Effect of paclobutrazol on growth retarding in potted chrysanthemum (*Dendranthema grandiflora*)

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Piphatwatthanakul, P., Boonkamjat, T. and Saetiew, K. (2024). Effect of paclobutrazol on growth retarding in potted chrysanthemum (*Dendranthema grandiflora*). International Journal of Agricultural Technology 20(2):731-748.

Abstract Paclobutrazol application was compared to the control. It showed a positive influence in reducing stem height, canopy width, internode length, node number, and flower size. Besides, methods of application were insignificantly found. As for the second experiment, decreased concentrations to 0, 100, 200, 300, and 400 ppm of paclobutrazol were tested, using the same manner of application as in the first experiment. It appeared that all concentrations of paclobutrazol decreased canopy width, stem length, internode length, and node number when compared with the control treatment, similar to results of the experiment 1. Nevertheless, it occurred in Prakaimat variety that both foliar spraying and soil drenching of paclobutrazol led to increasing chlorophyll content and carotenoid level. Meanwhile, Fiona chrysanthemum responded better to foliar spraying method than of soil drenching.

Keywords: Chlorophyll, Foliar spraying, Morphology, Plant growth regulator, Soil drenching

Introduction

The Chrysanthemum (*Dendranthema grandiflora*), belonging to the family Asteraceae, holds considerable economic importance as a prominent flowering plant species. Originally from China and Japan, this visually captivating and chromatically diverse flower has garnered noteworthy attention. The extensive assortment of its cultivars further underscores its botanical and high commercial value (Krasaechai, 1992). The floriculture industry has evolved into a multifaceted endeavor of significant importance, encompassing aesthetic, social, environmental, and economic dimensions. In recent years, it has become a lucrative agribusiness, driven by a growing desire for floricultural products spurred by improved living standards and an increased societal emphasis on eco-friendly environments (Vahoniya *et al.*, 2018). Commonly employed as a cut flower and potted plant. Beyond its ornamental applications, the chrysanthemum is a versatile resource for pot mums, border plants, and hanging baskets. Pot

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chrysanthemum holds a significant position within the European, North American, and Japanese floriculture markets as a favored option for enhancing residential environments, including living spaces, balconies, and patios. Rapid urbanization and evolving lifestyles notably influence the burgeoning demand for potted plants in Thailand.

Furthermore, instances arise where the quality produced does not align with market preferences, prompting the substantial importation of ornamental plants like chrysanthemums. Consequently, endeavors have been undertaken to augment the worth of chrysanthemums through their presentation as potted ornamental plants (Samyo and Atthalangrong, 2020). The production of potted ornamental plants encompasses various methodologies. These methods include deliberately choosing plant varieties developed explicitly for optimal potted plant cultivation. Additionally, techniques such as pinching, pruning, and applying plant growth retardants are employed to refine the growth and form of these plants (Hongpakdee, 2014). Pot mums exhibit versatility and aesthetic appeal, rendering them adaptable to diverse container sizes and types. Applying plant growth retardants becomes imperative to preserve their stature and compact form. These retardants are routinely employed to curtail stem elongation across numerous ornamental plant varieties. Mechanistically, plant growth retardants operate through the inhibition of cell division within the subapical meristem of the shoot (Ghosh and Rao, 2015).

Paclobutrazol (PBZ) functions as a plant growth retardant, exerting control over canopy size, peduncle length, and the gibberellin synthesis process responsible for elongating plant internodes (Saenthongkham et al., 2019). Such attributes hold potential for potted chrysanthemum production. Plant growth retardants, exemplified by paclobutrazol, feature prominently in managing plant height by inhibiting gibberellin biosynthesis (Karimi et al., 2014). PBZ has demonstrated efficacy in controlling plant stature across various flowering potted species. Its use in spray form regulates poinsettia height and flowering (Faust et al., 2001). Meanwhile, PBZ drench immersion proves more effective than spray for height control in *Dianthus carvophyllus* (Bañon et al., 2002). Applying PBZ to easter lily tubers at a 120 mg/l concentration reduces stem height without compromising 'Nellie White' easter lily flowering (Currey and Lopez, 2010). PBZ also stimulates early flower bud development in azaleas (Christiaens et al., 2012). and augments flowering in China aster (Mishra et al., 2005). However, delayed flowering has been noted, as seen in mini carnation (Atanassova et al., 2004). and hibiscus (Nazarudin, 2012). Studies indicate PBZ can potentially enhance antioxidant systems, such as vitamins C and E, under salinity or waterdeficit stress conditions (Jungklang et al., 2017). PBZ application methods encompass foliar spraying, propagule soaking, and substrate drenching, yielding superior outcomes while diminishing plant attributes. Notably, PBZ was low solubility grants it a persistence of up to 12 months in soil post-application. Efficacy hinges upon appropriate dosages, application techniques, plant varieties, cultivars, and environmental contexts (Phasri *et al.*, 2019). Notably, Prakaimat and Fiona chrysanthemum cultivars primarily utilized as cut flowers have not undergone potted plant treatment with paclobutrazol. Thus, the investigation determined the optimal paclobutrazol concentration and administration method for producing potted chrysanthemums.

