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The effects of gibberellic acid (GA_3) on the harvesting time of spray type *Chrysanthemum* cut flowers in medium land

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Abstract. *Shintiavira H, Sulistyaningsih E, Purwantoro A, Wulandari RA. 2020. The effects of gibberellic acid (GA_3) on the harvesting time of spray type* Chrysanthemum *cut flowers in medium land. Biodiversitas 21: 1723-1729.* The high temperature of the medium land causes a delay in harvesting time and also decreases the diameter of *Chrysanthemum* cut flowers (*Chrysanthemum morifolium* R.). It is, however, possible to accelerate and increase the flower diameter through the use of the Gibberellic acid (GA_3). This research, therefore, aimed to obtain the best GA_3 concentration to accelerate harvesting time using three varieties of spray type *Chrysanthemum* cultivated on a medium land. The experimental design involved the use of a split-plot with three replications. The main plot varied the GA_3 concentration between 0, 200, 400, and 600 mgL^{-1} while the subplot used the spray type *Chrysanthemum* varieties such us 'Yastayuki', 'Arosuka Pelangi', and 'Socakawani'. It was applied on the 56, 63, and 70 days after planting (DAP). The data were analyzed using Analysis of Variance (ANOVA) and a further test was conducted using Duncan Multiple Range Test (DMRT) at α=0.05. The results showed GA_3 at 400 mgL^{-1} was the best concentration to increase the harvesting time in all varieties. However, the harvesting time of spray type without secondary branching ('Arosuka Pelangi') was faster than spray type with secondary branching ('Yastayuki' and 'Socakawani'). The harvesting time was earlier 13, 9, and 7 days for 'Arosuka Pelangi','Yastayuki' and 'Socakawani' compare with control, respectively. The average diameter of the blooming flowers was also wider with 54.9 mm compared to the control treatment which was 38.7 mm.

Keywords: *Chrysanthemum*, endogenous hormones, harvesting time, gibberellic acid, medium land

Abbreviations: ANOVA: Analysis of Variance, ASL: above sea level, CV: coefficient of variations, DAP: days after planting, DMRT: Duncan Multiple Range Test, GA_3: gibberellic acid, IAA: Indole-3 acetic acid

INTRODUCTION

Chrysanthemum cut flowers (*Chrysanthemum morifolium* R.) have been widely cultivated in different areas of the world including Indonesia where its cultivation ranges from the high to low lands (Sanjaya et al. 2018). However, the temperature and light intensity in medium lands had been reported negative effects on the development of *Chrysanthemum*. Shintiavira et al. (2019) reported that the *C. morifolium* 'Yastayuki', 'Arosuka Pelangi' and 'Socakawani' grown in medium land to have shown prolonged harvesting time and decreased flower size compared to those in high land. This reduces the quality of the flower which can, however, be mitigated through the use of GA_3.

According to Su et al. (2001), exogenous GA_3 improved the development of the *Phalaenopsis hybrida* inflorescence under temperature above 30°C. It has also been discovered that a higher endogenous GA_3 in flower development compared to the initiation period for *C. morifolium* 'Jinba' indicates that the acceleration of the cell division and elongation of florets by this auxin (Jiang et al. 2010a). The use of exogenous GA_3 in the improvement of flower quality had been reported by many researchers, for example, Mori et al. (2013) showed its 200 mgL^{-1} concentration in flower bud 3.5 mm size accelerated the harvesting time and increased flower diameter compared to the control treatment of *C. morifolium* 'Sansui' in the subtropics area of Japan. Moreover, at 250- 500 mgL^{-1} concentrations, the diameter was found to have increased to 8.17-8.32 cm while the control was 6.56 cm in standard type *Chrysanthemum* cut flower cultivated in the medium land of Indonesia (Priambodo et al. 2014). However, there are no studies reported to have been conducted on the effect of different variations of GA_3 concentrations on spray type *Chrysanthemum* cut flower in medium land. Therefore, this research was conducted to obtain the best concentration of GA_3 required to improve the flower quality of *Chrysanthemum* varieties such us 'Yastayuki', 'Arosuka Pelangi', and 'Socakawani' cultivated in medium land Indonesia.

