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Vegetative Propagation in Everbearing Strawberry as Influenced by a Morphactin, GA_3, and BA[1]

María M. Biain de Elizalde and María R. Guitman[2]
Centro de Ecofisiología Vegetal, Serrano 661, 1414 Buenos Aires, Argentina

Additional index words. Fragaria X *ananassa,* methyl 2-chloro-9-hydroxy-fluorene-9-carboxylate, gibberellic acid, benzyladenine, runner

Abstract. **Applications of the morphactin IT 3456 (methyl-2-chloro-9-hydroxy-fluorene-9-carboxylate) (*Fragaria* X *ananassa* Duch., cv. Rabunda) at concentrations of 10 to 200 ppm promoted crown division; IT 3456 at 15 ppm with benzyladenine (BA) stimulated vegetative propagation, evaluated by adding the total number of runner plants to that of side-crowns. Gibberellic acid (GA_3) alone or morphactin with GA_3 had no significant effect on runnering or lateral branching.**

The octoploid everbearing cultivars of strawberry have been of interest to horticulturists and geneticists because they have a long period of fruit production. They are considered day neutral for flower induction (5), and flower continuously after a short winter rest. The most important disadvantage of some everbearing cultivars is limited production of runners.

The seasonal cultivars of *Fragaria* X *ananassa* are quantitative short day plants (5). Some authors have found an interaction between photoperiod and temperature on flower initiation and runner formation (3, 6, 7). Inductive conditions for flowering are 12 hr at 18°C, and for runnering 16 hr at 24°. There is significant variation among cultivars.

Gibberellin application to different species of *Fragaria* promotes runner formation (5, 11). Leshem (10) showed that temperature treatments that promote runnering lead to greater production of gibberellin-like substances.

Several methods have been suggested to increase planting stock of everbearing cultivars including flower removal (4), growth regulator application (8, 1), or a combination of these treatments (4, 13). The results, however are still controversial for commercial purposes. Rodríguez (14) applied gibberellic acid to 'Rabunda' plants during July and/or January and failed to increase runnering. The removal of flowers during October and November, however, increased the formation of runners by 36%.

The experiments reported here were devised to determine the effect of the morphactin, IT 3456 (E. Merck A. G., Darmstadt, Germany) in combination with GA_3 or BA on runnering and crown proliferation of the everberaring 'Rabunda' strawberry.

Materials and Methods

'Rabunda' was introduced to Argentina by the Estación Experimental Agropecuaria San Pedro (INTA) from Holland and is considered one of the best everbearing strawberries for commercial purposes, except for its reduced capacity for runnering.

Cold-stored single plantlets were rooted directly in black plastic pots containing 1 kg of soil from the horticultural area. Daily irrigation was supplemented twice a week with Hoagland nutrient solution. The flowers were continuously removed during the experiments.

In the first experiment we compared the effect of morphactin on the vegetative propagation of 'Rabunda' and of the seasonal 'Cambridge Favorite', a vigorous runner producer. On May 30, 1975, plants of 'Rabunda' were sprayed to drip with IT 3456 at 0, 10, 50, 100 and 200 ppm; 'Cambridge Favorite' was treated with only 0, 10 and 100 ppm. Each treatment was replicated 5 times for both cultivars and arranged in a completely randomized design. This experiment was carried out in a glasshouse at 10-20°C with 16 hr photoperiod using HPL lamps to supplement the natural daylight. The number of runners and sprouting buds were counted 70 days after treatment.

A second experiment was carried out in a glasshouse with temperatures between 18 and 30°C. 'Rabunda' plants were cold stored for 30 days and rooted as described above. Natural photoperiod was prolonged to 16 hr with white fluorescent lamps (110 W). On September 9, 1977, the following treatments were applied: 4 groups of 21 plants were sprayed to drip with IT 3456 at 0, 15, 30 or 60 ppm. Twenty days later, each group was divided into 3 subgroups. One was sprayed with 100 ppm GA_3, the second with 50 ppm BA and the third was not sprayed a second time. Fifteen days later, the GA_3 and BA treatments were repeated using 50 ppm GA_3 and 25 ppm BA, respectively. The experiment thus had 12 treatments with 7 replications.

Three months after the initial treatments were applied, the number of runners, runner plants and lateral-crowns were determined; the data were transformed to $\sqrt{x}$ or $\sqrt{x+3/8}$ (when the counts include zero values) to obtain homogeneity

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of variances. Duncan's multiple range test was performed on the transformed data but estimates of means are given (Table 1 and 2) in the untransformed scale (16).