Materials and methods

Plant material

The chrysanthemum, comprising the Prakaimat and Fiona cultivars, was cultivated from plant tissue cultures. Initially, they were transplanted into nursery trays with a planting substrate composed of peat moss. After three weeks, they were further transplanted into 8-inch pots with a planting mixture consisting of composted samenea leaves, finely chopped coconut husks, raw rice husks, and rice husk ash, with a ratio of 2:1:1:2. Subsequently, shoots were pinched from the chrysanthemums after two weeks to encourage branching. Three weeks later, 10 cm branches were excised from the parent plants, soaked in an anti-fungal solution for 5 minutes, and then planted in nursery trays. When these branches reached the age of 3 weeks, they were transplanted into 6-inch pots, with three plants per pot, using the same planting mixture as the parent plants. The cultivation process occurred in a greenhouse with LED lighting providing 4 hours of light, maintaining a relative humidity of 40-60%, and the diurnal temperature range encompasses approximately 32±3°C during the day and approximately 26±2 °C during the night. To regulate plant growth, 15% Paclobutrazol was administered through soil drenching twice or foliar spraying twice at various concentrations, with 50 ml applied per pot, when the chrysanthemums were 21 and 28 days.

Experiments set up

The experiment plans were performed as 5x2 factorial experiment in a completely randomized design, incorporating two primary factors. Experiment 1 comprised two factors, i.e., concentration levels, including 0, 400, 800, 1200, and 1600 ppm, and application methods, namely soil drenching and foliar spraying. In Experiment 2, the concentration levels were adjusted to 0, 100, 200, 300, and 400 ppm while maintaining the same application methods as in Experiment 1.

Data collection and analysis

Data collection encompassed growth parameters of chrysanthemum plants, encompassing stem height, canopy width, internode length, node number, flower number, and flower size. Paclobutrazol was administered biweekly for 13 weeks, with flower size measurements conducted once during the final week of the experiment. Statistical analysis involved SAS version 9.0 (SAS Institute, Inc., Cary, NC), and treatment comparisons were analyzed by using Duncan's new multiple test (DMRT) at a significance level of P = 0.05. When the interaction between two factors lacked significance or displayed consistent treatment responses, data from multiple replications were consolidated for analysis.

Results

Stem height and canopy width

The interaction between concentrations of PBZ and methods provided in two cultivars of chrysanthemum, i.e., Prakaimat and Fiona. The findings were observed in the Prakaimat for the growth retardation which observed at all concentrations, and it was significantly influenced in stem height and canopy width when compared to the control (Figure 1). Concurrently, the application method exhibited nonsignificant on plant growth retardation. However, through a comprehensive analysis of the interaction between these two factors, it became apparent that the application method involving soil drenching at 1600 ppm resulted in the most significant reduction in yield, stem height and canopy width of 14.69 cm and 8.27 cm, respectively (Table 1).

Similarly, in the Fiona at 1600 ppm by soil drenching reduced stem height and canopy width of 20.77 cm and 11.75 cm, respectively. Meanwhile, in the second experiment, the PBZ concentration level was observed to influenced stem height and canopy width retardation. Despite the application method, growth retardation remained unaffected in both cultivars. However, all concentrations and application methods of Prakaimat induced discernible effects on the growth including growth retardation, stem height, and canopy width. Applying 400 ppm concentration via soil drenching resulted to reduce stem height and canopy width of 12.83 cm and 7.88 cm, respectively. The soil drench method of Fiona at 400 ppm was significantly reduced in stem height. In contrast, a foliar spray method at 400 ppm reduced the canopy width by 13.50 cm (Table 2).