MATERIALS AND METHODS

Plant materials and growth conditions

The research was conducted at Samigaluh Village, Kulonprogo District, Special Region of Yogyakarta Province, Indonesia located on 07º40' S and 110º12' E at an altitude of 485 m above sea level (ASL) from July to November 2018. The spray type *C. Morifolium* cv 'Yastayuki', 'Arosuka Pelangi', and 'Socakawani' rooted cuttings with 3-4 full expand leaves were used as plant materials. The 'Yastayuki' and 'Socakawani' were spray type *Chrysanthemum* with secondary branching while 'Arosuka Pelangi' was whose without secondary branching. They were grown on a bench soil in the screen house with 10 cm spacing under long-day conditions for 4 weeks with supplemented lighting from 10.00 pm - 04.00 am. The soil medium was mixed with basic fertilizer consisted of 5 kgM^{-2} goat organic manure, 200 $kgHa^{-1}$ Urea (Kujang, Indonesia), 300 $kgHa^{-1}$ SP-36 (Petrokimia, Indonesia) and 350 $kgHa^{-1}$ KCL (Kujang, Indonesia) as basic fertilizer. Additional fertilizer was 2 gL^{-1} NPK 16: 16: 16 (Meroke Tetap Jaya, Indonesia), 1g.L^{-1} red KNO_3 (El Trovador, Chile), 2 gL^{-1} Gandasil D (Kalatham, Indonesia) were given during 0-35 DAP. Then, 2 gL^{-1} NPK, 1gL^{-1} white KNO_3 (El Trovador, Santiago, Chile), 2 gL^{-1} Gandasil B (Kalatham, Indonesia) were applied during 36 DAP until harvesting time. The fertilizer was mixtured and applied in watering. Moreover, the microclimate such as air temperature, relative humidity, and light intensity were 29.1°C, 68.0%, and 18,290 lux, respectively. Thereafter, the GA_3 solution was applied on flowers bud at 57 DAP according to the treatment while the apical and bottom flower buds were removed at 64 DAP leaving only those located 20 cm to the top from the apical meristem. It is important to state that the plants were maintained according to the procedure of the operational standard of *Chrysanthemum* cultivation (Indonesian Directorate General of Horticulture 2012).

Research design

The experimental design involved the use of a split-plot with three replications. The main plot was GA_3 concentration varied between 0, 200, 400, and 600 mgL^{-1} while the subplot was the *Chrysanthemum* varieties including 'Yastayuki', 'Arosuka Pelangi', and 'Socakawani'.

Procedures

The GA_3 solution was made by dissolving pure GA_3 (AGROGIBB 40 SL, Indonesia) in aquadest based on the concentrations required for each treatment, 0, 200, 400, and 600 mgL^{-1}. The application of the solution started 56 DAP or at the flower bud diameter of ±3-5 mm. The volume sprayed on each flower bud per plant was ± 0.9 ml and this was conducted every week till the 10th week. This means it was applied on the 56, 63, and 70 DAP.

Observations

Endogenous GA_3 and IAA in flower buds

All the flower buds were pinched at 71 DAP to determine the endogenous GA_3 by using Barendse's (1987) method with minor modification. This involved the crushing of a total of 0.5 g samples of flower buds using liquid nitrogen after which 5 ml of 65% methanol (v/v) was added and centrifuged at 4000 rpm for 30 minutes at 4°C. The supernatant flowed using the C18 Sep-Pak cartridge and 5-5 µL of its content was injected into the HPLC equipped with a UV-Vis spectrophotometer (Shimadzu, Japan). It is, however, important to note that the wavelength of GA_3 was 200 nm and the wavelength of IAA was 218 nm. Furthermore, the endogenous hormones per flower bud were obtained by dividing the results of the endogenous hormones with the number of flower buds.

