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Flower Induction of *Spathiphyllum patinii* by Gibberellin A_3 and Miniaturization of Flowering Plants

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Abstract

Treatment of *Spathiphyllum patinii*, cv. Merry, six months after planting their mericlone plantlets to pots with 500 and 1000 ppm gibberellin A_3 (GA_3) either by spraying onto the leaves or by dropping onto the uppermost leaf sheath induced flowering. Flower stalks developed not only from the uppermost leaf sheath of the main axis following development of 3 or 4 additional leaves but also from the lower leaf sheath. The control plants without GA_3 did not flower within the experimental period.

Furthermore, treatment with 100 to 1000 ppm GA_3 of the cut tuberous-stem pieces and mericlone plantlets produced miniature-flowering plants with new 2 leaves.

These results suggest that floral induction at the shoot apex of *Spathiphyllum* plants occurs soon after the treatment with GA_3 regardless of their age, size and growing seasons. The method employed in the present experiment should be useful for rapid production of small or miniature flowering *Spathiphyllum* plants which are used not only as a new interior ornament, but also as an experimental plant and a method for physiological study of flower initiation by gibberellins.

Key words: Flower induction · GA_3 · Miniature flower plant · *Spathiphyllum*

I. Introduction

Flowering potted plants of *Spathiphyllum* are shade tolerant and their flowering period of unfurling white bract in mucronate-elliptical shape lasts for one or two months at room temperature. The plants are usually propagated by means of tissue-cultured mericlone plantlets and can flower following growth in a greenhouse for 15 to 21 months[2]. Henny (1981)[1] found that flowering of *Spathiphyllum* cv. Mauna Loa was induced by foliar application of 500 and 1000 ppm gibberellic acid. Shibata and Endo (1990)[4] also ascertained the promotion of flowering by gibberellin A_3 (GA_3) in *Spathiphyllum* cv. Mauna Loa and reported that the effect of GA_3 varied depending on the growing conditions such as temperature and light intensity. Our preliminary results[2] also showed that flowering of *Spathiphyllum* cv. Merry was hastened by treatment with GA_3 or GA_{4+7}. However, these previous studies were carried out using old plants which had grown for more than one year[1,2] or which had ever flowered once[4]. In those reports, it has still remained to be elucidated whether the GA_3 treatment causes initiation of flower buds at the shoot apex or it accelerates only development of flower buds already initiated.

In the present experiments, we examined the effect of GA_3 on the flower induction of the main axis using

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young *Spathiphyllum* cv. Merry plants of 6-month age. The flower-inducing effect of GA_3 in very small plants derived from the cut tuberous-stem pieces and the mericlone plantlets was also studied.

II. Materials and Methods

1, *Six-month-old potted plants*

The *Spathiphyllum* cv. Merry were grown for 6 months after planting the mericlone plantlets in vinyl pots of 9 cm in diameter, filled with a mixture of fertile soil, akadama soil, perlite and vermiculite (4:4:1:1 v/v/v/v). Plant height was 18 to 22 cm and each plant had 2 to 3 juvenile type leaves and 3 to 4 adult type leaves. An aqueous solution of 50 to 1000 ppm GA_3 containing 0.05% Tween-20 was applied either by spraying to the leaves (0.5 ml/plant) or by application of a drop with a glasscapillary tube onto the uppermost leaf sheath (0.05 ml/plant). Both applications were repeated one week later. The control plants were treated with distilled water containing Tween-20. On the first day, the uppermost open leaf was marked so that the number of new leaves emerging after treatments could be counted. Plants were grown in a glasshouse shaded 50% with cheesecloth from September to May, and watered with tap water every morning. Temperature ranged from 10 to 25°C, and the light intensity at midday was 50 to 100 μmol $m^{-2}s^{-1}$ photosynthetically active radiation (PAR). The number of new leaves on the main axis, plant height and flowering were recorded at 15-day intervals. The appearance of the furled bract from the leaf sheath was regarded as flowering. The white elliptical bracts unfurled with appearance of the spadix 1 to 2 months after the flowering began. Flowering was expressed by both the percentage of plants flowering to the total number of plants used and the average number of flowers per plant. Ten potted plants were used for each treatment. The experimental results are presented as the averaged values with a standard error.

