

## Article

# Plant Growth Analysis of Maize (*Zea mays*): Impact of Second Paclobutrazol Application Timing

Yusril Syafrizal<sup>1</sup>, Gendro Indri Wahyuningsih<sup>2\*</sup>, Fajarani Ulfah<sup>3</sup>, Ananta Bayu Pratama<sup>4</sup>, and Cahyo Adileksana<sup>5</sup>

<sup>1</sup> Agri Services Department, Edufarmers International Foundation; [yusril.syafrizal@edufarmers.org](mailto:yusril.syafrizal@edufarmers.org)

<sup>2</sup> Agri Services Department, Edufarmers International Foundation; [gendro.wahyuningsih@edufarmers.org](mailto:gendro.wahyuningsih@edufarmers.org)

<sup>3</sup> Agri Services Department, Edufarmers International Foundation; [fajarani.ulfah@edufarmers.org](mailto:fajarani.ulfah@edufarmers.org)

<sup>4</sup> Agri Services Department, Edufarmers International Foundation; [ananta.pratama@edufarmers.org](mailto:ananta.pratama@edufarmers.org)

<sup>5</sup> Agri Services Department, Edufarmers International Foundation; [cahyo.adileksana@edufarmers.org](mailto:cahyo.adileksana@edufarmers.org)

\* Correspondence: [gendro.wahyuningsih@edufarmers.org](mailto:gendro.wahyuningsih@edufarmers.org)

**Abstract:** Maize is a vital source of carbohydrates and livestock feed in Indonesia, where domestic demand is rising despite inconsistent productivity. Innovative cultivation methods, such as paclobutrazol (PBZ) application, are being explored to boost maize productivity. PBZ affects maize's growth, with its effectiveness depending on the timing of application. This study was to figure out the optimal timing of PBZ application to improve the synchronization of cob and female flower emergence, thereby maximizing plant growth and yield. The experiment utilized a randomized complete block design (RCBD) comprising five treatments: a control and four intervals for the second PBZ application (3, 6, 9, and 12 days after the first application), each replicated three times. Data were analysed using Analysis of Variance (ANOVA) at a 5% significant threshold, succeeded by Tukey's test. Results showed that the timing of the second PBZ application significantly influenced maize growth and yield. A three-day interval after the first PBZ application produced the highest values in key morphological variables, such as green leaf index and total dry weight. This timing also optimized plant growth metrics, leading to higher maize yield. In conclusion, a three-day interval is recommended for maximizing maize productivity.

**Keywords:** Paclobutrazol; Maize Productivity; Plant Growth Analysis

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## 1. Introduction

Maize serves as the second most significant source of carbohydrates after rice and is considered a strategic commodity due to its role as a high-protein feed for livestock [1]. The domestic demand for maize in Indonesia is projected to increase from year to year; however, national productivity tends to fluctuate and even decreased by 2.73% in 2023 compared to the previous year [2]. This emphasizes the importance of enhancing maize productivity to meet the growing demand. Improving maize cultivation method through innovation or technology becomes a viable option for boosting the yield. For instance, the usage of Plant Growth Regulators (PGRs) which positively affect plant physiology by regulating hormone levels and enhancing stress resistance such as heat, drought, salinity, and flood [3]. Subsequently, this leads to a better maize plant growth and adaptation capability of crop growing under various environmental conditions.

PGRs are often mistaken for phytohormones, which are growth hormones naturally produced by plants to develop essential organs such as roots, stems, leaves, flowers, and fruits. The growth regulators are organic compounds synthesized by agrochemical industry that, at low concentrations, can act as bio-stimulants or bio-inhibitors of plant growth [4]. One of these PGRs, Paclobutrazol (PBZ), influences plant development by

altering the concentrations of endogenous hormones including gibberellin, abscisic acid, and cytokinin [5]. Plants subjected to this growth regulator undergo decreased shoot elongation, increased root growth, increased abscisic acid levels, higher cytokinin production [6]. Moreover, some studies reported that PBZ application enhances leaf greenness, chlorophyll content, leaf area, biomass accumulation, and subsequently improves crop yield in sesame, rice, and soybean crops [7–9]. It has also been reported that PBZ can change phytohormone levels, leading to an earlier flowering stage in horticultural crops [10].