experiment 1						
Concentration	Method of provide	Stem heig	ght (cm)	Canopy width (cm)		
of PBZ (ppm)		Prakaimat cv.	Fiona cv.	Prakaimat cv.	Fiona cv.	
0	Soil drench	46.71±6.70 ^a	59.63±1.66 ^a	27.00±6.09 ^a	$34.42{\pm}10.10^{a}$	
400	Soil drench	12.17±0.71 ^b	36.46±8.56 ^b	10.67 ± 0.65^{b}	12.17±13.90 ^{ab}	
	Foliar spray	16.40±1.75 ^b	23.79±0.52 ^{cd}	12.19±1.14 ^b	16.40±1.13 ^b	
800	Soil drench	13.46±6.20 ^b	24.79±2.32 ^{cd}	10.33±1.53 ^b	14.75±1.37bc	
	Foliar spray	15.94±6.20 ^b	26.21±2.42°	12.27±1.09 ^b	14.71±1.72 ^{bc}	
1200	Soil drench	15.02±6.20 ^b	23.42 ± 0.87^{cd}	9.23 ± 1.53^{b}	16.63±0.95 ^{bc}	
	Foliar spray	17.01±6.20 ^b	26.65±1.50°	9.17±2.25 ^b	16.92±2.86bc	
1600	Soil drench	14.69±6.20 ^b	20.77±2.63 ^d	8.27±1.39 ^b	11.75±3.50°	
	Foliar spray	17.02±6.20 ^b	22.90±3.27 ^{cd}	$9.36{\pm}0.77^{b}$	16.59±6.43 ^{bc}	
Conc.		**	**	**	**	
Method		ns	ns	ns	ns	
Conc. x Method		**	**	**	**	
CV (%)		16.03	10.27	22.06	34.02	

Table 1. The effect of paclobutrazol on stem height and canopy width in 'Prakaimat' and 'Fiona' cultivars of potted chrysanthemums for 13 weeks in experiment 1

**/highly significant */statistically significant ns/non-significant at P = 0.05; Means within column follows by the same letter are not significantly different as determined by Duncan's multiple range test

Table 2. The effect of paclobutrazol on stem height and canopy width in 'Prakaimat' and 'Fiona' cultivars of potted chrysanthemums for 13 weeks in experiment 2

Concentration	Method of	Stem heig	ght (cm)	Canopy width (cm)		
of PBZ (ppm)	provide	Prakaimat cv.	Fiona cv.	Prakaimat cv.	Fiona cv.	
0	Soil drench	28.79±3.39ª	52.58±0.55ª	18.46±1.61ª	21.19±1.25ª	
100	Soil drench	$19.25{\pm}1.66^{bcd}$	$36.83{\pm}5.21^{b}$	13.79±3.82 ^b	16.15±0.29 ^b	
100	Foliar spray	16.42±1.75 ^{de}	32.00±1.78°	11.02 ± 3.26^{bcd}	$15.92{\pm}1.16^{bc}$	
200	Soil drench	$15.73{\pm}0.94^{ef}$	31.83±1.73°	$10.92{\pm}0.65^{bcd}$	15.19 ± 1.16^{bcd}	
	Foliar spray	17.58±0.91 ^{cde}	30.98±4.17°	10.08±0.35 ^{cde}	15.23 ± 1.59^{bcd}	
200	Soil drench	14.58 ± 1.39^{ef}	$25.10{\pm}1.60^{d}$	$8.33{\pm}0.30^{de}$	$14.23{\pm}0.69^{cd}$	
300	Foliar spray	19.96±1.16 ^{bc}	30.02±1.04°	11.58±0.65 ^{bc}	14.38 ± 1.42^{bcd}	
400	Soil drench	$12.83{\pm}0.33^{\rm f}$	$23.38{\pm}1.53^d$	7.88±0.08 ^e	14.33 ± 1.31^{bcd}	
400	Foliar spray	21.33 ± 2.46^{b}	29.21±1.40°	11.58 ± 1.17^{bc}	$13.50{\pm}0.91^{d}$	
Conc.		**	**	**	**	
Method		**	ns	ns	ns	
Conc. x Method		**	**	**	**	
CV (%)		10.24	7.07	14.83	7.19	

**/highly significant */statistically significant ns/non-significant at P = 0.05; Means within column follows by the same letter are not significantly different as determined by Duncan's multiple range test