Results

In the first experiment IT 3456 released the lateral buds from apical dominance on both cultivars (Table 1). The number of sprouting buds in 'Rabunda' was significantly increased by 10 ppm IT 3456 but in maximum response was 50 ppm. In 'Cambridge Favorite' 10 ppm produced maximal response. There was no runner production in either cultivar.

Vegetative growth was proportionaly reduced by morphactin applications. The highest rates temporarily affected elongation of the shoot axis and reduced the leaf area. Some leaf and flower anomalies were also observed although the capability for final recovery was not lost. Root elongation was stimulated by morphactin and lateral root growth inhibited.

The second experiment was started using morphactin followed by the GA_3 or BA. Leaf and flower anomalies were noticed a few days after the morphactin treatments and growth was depressed in proportion to concentration. The number of runners was not significantly increased by any treatment although morphactin (15 ppm) and BA had the greatest number of runners. Higher rates of morphactin reduced runnering. GA_3 did not influence the response of the morphactin-induced plants. The greatest number of runner plants was found with GA_3 applied alone, but there was no significant increase over the control treatments. A tendency to increased runner plants was observed with 15 ppm IT 3456 applied in combination with BA.

Morphactin significantly increased the number of side-crowns at all concentrations. The increments were more than 125% over the control; without significant differences among rates. GA_3 or BA did not modify this response (Table 2). The capacity of vegetative propagation of 'Rabunda' plants could be measured by adding the total number of runner plants to the number of side-crowns. This was significantly enhanced by 15 ppm IT 3456 in combination with BA. There was a positive interaction between these regulators to induce an increment of about 130% in planting stock (Table 2).

Discussion

The development of side-crowns and runners in strawberry involves the release of apical dominance. Runners are axillary branches in which the internodes become greatly elongated, while in the absence of elongation the branch becomes a new crown on the parent plant or may remain small and undeveloped (5). In these experiments with 'Rabunda', an everbearing cultivar with reduced capacity for vegetative propagation, new side-crowns were formed following morphactin applications but the number of runners was not significantly increased. Similar results were found by Andrew et al. (1) in morphactin-induced plants of 2 everbearing strawberries. The morphactins are known to affect various physiological and morphogenetic processes e.g. inhibition of the apical bud and elongation of the main shoot, resulting in the emergence of lateral buds. This morphogenetic effect distinguishes the morphactins from other growth retardants. Its action may be antagonistic or synergistic with other regulators without having either a specific or a competitive interaction (15).

A tendency to increase runners formation by 15 ppm IT 3456 and BA applications was observed. BA may be acting by enhancing the enlargement of internodes of the newly emerged lateral branches induced to develope by the morphactin. Kender et al. (8) working with 'Geneva', a very poor runner producer, proposed that BA may play a permissive role when endogenous inhibitors blocked the gibberellin-mediated runnering response.

The combined treatment of 15 ppm IT 3456 and BA resulted in the greatest production of plantlets from runners and crown division. The developmental capacity of both types of plantlets has been considered similar (Rodriguez, personal communication). Boxus et al. (2) working with plant tissue cultures, have not observed any difference between meristems excised from lateral or apical buds of mother plants or from runners. The choice of meristems on the mother plant does not influence their further development.

GA_3 did not modify the response of morphactin-induced plants in relation with the number of runners of lateral crowns. GA_3 alone produced a greater number of runner plants though it was not statistically significant. GA_3 in combination with morphactin did not induce a similar effect. It may be that the rate of GA_3 was insufficient to overcome the depressive effect of morphactin on the longitudinal growth. Mann et al. (12) and Kohler (9) working with *Citrus* seedlings and *Pisum sativum*, found that GA_3 does have a concentration-dependent-counteracting effect on the morphactin-induced inhibition of the shoot elongation growth.

Vegetative growth during the second experiment was greatly improved, possibly due to a more appropriate set of environmental conditions (light and temperature). The external conditions may modify the vegetative response of the plants to exogenous applications of growth regulators.

One of the problems posed by the everbearing strawberry cultivation is the poor vegetative propagation. The use of the growth regulators as morphactin and BA could be a useful technique for the horticulturists as a means of increasing the number of runner plants and the lateral crowns.

