Growth of forest clove seedlings at different concentrations of paclobutrazol

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Abstract. The objective of the study is to reach shorter seeds of forest cloves through the application of growth inhibitors, namely paclobutrazol. The study used a randomized block design (RBD) one factor with three replications. The experimental factor was 5 concentrations of paclobutrazol which were 0 ppm (control), 25 ppm, 50 ppm, 75 ppm, 100 ppm. The results revealed that the concentration of paclobutrazol 75-100 ppm produces shorter forest clove seedlings with higher leaf chlorophyll content.

1. Introduction
Clove is native to Maluku islands and included in the family Myrtaceae [1,2]. In Maluku, there are one species of a wild type which the local community calls forest cloves [3]. Forest cloves in Maluku have been widely cultivated by people in their distribution areas, especially in the Hitulama and Hitumesing on Ambon island and in the Latu and Hualoi on Seram Island [4,5]. The characteristics of Forest Clove is rapid growth so that they can reach a height of 20 meters, especially when they are over 20 years [6,7]. The high tree causes farmers difficult to harvest the flowers. To overcome these problems, some forest clove farmers in Maluku cut some of the canopies at harvest time. This pruning causes canopy damage, which will reduce flower production in the next harvest season.

To overcome these problems, it needs research to produce shorter forest clove seedlings. One solution is to use growth inhibitors such as paclobutrazol. Paclobutrazol is a growth regulator from the retardant group. Paclobutrazol can suppress vegetative growth and increase leaf chlorophyll content without causing abnormal growth [8,9]. Paclobutrazol works by inhibiting the biosynthesis of gibberellins thus causing inhibition of cell elongation in the subapical meristem. When gibberellin production is inhibited, cell division still occurs but new cells do not extend [10]. The use of paclobutrazol to produce shorter clove seedlings has been reported in Zanzibar cloves [11], but it has not been studied at forest clove. Therefore, this study aims to obtain the best concentration of paclobutrazol to produce shorter forest clove seedlings.
2. Methods
The study used three months old forest clove seedlings and grown for three months in the nursery. The study used a randomized block design (RBD) one factor with three replications. The experimental factors consisted of five concentrations of paclobutrazol, namely 0 ppm (control), 25 ppm, 50 ppm, 75 ppm, 100 ppm. Observation studies are seedling height, the number of leaves, stem diameter and leaf chlorophyll content. Measurement of seedling height using a measuring bar, stem diameter using a calliper, and chlorophyll content using SPAD-502 plus chlorophyll meter which is converted using the equation \( y = 0.0007x - 0.005 \), where \( y \) = chlorophyll content and \( x \) = the result of SPAD-502 plus chlorophyll measurement meter [12].

The data were analyzed using analysis of variance (F test) at the \( \alpha \) level of 5%. If the treatment has a significant effect then it is followed by the Duncan Multiple Range Test (DMRT) [13]. To see the response pattern of the variable to the level of paclobutrazol, orthogonal polynomials are used. Overall analysis performed using STAR 2.0.1 software.

3. Results and Discussion

3.1. Seedling growth
The performance of paclobutrazol concentration had a significant effect on seedling height, the number of leaves, and stem diameter of forest clove seedlings at the end of the observation (table 1). The pattern of the response of seedling height, number of leaves, and the diameter of seedlings at various concentrations of paclobutrazol follows a linear regression pattern that tends to decrease (figure 1).

Table 1. Seedling height, number of leaves, and stem diameter of forest clove seedlings at various concentrations of paclobutrazol

| Concentration of paclobutrazol (ppm) | Seedling height (cm) | Number of leaves | Stem diameter (mm) |
|--------------------------------------|----------------------|------------------|-------------------|
| 0                                    | 21.11 ± 0.65a        | 7.33 ± 0.58a     | 2.46 ± 0.05a      |
| 25                                   | 19.39 ± 0.69ab       | 6.67 ± 0.46b     | 2.31 ± 0.12b      |
| 50                                   | 18.59 ± 0.39b        | 6.47 ± 0.50bc    | 2.27 ± 0.08b      |
| 75                                   | 18.31 ± 0.36b        | 6.13 ± 0.31cd    | 2.20 ± 0.02bc     |
| 100                                  | 17.39 ± 1.17b        | 5.87 ± 0.31ad    | 2.14 ± 0.05c      |

F test ** ** **
Response pattern L** ** ** **

Note: The numbers followed by the same letters in the same column show no significant difference using the DMRT test at \( \alpha \) of 5%. **: significantly different at \( \alpha \) by 1%.

