

The potential of Central Java local black rice and red rice as drought tolerant cultivars

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Abstract. Black and red rice contain nutritional values not found in white rice. They contain anthocyanins which are beneficial to health. Drought stress can potentially reduce rice yield. So, it is necessary to develop local black and red rice into tolerant cultivars. This study used a completely randomized design (CRD) with cultivar factor (V) and drought stress level (K). Drought stress treatment was carried out when the plants were 35-95 DAP (days after planting) using the modified gravimetric method. Plant destruction was conducted at 7 and 21 DAT (days after treatment) to measure the growth analysis component. The observation of the growth analyses was plant dry weight, leaf area (LA), specific leaf area (SLA), relative growth rate (RGR), and net assimilation rate (NAR). Agronomic characters by observing shoot-root ratio and flowering age. The results showed drought stress decreased plant dry weight, leaf area, and relative growth rate on Sragen black rice. Jelitheng Karanganyar showed high RGR and NAR under drought stress and the fastest flowering age response. Wonogiri black rice gave the growth response by reducing specific leaf area. The highest shoot-root ratio was shown on Wonogiri black rice.

1 Introduction

Rice is a staple food source for more than half of the world's population and more than 90% of Asians, and it is also the main source of income for most of the world's people [1], [2]. Increasing community welfare causes a shift in human's lifestyles. Nowadays, people pay more attention to health, so black and red rice are increasingly in demand. Black and red rice contain high nutritional value and they are good for people with diabetes. These nutritional values are not found in white rice, including amino acids and beneficial phytochemical compounds [3]. Several previous studies, black and red rice have bioactive compounds as antioxidants [4], anti-inflammatory [5], anti-diabetic [6], dan anti-obesity [7]. Anthocyanins are bioactive compounds that play a role in giving rice pigment color [8].

Rice production in Indonesia in 2019 decreased by 7.76% compared to 2018 [9]. The decline in rice production in 2019 was caused by weather factors (drought), crop failure, and the late start of the rice planting period. Climate change that causes drought has the

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potential to threaten rice production. Drought stress with varying intensity affects the rainfed land area of approximately 34 million ha and 8 million ha of upland rice in Asia [10]. According to data from the Statistic of Central Java [11], the area of rice land in Central Java Province in 2018 that was affected by drought was 42,156 ha. Drought also caused about 10 thousand ha of paddy field to collapse. Drought significantly reduces grain yield and vegetative growth, thus threatening food security [12]. Therefore, it is important to increase the availability of rice to maintain food security and meet food needs.

Drought stress up to 30% of field capacity reduces the seed filling period by up to 38% and inhibits assimilate translocation, which can lead to yield reduction [13]. Different mechanisms allow plants to survive and thrive under water-limited conditions, such as maximizing water uptake by a deep and dense root system, reducing water loss by reducing leaf area and stomatal closure, and osmotic adjustment [14]. Efforts to increase plant adaptation to abiotic stress in suboptimal land need to be carried out to increase plant productivity in conditions of water deficit [15], [16]. According to [17], local black and red rice has the potential to be a source of genes that control rice's superior traits, such as tolerance to abiotic stress. Black and red rice, which are rich in nutrients, have the potential to be developed into drought-tolerant cultivars. This study aimed to study the growth response of black rice and red rice under drought stress conditions.

2 Materials and method

The research was conducted from June to December 2021 at IP2TP experimental garden screen house, Depok Village, Kandeman District, Batang, Central Java. Field capacity testing was carried out at the Soil Physics Laboratory, Faculty of Agriculture, Universitas Sebelas Maret. This study used a CRD (Complete Randomized Design) with cultivar and drought stress levels factors. The rice cultivars tested were Wonogiri black rice (V1), Jelitheng Karanganyar (V2), Salatiga black rice (V3), Sragen black rice (V4), Wonogiri red rice (V5), and Karanganyar red rice (V6). Drought stress by watering 100% field capacity (K1 or control), 70% field capacity (K2 or moderate drought), and 40% field capacity (K3 or severe drought). There were 18 treatment combinations with three replications, and each unit contained three plants. The modified gravimetric method used drought stress treatment when the plants were 35-95 DAP (days after planting). Plant destruction was carried out on 18 plants at 7 and 21 DAT (days after treatment) to measure plant dry weight, LA (Leaf Area), SLA (Specific Leaf Area), RGR (Relative Growth Rate), and NAR (Net Assimilation Rate). Calculation of LA, SLA, RGR, and NAR using the formula:

