# Growth and Cytokinin Content of Black Rice Plants ( *Oryza sativa* L."Aen Methane") Results Treatment Paclobutrazol and Organic NPK

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#### Abstract

Black rice is one of the plant foods which is less cultivated in Indonesia. Today it is interesting because it contains anthocyanin which is good for health. The aim of this study was to evaluate the effect of packlobutrazol and NPK organic fertilizer to growth, endogenous cytokinin, yield and anthocyanin content in black rice (O. sativa 'Aer Mentan'). This research uses Factorial block design 4 x4. First factor was paclobutrazol with 0 ppm, 12.5 ppm, 25 ppm, 50 ppm concentration. The second factor was fertilizer with dose 0 g, 2 g, 4 g, and 6 g with 4 replication. Statistical analysis used Anova (0.05) and to determine significant difference among treatments performed used DRMT. This research found that paclobutrazol 50 ppm reduced stem black rice, increased diameter, increased panicle length, tiller number. Combination between paclobutrazol 50 ppm 4 gr/pot and 6 gr/pot organic NPK fertilizer 100 weight grain, Combination of paclobutrazol 12.5 ppm and NPK organic fertilizer 6 gr/pot increased root length. Paclobutrazol treatment 25 ppm could increase endogenous cytokinin.

Keywords: paclobutrazol, black rice, NPK organic fertilizer, cytokinin endogenous

#### Introduction

Black rice is a local variety that contains pigments, unlike white rice or other colored rice (Suardi *et al.*, 2009). Black rice contains The high anthocyanin content functions as an antioxidant (Meng *et al.*, 2005), which states that black rice is used as an alternative healthy food in treating diseases. Black rice extract can inhibit the growth of cancer cells and lower cholesterol in blood plasma (Zawistoew Kiest *et al.*, 2005).

Cytokinins are known to play a crucial role in photosynthesis transport, a process linked to vascular strength and vascular activity (Roitsch and Ehneb, 2000). Vascular strength is a combination of vascular activity and vascular size. Cytokinins generally stimulate cell division, so that as more cells are formed, both vascular size and vascular activity increase. Increased vascular activity results in photosynthesis being allocated to the vascular bed. According to Cheng et al. (2014), nitrate increases cytokinin, which in turn regulates panicle development, increasing the number of flowers and branches in rice. Paclobutrazol is a retardant and is often used to inhibit plant growth. The results of research by Tumewu et al. (2007) showed that the application of paclobutrazol to sweet corn plants can reduce plant height. Application of paclobutrazol with a concentration of 1 mgL <sup>-1</sup> on the roots or 25 mgL<sup>-1</sup> on the leaves of tomato plants can suppress stem height growth, increase plant stem thickness, accelerate root growth, contribute to improving seed quality in plants, accelerate fruit formation, and increase fruit yield (Berova and Zlatev, 2004). Compound fertilizer is an artificial fertilizer that contains substances especially hara NPK. The percentage of nutrients in it varies. Based on the

description above, an application evaluation will be carried out. paclobutrazol and NPK fertilizer application Organic granules on growth, cytokinin content and yield of black rice plants Aen Metan

#### **Method Study**

#### Material

Material Which used on study This seed paddy black Which sown in the tray doused with paclobutrazol 12.5, 25,50 ppm, age paddy 14 days later, they were transferred into pots containing garden soil and compost in a 2:1 ratio. After the rice plants were 1, 2, and 3 months old, they were given organic fertilizer of 2 grams/pot, 4 grams/pot, and 6 grams/pot. The material used for the chlorophyll test was 80% acetone. The material used for the cytokinin hormone test was methanol, ethyl acetate, phosphoric acid, acetonitrile, zeatin standard, NaHCO 3. The solution used for total anthocyanin was ethanol and phosphate buffer solution. Material Which used for anatomy is FAA (Formalin: Glacial acetic acid; alcohol), alcohol 70% level, 80, 90%.

#### **Procedure Work**

The soil medium used was the Faculty of Biology's garden soil, black rice seeds were spread on a tray containing garden soil sprinkled with water or paclobutrazol at a concentration of 12.5 ppm, 25 ppm, or 50 ppm. The plastic pots had a diameter of 30 cm. Pots containing planting media were placed in an experimental field at the Faculty of Biology. Granular organic NPK fertilizer was applied when the plants were 1, 2, and 3 months old, according to the recommended dosages of 2 g/pot, 4 g/pot, and 6 g/pot.

#### **Analysis Data**

Data Which obtained analyzed using analysis variance two way ANOVA.

