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## The growth of 'Crystal' guava seedling in response to pinching and dormancy breaking chemicals

**Abstract.** Pinching and dormancy breaking chemicals (DBC) application are potentially used to regulate the growth of plant. This study aims to evaluate the growth response of 'Crystal' guava (*Psidium guajava* L.) seedling in response to pinching and DBC application. This experiment was carried out in the Integrated Field Laboratory, Faculty of Agriculture, Universitas Lampung from March to June 2021, with a randomized completely block design (RCBD) with 2 factors (pinching and DBC) and repeated four times. The results showed that pinching could reduce the increase of height of guava seedling. The combination of pinching with DBC could significantly increase the number of new emerging leaves, branches, and shoots as well as the length of new shoots. The leaf area on new emerging leaves was not affected by pinching and DBC factors. The application of DBC to non-pinched plants inhibited vegetative growth and precisely increased generative growth, as indicated by the increase of the number of flowers produced. The most recommended treatment to improve the vegetative growth of guava plant seedlings was a combination of pinching and  $\text{KNO}_3$  40 g  $\text{L}^{-1}$ .

**Keywords:** 'Crystal' guava · Dormancy breaking chemicals ·  $\text{KNO}_3$  · Pinching · Vegetative growth

## Respon pertumbuhan bibit jambu biji 'Kristal' terhadap aplikasi pinching dan zat pemecah dormansi

**Sari.** Pinching dan pemberian zat pemecah dormansi berpotensi untuk digunakan sebagai pengatur pertumbuhan tanaman. Penelitian ini bertujuan untuk mengevaluasi respon pertumbuhan bibit tanaman jambu biji (*Psidium guajava* L.) 'Kristal' terhadap perlakuan pinching dan pemberian zat pemecah dormansi (ZPD). Percobaan ini dilaksanakan di lahan Laboratorium Lapang Terpadu, Fakultas Pertanian, Universitas Lampung dari bulan Maret hingga Juni 2021, dengan rancangan acak kelompok (RAK) faktorial (pinching dan ZPD) yang diulang sebanyak 4 kali. Hasil penelitian ini menunjukkan bahwa teknik pinching dapat menurunkan pertambahan tinggi tanaman jambu. Namun hal ini justru dapat memperbaiki pertumbuhan bibit jambu. Kombinasi pinching dengan ZPD dapat meningkatkan jumlah daun, cabang, dan tunas baru serta panjang tunas baru secara signifikan. Luas daun baru tanaman jambu tidak dipengaruhi oleh faktor pinching dan ZPD. Pemberian ZPD pada tanaman yang tidak dipinching menghambat pertumbuhan vegetatif, sebaliknya meningkatkan pertumbuhan generatif dengan indikator peningkatan jumlah bunga yang diproduksi. Perlakuan yang paling direkomendasikan untuk memperbaiki pertumbuhan vegetatif bibit tanaman jambu adalah kombinasi antara pinching dan  $\text{KNO}_3$  40 g  $\text{L}^{-1}$ .

**Kata kunci:** Jambu biji 'Kristal' ·  $\text{KNO}_3$  · Pertumbuhan vegetatif · Pinching · Zat pemecah dormansi

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## Introduction

Guava (*Psidium guajava* L.) is a fruit tree species from the genus of *Psidium* that is originated from the Central American region prior to widely spread throughout the world (Aravalo-Marin *et al.* 2021), including Indonesia (Widyastuti *et al.*, 2019a; 2019b). Guava fruit can be used both as table fruit and raw material for making fruit juice. Guava fruit is rich in natural fiber, antioxidant compounds and vitamin C (Guntarti and Hutami 2019; Hartati *et al.*, 2020; Susanto *et al.*, 2019). Based on the results of previous studies, varietal factors can cause differences in the nutritional and phytochemical content of guava fruit (Suwanwong and Boonpangrak, 2021; Omayio *et al.*, 2022).