According to studies, paclobutrazol application prevents leaf death and chlorophyll degradation, while promoting root formation in maize [11,12]. This leads to enhanced photosynthetic activity, better nutrient and water uptake, and ultimately augmented grain yield [12]. Our previous study showed that 400 ppm becomes the minimum effective concentration of PBZ needed to promote morphological and yield characters in maize. Additionally, we observed that maize plants treated with PBZ had the potential to produce more than two cobs. However, this was not optimal, as the extra cobs appeared too late and did not fully pollinate, which adversely affected the grain-filling process. Another study indicated that effectiveness of PBZ application varies based on the plant developmental stages; for instance, applying 400 mg L<sup>-1</sup> of PBZ during the early growth stage produced the highest yield [13]. Therefore, we conducted further research to identify the optimal timing for PBZ application in maize, with the goal of better synchronizing the emergence of extra cobs and female flowers for optimal pollination. Consequently, maize plant growth and productivity can be maximized through proper PBZ application timing.

## 2. Materials and Methods

This investigation took place at the experimental field of Edufarmers International Foundation between January 2024 to May 2024. The site was in Candirejo Village, Ngawen District, Klaten Regency, Central Java Province, at an elevation of 176 meters above sea level (masl). The materials used during the research were maize seed (ADV Jago 20 kg.ha<sup>-1</sup>), NPK Phonska (350 kg.ha<sup>-1</sup>), Urea fertilizer (200 kg.ha<sup>-1</sup>), ZA fertilizer (50 kg.ha<sup>-1</sup>), additional fertilizer (10 kg.ha<sup>-1</sup>), and paclobutrazol (960 ml.ha<sup>-1</sup>). The research was carried out using a single factor Randomized Complete Block Design (RCBD) featuring 5 different treatments and 3 replications, resulting in a total of 15 experimental units. The factor investigated was the various interval timings of second paclobutrazol (PBZ) application, categorized as follows: 1) IP0 = spraying interval 0 day; 2) IP3 = spraying interval 3 days; 3) IP6 = spraying interval 6 days; and 4) IP12 = spraying interval 12 days.

Growth and yield observations of the maize crop were performed by both destructive and non-destructive methods, utilizing 3 samples for each treatment at 30, 55, and 98 days after planting (DAP). The observed variables included morphological characteristics: a) green leaf index (measured using SPAD Minolta 502), b) leaf area (estimated using software ImageJ), c) total plant dry weight, and d) crop yield. These data were then calculated to obtain crop growth analysis data, including Leaf Area Index (LAI), Net Assimilation Rate (NAR), Crop Growth Rate (CGR), and Relative Growth Rate (RGR). The calculation refers to the following formula [14]:

$$LAI = \frac{\text{Total leaf area}}{\text{Ground area}} \quad (1)$$

$$NAR = \frac{(W2 - W1)(\text{Log } L2 - \text{Log } L1)}{(T2 - T1)(L2 - L1)} \quad (2)$$

$$CGR = \frac{W2 - W1}{Ga(T2 - T1)} \quad (3)$$

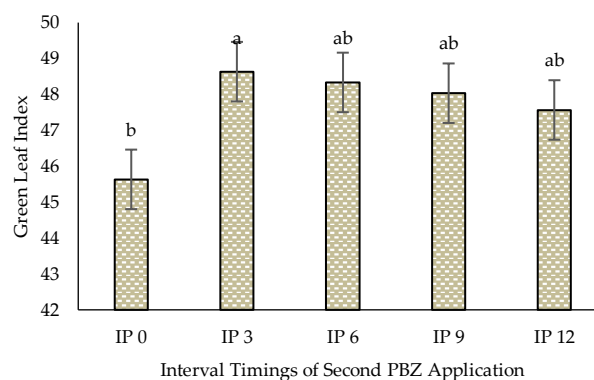
$$RGR = \frac{\ln W2 - \ln W1}{T2 - T1} \quad (4)$$

Where, W1 = total dry weight of plant measured at time T1 (g), W2 = total dry weight of plant measured at time T2 (g), T1 and T2 were the interval of observation time, Ga = ground area (cm<sup>2</sup>), and ln = natural log. NAR, CGR, and RGR are expressed as g/cm<sup>2</sup>/week, g/m<sup>2</sup>/week, and g/week, respectively. Finally, all the collected data were analysed using analysis of variance (ANOVA) at a 5% significance threshold to identify any significant differences, subsequently followed by a Tukey test for further comparison.

