Effects of Gibberellin Application at Different Growing Stages on Growth and Development of *Curcuma alismatifolia* Gagnep.

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Summary

A rhizome of *Curcuma alismatifolia* Gagnep. was cultivated in a pot using soil, sand, rice husk and rice husk charcoal mixed at the ratio of 1:1:1:1 by volume. The experiment was conducted to study growth and development of plants when 100 ml of GA_3 solution (100 mgL^{-1}) was applied by drenching to soil at different growth stages, i.e., shoot emergence stage (4 weeks after planting (WAP)) , 1st-leaf unfolding stage (7 WAP) , 2nd-leaf unfolding stage (10 WAP) and 3rd-leaf unfolding stage (11 WAP) compared with control plants supplied with distilled water. The results showed that the GA_3 application at later stage tended to increase plant height and scape length but decreased the number of shoot, inflorescence and rhizome per cluster. The flowering date was delayed by GA_3 application. The control plants flowered at 103 days after planting. The flowering date was most delayed at 114 days after planting, when GA_3 solution was supplied at 3rd-leaf unfolding stage. The concentrations of nutrient (NPK) were analyzed in the underground organs and aboveground organs at flowering stage (15-16 WAP). The GA_3 application at different growing stages had effects on nutrient concentrations in the aboveground parts. When GA_3 solution was supplied at 3rd-leaf unfolding stage, the N, P and K concentrations were the lowest compared with the application at other stages. However, the GA_3 application in all treatments had no effect on N, P and K concentrations in underground organs.

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Curcuma alismatifolia Gagnep. or Siam tulip, a member of the ginger family of Zingiberaceae. *Curcuma* species native to indo-china (Gagnepain, 1908), has become popular in the international market as a new ornamental bulb plant with a beautiful inflorescence resembling that of a tulip hence it became the common name. It is used as a bedding plant in tropical countries, as a pot plant through the world, and as a cut flower. Most of *Curcuma* plants grow in the rainy season and rhizomes are in dormancy on dry season in Thailand. The products (flowers and rhizomes) of *C. alismatifolia* come to the market at the same time all over the country, resulting in the low price of flowers and rhizomes. In some cases, it is not profitable to harvest them due to very low price, some farmers lift and leave them away in the field due to unprofitability. The control of flowering time of *C. alismatifolia* are the way to prevent losing money or to increase a profit. Furthermore, flowers and rhizomes should be standard and healthy without disease.

Phytohormones play a dominant role in the regulation of the growth and development of higher plants. Gibberellins (GAs) are a family of plant hormones controlling plant growth and development including stem elongation, germination and the transition from vegetative growth to reproductive growth (Stephan *et al.,* 2005). In *Dahlias,* the tuberization is induced by short day period, and GA enhanced thickening at the petiole base of budless cuttings. In addition, GA and ABA directly control tuberization by determining the site of the sink for assimilates (De Hertogh and Le Nard, 1993). Application of gibberellin promotes flowering in *Zantedeschia*. Both GA_3 and GA_{4+7} increase flower production through increasing in the number of buds emerging and the proportion of shoots flowering (De Hertogh and Le Nard, 1993). In our previous study, we found that gibberellin increased the plant height and flower stalk length and also delayed the flowering date (Khuankaew *et al.,* 2008). However, the suitable stage of application GA_3 is important to study. The objective of this research was to investigate the effects of GA_3 application in different growth stages on growth and development of curcuma including nutrient concentration in both aboveground and underground organs.

MATERIALS AND METHODS

Rhizomes of *Curcuma alismatifolia* Gagnep. with the average rhizome diameter at 1.82 cm and 6-7 storage roots were cultivated in off-season on 21 February in a 6 x 12 (diameter x height) inch pot using soil, sand, rice husk and rice husk charcoal ratio 1:1:1:1 (by volume). After shoot sprouted, plants were supplied with the complete solution consisted of N, 200; P, 50; K, 200; Mg, 25; Ca, 136; B 0.216, Mn, 0.812; Zn, 0.262; Cu, 0.025; Mo, 0.0435; Fe, 0.405 (in mgL^{-1}). The experiment was completed randomized design with four replications per treatment. All treatments were supplied with

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GA_3 100 mgL^{-1}, 100 ml each, by drenching to soil at different stages i.e.;