2. *Tuberous stem pieces*

Tuberous stems were obtained from the main axis with several lateral shoots of 15-month-old plants grown in a plastic pot of 18 cm in diameter filled with the soil mixture. The tuberous stems were washed thoroughly with tap water, and then cut lengthwise into 4 or 5 pieces (about 2.5 g fresh weight per piece). The stem pieces were sterilized with 70% ethanol and 1% perchlorite solution, and then placed in a petri dish containing an aqueous solution of GA_3 at 10 to 500 ppm for 20 h in fluorescent light of 30 μmol $m^{-1}s^{-2}$ PAR at 25°C. As the control, the stem pieces were treated with distilled water. These stem pieces were each planted in vinyl pots of 9 cm in diameter filled with the soil mixture, and grown under 50 to 250 μmol $m^{-1}s^{-2}$ PAR at midday of 25 to 28°C in a glasshouse and watered daily with tap water. About 2 weeks later the uppermost bud present on the stem pieces began to sprout and the leaves emerged. Three months later, the number of plants with a bract was counted and the percentage of plants flowering was determined. The number of leaves on the shoot also was counted. Shoot length below the spadix in the flowering plants and that below the basal end of the top leaf blade in the non-flowering plants were measured and referred to as plant height. Fifteen to 20 stem pieces were used for each treatment.

3. *Mericlone plantlets*

Mericlone plantlets having 3 or 4 small leaves were taken out of the tissue culture bottles and their roots were washed thoroughly with tap water. Water on the surface of the leaves and roots was removed by placing

the plants on filter paper for a few minutes. Then the plants were placed in a petri dish containing an aqueous solution of GA_3 at 10 to 1000 ppm for 20 h in the light of 30 μmol $m^{-1}s^{-2}$ PAR at 25°C. The control plantlets were treated with distilled water. Those plants were planted in lines 3×5 cm apart in a plastic box filled with peatmoss, perlite and vermiculite (1 : 1 : 1 v/v/v). They were grown at 28°C under continuous fluorescent light of 70 μmol $m^{-1}s^{-2}$ PAR at leaf level and watered daily with tap water and sometimes with Hyponex solution. Two and a half months later, number of plants flowering, stage of flower development, number of newly emerged leaves and plant height were measured. The stage of flower development was determined according to the score of floral stages: 0. vegetative state; 1. appearance of bract from leaf sheath ; 2. entire bract emerged out of the leaf sheath ; 3. elongated flower stalk ; 4. unfurling bract ; 5. bract unfurled with the appearance of spadix. Fifteen to 20 mericlone plantlets were used for each treatment.

III. Results

1. *Flowering of six-month-old potted plants*

As shown in Figs. 1-a, A and B, and 1-b, A and B, flowering began in a few plants at the beginning of November, about two months after treatment, with 500 and 1000 ppm GA_3 by either method of application.