One of the popular guava varieties today is 'Crystal'. This variety comes from Taiwan whose pedigree is the result of the selection of the mother tree in the experimental garden of Taiwan technical Mission, Delanggu, Mojokerto, East Java (Ministry of Agriculture 2007). 'Crystal' guava has superior characteristics based on consumer likeliness such as sweet fruit flesh, high edible fruit portion, crispy, few seeds, and high nutritious content, e.g., vitamin C (18.73 mg per 100 g) (Ministry of Agriculture 2007). Due to its importance, the guava production improvement through agricultural intensification is highly required.

Concerning agricultural intensification, several culture techniques are potentially applied to gain fruit production in tropical areas. Earlier studies reported the regulation of plant growth and development by selecting the suitable growing area (Widyastuti *et al.*, 2022; Widyastuti 2019b), and employing certain technology of shading (Budiarto *et al.*, 2019b; Budiarto *et al.*, 2022), application of inorganic, biofertilizers (Dheware *et al.*, 2020), and organic fertilizers (Musyarofah *et al.*, 2020; Luna-Jiménez *et al.*, 2020). In addition, physical manipulation is also reported to be able to regulate fruit tree plant grow and development, namely shoot pruning (Bhagawati *et al.*, 2015; Budiarto *et al.*, 2018; Susanto *et al.*, 2019; Widyastuti *et al.*, 2019c), flower pruning (Suman and Bhatnagar 2019), root pruning (Budiarto *et al.*, 2019a), and stem strangulation (Widyastuti 2019a).

One of several physical manipulation techniques that has not been widely applied to guava is pinching. Pinching can be defined in

general as the light level of hand pruning on the apical bud, also known as apical shoot removal, that aims to stimulate the emergence of lateral shoot. Previous experiments on kaffir lime proved that pinching give significant effect on the vegetative growth performance (Budiarto *et al.*, 2019b).

Further growth improvement potentially to be achieved by plant growth regulators (PGR) application. PGR can be defined as a group of artificial compounds that can stimulate or inhibit the process of plant growth with a mode of action similar to phytohormones. Phytohormones such as auxin and cytokinin are reported to involve in the formation of cambium growth (Bagdassarian *et al.*, 2020).

One of the PGR from the cytokinin group that is commonly used is Benzyl amino purines (BAP). BAP can be used as a dormancy breaking chemical (DBC). Previous research stated that giving BAP 100 ppm can increase the percentage of success of side grafting, accelerate the dormancy breaking time and increase the length of guava shoots (Pratomo, 2018). In addition to BAP,  $KNO_3$  is also often used as DBC. According to Hendrajaya (2019) the provision of  $KNO_3$  (40 g per plant) which causes an increase in plant growth performance continues to increase crop yields.

The research on pinching and DBC application is still not widely performed in the 'Crystal' guava plant. Therefore, this study aims to examine the growth of the guava seedling in response to pinching and application of DBC in the form of BAP and  $KNO_3$ .

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## Materials and Methods

This experiment was carried out in the Integrated Field Laboratory Complex of the Universitas Lampung (120 m above sea level), starting from March to June 2021. The planting material used is a 1-year-old 'Crystal' seedling, with a high uniformity level and free from pest and disease attacks.

This study used a two-factor of Randomized Completely Block Design (RCBD). The first factor was (P) pinching which consisted of two levels, i.e., ( $P_1$ ) with pinching and ( $P_2$ ) without pinching. Pinching was applied manually by hand, on the apical bud of guava seedling. Afterward, the DBC was applied on the guava seedling according to the treatment.