Treatment 1 (T1): no GA_3 (supplied only water as control)
Treatment 2 (T2): at shoot emergence stage (4 WAP)
Treatment 3 (T3): at first leaf unfolding stage (7 WAP)
Treatment 4 (T4): at second leaf unfolding stage (10 WAP)
Treatment 5 (T5): at third leaf unfolding stage (11 WAP)

Plant growth and development, and the quantity and quality of flowers and rhizomes were measured. Sampling was done at flowering stage (the first floret opened). Above ground and underground parts were separated and washed with tap water and deionized water then dried and ground into powder. Samples were digested using a Kjeldahl digest solution (Ohyama *et al.*, 1985, 1991) to determine N concentration by a modified indophenol method. The P concentration was determined by the ammonium molybdate method (Davidescu and Davidescu, 1972). The K concentration was determined by atomic absorption spectrophotometry using a $HClO_4$-HNO_3 digestion modified method (Mizukoshi *et al.,* 1994).

RESULTS

Plant growth and development

The results showed that the GA_3 application at different growth stages from shoot emergence to 3rd-leaf unfolding stage had affects on plant growth and development. The height of control plants without GA_3 treatment (T1) increased slowly during 4 to 18 WAP. However, plant height of all GA_3 treatments (T2, T3, T4, T5) increased rapidly after the GA_3 application (**Fig. 1**). In addition, at 18 WAP, T4 and T5 treatment gave the highest plants at 49.3 and 48.3 cm respectively (**Fig. 1**).

The GA_3 application did not only affect on plant height but also affected on the number of shoots per cluster. We found that the application of GA_3 solution decreased number of shoots per cluster and delayed time to shoots sprouting (Khuankaew *et al.*, 2008). Moreover, the applications of GA_3 solution during growing period also tended to decrease the number of shoots per cluster (**Table 1**). In addition the application of GA_3 solution delayed the flowering date (103 days after planting) compared with control plants. The application in different stages had also affected on flowering date, which the flowering date was the most delayed (114 days after planting) in T4 (**Table 1**).

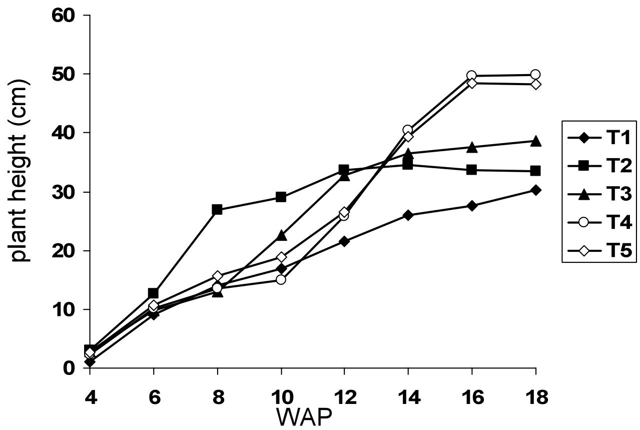

Fig. 1. Effect of GA_3 100 mgL^{-1} application in difference growth stages on plants height of *Curcuma alismatifolia* Gagnep.
T1: untreated plants, T2: shoot emergence stage, T3: 1st-leaf stage, T4: 2nd-leaf stage, T5: 3rd-leaf stage.

Quality and quantity of flowers

The *C. alismatifolia* supplied with GA_3 solution at different stages had effects on spike stalk length (**Table 2**). The results showed that T4 and T5 had taller spike stalks than T1, T2 and T3 treatments. On the other hand, supplying GA_3 solution at shoot emergence stage (T2) to third leaf unfolding stage (T4) decreased the number of spikes per cluster (**Table 2**). The number of pink bracts (coma bracts) and green bracts tended to decrease by later application of GA_3 solution.

Quality and quantity of rhizomes

Thirty weeks after planting, new rhizomes were harvested then cleaned and the size and weight were measured. The GA_3 application had affects on new rhizome fresh weight per cluster, we found that control plants which were not supplied with GA_3 gave the best rhizome fresh weight at 83.7 g per cluster, and heavier than all treatments (**Table 3**). Furthermore, the storage roots length (t-root) were shorted when using GA_3. The rhizome diameter was almost the same among treatments. The numbers of storage roots per rhizome tended to decrease by the application of GA_3 solution

Table 1. Effects of GA_3 application at difference growth stages on growth and development of *Curcuma alismatifolia*

Treatment	GA_3 application stage	Number of leaves per plant (leaf)[NS]	Number of shoots per cluster (shoot)[1/]	Number of days to flower (days)[1/]
T1	No application	5.2	2.6 a	102.6 c
T2	Shoot emergence	5.2	1.4 b	112.2 ab
T3	1st-leaf	5.8	1.2 b	110.6 ab
T4	2nd-leaf	5.4	1.0 b	113.6 a
T5	3rd-leaf	5.6	0.8 b	106.2 bc

[1/] Means with the same letter within column are not significant difference at $p<0.05$ by least significant difference.
[NS] no significant difference.