Growth parameters and quality traits of Chrysanthemum cut flowers

Growth parameters involved the diameter and number of flower buds as well as the length of pedicels measured every week. The variables of the quality traits of *Chrysanthemum* cut flowers included (i) the harvesting time which is defined as the state where more than 60% of the total half of the flower has opened at 45°, (ii) the length of the pedicel was measured by calculating the average length of the peduncle located 20 cm from the top of the plant, (iii) the total number of flowers was measured by calculating the total number of buds and blooming flowers located 20 cm from the top of the plant, (iv) the total number of blooming flowers was measured by calculating the total flowers opening at 45°, (v) the percentage of the blooming flowers were measured by dividing their total number by the total number of flowers in percent units, (vi) the diameter of the blooming flower was measured by calculating the length of the flower head from the left to the right petal, (vii) the length of the ray floret was measured by calculating the length of the detached ray floret from the base up to the top, (viii) the width of the ray floret was measured by calculating the width of those detached in the middle, (ix) The thickness of the ray floret was measured by calculating the thickness average of basal, middle and tip of 30 ray florets using a digital micrometer, and (x) The number of the ray floret per flower was measured by calculating the detached ray petal per flower.

Statistical analysis

The data were analyzed using ANOVA and if a significant difference was observed among the treatments, the DMRT method was applied at $\alpha = 0.05$. Moreover, the correlation test was used to determine the relationship between variables. It is important to note that all the statistical analyses were conducted using SAS 9.12. (Abebe 2000).

RESULTS AND DISCUSSION

The effect of exogenous GA_3 concentration on endogenous GA_3 contents of flower bud

The results showed that there was no interaction between the concentration of exogenous GA_3 and varieties of endogenous GA_3 of flower bud at 71 DAP. However, there was interaction between the concentration of exogenous GA_3 and varieties of endogenous IAA of flower bud at 71 DAP (Table 1). An increase in endogenous GA_3 was followed by an increase in endogenous IAA in the 'Yatsayuki' and 'Socakawani'. While, an increase in endogenous GA_3 was followed by a decreased in endogenous IAA in 'Arosuka Pelangi'.

The effect of exogenous GA_3 level on the diameter of flower bud, length of the pedicel, and the number of flower buds

The 200-600 mgL^{-1} GA_3 applied significantly increased the length of the pedicel after 70 DAP and this is associated with the excess solution applied at 63 DAP as shown in Figure 1. However, the first application of the solution at 56 DAP was discovered not to have any influence on the total number of flower buds at 63 DAP while the second caused the pinching of flower buds at 20 cm from the top. This, therefore, led to the reduction of the number of flower buds at 70 DAP. Moreover, the third application of GA_3 decreased the total number for both the bud and those blooming up to the harvesting time. Figure 2 shows the endogenous GA_3 accumulated on the bud from the 63 DAP has the ability to inhibit the growth of the axillary flower bud. Furthermore, the diameter of the bud required more GA_3 application compared to the control from 56 to 84 DAP while 200-600 mgL^{-1} concentrations accelerated the opening of ray florets after 84 DAP and this was followed by the increase in the diameter of the blooming flowers. This, therefore, showed that an increment in the endogenous GA_3 at 71 DAP did not have any significant influence on the size of the flower bud diameter. However, those accumulated on the flower bud at 71 DAP accelerated the expansion of the ray floret after 84 DAP as shown in Figure 3.

Table 1. The average endogenous GA_3 and IAA of flower buds in varieties of exogenous GA_3 at 71 DAP

Variables	Conc. exogenous of GA_3 (mgL^{-1})	Varieties			Average
		Yastayuki	Arosuka Pelangi	Socakawani	
Endogenous GA_3 (ng)	0	15.18	16.12	43.33	24.88 b
	200	255.10	393.46	360.31	336.29 a
	400	492.50	353.85	355.56	400.63 a
	600	289.34	377.15	343.70	336.73 a
	Average	263.03 a	285.14 a	275.73 a	(-)
Endogenous IAA (ng)	0	343.56 de	601.82 b	83.06 f	342.84
	200	487.20 bc	441.28 cd	568.46 bc	498.98
	400	583.71 bc	449.42 cd	310.74 de	447.96
	600	237.75 e	529.46 bc	753.27 a	506.83
	Average	413.06	505.51	428.88	(+)

Note: CV= 29.74%. The means with the same letters are not significantly different based on DMRT, α=0.05. However, (+) sign showed there was an interaction between the concentration of GA_3 and varieties while (-) sign means there was none.