Internode length and node number

Application method, concentrations of PBZ and interaction between these factors had significant effects on internode length and node numbers, PBZ effecting growth in the Prakaimat when employing soil drenching at 800 ppm and foliar spraying at 400 ppm led to the most effect on internode length of 0.44 cm. Additionally, foliar spraying with PBZ of 1200 ppm resulted in the lowest node number of 14.71 per plant. Similarly, the lowest internode length and node number decreased in Fiona at 1200 and 1600 ppm by soil drenching of 0.43 cm and 21.92 per plant, respectively (Table 3). However, it was found that the methods were not significant, probably due to the high concentration in Experiment 1, causing the methods not to affect reducing the internode length and node number in both cultivars. In the second experiment, the interaction between PBZ concentrations and application methods influenced retardation in yield, internode length and node numbers. The Prakaimat, applying soil drenching at 400 ppm, resulted in the most significant of internode length, and node number, with notable reductions of 0.38 cm and 14.33 per plant. The soil drench method of Fiona at 400 ppm leads to a reduction in internode length and node numbers of 0.54 cm and 24.50 per plant, respectively (Table 4).

Table 3. The effect of paclobutrazol on internode length and node number in 'Prakaimat' and 'Fiona' cultivars of potted chrysanthemums for 13 weeks in experiment 1

Concentration of	Method of	Internode length (cm)		Node number/plant	
PBZ (ppm)	provide	Prakaimat cv.	Fiona cv.	Prakaimat cv.	Fiona cv.
0	Soil drench	1.97±0.13ª	1.98±0.13ª	26.25±1.66 ^a	28.21 ± 1.54^{a}
400	Soil drench	0.48 ± 0.03^{b}	0.73 ± 0.19^{b}	15.75±1.40 ^b	23.17±0.58bc
	Foliar spray	$0.44{\pm}0.03^{b}$	0.53±0.03°	16.04±0.89 ^b	23.25±1.29 ^{bc}
800	Soil drench	0.44 ± 0.06^{b}	0.49±0.03°	15.50±0.79 ^b	24.34±1.25 ^b
	Foliar spray	0.46 ± 0.05^{b}	0.52±0.03°	15.79±1.23 ^b	23.50±0.69 ^{bc}
1200	Soil drench	0.47 ± 0.02^{b}	0.43±0.03°	14.88 ± 0.83^{b}	23.58±0.50 ^{bc}
	Foliar spray	0.45 ± 0.04^{b}	0.56±0.06°	14.71±1.57 ^b	23.17±0.58bc
1600	Soil drench	0.48 ± 0.03^{b}	0.43±0.03°	15.50±2.04 ^b	21.92±1.23°
	Foliar spray	0.46 ± 0.04^{b}	0.49±0.04°	15.59±1.23 ^b	22.00±0.55°
Conc.		**	**	**	**
Method		ns	ns	ns	ns
Conc. x Method		**	**	**	**
CV (%)		8.85	10.63	7.86	4.37

**/highly significant */statistically significant ns/non-significant at P = 0.05; Means within column follows by the same letter are not significantly different as determined by Duncan's multiple range test

Concentration	Method of	Internode length (cm)		Node number/plant	
of PBZ (ppm)	provide	Prakaimat cv.	Fiona cv.	Prakaimat cv.	Fiona cv.
0	Soil drench	1.83±0.13ª	2.25±0.12 ^a	24.50±0.43ª	31.46±1.23ª
100	Soil drench	$0.57{\pm}0.05^{bc}$	$1.30{\pm}0.08^{b}$	$20.42{\pm}0.50^{b}$	27.71 ± 0.44^{b}
100	Foliar spray	0.60±0.03 ^b	1.35±0.09 ^b	18.17±1.97°	26.67 ± 0.82^{bc}
200	Soil drench	$0.55 {\pm} 0.03^{bc}$	1.06±0.04°	18.46±0.42°	27.63±0.55 ^b
	Foliar spray	0.58 ± 0.03^{bc}	1.15 <u>+</u> 0.12 ^c	20.50 <u>+</u> 0.58 ^b	26.29 <u>+</u> 0.97 ^{cd}
300	Soil drench	$0.44{\pm}0.02^{de}$	0.80±0.09°	15.67 ± 0.61^{d}	25.33±0.27 ^{de}
	Foliar spray	$0.52{\pm}0.03^{bcd}$	$0.87{\pm}0.05^{de}$	21.75±0.50 ^b	26.21±0.37 ^{cd}
400	Soil drench	0.38±0.03e	$0.54{\pm}0.03^{\rm f}$	14.33±0.47 ^e	24.50±1.04e
	Foliar spray	$0.48{\pm}0.02^{cd}$	0.69±0.06e	21.42±1.55 ^b	26.50±0.43 ^{bcd}
Conc.		**	**	**	**
Method		**	**	**	ns
Conc. x Method		**	** ** **		**
CV (%)		8.15	7.12	4.55	2.96