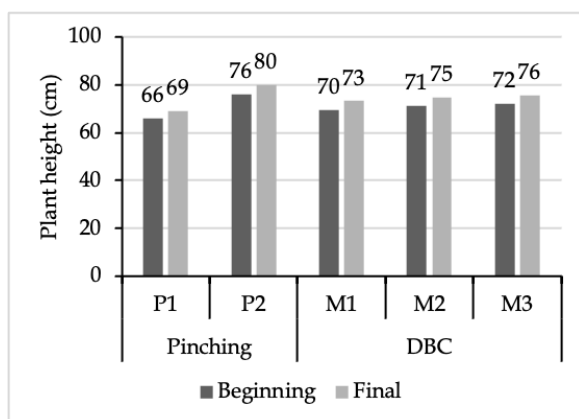
The second factor was the (M) DBC that consisted of 3 levels, namely (M<sub>1</sub>) without DBC, (M<sub>2</sub>) KNO<sub>3</sub> 40 g L<sup>-1</sup>, and (M<sub>3</sub>) BAP 100 ppm. From these two factors, six combinations were obtained (P<sub>1</sub>M<sub>1</sub>, P<sub>1</sub>M<sub>2</sub>, P<sub>1</sub>M<sub>3</sub>, P<sub>2</sub>M<sub>1</sub>, P<sub>2</sub>M<sub>2</sub>, P<sub>2</sub>M<sub>3</sub>) and each combination treatment was repeated four times so that there were 24 experimental units.

The measured growth components were the increase of plant height, the number of new emerging leaves, branches and shoots per plant, the length of new emerging shoots, the leaf area, and the number of flowers. The data obtained were tested with analysis of variance at the level of 5% and then continued by Least Significance Difference test at 5%.

## Results and Discussion

**Plant Height.** The pinching treatment had a significant effect on the height of 'Crystal' guava plant. Meanwhile, the treatment of DBC and its interaction with pinching had no significant effect on the plant height at 11 weeks after treatment (WAT).

The mean of beginning plant height was 71 cm, while the mean of final plant height was 75 cm. The tallest plants observed at 11 WAT was the treatment without pinching (P<sub>2</sub>) for about 79 cm, while the shortest plant was observed in the pinching (P<sub>1</sub>) for about 69 cm (Figure 1).



**Figure 1.** The 'Crystal' guava plant height at beginning and final observation on 11 weeks after treatment (WAT).

Note: DBC - Dormancy breaking chemical, P<sub>1</sub> - pinching, P<sub>2</sub> - No pinching, M<sub>1</sub> - No DBC, M<sub>2</sub> - KNO<sub>3</sub> 40 g L<sup>-1</sup>, M<sub>3</sub> - BAP 100 ppm.

**Table 1.** The effect of pinching and dormancy breaking chemicals on the increase of plant height from beginning to 11 WAT

Treatment	The increase of plant height (cm)
Pinching	
Pinching (P <sub>1</sub> )	3.05 b
No Pinching (P <sub>2</sub> )	3.96 a
Dormancy breaking chemical (DBC)	
No DBC (M <sub>1</sub> )	3.69 a
KNO <sub>3</sub> 40 g L <sup>-1</sup> (M <sub>2</sub> )	3.35 a
BAP 100 ppm (M <sub>3</sub> )	3.48 a

Note: the mean followed by difference alphabet is significantly different among the same factor based on the Least Significant Difference test at 5%.

The result of present experiment revealed the reduction of seedling height at the beginning observation period due to the apical removal in pinched plant rather than control. Afterward, the seedling experienced an increase of height, during 3 WAT up to 11 WAT. However, the increase of height in pinched seedling was significantly lower than those in no pinching seedling, i.e., 3.05 cm compared to 3.96 cm, respectively (Table 1). The lower increase in pinched plant, rather than control, could be associated with the removal of apical buds which could grow vigorous upward leading to the increase of plant height. Meanwhile, the average of the increase of plant height in treatments without DBC, with KNO<sub>3</sub> 40 g L<sup>-1</sup> and with BAP 100 ppm were 3.69, 3.35, and 3.48 cm, respectively (Table 1).

**Number of Leaves.** The interaction of pinching and DBC had a significant effect on the number of new emerging leaves at 11 WAT. In the combination of pinching (P<sub>1</sub>) and KNO<sub>3</sub> (M<sub>2</sub>), the guava could produce more leaves, thus make this treatment as the best treatment to increase leaves number compared to others. This best combination treatment of P<sub>1</sub>M<sub>2</sub> had 6 leaves more than the control or P<sub>2</sub>M<sub>1</sub> (Table 2).