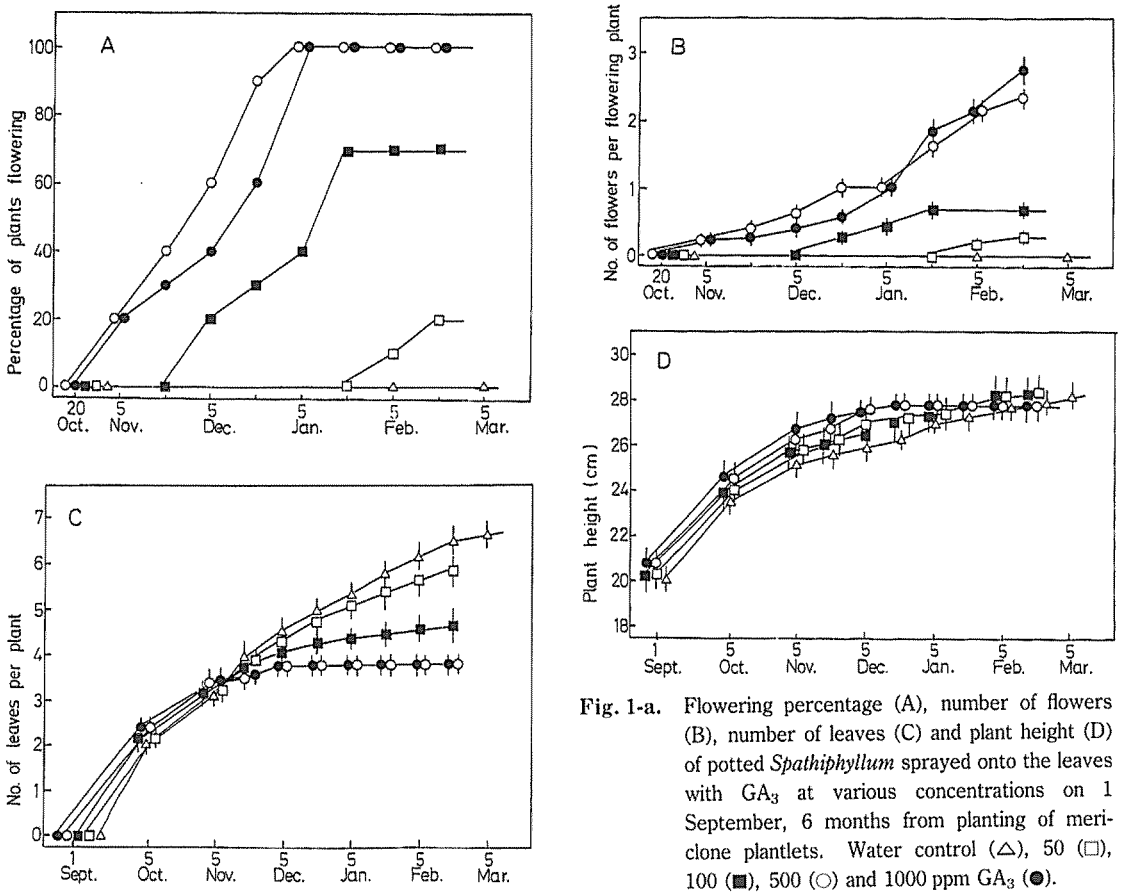

Fig. 1-a. Flowering percentage (A), number of flowers (B), number of leaves (C) and plant height (D) of potted *Spathiphyllum* sprayed onto the leaves with GA_3 at various concentrations on 1 September, 6 months from planting of mericlone plantlets. Water control (△), 50 (□), 100 (■), 500 (○) and 1000 ppm GA_3 (●).

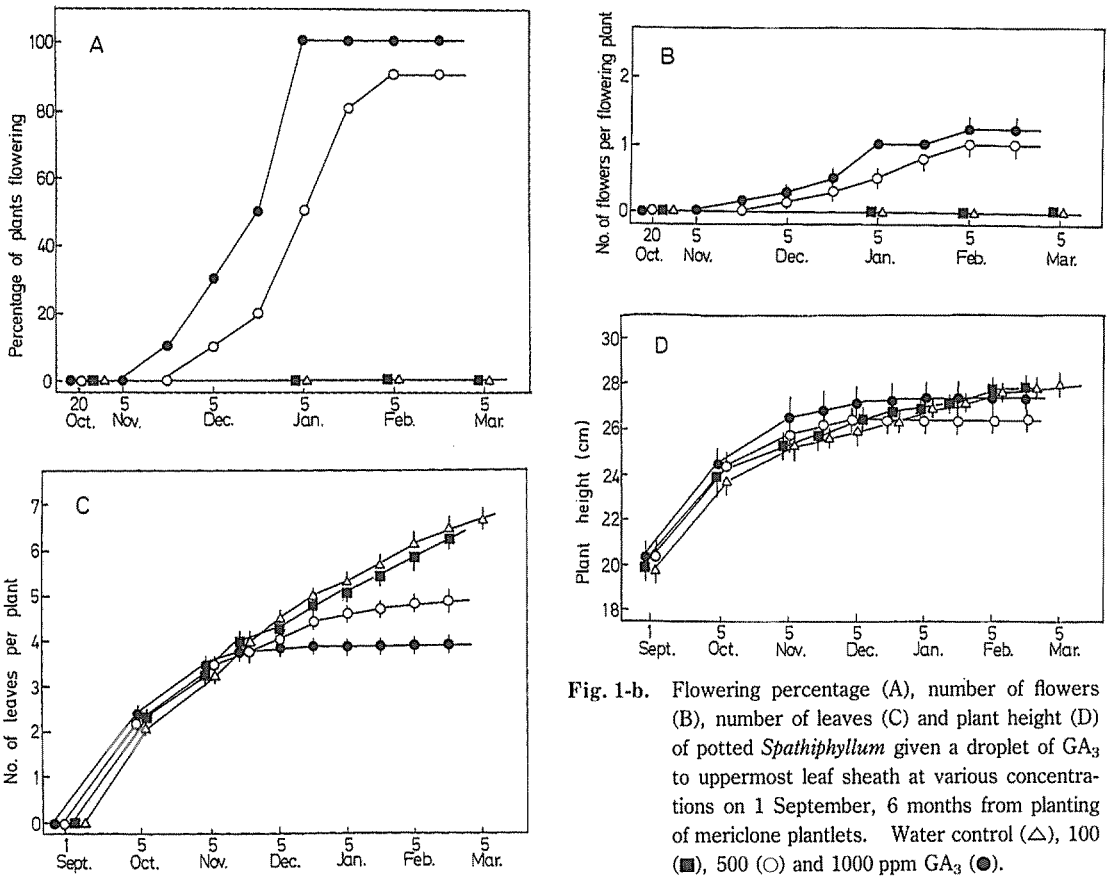

Fig. 1-b. Flowering percentage (A), number of flowers (B), number of leaves (C) and plant height (D) of potted *Spathiphyllum* given a droplet of GA_3 to uppermost leaf sheath at various concentrations on 1 September, 6 months from planting of mericlone plantlets. Water control (△), 100 (■), 500 (○) and 1000 ppm GA_3 (●).