Table 4. The effect of paclobutrazol on internode length and node number in 'Prakaimat' and 'Fiona' cultivars of potted chrysanthemums for 13 weeks in experiment 2

**/highly significant */statistically significant ns/non-significant at P = 0.05; Means within column follows by the same letter are not significantly different as determined by Duncan's multiple range test

Flower number and flower size

The study in experiment 1, it was found that in Prakaimat, soil drenching and foliar spraying at 800 ppm has the highest flower number at 59.17 per plant and the lowest at 29.83 per plant. Moreover, flower size decreased at 1600 ppm by soil drenching, and the smallest flower size was 2.58 cm. However, the concentration levels of PBZ were not significant in the flower number. Meanwhile, both methods significantly increased the flower number and reduced the flower size in Prakaimat. Which differs from Fiona, the concentration is significant in yield. Meanwhile, the methods do not affect the flower number and flower size. Upon examining the interaction between the two factors, it was observed that employing soil drenching at 1600 ppm and 0 ppm led to the lowest and highest flower numbers of 39.96 per plant and 54.25 per plant, respectively. Applying PBZ at 1600 ppm by soil drench method affects the flower size yield, causing the flower size to decrease to 3.57 cm (Table 5) (Figure 2). In addition, it was observed that chrysanthemums responded differently to PBZ in Prakaimat when exposed to high concentrations, causing the flower number to decrease more than in low and moderate concentrations, and Fiona, it was found that when exposed to all concentrations, causing the flower number to decrease than the control. In the second experiment, when the concentration level of PBZ was adjusted reducing, it was found that the concentration and method showed consistent results in the Prakaimat, employing the foliar spraying method gave a better yield in the flower number than the soil drenching method, as observed at 400 ppm by the foliar spraying method had the highest yield, 44.17 per plant. Conversely, the soil drenching method led to a notable reduction in flower size, with the smallest size by 2.74 cm at 400 ppm (Table 6).

Table 5. The effect of paclobutrazol on flower numbers and flower size in 'Prakaimat' and 'Fiona' cultivars of potted chrysanthemums for 13 weeks in experiment 1

Concentration	Method of	Flower nur	nber/plant	Flower size (cm)	
of PBZ (ppm)	provide	Prakaimat cv.	Fiona cv.	Prakaimat cv.	Fiona cv.
0	Soil drench	37.83±9.45 ^{bc}	54.25±6.70 ^a	4.83±0.13ª	4.73±0.23ª
	Soil drench	35.83 ± 13.28^{bc}	$42.50{\pm}5.57^{ab}$	3.13±0.24°	$3.75{\pm}0.12^{b}$
400	Foliar spray	43.17 ± 10.04^{abc}	44.33±4.84 ^b	3.43±0.21 ^b	$3.70{\pm}0.10^{b}$
800	Soil drench	29.83±9.08°	$40.00{\pm}10.25^{b}$	3.09±0.32°	$3.64{\pm}0.14^{b}$
	Foliar spray	59.17±10.19 ^a	41.92±7.67 ^b	2.99±0.08°	$3.62{\pm}0.08^{b}$
1200	Soil drench	38.13 ± 11.60^{bc}	46.09±6.42 ^{ab}	2.96±0.14°	$3.63{\pm}0.18^{b}$
	Foliar spray	49.92±19.32 ^{ab}	50.09±4.96 ^{ab}	3.03±0.17°	3.60±0.12 ^b
1600	Soil drench	38.96 ± 7.37^{bc}	39.96±8.37 ^b	$2.58{\pm}0.12^{d}$	$3.57{\pm}0.15^{b}$
	Foliar spray	42.96±14.11 ^{abc}	47.04±6.01 ^{ab}	2.93±0.13°	3.59±0.13 ^b
Conc. Method Conc. x Method		ns ** *	* ns *	** ** **	** NS **
CV (%)		28.64	15.05	5.34	4.04

**/highly significant */statistically significant ns/non-significant at P = 0.05; Means within column follows by the same letter are not significantly different as determined by Duncan's multiple range test

From observations in the Fiona, it was found that only the PBZ concentration caused the flower number yield to increase. Furthermore, the interaction between the concentrations and methods found that the soil drenching at 400 ppm had the highest of 44.83 per plant (Table 6). However, data regarding flower size in Fiona are not available due to unsuitable environmental conditions,

the bloom stage of the flowers stopped developing, and data could not be recorded.