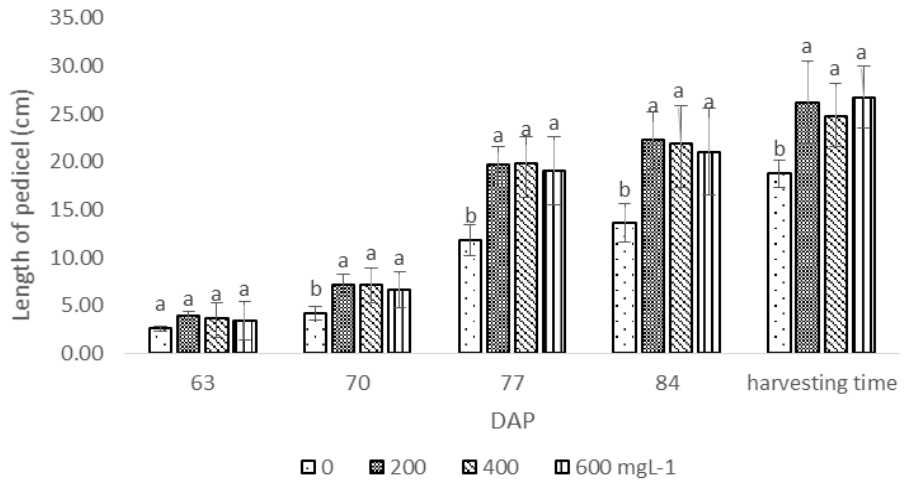

Figure 1. The average length of pedicels after exogenous GA_3 applied in three varieties of *Chrysanthemum*. The means with the same letters are not significantly different based on DMRT, α=0.05.

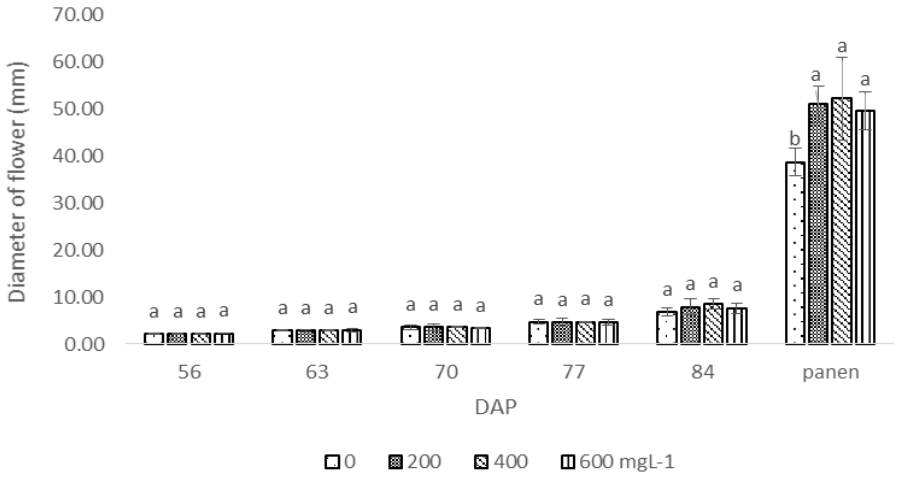

Figure 2. The number of flowers in exogenous GA_3 concentration in three varieties of *Chrysanthemum*. The means with the same letters are not significantly different based on DMRT, α=0.05