**Table 2.** The effect of pinching and dormancy breaking chemicals on the number of new emerging leaves at 11 WAT

DBC	Pinching (P)	
	P <sub>1</sub>	P <sub>2</sub>
M <sub>1</sub>	45.5 A a	53.0 A a
M <sub>2</sub>	59.75 A a	36.25 B b
M <sub>3</sub>	45.5 A a	36.5 A b

Note: The mean followed by difference uppercase and lowercase alphabet is significantly different within the same line and the same column based on the Least Significant Difference test at 5%, respectively. DBC – Dormancy breaking chemical, P<sub>1</sub> – pinching, P<sub>2</sub> – No pinching, M<sub>1</sub> – No DBC, M<sub>2</sub> – KNO<sub>3</sub> 40 g L<sup>-1</sup>, M<sub>3</sub> – BAP 100 ppm.

**Number of Branches.** The data analysis showed that there was a significant interaction effect between pinching and DBC on the increase of branches number. It was known that the highest result of the number of branches was found in the P<sub>1</sub>M<sub>3</sub>, i.e., 6.25 branches, while the lowest result was found in the control treatment or P<sub>2</sub>M<sub>1</sub>, i.e., 3.75 branches (Table 3).

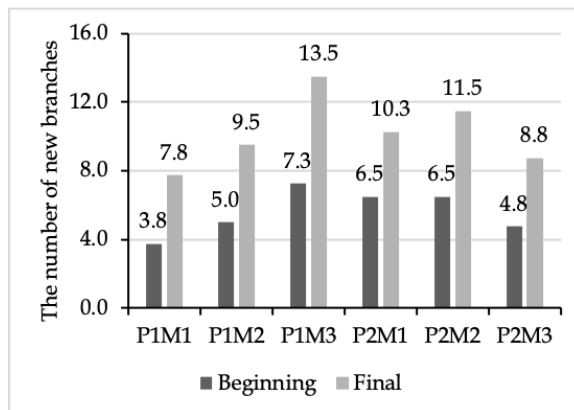
It was likely that the addition of BAP for about 100 ppm resulted in the largest increase in the number of branches. In pinched plants (P<sub>1</sub>), the administration of BAP (M<sub>3</sub>) could result in the largest increase in the number of branches. In plants that were not treated with pinching (P<sub>2</sub>), the administration of BAP (M<sub>3</sub>) and KNO<sub>3</sub> (M<sub>2</sub>) could improve branching response by producing a greater increase compared to no DBC.

The average of the branches number at the beginning varies from 3.8-7.3 branches, while the average of branches number at the end of the observation (11 WAT) varied from 7.8-13.5 branches (Figure 2). Although it had variations in terms of branches number, as a result of the pinching and DHC, broadly speaking the increase of branches number was about 45% over 11 observation weeks.

**Table 3. The effect of pinching and dormancy breaking chemicals on the increase of branches number from beginning to 11 WAT**

DBC	Pinching (P)	
	P <sub>1</sub>	P <sub>2</sub>
M <sub>1</sub>	4.00 A	3.75 A
	B	B
M <sub>2</sub>	4.5 A	5 A
	B	a
M <sub>3</sub>	6.25 B	4 A
	a	b

Note: The mean followed by difference uppercase and lowercase alphabet is significantly different within the same line and the same column based on the Least Significant Difference test at 5%, respectively. DBC – Dormancy breaking chemical, P<sub>1</sub> – pinching, P<sub>2</sub> – No pinching, M<sub>1</sub> – No DBC, M<sub>2</sub> – KNO<sub>3</sub> 40 g L<sup>-1</sup>, M<sub>3</sub> – BAP 100 ppm.



**Figure 2. The difference of branches number at beginning and final observation on 11 weeks after treatment.**

Note: P<sub>1</sub> – pinching, P<sub>2</sub> – No pinching, M<sub>1</sub> – No DBC, M<sub>2</sub> – KNO<sub>3</sub> 40 g L<sup>-1</sup>, M<sub>3</sub> – BAP 100 ppm

**Number of New Emerging Shoots.** The number of new shoots was also affected by the interaction between the pinching and DBC. According to Table 4, the pinching, with or without DBC application, results in insignificantly different number of new shoots.