Table 6. The effect of paclobutrazol on flower numbers and flower size in 'Prakaimat' and 'Fiona' cultivars of potted chrysanthemums for 13 weeks in experiment 2

Concentration of	Method of	Flower nun	Flower size (cm)	
PBZ (ppm)	provide -	Prakaimat cv.	Fiona cv.	Prakaimat cv.
0	Soil drench	29.83±2.38°	34.25±1.42 ^{cd}	4.50±0.34ª
100	Soil drench	$40.42{\pm}1.07^{ab}$	$31.92{\pm}1.95^{d}$	3.43±0.11°
	Foliar spray	37.25±7.34 ^{abc}	36.33±3.14 ^{bc}	3.75±0.12 ^b
200	Soil drench	33.50±5.09 ^{bc}	34.04±2.77 ^{cd}	3.27±0.12°
	Foliar spray	37.75±7.22 ^{bc}	36.13±1.65 ^{bcd}	3.38±0.13°
300	Soil drench	33.67 ± 5.04^{bc}	37.58±3.77 ^{bc}	2.88±0.07°
	Foliar spray	42.67 ± 6.47^{a}	38.79 ± 3.98^{ab}	3.13±0.12 ^{cd}
400	Soil drench	29.17±3.66°	41.83±2.22 ^a	2.74±0.21 ^e
	Foliar spray	44.17±7.29 ^a	38.75±2.81 ^{ab}	$2.98{\pm}0.09^{cd}$
Conc.		**	**	**
Method		**	ns	**
Conc. x Method		**	**	**
CV (%)		14.72	7.32	5.52

**/highly significant */statistically significant ns/non-significant at P = 0.05; Means within column follows by the same letter are not significantly different as determined by Duncan's multiple range test

Chlorophyll a and b content

The effect of PBZ on the amount of pigment in Prakaimat was found that at concentrations of 100, 200, and 300 ppm, the amount of chlorophyll *a* increased by 49.58, 51.08, and 47.39 µg/gFW, respectively. Also, notable enhancements in chlorophyll *b* content were 28.69, 28.35, and 26.53 µg/gFW, respectively. Furthermore, a comparison of application methods demonstrated that foliar spraying increased in yield, chlorophyll *a* and *b* at 48.60 and 27.12 µg/gFW. However, applying 100 ppm of PBZ through foliar spraying yielded the most increases significantly in chlorophyll *a* and *b*, 55.65 and 33.06 µg/gFW, respectively (Figure 1A and 1B). In Fiona, the highest chlorophyll *a* and *b* by 46.82 and 25.75 µg/gFW, respectively, with the application of PBZ at 400 ppm. It was observed that the foliar spray had higher than the soil drench amounts of chlorophyll *a* and *b* by 43.64 and 25.29 µg/gFW, However, in the interaction between concentrations and both methods at 100 and 200 ppm by foliar spraying, the highest significant levels of chlorophyll *b* were 27.55 and 27.85 μ g/gFW, respectively. Furthermore, at 400 ppm of PBZ through a foliar spray, the most chlorophyll *a* and *b* were 49.73 and 27.79 μ g/gFW, respectively (Figure 1D and 1E).



Figure 1. Effect of paclobutrazol concentration and methods on chlorophyll a, b and carotenoid content in Prakaimat (A, B, C) and Fiona (D, E, F) at 13 weeks after treatment. ---/The dashed line (control) represents the chlorophyll a, b, and carotenoid content in chrysanthemum plants untreated with PBZ (0 ppm)

Carotenoid content

Carotenoid content tended to increase in both cultivars when applied to PBZ, especially at 100 and 200 ppm, resulting in carotenoid content increased to 9.42 and 9.80 μ g/gFW, and from the observation that the foliar spray tends to have a higher carotenoid content than the soil drench in the Prakaimat (Figure 1C). In Fiona, the highest carotenoid content was 9.49 μ g/gFW at 400 ppm of PBZ. In particular, the foliar spray at 400 ppm resulted significantly in carotenoid content as high as 9.91 μ g/gFW, and it was observed that the foliar

spray tended to increase the carotenoid content than the soil drench, similar to the Prakaimat (Figure 1F).