The results showed that increasing concentrations of paclobutrazol caused seedling height, number of leaves, and stem diameter of seedlings to be lower than controls. The 100-ppm paclobutrazol produces the lowest seedling growth then followed by the concentration below it. The paclobutrazol concentration of 100 ppm produced 17.62% shorter seedling height compared to control and was not different from the paclobutrazol concentration of 25, 50, 75 ppm. The concentration of 100-ppm paclobutrazol produced 19.91% fewer leaf counts than the control and did not differ from the concentration of 75 ppm. The concentration of 100-ppm paclobutrazol also produced a stem diameter of 13.00% lower than the control and did not differ from the concentration of 75 ppm. Controls in the study resulted in higher seedlings, more leaf counts, and higher stem diameters. Inhibition of vegetative growth due to the administration of paclobutrazol in research related to the inhibition in gibberellins biosynthesis in plant tissues. Some research used growth regulator showed best product for seedling [14,15]. Paclobutrazol is a growth-inhibiting agent which inhibits three stages in the terpenoid pathway to produce gibberellins [16]. Paclobutrazol inhibits the biosynthesis of gibberellins.
thereby causing inhibition of cell elongation in the subapical meristem. Inhibition of biosynthesis of gibberellins occurs through inhibition of the oxidation of caurene to Cauenic acid [17]. Gibberellins are phytohormones that stimulate vegetative growth, where if the production of gibberellins is inhibited, the cells continue to divide but the new cells do not elongate.

Regression analysis showed that increasing the dose of paclobutrazol caused a decrease in vegetative growth of forest clove seedlings. The response pattern shows a tendency to decrease plant height, number of leaves and stem diameter along with increasing concentrations of paclobutrazol. The pattern of response of seedling height on the paclobutrazol concentration followed the equation $y = -0.0342x + 20.665$, with $R^2 = 0.7616$; the number of leaves is $y = -0.0139x + 7.1867$, with the value $R^2 = 0.6271$, and the diameter of the stem is $y = -0.003x + 2.4251$, with the value $R^2 = 0.7373$. Inhibition of vegetative growth by increasing the concentration of paclobutrazol related to the work of paclobutrazol in inhibiting the biosynthesis of gibberellins. Previous studies have reported that the application of paclobutrazol can inhibit the biosynthesis of gibberellins and delay the transfer of energy to the mitochondria so that the cell elongation process is inhibited. Inhibition of vegetative growth will result in shorter plants [11,18,19].

3.2. Chlorophyll content
Paclobutrazol concentration has a significant effect on the chlorophyll content of leaves of forest clove seedlings (table 2). The response pattern of leaf chlorophyll content at various concentrations of paclobutrazol follows a linear regression pattern that tends to increase (figure 2).

**Table 2.** Chlorophyll content of leaves of forest clove plant seeds at different concentrations of paclobutrazol

| Concentration of paclobutrazol (ppm) | Chlorophyll (mg cm$^{-2}$) |
|--------------------------------------|-----------------------------|
| 0                                    | 0.0193 ± 0.0005b            |
| 25                                   | 0.0199 ± 0.0006b            |
| 50                                   | 0.0209 ± 0.0005b            |
| 75                                   | 0.0237 ± 0.0008a            |
| 100                                  | 0.0244 ± 0.0008a            |
The results showed an increase in leaf chlorophyll content after applied with paclobutrazol. The concentration of 100-ppm paclobutrazol produces a higher chlorophyll content and is not different from the concentration of 75 ppm. Application of 100 ppm and 75-ppm paclobutrazol concentrations resulted in higher chlorophyll content of 26.42% and 22.79% compared to controls. Paclobutrazol can inhibit the vegetative growth of plants as well as increase the green color of the leaves without causing abnormal growth [8]. Chlorophyll acts as an absorber, which converts solar radiation energy into chemical energy through photosynthesis. Increasing chlorophyll content will increase photosynthesis capacity.

Regression analysis showed that an increase in the concentration of paclobutrazol increased the chlorophyll content of the leaves of the forest clove seedlings. The response pattern of leaf chlorophyll content in the application of paclobutrazol concentration followed the equation $y = 6E^{-05}x + 0.0189$, with a value of $R^2 = 0.87$. Increased chlorophyll content due to the application of paclobutrazol has been widely reported in the Zanzibar clove [20] and oranges fruit [21].

![Figure 2. Response pattern of chlorophyll content of leaves of forest clove seedlings](image)

### 4. Conclusion
Paclobutrazol decreased vegetative growth of forest clove seeds and increased chlorophyll content of leaves compared to controls. The paclobutrazol concentration of 75-100 ppm is the best concentration to produce shorter forest clove seedlings.

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