#### Results and Discussion

#### Amount offspring black rice

Paclobutrazol concentration

Table 1. Average amount offspring plant paddy black with various Treatment of paclobutrazol concentration and organic granular NPK fertilizer

|                | Con  | ncentratio | rganic |                |                |                |
|----------------|------|------------|--------|----------------|----------------|----------------|
| verage<br>(ppm | 1)   | 0          | 2      | 4              | 6              |                |
|                | 0    | 0          | 0      | 0              | 2              | 0 <sup>z</sup> |
| Sunday         | 12.5 | 0          | 0      | 1              | 2              | 1 <sup>y</sup> |
| The 3rd        | 25   | 0          | 0      | 2              | 2              | 1 <sup>y</sup> |
|                | 50   | 1          | 2      | 2              | 2              | 2 <sup>x</sup> |
| Average        |      | 0 r        | 0 r    | 1 <sup>q</sup> | 2 <sup>p</sup> | -              |

|         | 0    | 0              | 1              | 1              | 3              | 1 <sup>z</sup>        |
|---------|------|----------------|----------------|----------------|----------------|-----------------------|
| Sunday  | 12.5 | 1              | 1              | 2              | 3              | 2 <sup>y</sup>        |
| 6th     | 25   | 1              | 1              | 3              | 4              | 2 <sup>y</sup>        |
|         | 50   | 2              | 2              | 3              | 3              | 3 <sup>x</sup>        |
| Average |      | 1 <sup>r</sup> | 1 <sup>r</sup> | 2 q            | 3 <sup>p</sup> |                       |
|         | 0    | 2              | 3              | 3              | 3              | 3 <sup>y</sup>        |
| Sunday  | 12.5 | 2              | 3              | 3              | 3              | 3 <sup>y</sup>        |
| 9th     | 25   | 3              | 3              | 4              | 4              | <b>4</b> <sup>x</sup> |
|         | 50   | 4              | 4              | 5              | 4              | 4 <sup>x</sup>        |
| Average |      | 3 q            | 3 <sup>q</sup> | 4 <sup>p</sup> | 4 <sup>p</sup> | -                     |

Description: Different letters in the same column and row indicate significant differences between treatments at a 5% error level (n=4)

Based on Table 1, it was found that the application of 50 ppm paclobutrazol had an effect on the average number of black rice plant tillers at the age of 3 weeks and 6 weeks. Meanwhile, in week 9, there was no significant difference between 25 ppm and 50 ppm paclobutrazol, but it was significantly different from the control. This may be because paclobutrazol can cause changes in the balance. hormone endogen in the plant paddy black, especially gibberellin,

cytokinins Rosita *et et al.*, (2006), Also reported that administration of 250 ppm paclobutrazol increased the number of kencur plant shoots was 30% compared to the control. Granular organic NPK At a dose of 6 grams at 3 and 6 weeks of age, the number of offspring increased. This is related to interactions between hormones and substance Hara on plant. Whereas on week to 9, application of granular organic NPK did not cause any difference in the number of tillers. The results of the study Ciptadi *et al*, (2009), showed that the application of organic fertilizer to rice plants can increase the average number of tillers.

#### Average long panicle plant paddy black.

Table 2. Average long panicle (cm) plant paddy black with various treatments concentration of paclobutrazol and organic granular NPK fertilizer

| Coı | ncentration   | Concentr | ation           |         |        | ferti             | lizer granul o           | organic     |
|-----|---------------|----------|-----------------|---------|--------|-------------------|--------------------------|-------------|
|     | paclobutrazol |          | (gr)            | Average |        | 9                 | _                        |             |
|     | •             |          |                 |         |        |                   | _                        | (ppm)       |
|     | 0             | 2        | 4               | 6       |        |                   |                          |             |
|     |               | 0        | 26.2            | 28.2    | 28.5   | 27.7              | 27.6 <sup>z</sup>        | <del></del> |
|     | 1             | 2.5      | 27.5            | 31.7    | 30.7   | 28                | <b>29.4</b> <sup>x</sup> |             |
|     |               | 25       | 27.2            | 30.5    | 27.5   | 28.5              | 28.4 <sup>y</sup>        |             |
|     |               | 50       | 32              | 29.7    | 28.5   | 28.5              | <b>29.8</b> <sup>x</sup> |             |
|     | Avera         | age      | 28 <sup>q</sup> | 30 p    | 28.8 q | 28.1 <sup>q</sup> | _                        |             |