Table 1. Effect of IT 3456 on the number of lateral buds growing per plant of 2 strawberry cultivars.[z]

IT 3456 (ppm)	No. of buds growing	
	Rabunda	Cambridge Favorite
0	1.5 e[z]	1.8 c
10	4.5 d	5.9 ab
50	7.8 a	–
100	7.0 abc	7.0 a
200	7.5 ab	–

[z]Means separation within columns by Duncan's multiple range test, 5% level.

Table 2. Effect of morphactin, GA_3 and BA on runnering and crown-division of 'Rabunda'.

Treatments	IT 3456			
	0 ppm	*15* ppm	*30* ppm	*60* ppm
	No. runners			
Control	1.00 abc[z]	1.14 abc	0.29 bc	0.14 c
GA_3	1.57 ab	1.43 abc	0.57 bc	1.00 bc
BA	1.43 abc	2.43 a	1.00 abc	0.43 bc
	No. runner plants			
Control	1.86 abcde	3.00 abcd	1.29 de	0.0 e
GA_3	5.43 a	2.71 abcde	0.71 de	1.57 bcde
BA	3.86 abc	4.00 ab	2.00 cde	1.00 cde
	No. side-crowns			
Control	1.14 g	2.57 abcd	2.71 abcde	3.43 ab
GA_3	1.00 g	2.71 abc	3.57 ab	3.00 abc
BA	1.00 g	3.00 abc	3.43 ab	2.57 abcdef
	No. runner plants + side-crowns			
Control	3.00 b	5.57 ab	4.00 ab	3.43 ab
GA_3	6.43 ab	5.42 ab	4.28 ab	4.57 ab
BA	4.86 ab	7.00 a	5.43 ab	3.57 ab

[z]Mean separation in columns and rows for each parameter by Duncan's multiple range test, 5% level.

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Effects of Inorganic Cations on Ethephon-induced Increases in Membrane Permeability[1]

B. W. Poovaiah

Department of Horticulture, Washington State University, Pullman, WA 99164

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Abstract. **Treatment of beet root slices (*Beta vulgaris* L.) with (2-chloroethyl)phosphonic acid (ethephon) at 100 ppm increased membrane permeability 7-fold as indicated by betacyanin leakage. Leakage was much greater at higher concentrations of ethephon. Divalent and trivalent cations (Ca^{++}, Mg^{++}, La^{+++}) prevented the effects of ethephon on membrane leakage without altering the rate of ethylene evolution. The monovalent cations (K^+, Na^+, NH_4^+) were not effective in relieving the ethephon effect.**

The influence of ethephon on flowering, vegetative growth, dormancy, abscission, ripening, senescence, latex flow and disease, and frost resistance are well established (1, 2, 3, 4, 14, 15, 16, 17). These effects are dramatic and commercially important (4, 21).

When ethephon breaks down, it releases ethylene gas, chloride and phosphate ions (2, 23). The release of ethylene is pH-dependent and is enhanced at higher pH (23). Physiologically ethephon is effective at concentrations between 100 and 10,000 ppm (4). High concentrations of ethephon (1,000-10,000 ppm) cause leaf abscission, senescence, terminal dieback, enlarged lenticels, splitting of bark and excessive gummosis (1, 4, 14). Ethylene-induced increases in membrane permeability have been recently reported in bulb tissues (12) and flowers (6). This investigation explores the possibility of reducing membrane damage caused by ethephon.

Materials and Methods

Beet roots were cut into sections 1 cm in diameter and 1 to 2 mm thick. The sections were washed for 2 hr in several changes of aerated, distilled water. Five sections were then transferred to 10 ml of test solution. The loss of membrane integrity was assessed by spectrophotometric determination of betacyanin pigment that leaked from the vacuole into the treatment solution during 12 hr. Each treatment was replicated 4 times and each experiment repeated 4 times.

For ethylene determination, 20-ml vials containing 10 ml of test solution and 5 beet slices were sealed with a serum cap for 4 hr. One ml gas samples were then removed and injected into a gas chromatograph equipped with a flame ionization detector as previously described (10). The pH was monitored at the beginning and at the completion of each experiment.

Results and Discussion

Ethephon treatment increased pigment leakage from root sections, but the addition of $CaCl_2$ at 10^{-3} to 10^{-2}M reduced the leakage to that of the water control. Since stabilization and destabilization effects of various cations might alter the response to ethephon, other cations were tested alone and in combination with ethephon. Ethephon caused the greatest leakage, and Ca^{++} depressed leakage more than 5-fold (Table 1).

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