$$LA (m^2) = \frac{Wr}{Wt} \times 0.01 m^2 \quad (1)$$

$$SLA (m^2/g) = \frac{Leaf Area (m^2)}{plant dry weight (g)} \quad (2)$$

$$RGR (g/day) = \frac{\ln(W_2) - \ln(W_1)}{t_2 - t_1} \quad (3)$$

$$NAR (g/m^2/day) = ((W_2 - W_1)/(t_2 - t_1)) ((\ln A_2 - \ln A_1)/(A_2 - A_1)) \quad (4)$$

Where Wr is the weight of the replica leaf paper (g), Wt is the total weight of the paper (g), W_1 and A_1 were the total dry weight and leaf area of the plant at T_1 or 7 DAT, while W_2 and A_2 were the total dry weight of the plant at T_2 or 21 DAT. Observation of shoot-root ratio and flowering age on 54 randomly selected plant samples. The measurements of field capacity conditions to determine the watering volume to be given to plants. Calculation of field capacity using the formula:

$$Field\ Capacity\ (FC) \approx \frac{Ww - Dw}{Dw} \times 100\%$$

(5)

Ww is the wet weight of soil (g), while *Dw* is the dry weight of soil (g).

3 Results and discussion

3.1 Plant Dry Weight

Plant productivity is partly determined by the allocation of assimilation to plant organs. According to [18], drought stress can decrease photosynthesis and dry matter accumulation. Moreover, it can reduce net assimilation rate (NAR). It indicates drought stress causes stomatal closure. Drought stress affects the development of root cells, which may interfere with nutrient absorption and photosynthesis [19]. It is related to biomass accumulation; therefore, on shoot and root will also be reduced. Table 1 showed a decrease in plant dry weight due to increased drought stress in Sragen black rice. Despite drought stress, Wonogiri black rice and Jelitheng Karanganyar had increased plant dry weight at 21 DAT. At 7 DAT, the highest plant dry weight (19.80 g) was in Wonogiri black rice with 70% FC, while at 21 DAT, it was shown by Jelitheng Karanganyar with 40% FC (37.40 g).

Table 1. Plant dry weight of local black and red rice cultivars at various levels of drought stress (g)

| Cultivars | 7 DAT | | | 21 DAT | | |
|-----------|---------|--------|--------|---------|--------|--------|
| | 100% FC | 70% FC | 40% FC | 100% FC | 70% FC | 40% FC |
| V1 | 4.30 | 19.80 | 7.50 | 4.70 | 11.60 | 26.40 |
| V2 | 12.40 | 1.50 | 5.00 | 25.80 | 36.70 | 37.40 |
| V3 | 14.30 | 1.50 | 12.70 | 28.00 | 16.70 | 21.50 |
| V4 | 4.70 | 2.40 | 0.60 | 25.80 | 14.30 | 7.20 |
| V5 | 15.00 | 16.10 | 8.90 | 19.00 | 16.50 | 21.00 |
| V6 | 3.70 | 15.00 | 14.60 | 27.90 | 33.00 | 19.80 |

Note: V1= Wonogiri black rice, V2= Jelitheng Karanganyar, V3= Salatiga black rice, V4= Sragen black rice, Wonogiri red rice= V5, and V6= Karanganyar red rice.

3.2 Leaf Area

Rice's vegetative growth is a determinant of the production's success. Leaf area is one factor that affects photosynthesis's production of biomass. Leaves will reduce cell expansion, decrease cell division, apical death, and leaf curl so that leaf area decreases significantly during drought stress [18]. Leaves play an essential role in the drought tolerance of plants.