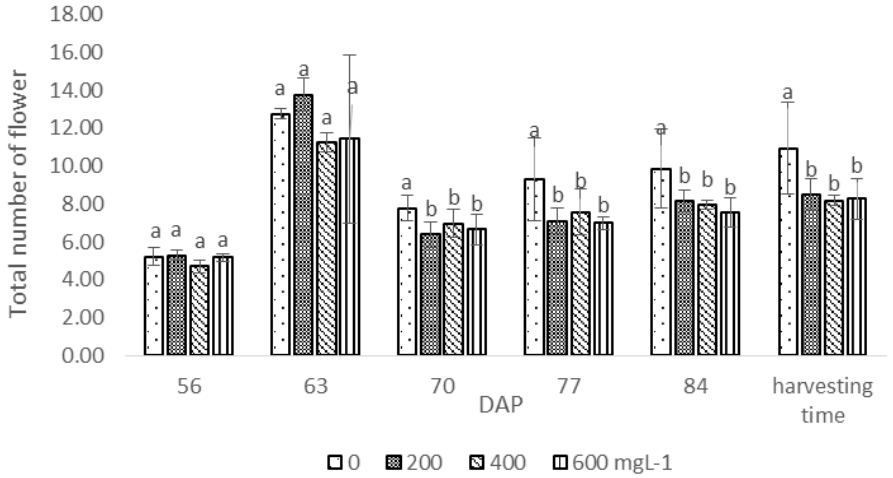

Figure 3. The diameter of flower buds (56-84 DAP) and diameter of blooming flower at harvesting time in exogenous GA_3 levels in the three *Chrysanthemum* varieties. The means with the same letters are not significantly different based on DMRT, α=0.05

Effect of exogenous GA_3 level on quality traits of *Chrysanthemum* cut flower

The harvesting time for all the varieties without GA_3 was 105 days but was significantly accelerated by the exogenous application of 200 mgL^{-1} solutions to 98, 96, and 100 DAP in 'Yastayuki', 'Arosuka Pelangi', and 'Socakawani' respectively. Furthermore, there was a significant acceleration to 96, 92, and 98 DAP at 400 mgL^{-1} and 600 mgL^{-1}. The effect of the GA_3 on the shortening of harvesting time supported the quality traits of *Chrysanthemum* as discovered with the increased length of pedicels and reduction in the total number of flowers at 400 mgL^{-1}. However, the percentage of the blooming flowers was not significantly different at all the GA_3 concentrations but was found to have increased its diameter and the value was observed to be 54.20 mm for all the varieties at 400 mgL^{-1}. Moreover, the opening of the ray floret has an influence on its length, width, thickness, and number. At 200-600 mgL^{-1}, the width and thickness were found to be decreasing while the length increased. However, the number of ray florets was dependent on the varieties with a significant difference found between the concentrations in 'Socakawani' while none was observed in 'Yastayuki' and 'Arosuka Pelangi'. The application of GA_3 at 200, 400, and 600 mgL^{-1} were increased the number of ray florets compared to the control for 'Socakawani'.

Correlation analysis between endogenous hormones and the quality of *Chrysanthemum* cut flower

The exogenous GA_3 was found to be positively and significantly correlated to endogenous GA_3 with 0.64. Moreover, the endogenous GA also positively and significantly correlated to the length of pedicels with 0.62, the diameter of blooming flowers with 0.80, and the length of the ray floret with 0.51. However, a negatively significant correlation was discovered with shortening harvesting time at -0.72, the thickness of the ray floret at -0.52, the total number of flowers at -0.53, and total blooming flower at -0.59 as shown in Table 3.

Table 2. The average harvesting time, length of pedicels, total number of flower, number of blooming flowers, percentage of blooming flowers, the diameter of blooming flower as well as the length, width, thickness, and the number of ray florets