Most treated plants had developed flower stalks from the uppermost leaf sheath by the end of December, about 4 months after treatment, and thereafter also from the lower leaf sheaths and from some lateral shoots. Flowering was more stimulated with higher concentrations of GA_3. GA_3 application by the drop method was less effective than that by spraying at all concentrations tested. As shown in Figs. 1-a, C and 1-b, C, the flowering occurred after 3 or 4 new leaves had emerged with 500 or 1000 ppm GA_3 treatment. The application of GA_3 promoted plant-height increase only slightly before flowering but the increase of plant height diminished after flowering (Figs. 1-a and 1-b: D). The control plants did not flower within the experimental period. However, a few control plants flowered at the end of May when the plants were 31.5 cm $\pm$ 1.2 in height and had 9.5 $\pm$ 0.3 leaves.

2. *Flowering of stem pieces*

Fig. 2 shows the percentage of plants flowering, number of leaves and plant height for the plantlets grown from the tuberous stem pieces treated with GA_3 at various concentrations. The higher the GA_3 concentration, the greater was the flowering response with fewer number of leaves and smaller plant height. The treatment with 10 to 1000 ppm GA_3 produced very small flowering plants, 6 to 8 cm in height with only 2 or 3 leaves. The control plants were still vegetative 12 months after treatment. Similar results of flowering were also obtained in

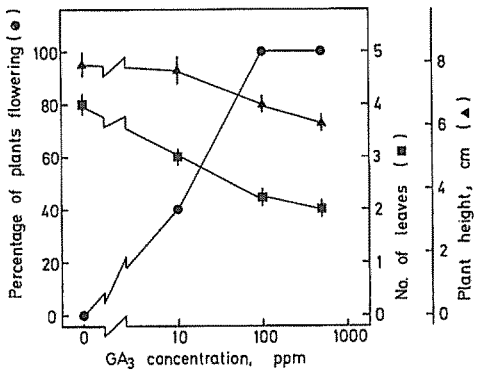

Fig. 2. Flowering percentage (●), number of leaves (■) and plant height (▲) of *Spathiphyllum* plantlets sprouted from the stem pieces of the main axis and treated with GA_3 at various concentrations. Measurements were made 3 months after the treatment with GA_3.

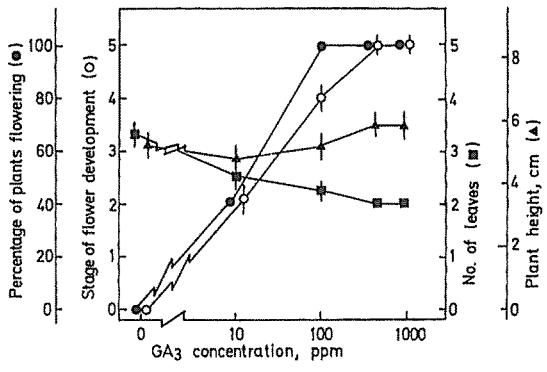

Fig. 3. Flowering percentage (●), stage of flower development (○), number of leaves (■) and plant height (▲) of *Spathiphyllum* mericlone plantlets treated with GA_3 at various concentrations. Measurements were made 2 and half months after the treatment with GA_3.

the experiment with the stem pieces of the lateral plants from the same plants (data not shown).

3. *Flowering of mericlone plantlets*

Fig. 3 shows the percentage of plants flowering, stage of flower development, number of newly emerged leaves and plant height for the mericlone plantlets treated with GA_3 at various concentrations. The treatment with 100 to 1000 ppm GA_3 provoked flowering in all plants with an unfurled bract, and the plants were very small having only 2 newly emerged leaves. The control plants remained vegetative even 12 months.