Figure 2. Effect of PBZ on qualities of chrysanthemum, untreated (A) and treated (B) in Prakaimat, and untreated (C) and treated (D) in Fiona



Figure 3. Leaves of the chrysanthemum i.e., Prakaimat (A) and Fiona (B) cultivars after being treated with PBZ

Discussion

The study showed that paclobutrazol inhibits vegetative plant growth for the two cultivars of chrysanthemum. In Experiment 1, when the chrysanthemums were applied to an overdose of PBZ concentration, the method did not affect retarding their growth and causing no difference in all parameters. Thus, the concentration was adjusted to be lower in experiment 2. It was found that applied PBZ at 300-400 ppm by soil drench could retardant vegetative growth, i.e., stem height, canopy width, internode length, and node number were finest when compared to the control, due to paclobutrazol (PBZ) effectively inhibit the growth of bud shoots as a function of mediates changes in plant hormone levels, such as gibberellin (GA), a hormone that develops node stretching, affects isoprenoid processes and changes their state by inhibiting cytochrome P450dependent oxygenase inactivation (Soumya, 2017), which mediates oxidative dimethylation reactions, including those which are necessary for the synthesis of ergosterol and the conversion of kaurene to kaurenoic acid in the gibberellins biosynthetic pathway (Fletcher et al., 2000) causing to inhibition of cell enlargement, stem shortening is also caused by inhibition of cell division and elongation of the subapical meristems. Consistent with the previous study PBZ by soil drenching at 120 mg/l, the plant height decreased on average by 28% in chrysanthemum (Chauhan et al., 2020), and reported in Cymbidium hybridum shows applying PBZ by spray at 400 mg/l reduced plant height to 90% (Li et al., 2019). Similarly, applying PBZ controls the growth of potted sunflowers (Helianthus annus L. 'Pacino') by soil drenches were applied at 0-32 mg a.i./pot had plant height shorter than untreated plants, especially at 16 a.i./pot. However, provided that the concentration is inappropriate, it could exhibit phytotoxicity symptoms, including crinkled leaves, stunted growth, and smaller and greener leaves (Shravan et al., 1998). In terms of canopy width, this study found that when both cultivars of chrysanthemums were exposed to PBZ, the canopy width became smaller and more compact form, consistent with reports of the use of PBZ in Pachira aquatica Aubl., at 150 mg/l, with a single foliar spray, caused the canopy width to decrease (Li et al., 2009). In addition, PBZ affects internode length, causing the internode length to decrease, which is consistent with the previous study, the effect of PBZ in two manner, i.e., foliar spray and soil drench, found that soil drenching at 2.500 mg a.i./plant resulted in shorter internode length than foliar spraying in Lupinus varius L. (Karaguzel et al., 2004), Similarly, in node number from the report, using PBZ at 50 g a.i./ha by foliar spraying three times reduced the node number (Koutroubas and Damalas, 2015) Moreover, this study showed that the soil drench method could retard growth in terms of vegetative growth better than the foliar spray method, which is