Description: Different letters in the same column and row indicate significant differences between treatments at a 5% error level (n=4)

Based on Table 2, it was found that paclobutrazol application generally increased the average panicle length of black rice plants. Research (Starman and Wiliam, 2000) reported that the application of 80 mg/L paclobutrazol to *Scaevolaaemula* and *Scaevolaalbida plants* increased the average flower stalk length compared to the control. Application of NPK fertilizer organic The granules

increased the average panicle length at a 2 g dose compared to the control. However, the 4 g and 6 g doses did not result in a significant difference in average panicle length compared to the control. Granular organic NPK fertilizer may increase the endogenous auxin content in black rice plants, thereby inhibiting the expression of the Os IPT4, Os IPT5, Os IPT7 and Os IPT8 genes which play a role, increase average length panicle on black rice (Seon) et al, 2012).

### Average long root plant black rice

**Paclobutrazol concentration** 

Average

12.2

11.2 r

Table 3. Average root length (cm) of black rice plants with various concentration treatments of paclobutrazol and organic granular NPK fertilizer

#### Concentration fertilizer granul organic (gr) Average (ppm) 0 2 4 6 10.7 15 11.2 15.2 13.0 <sup>y</sup> 12.5 12.2 13.2 19.5 11.5 14.1 <sup>x</sup> 25 9.5 15 12.7 xz 13.7 12.7 50

12.2

12.7 q

Information: Letter Which different on column And line Which same shows different real between treatments at an error level of 5% (n=4)

9 14.3 <sup>p</sup> 14.7

13.5 <sup>p</sup>

 $12.0^{z}$ 

Based on Table 3, the root length of the plants that received The 12.5 ppm paclobutrazol treatment had a higher value than the control. Meanwhile, the 25 ppm and 50 ppm paclobutrazol treatments causes shorter roots compared to the control. Application of granular organic NPK fertilizer The doses of 4 gr and 6 gr increased root length the most. In accordance with the statement of Pinto et al. (2005), who also reported that paclobutrazol suppressed stem length by inhibiting the activity of physiological but does not inhibit the production and translocation of assimilates to other plant organs. Administration of paclobutrazol to plants inhibits gibberellin biosynthesis. Gibberellin interacts with auxin in influence physiological plants, gibberellins more push stem elongation, while auxin in plant organs promotes root and shoot formation. This proves that organic NPK fertilizer affects root length. The higher the dose of NPK fertilizer used, the longer the plant roots. In this study, the combination of 12.5 ppm paclobutrazol and granular organic NPK fertilizer at a dose of 6 grams further increased the growth rate. root length in black rice plants compared to control and treatment.

### Average heavy wet 100 seed content

**Paclobutrazol concentration** 

Table 4. Average wet weight of 100 filled seeds (gr) of black rice plants with various treatments concentration of paclobutrazol and organic granular NPK fertilizer

|               | Concenti |     |     |     |                  |
|---------------|----------|-----|-----|-----|------------------|
| Average (ppm) | 0        | 2   | 4   | 6   |                  |
| 0             | 1.8      | 1.8 | 2.3 | 1.9 | 1.9 <sup>x</sup> |

| 12.5    | 2.1              | 2.1   | 2.0              | 2.0              | <b>2.0y</b>      |
|---------|------------------|-------|------------------|------------------|------------------|
| 25      | 2.1              | 2.3   | 2.1              | 2.2              | 2.2 <sup>x</sup> |
| 50      | 2.1              | 1.9   | 2.2              | 2.5              | 2.2 <sup>x</sup> |
| Average | 2.0 <sup>q</sup> | 2.0 q | 2.1 <sup>p</sup> | 2.1 <sup>p</sup> | -                |

Description: Different letters in the same column and row indicate significant differences between treatments at a 5% error level (n=4)

The results presented in table 4, show that paclobutrazol treatment with concentration 12.5, 25 or 50 ppm A little increase average heavy wet

100 filled seeds compared to the control. Paclobutrazol application inhibits gibberellin synthesis but may stimulate cytokinin activity. Cytokinins function in transporting photosynthate products into the pore, which involves transport through the symplast and apoplast (Okura *et al.*, 2004). Cytokinins have been reported to increase sucrose absorption capacity by stimulating the expression of the *OsCINI gene*. This gene increases the activity of the invertase enzyme, which functions to fill rice grains, and also increases the expression of the *Os* SUT gene, which activates the sucrose transporter (Lee and Hing, 2014). Application of organic granulated NPK at a dose of 4 grams or 6 grams slightly increased the weight of 100 grains compared to the control. Application of paclobutrazol treatment at 25 ppm and 50 ppm doses of organic NPK fertilizer 4 gr/pot 6 gr/pot higher than control.