IV. Discussion

The present results demonstrated a flower inductive effect of GA_3 on the 6-month-old *Spathiphyllum* plants which were too young to flower under natural conditions. Spraying of GA_3 onto the leaves was more effective in promoting flowering than drop application of GA_3 onto the leaf sheath (Compare Figs. 1-a with 2-a: A and B). This may be not caused by the difference of GA_3 action on the floral processes in the leaves due to the site of its application but rather caused by the fact that floral induction requires a larger amount of GA_3 at the apex. In fact, the GA_3 solution volume and absolute amount provided to each plant by spraying was ten times greater than that by drop application. Furthermore, the effect of GA_3 on flowering was also greater at a higher concentration and larger absolute amount by either method of application (Figs. 1-a and 1-b: A and B).

The application of GA_3 resulted in flowering from the third or fourth leaf sheath on the main axis after the treatment (Figs. 1-a and 1-b: C). Quite similar result of flowering, the appearance of bract following emergence of a few leaves on the main shoot with GA_3, was also obtained in another experiment using 12-month-old plants growing in summer[3]. These results also suggest that floral induction at the shoot apex actually occurs soon after treatment with GA_3 regardless of plant age and growing season. Flower initiation at the apex stopped the subsequent production of leaves on the main axis (Figs. 1-a and 1-b: C, Figs. 2 and 3). This explains why the

flowering plants are shorter in height and have fewer leaves than the non-flowering plants (Figs. 1-a and 1-b: D, Figs. 2 and 3). The present results also show that GA_3 treatment can induce flowering of *Spathiphyllum* plants within a shorter growing period after planting mericlon plantlets to pots. The GA_3 treatment of the cut pieces of stems or the mericlone plantlets produced very small flowering plants with only 2 leaves or 2 additional leaves (Figs. 2 and 3). These also are evidence that exogenous GA_3 can induce flower initiation immediately after the application.

Exogenous GA_3 at a higher dosage such as 500 or 1000 ppm was much more effective on floral induction of the plants at all ages from mericlone plantlets (Fig. 3) to 6-month-old plants (Fig. 1-a) and 15-month-old plants[2]. This evidence suggests not only the requirement of a large amount of GA_3 for initiation of the flower buds at the apex of this species but also no difference in sensitivity of the apex to the amount of GA_3 regardless of developmental stage of the plants. It is interested to examine further the level of endogenous gibberellins at the shoot apex of *Spathiphyllum* plants to bring about the flower induction.

The bracts of the miniature flowering *Spathiphyllum* plants produced by GA_3 treatment in the continuous fluorescent light (Figs. 3 and 4) were usually whitish leafy with green midrib or rarely small orbicular unlike elliptical shape of entire white bracts which formed usually in the plants at 6 months old grown in the greenhouse. The deformation of bracts formed at the earliest stage in the development of the flower buds is an interesting physiological aspect of the initiation of flower buds at the shoot apex.

The bracts of these small flowering plants have retained their whitish colour for one month or more at room temperature. These small potted flowering plants may serve to create an atmosphere of the room interior different from large flowering plants.

Fig. 4. Flowering plant of *Spathiphyllum* derived from mericlone plantlet treated with 500 ppm GA_3 and bar scaled in cm (right), and non-flowering plant with distilled water (left). A bract unfurled on the main axis and bract unfurling on the lower axillary shoot are seen in the GA_3-treated plant. The plants were transplanted into plastic pots of 8 cm in diameter 3 months after treatment and photographed.

The present method of GA_3 treatment can produce rapidly such small or miniature flowering *Spathiphyllum* plants which may be used not only for a potted flower ornament but also as an experimental plant and a method for physiological study of flower initiation by gibberellins.

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スパティフィラムの花成誘導に及ぼすジベレリン A_3 の作用と小型の開花植物の育成

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スパティフィラムのメリクロン苗を培養土を入れた鉢に植え6か月間栽培した展開本葉5－7枚の草丈18–22 cmの植物を用いて，500あるいは1000 ppmのジベレリン A_3 (GA_3) 溶液を葉面散布または最上葉の葉鞘内に滴下すると，その後展開した3あるいは4枚めの葉の葉鞘から白色楕円形の苞葉が出現し，花茎が伸長して開花した。また，下位葉の葉鞘からも花茎が伸長して開花した。

肥大根茎の切断片およびメリクロン苗を，10から1000 ppm GA_3 の溶液に20時間ひたして培養土に植えると，葉を2枚を持った小さい開花植物を生じた。

これらの実験結果は，GA_3 を処理すると，スパティフィラムの花成が，草齢や植物体の大きさに関係なくすぐに誘導されることを示している。GA_3 による若齢のスパティフィラムの花成誘導は，小型の鉢花生産にまたジベレリンによる花成誘導の生理学的研究に利用できる。

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