The experimental procedures started land sanitation and preparation, followed by seed planting, regular plant maintenance such as fertilization, plant watering, weeding, pest and disease control. Paclobutrazol was then applied according to the treatment intervals. For each experimental unit, a planting hole was created with a spacing of 70 cm x 20 cm, and then one seed was placed in each hole at a depth of 3 to 5 cm. Fertilization was done three times, i.e. 10 DAP using NPK phonska = 100 kg.ha<sup>-1</sup>, ZA = 50 kg.ha<sup>-1</sup>, and Ultradap = 5 kg.ha<sup>-1</sup>; 25 DAP using NPK Phonska = 150 kg.ha<sup>-1</sup> and Urea = 100 kg.ha<sup>-1</sup>; and 44 DAP using NPK phonska = 100 kg.ha<sup>-1</sup>, Urea = 100 kg.ha<sup>-1</sup>, and MKP = 5 kg.ha<sup>-1</sup>. Meanwhile, 600 ppm of paclobutrazol was applied to the plants at specified intervals, with the initial treatment given at 45 days after planting.

### 3. Results and Discussion

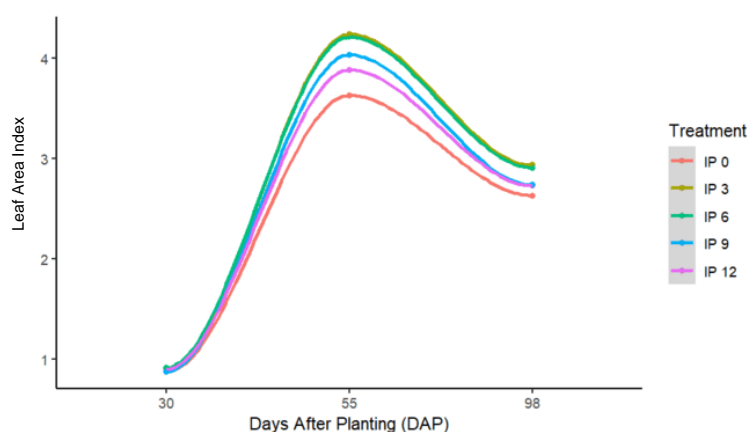
#### 3.1. Green Leaf Index



**Figure 1.** Effect of different interval timings for the second PBZ application on the green leaf index of maize. Bars with distinct letters denote significant differences among treatments ( $P < 0.05$ ). IP0, IP3, IP6, IP9, and IP12 stand for control and spraying intervals of three, six, nine, and twelve days, respectively.

The result of ANOVA analysis indicates that different interval timing application of PBZ significantly influences green leaf index (GLI). Generally, PBZ treated plants had significantly higher value of GLI compared to untreated plants. A previous study suggested that PBZ application could influence the concentration of chlorophyll, the pigment responsible for the green colour of plants [15]. Moreover, PBZ can enhance nitrogen uptake, which is a crucial element in the formation of chlorophyll and the RuBisCo enzyme [16]. Further analysis indicated that 3 days spraying interval treatment significantly increases GLI value by 6.2% compared to control. During this period (45-48 DAP), maize plants are in the late vegetative stage, resulting in a greater rise in chlorophyll concentration relative to other treatments. It assumes that elevated chlorophyll concentration in maize leaves correlates with the late vegetative and early reproductive phases, as the crops attain photosynthetic maturity during this period to facilitate grain filling process [17]. Another study indicated that PBZ exhibits anti-gibberellin activity, which can promote the flowering stage [6]. This process halts vegetative growth in plants and enhances GLI in anticipation of the generative phase. Therefore, PBZ application during key growth stages can optimize both growth regulation and chlorophyll production.