Meanwhile, without pinching treatment, the number of new shoots seemed to improve by the application of KNO<sub>3</sub>. It was proof that KNO<sub>3</sub> as a DBC could improve the plant budding response. It also confirmed that the pinching treatment could significantly increase the number of new shoots of 'Crystal' guava (Table 5). At 11 WAT, pinching plant experienced an increase in the number of new shoots as much as 17% compared to plants without pinching.

**Table 4. The effect of pinching and dormancy breaking chemicals on the number of new emerging shoots at 11 WAT**

DBC	Pinching (P)	
	P <sub>1</sub>	P <sub>2</sub>
M <sub>1</sub>	16.25 A	13.5 A
	a	b
M <sub>2</sub>	16.5 A	17.25 A
	a	a
M <sub>3</sub>	19.5 A	14 B
	a	b

Note: The mean followed by difference uppercase and lowercase alphabet is significantly different within the same line and the same column based on the Least Significant Difference test at 5%, respectively. DBC – Dormancy breaking chemical, P<sub>1</sub> – pinching, P<sub>2</sub> – No pinching, M<sub>1</sub> – No DBC, M<sub>2</sub> – KNO<sub>3</sub> 40 g L<sup>-1</sup>, M<sub>3</sub> – BAP 100 ppm.

**Table 5. The effect of pinching on the number of new emerging shoots at 11 WAT**

Pinching	Mean
P <sub>1</sub>	17.42 a
P <sub>2</sub>	14.92 b

Note: The mean followed by difference lowercase alphabet is significantly different within the same column based on the Least Significant Difference test at 5%. P<sub>1</sub> - pinching, P<sub>2</sub> - No pinching.

**The length of the new shoot.** In addition to the number of new shoots, the length of new shoots was also influenced by the interaction between pinching and DBC. The longest shoot was observed in guava plants treated with pinching and KNO<sub>3</sub>, by producing 83% longer shoot than control (Table 6). On average, the number of new shoots of pruned plants was 36% greater than without pinching. The average of shoots length in plant with pinching treatment was 12.07 cm, whereas the one without pinching was only 8.85 cm.

**Table 6. The effect of pinching and dormancy breaking chemicals on the length of new emerging shoots at 11 WAT**

DBC	Pinching (P)	
	P <sub>1</sub>	P <sub>2</sub>
M <sub>1</sub>	13.17 A a	7.23 B b
M <sub>2</sub>	13.24 A a	7.03 B a
M <sub>3</sub>	9.8 A b	12.29 A a

Note: The mean followed by difference uppercase and lowercase alphabet is significantly different within the same line and the same column based on the Least Significant Difference test at 5%, respectively. DBC - Dormancy breaking chemical, P<sub>1</sub> - pinching, P<sub>2</sub> - No pinching, M<sub>1</sub> - No DBC, M<sub>2</sub> - KNO<sub>3</sub> 40 g L<sup>-1</sup>, M<sub>3</sub> - BAP 100 ppm.

**Table 7. The effect of pinching and dormancy breaking chemicals on the leaf area**

Treatment	Leaf area (cm <sup>2</sup> )
P <sub>1</sub> M <sub>1</sub>	35.91 a
P <sub>1</sub> M <sub>2</sub>	31.99 a
P <sub>1</sub> M <sub>3</sub>	30.30 a
P <sub>2</sub> M <sub>1</sub>	30.76 a
P <sub>2</sub> M <sub>2</sub>	35.76 a
P <sub>2</sub> M <sub>3</sub>	33.74 a

Note: The mean followed by difference lowercase alphabet is significantly different within the same column based on the Least Significant Difference test at 5%. P<sub>1</sub> - pinching, P<sub>2</sub> - No pinching, M<sub>1</sub> - No DBC, M<sub>2</sub> - KNO<sub>3</sub> 40 g L<sup>-1</sup>, M<sub>3</sub> - BAP 100 ppm.