Table 2. Effects of GA_3 application at difference growth stages on quality and quantity of flowers

Treatment	GA_3 application stage	Spike stalk length (cm)[1/]	Spike length (cm)[NS]	Number of pink bracts[NS]	Number of green bracts[1/]	Number of spikes per cluster[1/]
T1	No application	33.00 d	12.90	10.0	8.4 b	1.8 a
T2	Shoot emergence	38.26 bc	12.10	10.4	9.8 a	1.7 a
T3	1st-leaf	35.75 cd	11.89	9.4	8.6 ab	1.5 ab
T4	2nd-leaf	42.22 b	13.27	9.2	8.6 ab	1.0 c
T5	3rd-leaf	51.83 a	12.72	8.6	7.6 c	1.2 bc

[1/] Means with the same letter within column are not significant difference at $p<0.05$ by least significant difference.
[NS] no significant difference.

Table 3. Effects of GA_3 application at difference growth stages on quality and quantity of rhizomes

Treatment	GA_3 application stage	Rhizome weight per cluster (g)[1/]	Rhizome diameter (cm)[NS]	Rhizome length (cm)[NS]	Number of storage roots per rhizome[NS]
T1	No application	83.66 a	2.12	2.22	6.8
T2	Shoot emergence	56.58 b	1.97	2.52	5.8
T3	1st-leaf	51.93 b	2.21	2.50	6.2
T4	2nd-leaf	44.85 b	2.20	2.29	5.0
T5	3rd-leaf	38.36 b	2.30	2.17	4.0

[1/] Means with the same letter within column are not significant difference at $p<0.05$ by least significant difference.
[NS] no significant difference.

Table 4. Effects of GA_3 application at difference growth stages on nutrient concentrations in plant organs.

Treatment	GA_3 application stage	Nutrient concentration (mg g DW^{-1})					
		Aboveground parts			Underground parts		
		N[1/]	P[1/]	K[1/]	N[NS]	P[NS]	K[NS]
T1	No application	17.2 a	9.7 ab	66.0 a	12.2	22.3	82.2
T2	Shoot emergence	17.2 a	9.2 bc	59.2 bc	13.1	19.6	84.0
T3	1st-leaf	17.6 a	10.4 a	61.9 ab	11.9	22.0	85.3
T4	2nd-leaf	15.6 b	9.1 bc	63.2 ab	12.4	20.5	85.9
T5	3rd-leaf	15.4 b	8.3 c	56.9 c	12.8	20.5	79.1

[1/] Means with the same letter within column are not significant difference at $p<0.05$ by least significant difference.
[NS] no significant difference.

although statistically not significant.

Nutrient concentration in plant organ at flowering stage

At flowering stage (15-16 WAP), underground part organs and aboveground part organs were analyzed in the term of nutrient concentrations. The GA_3 application at different growth stages had effects on nutrient (N, P and K) concentrations in aboveground parts. The results showed that T2 and T3 treatments did not give the effect on N concentration but when using GA_3 applications at T4 and T5, the N concentration was decreased to 15.6 and 15.4 mg g DW^{-1}, respectively, compared with control plant at 17.2 mg g DW^{-1} (**Table 4**). Moreover, T5 treatment gave the lowest P concentration in aboveground parts was 8.3 mg g DW^{-1} compared with control plant at 9.7 mg g DW^{-1}. This experiment showed that *C. alismatifolia* plants which were supplied with GA_3 solution had effects on decreasing K concentration at all treatment GA_3 applied, particularly, at T5 treatment was the lowest K concentration as well as P concentration.

The GA_3 application at different stages did not give the effects on the concentration of N, P and K in the underground part organs. The results showed that the average nutrient concentrations were 12.5, 21.0 and 83.3 mg g DW^{-1} of N, P and K respectively, however, the N, P and K concentrations in the underground parts were higher than the aboveground parts in all treatments.