Variables	Conc. exogenous of GA_3 (mgL^{-1})	Varieties			Average
		Yastayuki	Arosuka pelangi	Socakawani	
Harvesting time	0	105.0 a	105.0 a	105.0 a	105.0
(days)	200	98.00 c	96.0 d	100.0 b	98.00
	400	96.00 d	92.0 e	98.0 c	95.33
	600	96.00 d	92.0 e	98.0 c	98.33
CV=0.00 %	Average	98.8	96.3	100.3	+
Length of pedicels	0	19.52	17.16	19.69	18.79 b
(cm)	200	25.47	20.98	27.83	24.75 a
	400	26.43	21.80	30.27	26.24 a
	600	27.56	23.25	29.47	26.76 a
CV=5.95 %	Average	24.79 b	20.79 c	26.82 a	-
Total number of flowers	0	13.67 a	9.00 bc	10.12 b	10.93
	200	8.59 bc	7.58 c	9.28 bc	8.49
	400	8.20 bc	7.91 bc	8.43 bc	8.18
	600	8.72 bc	7.06 c	9.00 bc	8.26
CV=13.24%	Average	9.79	7.88	9.20	+
Number of blooming flowers	0	10.58	7.50	8.17	8.75 a
	200	6.50	6.25	6.78	6.74 b
	400	6.55	7.00	6.67	6.51 b
	600	7.14	6.78	6.22	6.71 b
CV=13.04%		7.96 a	6.88 a	6.95 a	-
Percentage of blooming flowers	0	77.96	84.21	80.37	80.85 a
(%)	200	76.49	82.44	74.50	77.81 a
	400	80.63	88.87	80.65	82.47 a
	600	82.25	96.00	69.37	82.54 a
CV=8.12%	Average	79.34 b	87.88 a	75.82 b	-
The diameter of blooming flower	0	41.70	35.80	38.67	38.72 c
(mm)	200	52.33	45.00	51.37	49.56 b
	400	61.63	47.77	55.43	54.94 a
	600	49.77	44.10	51.00	48.29 b
CV=5.09%	Average	51.36 a	43.17 c	49.12 b	-
Length of the ray floret	0	2.433	2.176	1.953	2.187 b
(mm)	200	2.663	2.383	2.327	2.457 a
	400	2.913	2.230	2.437	2.527 a
	600	2.783	2.300	2.213	2.432 a
CV=7.15%		2.698 a	2.272 b	2.232 b	-
Width of the ray floret	0	0.867	1.267	0.796	0.977 a
(mm)	200	0.766	1.180	0.823	0.923 ab
	400	0.706	0.970	0.840	0.836 bc
	600	0.650	0.970	0.733	0.797 c
CV=10.71%		0.745 b	1.097 a	0.808 b	-
Thickness of ray floret	0	0.089	0.128	0.124	0.113 a
(mm)	200	0.103	0.098	0.107	0.100 ab
	400	0.065	0.078	0.068	0.071 c
	600	0.079	0.091	0.091	0.087 bc
CV=14.32		0.008 b	0.009 a	0.009 a	-
Number of ray floret per flower	0	57.00 ed	33.33 e	95.00 c	61.78
	200	61.00 d	32.33 e	126.0 b	73.22
	400	64.47 d	31.33 e	152.33 a	82.78
	600	61.33 d	31.00 e	172.00 a	88.00
CV=18.91%		61.00	32.00	136.33	+

Note: CV: coefficient of variations. The means with the same letters are not significantly different based on DMRT, α=0.05. However, (+) sign showed there was an interaction between the concentration of $GA_3$and varieties while (-) sign means there was none

Table 3. Correlation between endogenous GA_3 and the quality of *Chrysanthemum* cut flower

Variable		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
Exogenous GA3	1	1.00														
Endogenous GA3	2	0.64*	1.00													
Harvesting time	3	-0.80*	-0.72*	1.00												
Length of pedicels	4	0.65*	0.62*	-0.40	1.00											
Total number of flowers	5	-0.48	-0.53*	0.63*	-0.12	1.00										
Number of blooming flower	6	-0.48	-0.59*	0.54*	-0.37	0.85*	1.00									
Percentage of blooming flower	7	0.12	0.02	-0.32	-0.39	-0.46	0.08	1.00								
Diameter of blooming flower	8	0.53*	0.80*	-0.49	0.76*	-0.29	-0.43	-0.17	1.00							
Number of flower bud	9	-0.25	-0.20	0.45	0.25	0.73*	0.26	-0.93*	0.02	1.00						
Diameter of flower bud	10	-0.43	-0.33	0.35	-0.28	0.17	0.08	-0.22	-0.24	0.21	1.00					
Vase life	11	0.32	0.24	0.06	0.77*	0.04	-0.23	-0.48	0.48	0.37	-0.06	1.00				
Length of ray floret	12	0.30	0.51*	-0.30	0.39	-0.08	-0.15	-0.08	0.61*	0.05	-0.02	-0.03	1.00			
Width of ray floret	13	-0.36	-0.27	0.10	-0.59*	-0.16	0.01	0.33	-0.57*	-0.30	0.41	-0.49	-0.25	1.00		
Thickness of ray floret	14	-0.54*	-0.52*	0.54	-0.49	0.05	0.04	-0.02	-0.58*	0.04	0.31	-0.17	-0.45	0.37	1.00	
Number of ray floret	15	0.20	0.10	0.19	0.64*	0.09	-0.19	-0.51*	0.36	0.40	0.03	0.95*	-0.15	-0.44	-0.05	1.00