### 3.2. Leaf Area Index



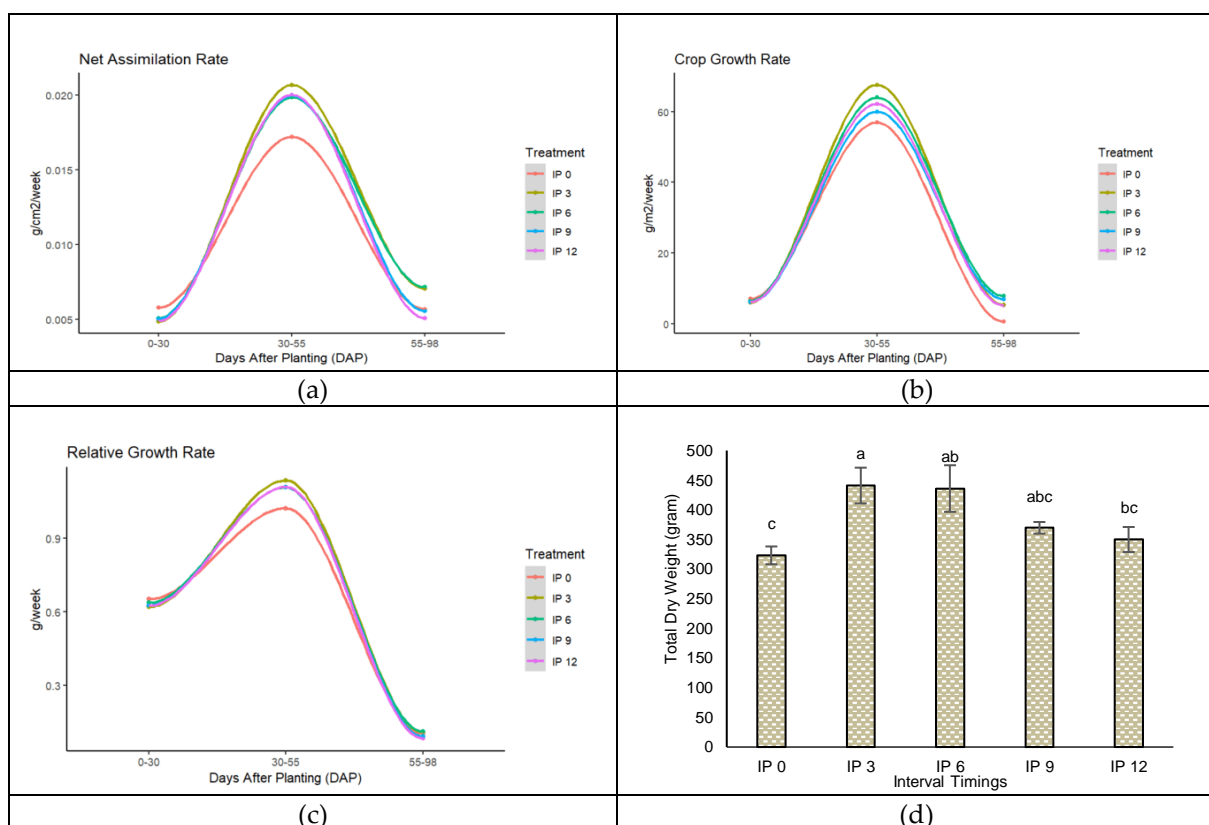
**Figure 2.** Leaf area index of maize influenced by the different interval timings of second PBZ application. IP0, IP3, IP6, IP9, and IP12 stand for control and spraying intervals of three, six, nine, and twelve days, respectively.

Leaf area index (LAI) is closely related to photosynthesis, productivity, and growth rate, serving as a crucial indicator of plant development and influencing overall crop yield and efficiency [18]. The graph below (Figure 2) illustrates the trend of LAI in maize as affected by distinct PBZ application intervals. Generally, the trend shows that after the first PBZ application, maize LAI significantly increased and then decrease as the plants entered the reproductive stage. Moreover, at 55 and 98 DAP, untreated plants exhibited the lowest LAI value compared to the other treatments. A prior study indicated that the application of PBZ enhanced the photosynthetic rate, leading to higher photosynthate production, which promotes plant growth, including leaf expansion [19]. Furthermore, the three-day and six-day interval applications resulted in the highest LAI, likely due to their higher GLI (Figure 1). The increase in LAI is often associated with higher photosynthetic rates [20], and it has been previously noted that this physiological variable can be influenced by leaf greenness. Additionally, photosynthate production during this period (45 – 51 DAP) was still being utilized for vegetative growth, whereas during the nine-day and twelve-day interval treatments, the plants had been in the reproductive stage.

### 3.3. Crop Growth Analysis

Measuring crop growth parameters is crucial to assess plant development under different environmental conditions. In this study, three key parameters were measured: net assimilation rate (NAR), crop growth rate (CGR), and relative growth rate (RGR). NAR signifies the augmentation of dry matter per unit leaf area, demonstrating the plant's ability to assimilate nutrients. CGR shows the augmentation of dry matter per unit area, emphasizing the plant's total production. RGR indicates the plant's ability to increase dry matter over time, which is closely related to its initial weight. Thus, dry matter accumulation is a key variable for analysing plant growth quantitatively [21].