DISCUSSION

Plant growth and development

In this study, the 100 ml of GA_3 solution (100 mgL^{-1} in water) was applied by drenching to the soil at different growth stages, i.e., shoot emergence, 1st-leaf unfolding, 2nd-leaf unfolding and 3rd-leaf unfolding stages compared with control plants (supplied only with distilled water). The GA_3 application at growing period such as 2nd-leaf and 3rd-leaf stages, the plant height and scape length were higher than the control and other treatment. It might be that during that stages, plant growth and development in both parts of underground (fibrous roots) and aboveground part (leaves) were more grown-up than other treatments. Therefore it gave more response on effects of GA_3 application.

The GA_3 application delayed flowering date as well as application with different stage, showed that at T2, T3 and T4 have more responsible on GA_3 application than at T5. In *Aquilegia x hybrida* Sims., GA_{4+7} had effects on plants height, however, the response depended more on the concentration than the stages of application (Gianfagna and Merritt, 2000). The number of shoots and flowers per cluster was decreased by GA_3 supplied. In this experiment, almost of new shoots produced a flower and number of spikes and shoots was almost the same. It might be that the plants treated with GA_3 had more active growth (expanding leaves and length of spike stalk) than untreated plants. Therefore, the elongation of leaves needs to use chemical materials for growing up, transferred from the source organ to the sink organ, as a result the number of shoots and flowers might be decreased.

For new shoots sprouting plants, low or not enough nutrient supply will depress the shoot sprouting. The other factor, the GA_3 application (exogenous PGR) might be involved in endogenous hormones balance in plants. Fresh rhizome weight per cluster tended to be reduced by GA_3 application at different stages as well as according to above more growth in aboveground part less growth in underground part. Vreugdenhil and Sergeeva (2006) studied the effect of the application of gibberellins on tuberization in potato, the exogenously applied gibberellins inhibited tuber formation, whereas applying inhibitors of gibberellin biosynthesis had the opposite effect. Foliar sprays of the solution containing GA greatly increased the shoot growth of carrot plant, but expense of the growth of the storage root (Linser *et al.,* 1974). This means that there is a sink competition between shoot and root and phytohormones affect on the sink strength of tissues and organs (Marschner, 1996).

Nutrient concentration

The GA_3 application at different stages not only effected on plant growth and development but also effected on nutrient concentration. The results showed that GA_3 application at different growth stages tended to reduce the N, P and K concentrations in aboveground part organs especially supplying at T5 (11 WAP). The application of GA_3 at T5 gave the lowest concentration of N, P and K, at 15.4, 8.3 and 56.9 mg g DW^{-1} respectively. It has been reported that the application of GA reduced the uptake of nitrogen and phosphorus relative to the cations, and this is a common response in pea (Garcia and Guardiola, 1981). Therefore, when GA_3 solution was supplied at T5 stage, the effect of GA_3 on cell elongation and cell expansion might be more effective than the other treatment, as a result the GA_3 application at T5 reduced the uptake of nutrients.

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クルクマ アリスマティフォリア Gagnep. の生長と分化に対する生育時期別ジベレリン施用の影響

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要　約

クルクマ アリスマティフォリア Gagnep. の塊茎を土壌:砂:モミ殻:炭化モミ殻（体積当たり１:１:１:１）の培地を入れたポットで栽培した。本実験では、ジベレリン（GA_3）の施用時期（出芽期（植え込み４週後）、第一葉展開期（植え込み９週後）、第二葉展開期（植え込み10週後）、第三葉展開期（植え込み11週後））がクルクマの生長と分化に及ぼす効果を調べた。GA_3水溶液（100mg L^{-1}）を各ポット当たり100mL 土壌に添加した。対照区では、蒸留水を100mL 添加した。GA_3を生育後期に与えた区では、草丈と花茎が増加したが、茎葉部、花序、塊茎の生体重が低下した。また、GA_3の添加で開花時期が遅くなった。対照区では、植え込み後、102.6日で開花したが、第三葉展開期にGA_3を添加した区では、開花日数は113.6日と遅くなった。開花期（植え込み15-16週間後）における地上部、地下部各器官の、窒素、カリウム、リン濃度と株当たり含有量を測定した。GA_3施用は、施用時期に係らず、地上部の養分濃度に影響を与えた。特に、第三葉展開期に投与した区では、N、P、K 濃度とも最も低下した。しかしながら、GA_3施用は、地下部の N、P、K 養分濃度には影響を与えなかった。

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