**Leaf Area.** The pinching, DBC and the interaction of both factors had no significant effect on the leaf area of the 'Crystal' guava plant. The average leaf area at 11 MST was 33.08 cm<sup>2</sup>.

**Number of Flowers.** This study also presented data on the number of flowers that appeared from the 5<sup>th</sup> to 11<sup>th</sup> WAT, although no further tests were carried out on this data. Flowers were mostly found in the non-pinching treatment given by DBC. The flower produced by non-pinching combined with DBC in form of KNO<sub>3</sub> 40 g L<sup>-1</sup> and BAP 100 ppm was higher than pinching plant, with and without DBC (Table 8).

**Table 8. The effect of pinching and dormancy breaking chemicals on the number of flowers from 5 to 11 WAT**

Treat ment	Weekly observation						
	5	6	7	8	9	10	11
P <sub>1</sub> M <sub>1</sub>	0	0	1	1	1.5	1.75	1.75
P <sub>1</sub> M <sub>2</sub>	0	0	1.25	1.5	2.5	2.5	3
P <sub>1</sub> M <sub>3</sub>	0.25	0.25	0.75	1.25	1.25	1.5	1.5
P <sub>2</sub> M <sub>1</sub>	0.5	0.75	1	1.25	1.25	1.25	1.25
P <sub>2</sub> M <sub>2</sub>	0.75	0.75	2.25	3.5	5.25	6.75	6.75
P <sub>2</sub> M <sub>3</sub>	0	0.25	1.75	2.5	3.75	4.75	6

Note: P<sub>1</sub> - pinching, P<sub>2</sub> - No pinching, M<sub>1</sub> - No DBC, M<sub>2</sub> - KNO<sub>3</sub> 40 g L<sup>-1</sup>, M<sub>3</sub> - BAP 100 ppm.

**Discussion.** Pinching is defined as the light pruning by hand on the certain apical bud. Plants without pinching favor the dominant growth of apical shoots, as evidence of the occurrence of apical dominance phenomenon. The dominant growth in the vertical direction caused a significant increase in the height of plant in control or no pinching treatment.

The applied pinching in guava seedling generally aimed to decline apical dominance, in similar to earlier studies result (Budiarto, 2018). The high production of auxin hormone in apical shoots that were actively dividing caused a high translocation of auxin to prospective lateral buds, resulting in high concentrations of auxin in that area. At such high concentrations, auxin actually made dormant lateral bud. The cutting of apical buds caused a decrease in the supply of auxins so that it gradually decreased the concentration of auxins in the lateral meristem, which induced the breakdown of dormancy and the growth of lateral shoots.

The absence of apical dominance resulted more lateral bud grow and subsequently formed wider canopy. The wider canopy in guava

seedling was important for increasing source capacity of plant prior to shift to generative stage. The success of pinching to create more branches and new emerging shoot was also proved in present experiment. Apart from the pinching treatment, the administration of DBC, such as  $\text{KNO}_3$  could also improve the branching response in plants that are not pinched.

Even in the pinching treatment, the combination with  $\text{KNO}_3$  could increase the number of leaves and the length of new shoots compared to only the  $\text{KNO}_3$  treatment. The results of present study provided an additional knowledge that the  $\text{KNO}_3$  treatment still need to be supplemented with other practical culture treatments to improve growth performance, such as pinching.

The provision of DBC in the form of  $\text{KNO}_3$  and BAP was proven to induce flowering, if not accompanied by pinching techniques. Flowering in the early stages of 'Crystal' guava seedlings was a detrimental thing. At the beginning of the planting period, guava plant seedling should be directed towards vegetative growth to form massive leaves, so that the canopy increases in size and area. A large canopy could support a high number of harvested fruits, and vice versa.

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## Conclusion

The pinching could improve the growth performances of guava seedling. The combination of pinching with DBC, both ( $M_2$ )  $\text{KNO}_3$  40 g  $\text{L}^{-1}$ , and ( $M_3$ ) BAP 100 ppm, could increase the number of new emerging leaves, branches, and shoots as well as the length of new shoots. The application of DBC to non-pinched plants inhibited vegetative growth and actually increased the number of flowers production.

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