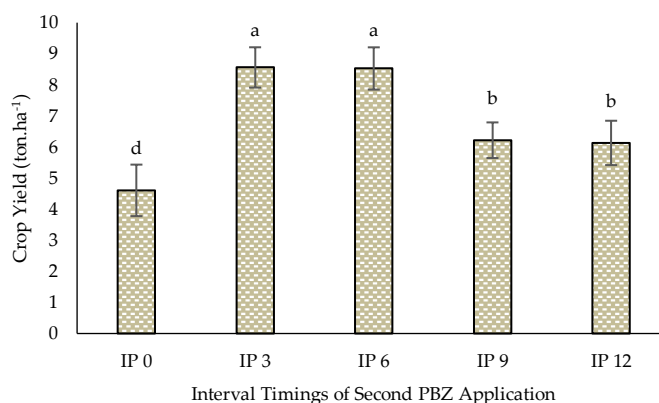
As shown in Figure 3 (D), PBZ application augmented the total dry weight of maize, with the three-day interval for the second application resulting in the most significant increase (26.7%), raising the total dry weight from 323.17 to 440.96 grams. This effect is likely due to the enhanced LAI observed in this treatment, as rapid canopy expansion contributes to greater above-ground biomass accumulation [22]. The results of the crop growth parameters in this study followed a similar trend, with untreated plants showing the lowest NAR, CGR, and RGR, while the three-day interval treatment demonstrated the most significant growth rates relative to other intervals. The increase in NAR observed in plants with a three-day interval of second PBZ application suggests high photosynthetic efficiency, as evidenced by the rise in total plant dry weight. A prior study noted that during the 12-leaf stage, which is three-day interval treatment, maize crop will experience a significant increase in nutrient accumulation and dry weight [23]. Additionally, the highest CGR was observed in the same treatment, aligning with previous findings that NAR is positively correlated with CGR and LAI [23]. Besides the improvements in morphological traits such as the green leaf index and leaf area index, paclobutrazol application during this period also enhances root growth, optimizing water and nutrient absorption critical for plant photosynthesis [11].



**Figure 3.** Net assimilation rate (A), crop growth rate (B), relative growth rate (C), and total dry weight (D) of maize as affected by the different interval timings of second PBZ application. Bars with distinct letters denote significant differences among treatments ( $P < 0.05$ ). IP0, IP3, IP6, IP9, and IP12 stand for control and spraying intervals of three, six, nine, and twelve days, respectively.

### 3.4. Crop Yield

The ANOVA results indicate that different interval timings for the second PBZ application significantly affect maize yield. Overall, the control treatment yielded the least amount (4.6 ton.ha<sup>-1</sup>), while the three-day and six-day intervals resulted in the highest yields (8.56 and 8.53 ton.ha<sup>-1</sup>, respectively). The rise in GLL, LAI, and aboveground biomass may be linked to improved maize productivity. This finding aligns with prior research, which demonstrated that PBZ application in maize increases grain yield by enhancing chlorophyll content, improving the photosynthesis rate, strengthening antioxidant defenses, and delaying leaf senescence [12]. Moreover, LAI reflects the plant's capacity to intercept light, which impacts the photosynthesis rate [22]. This increase in photosynthetic activity leads to higher production of plant assimilates, which are subsequently utilized for grain filling.



**Figure 4.** Effect of different interval timings for the second PBZ application on the crop yield of maize. Bars with distinct letters denote significant differences among treatments ( $P < 0.05$ ). IP0, IP3, IP6, IP9, and IP12 stand for control and spraying intervals of three, six, nine, and twelve days, respectively.

## 4. Conclusions

In conclusion, optimizing paclobutrazol application intervals can significantly enhance maize productivity. The study revealed that different PBZ application timings affect key growth parameters and yield. Notably, three-day and six-day intervals resulted in the highest yields, largely due to improved LAI, which is closely linked to the plant's ability to capture light for photosynthesis. Despite both intervals improving productivity, the three-day interval also demonstrated superior GLL value, total dry weight, and crop growth metrics. Thus, adjusting PBZ application timing to every three days after the initial treatment is recommended to maximize both maize productivity and growth performance.

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