Note: * Significant at 5% based on Pearson Correlation Test

Discussion

There was a positive correlation between endogenous GA_3 and the length of pedicels. Moreover, 200-600 mgL^{-1} was applied at 56, 63, and 70 DAP on flower buds and the concentration was further increased at 71 DAP. However, the first response observed was the increment of the pedicels starting from 70 DAP due to the absorbtion of the solution in the flower buds leading to the elongation of its cells. According to the Law (1987), gibberellins increased the regulation of L-tryptophan to D-tryptophan which was later converted to auxin to cause the acidification of cell walls required to initiate the process of cell enlargement (Rayle and Cleland 1992). The effect of the GA_3 on the total number of flowers was also observed at 70 DAP. The total number of flowers, including the bud and blooms, decreased up to the harvesting time after the application of GA_3 at 400 mgL^{-1}.

The exogenous GA_3 increased endogenous GA_3 and IAA in 'Yastayuki' flower bud. Jiang et al. (2010b) indicated that endogenous GA_3 and IAA played a role in the formation of axillary buds. According to Huh et al. (2011), increasing IAA in shoot apex has the ability to decrease branching in *Chrysanthemum*. However, a high concentration of this solution in the apex flower bud could inhibit its lateral growth (Chen et al. 2013; Dierck et al. 2018). Moreover, at 400 mgL^{-1}, the flower diameter was affected such that the reduction in the number of flowers was observed to be inversely proportional to the diameter of the blooming flower due to the assimilation competition between the flower buds (Kozlowska et al. 2011). The size of the flower diameter was larger when the number of total flower buds was reduced (Carvalho et al. 2006). The results showed the harvesting time for all the varieties with 0 mgL^{-1} GA_3 concentration was 105 days while the exogenous application of GA_3 at 400 mgL^{-1} significantly accelerated it up to 96 DAP for 'Yastayuki'. However, the exogenous GA_3 increased endogenous GA_3 and IAA in 'Socakawani' flower bud but it did not decrease the number of flower buds. It might be, transportation of endogenous IAA to axillary has occurred after 71 DAP. This condition caused the harvesting time was longer than 'Yastayuki' (98 DAP). Meanwhile, the exogenous GA_3 increased endogenous GA_3 and decreased endogenous IAA in 'Arosuka Pelangi' flower bud. It showed that there was transportation of endogenous IAA to axillary bud. Therefore, there was no signifcantly decreased the number of flower bud. Moreover, the harvesting time of 'Arosuka Pelangi' was fastest (92 DAP) than 'Yastayuki' and 'Socakawani'.

This, therefore, proved the positive correlation between endogenous GA_3 and the diameter of blooming flowers at harvesting time. The average flower bud size at 84 DAP was 8 mm and it has been reported by Qi et al. (2016) that after this period, flower development moves to the pigmentation phase or S3-S4 and this means the harvesting time would be accelerated. According to Jiang et al. (2010b), cell of flower buds undergoes a differentiation process that requires more cytokinin compared to gibberellins that assist the expansion of *Chrysanthemum* ray florets. Therefore, the rate of blooming and increment in flower diameter was due to the speed of expansion and opening of the rolling ray florets (Martin and Gerats 1993) which further increased the number of ray florets depending on the varieties. Furthermore, the GA_3 treatment did not influence the increase in the number of ray florets per flower in 'Yastayuki' and 'Arosuka Pelangi' but affected 'Socakawani'. Another factor affecting the flower diameter was the length of ray florets. In line with Zhang et al. (2012) and Li et al. (2015), the application of exogenous GA_3 increased the length of the ray floret by driving the cell elongation of *Gerbera hybrida*. However, an increase in the length was followed by a reduction in the width and thickness and this is in agreement with the positive correlation established between endogenous GA_3 and length and the negative relationship it has with the width and thickness of the ray florets.

From this study, 400 mgL^{-1} GA_3 was discovered to have the best concentration required to accelerate the harvesting time in all varieties with different mechanisms.

The harvesting time of spray type without secondary branching ‘ (Arosuka Pelangi’) was faster than spray type with secondary branching (‘Yastayuki’ dan ‘Socakawani’).

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