. (2018), who reported significant reductions in stem elongation in *Jasminum sambac* and 'Florenza' *Helianthus annuus* following paclobutrazol application, respectively.

Although the reduction in capitulum diameter was significant, it did not compromise the visual standard required for commercialization, which is favorable from an ornamental standpoint. Studies such as that of Wanderley et al. (2014) reinforce this potential.

Conversely, Guimarães et al. (2021) showed that, under saline stress conditions, paclobutrazol positively influenced the inflorescence development of 'Sol Noturno' cultivar, highlighting its versatility as an abiotic stress mitigator.

The reduction in leaf area follows a pattern similar to that reported by Tellez et al. (2024) in ornamental pineapple and may be explained by the effect of paclobutrazol on limiting cell elongation and leaf expansion (Desta and Amare, 2021).

With respect to leaf number, the decrease starting at 300 mg L<sup>-1</sup> partially contrasts with the findings of Brito et al. (2016), who observed no significant changes in the 'Multissol' cultivar. Similar results were reported by Lailaty and Nugroho (2021) for chrysanthemums.

The absence of a consistent model for stem diameter was also observed in studies by Brito et al. (2016) and Almeida et al. (2024), suggesting genotypic variations in response to the regulator.

Regarding leaf and stem fresh and dry biomass, the results indicate that higher PBZ doses negatively affect biomass accumulation. However, the quadratic trend in fresh leaf biomass suggests the existence of a balance point. This is consistent with findings by Tellez et al. (2024) and Wanderley et al. (2014), who also reported reductions in biomass as PBZ concentrations increased.

For root development, the increase in biomass at intermediate doses aligns with results from Berova and Zlatev (2000) and Shahrokhi et al. (2011), showing that moderate concentrations can stimulate root growth, while higher doses tend to inhibit it.

Sarkar (2023) highlighted that soil-applied PBZ is absorbed by the roots and transported exclusively through xylem tissue, which explains its primary effect on shoot growth rather than root tips.

The increased levels of chlorophyll *a* and *b* are similar to those reported by Santos Filho et al. (2022) and Sarkar (2023), suggesting enhanced photosynthetic efficiency in compact leaves, possibly due to a higher concentration of chloroplasts per leaf area.

### Postharvest Quality Analyses

The reduction in inflorescence diameter at higher paclobutrazol doses confirms the regulator potential for producing compact plants without negatively affecting their ornamental quality, a desirable trait for the potted plant market.

The lack of significant differences in *L\** and *b\** parameters suggests that paclobutrazol does not substantially interfere with brightness or blue-yellow coloration, thus preserving visual quality. However, the decrease in *a\** values indicates a lower intensity of reddish tones, showing that the regulator selectively affects certain chromatic aspects of the plant.

The average visual rating of 3 indicates a commercially viable standard, even at the highest PBZ doses. Although no specific analyses of ligule senescence were conducted, the good postharvest visual conservation suggests that paclobutrazol may contribute to improved inflorescence longevity, which is advantageous from both logistical and commercial perspectives.

The increased leaf blade thickness in response to PBZ is consistent with the findings of Lailaty and Nugroho (2021) in chrysanthemums. This thickening may be explained by the formation of additional cell layers, resulting from structural changes induced by the regulator.

Therefore, despite the reduction in total leaf area, the compensatory thickening of the leaf blade may be understood as a functional adaptation. This anatomical reorganization can preserve the photosynthetic capacity

of the plants, supporting their physiological balance. Similar findings were reported by Suizu et al. (2021), who demonstrated that interferences in the gibberellin pathway via paclobutrazol altered both leaf anatomy and photosynthetic biochemistry in *Eleocharis vivipara*, reinforcing that structural modifications may occur independently of biochemical expression. In this context, the adjustments promoted by PBZ contribute to maintaining ornamental quality and favorable postharvest performance.

## Conclusions

The use of paclobutrazol as a growth regulation strategy for 'Vincent's Choice' ornamental sunflower proved to be effective. The concentration of 500 mg L<sup>-1</sup> was the most efficient in reducing plant height, leading to a 74.15% decrease compared to the control treatment. This dosage is recommended for growth containment of this cultivar, favoring the production of more uniform, robust plants with greater postharvest durability, attributes highly valued in the ornamental market that contribute to improved logistics and commercial acceptance.

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### Author Contribution

**MESS:** Conceptualization, Investigation, Formal Analysis, Writing – Original Draft. **LMC:** Investigation, Writing – Original Draft, Writing – Review & Editing. **AMPN:** Formal Analysis, Writing – Original Draft, Writing – Review & Editing. **GSM:** Formal Analysis, Writing – Original Draft, Writing – Review & Editing. **GMPC:** Investigation. **PRV:** Investigation. **SNO:** Writing – Review & Editing. **FCN:** Conceptualization, Writing – Original Draft, Writing – Review & Editing.

### Conflict of interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

### Data availability

All the research data is contained in the manuscript.

### Declaration of generative AI and AI-assisted technologies in the writing process

The authors declare that the use of AI and AI-assisted technologies was not applied in the writing process.

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