consistent with the reported effect of PBZ on easter lilies and sea marigolds (Gianfagna and Wulster, 1986; Carver et al., 2014). Although the properties of PBZ can be absorbed through roots and leaves, if applied by foliar spray, PBZ migrates through the phloem and accumulates in the leaf region, where the slow translocation is less effective than the direct root through the xylem in chrysanthemums (Menhenett, 1984; Desta and Amare, 2021). Furthermore, this study found that using PBZ affected the growth of the reproductive stage, increasing the flower number, especially through the foliar spray at 400 ppm in experiment 2. PBZ stimulates flower initiation by decreasing gibberellin levels and increasing auxin and cytokinin levels (Yadav et al., 2005). Consistent with a previous study has found that foliar spraying can retarding plant growth and cause the flower number to increase in Daphne genkwa (Zhang et al., 2023). Although, the PBZ impact extends to inducing a delay in flowering time (Ghatas, 2016). Notably, its influence on flowering is linked to inhibiting gibberellic acid activity, resulting in decreased flower diameter (Lailaty and Nugroho, 2021a). PBZ, also increases the delayed flowering days in geraniums (Abd El-Aal and Mohamed, 2017). However, from Experiment 1, the response to PBZ in Prakaimat showed that at 800 ppm, the highest flower number was produced compared to the high concentration, consistent with previous study reports using PBZ at low to moderate concentrations resulted in an increase in the flower number than the high concentration in Bougainvillea glabra 'Sanderiana' (Karaguzel and Ortacesme, 2002). The flower size was decreased using PBZ, which might be due to its inhibiting influence on cell enlargement and shoot expansion. A previous study using PBZ by soaking the tubers at 30 ppm for 30 minutes, shows the flower size decreased in the iris (Demir and Celikel, 2018). In select cases, the influence of PBZ lower concentration led to an increase in flower diameter in *Paeonia lactiflora* Pall. (Wu et al., 2022). In addition, using PBZ made the leaves smaller and darker green than untreated PBZ (Figure 3). It was observed in the treated plant leaves, which were smaller than those of the control, that could be due to the inhibition effect of PBZ on the cell elongation on the leaf in Syzygium myrtifolium (Ahmad Nazarudin et al., 2012) Other than, apply of PBZ retardation to the growth in the vegetative phase, causing also increases the accumulation of yield, increasing the amount of inulin in the jerusalem artichoke (Phasri et al., 2019). The present investigation has yielded noteworthy findings about the augmentation of chlorophyll content ascribed to the compelling influence of PBZ, which engenders distinct changes in the leaves manifested as a deepening of the shade to a darker green and a pronounced increase in leaf thickness affects is attributed to the expansion of the palisade and spongy tissues, accompanied by the development of a notably denser epicuticular layer, an observation drawn from previous research involving in potato (Tsegaw

et al., 2005) and chrysanthemum (Burrows et al., 1992; Zhang et al., 2020). Similarly, in chrysanthemums, a reduction in leaf area has been correlated with the apparent compaction of chloroplasts, thereby contributing to the observed escalation in chlorophyll content. That, intricate interaction between anatomical alterations and physiological responses extends further as a thicker mesophyll layer is recognized for its role in augmenting the rate of photosynthesis (Lailaty and Nugroho, 2021b). The efficacy of PBZ extends beyond these physiological adjustments, encompassing the modulation of cytokinin synthesis, an attribute that enhances chloroplast synthesis and attenuates chloroplast degradation. The consequential implications of this activity are manifest in sunflower plants, where PBZ action is recognized for its potential to affect chloroplasts during the senescence phase, resulting in a decline in their degradation rate (Sarkar, 2023; Desta and Amare, 2021). That, in tandem with other mechanisms, may explain the discernible increase in chlorophyll content within leaves treated with PBZ compared to their untreated counterparts. Furthermore, the application of PBZ is accompanied by an appreciable rise in carotenoid content across chrysanthemum specimens, an observation consistent with earlier investigations (Abou Elhassan et al., 2021). Intriguingly, these findings are not universal, as certain instances involving tomato plants revealed the agent's ineffectiveness in influencing carotenoid content (Berova and Zlatev, 2000). It is posited that the enhancement of abscisic acid (ABA) and cytokinin levels, prompted by PBZ exposure, may contribute to the heightened pigment content observed within PBZ-treated chrysanthemums, intricate interaction of physiological responses underscores the multifaceted impacts of PBZ on pigment modulation and broader plant physiology (Kishorekumar et al., 2007). Demonstrating a coherent relationship, the study revealed a correlation between the impacts of PBZ, ABA, and other phytohormones (Xu et al., 2020). Nonetheless, in Experiment 2, the unavailability of data about flowering can be attributed to the influence of exceptionally harsh weather conditions. As a result, its suitability for elucidating flower initiation development is limited.

In conclusion, PBZ demonstrated a growth-inhibiting impact, leading to reduced plant height and decreased canopy width and peduncle length. These findings offered insights into potential applications for advancing potted chrysanthemum cultivation. In both experiments, a significant relationship was observed between the application method and the PBZ concentration. Specifically, the soil drench application method combined with a concentration range of 300-400 ppm, is emerged as a promising guideline for future potted chrysanthemum production. However, due to variances in plant types, careful evaluation of variables, including concentration levels and application methods, are warranted and given the distinct responses exhibited by diverse plant species.

Acknowledgments

This work is financially supported by King Mongkut's Institute of Technology Ladkrabang Research and Innovation Services, KRIS (KREF016509) KMITL.

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(Received: 20 November 2023, Revised: 2 March 2024, Accepted: 12 March 2024)