### Heavy dry 100 seeds content

Table 5. Average dry weight of 100 filled seeds (gr) of black rice plants with various treatments concentration of paclobutrazol and organic granular NPK fertilizer

# Paclobutrazol concentration

# Concentration fertilizer granul organic

|                 |                  | (gr)             |       |                  |                  |
|-----------------|------------------|------------------|-------|------------------|------------------|
| verage<br>(ppm) | 0                | 2                | 4     | 6                |                  |
| 0               | 1.6              | 1.6              | 2.1   | 1.7              | 1.7 <sup>z</sup> |
| 12.5            | 1.9              | 2.0              | 1.9   | 1.9              | 1.9 <sup>y</sup> |
| 25              | 2.0              | 2.2              | 2.0   | 2.0              | 2.0 <sup>x</sup> |
| 50              | 1.9              | 2.0              | 2.0   | 2.3              | 2.0 x            |
| Average         | 1.8 <sup>r</sup> | 1.9 <sup>q</sup> | 2.0 p | 1.9 <sup>q</sup> | -                |

Description: Different letters in the same column and row indicate significant differences between treatments at a 5% error level (n=4)

Based on Table 7 Application of paclobutrazol 12.5, 25 or 50 ppm causes The average dry weight of 100 filled seeds was slightly higher than the control. Research by Ducsay and Varga (2003) showed that the application of NPK fertilizer (2:1:5) at a dose of 90.9 kg/ha In Chinese cabbage plants (*Brassicapekinensis* (Lour) Rupr.) can increase biomass production. In soybean plants, if given NPK fertilizer with a dose of 300 kg.ha mixed with KNO 3 150 kg.ha <sup>-1</sup> sprayed on the leaves can increase production in the form of the number of pods per plant, and seeds increase by 3 or 2 seeds per pod. The combination of 25 ppm paclobutrazol treatment and 6 grams of organic fertilizer causes the average weight of dry seeds to increase. slightly higher than other treatments and the control.

#### **Comparison Hormone Cytokinins Endogen**

| No | Sample Code | Zeatin  | Cis Zeatin | Zeatin<br>Riboside |
|----|-------------|---------|------------|--------------------|
| 1  | P0C3        | 0.70368 | 0.76366    | 4.87790            |
| 2  | P1C3        | 1,19362 | 0.97722    | 6,09258            |
| 3  | P2C3        | 1.84774 | 1.38056    | 7.26369            |
| 4  | P3C3        | 1.24283 | 0.77447    | 6,31392            |

Based on table 6, it is known that the endogenous cytokinin content in black rice plants that received paclobutrazol treatment using HPLC results is most high is zeatin riboside, followed zeatin and cis zeatin. Based on Table 8. The ratio of endogenous cytokinins is known 25, 50, and 12.5 ppm increased cytokinin content compared to the control. Cytokinins are compounds derived from adenine, a purine base in nucleic acids. Some natural cytokinins are kinetin and zeatin (Kassem *et al.*, 2011).

The application of NPK fertilizer increases the availability of NPK elements in plants, which will increase plant growth. Nitrogen in Plants act as components of DNA and mRNA which functions in protein synthesis. NPK in plants can increase the synthesis of IAA, cytokinin, and gibberellin hormones, which function to stimulate and regulate vegetative and generative plant growth (Greadly *et al.*, 2007).

#### **Conclusion**

The combination of paclobutrazol and organic NPK can generally increase amount offspring, heavy wet seed content 100, average chlorophyll, Anthocyanin content and leaf epidermis diameter, cortex diameter and thickness of vascular bundles in leaves and root epidermis and root cortex. In a combination study of 50 ppm paclobutrazol and 6g/pot organic NPK applied to black rice plants visible effect that caused Aen Metan black rice.

Research results show that the application of 50 ppm paclobutrazol can reduce the height of black rice stems, increase the thickness of the root cortex, the thickness of the leaf vascular bundles, the number of tillers, and the thickness of the leaf mesophyll. The combination of 50 ppm paclobutrazol and organic NPK fertilizer. dose 4 gr/pot or 6 gr/pot weight 100 seeds, height chlorophyll b and total chlorophyll content, anthocyanin levels, and root epidermis thickness. Application of 12.5 ppm paclobutrazol and 6 g/pot organic NPK increased root length, and 25 ppm paclobutrazol increased cytokinin hormones.

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