Table 2 showed a decrease in leaf area starting from 70% drought stress on Jelitheng Karanganyar, Salatiga black rice, Sragen black rice by 44.23%, 55.55% and 27.08% at 7 DAT. However, in Wonogiri black rice, Wonogiri red rice, and Karanganyar red rice, there were an increase in leaf area starting at 70% FC. The leaf area of Sragen black rice cultivar at 21 DAT decreased with increasing drought stress conditions in field capacity. The highest decrease in the leaf area at 21 DAT occurred in Sragen black rice with 40% FC drought treatment of 41.97%. Morphological acclimatization to drought in this study differed for each cultivar, namely the reduction and increase in leaf area. The decrease of leaf area is one of the mechanisms to avoid a deficit. So, the water loss during transpiration

occurs is less. It is expected to be due to the inhibition of leaf expansion so that cell division will decrease due to the decline of cell turgor pressure [20].

Table 2. Leaf area (m²) of local black and red rice in Central Java under drought stress at 7 and 21 days after treatment.

| Cultivars | 7 DAT | | | 21 DAT | | |
|-----------|---------|--------|--------|---------|--------|--------|
| | 100% FC | 70% FC | 40% FC | 100% FC | 70% FC | 40% FC |
| V1 | 0.07 | 0.16 | 0.12 | 0.04 | 0.06 | 0.10 |
| V2 | 0.13 | 0.07 | 0.08 | 0.14 | 0.19 | 0.19 |
| V3 | 0.12 | 0.06 | 0.22 | 0.17 | 0.11 | 0.15 |
| V4 | 0.12 | 0.09 | 0.10 | 0.14 | 0.10 | 0.06 |
| V5 | 0.15 | 0.25 | 0.16 | 0.11 | 0.19 | 0.16 |
| V6 | 0.13 | 0.18 | 0.17 | 0.13 | 0.16 | 0.19 |

Note: V1= Wonogiri black rice, V2= Jelitheng Karanganyar, V3= Salatiga black rice, V4= Sragen black rice, Wonogiri red rice= V5, and V6= Karanganyar red rice.

3.3 Specific Leaf Area (SLA)

Based on Figure 1, the specific leaf area in all treatments tended to decrease at 21 DAT. The lowest SLA at 7 DAT was shown by Wonogiri black rice with 70% FC watering. It indicates that Wonogiri black rice has a higher sensitivity than other rice cultivars. At 21 DAT, Wonogiri black rice showed the highest SLA decrease, namely 44.77% at 40% FC drought stress. When soil conditions are drier, ion loss generally occurs more rapidly, and ion diffusion to the roots is hampered. According to [21], drought stress also declines specific leaf area (SLA) on *Amaranthus tricolor* at moderate (50% FC) and severe (25% FC) drought stress up to 20.20%.

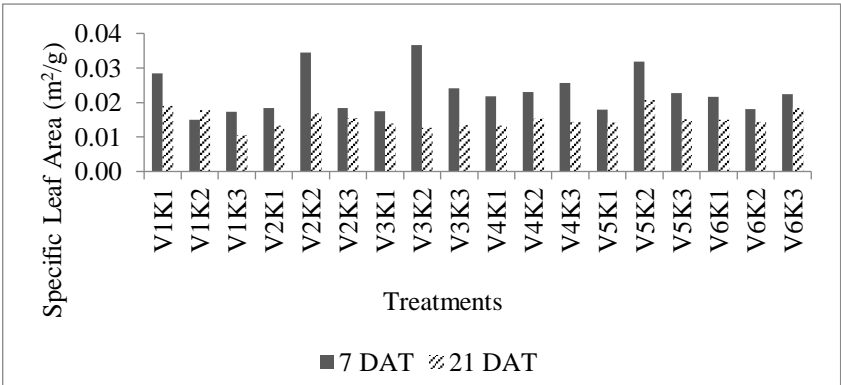


Fig. 1. Specific leaf area of six local Central Java rice cultivars at various levels of drought stress on 7 DAT and 21 DAT (days after treatment). (V1 = Wonogiri black rice, V2 = Jelitheng Karanganyar, V3 = Salatiga black rice, V4 = Sragen black rice, V5 = Wonogiri red rice, V6 = Karanganyar red rice; K1 = 100% FC, K2 = 70% FC, dan K3 = 40% FC).

3.4 Relative Growth Rate (RGR)

Sragen black rice and Karanganyar red rice cultivars experienced a decrease in RGR (relative growth rate) and an increase in drought stress. The most significant reduction in RGR was in Wonogiri black rice with 70% FC drought treatment. The RGR value decreases due to the removal of soil water content during the growth process, so the RGR value decreases. Research by [22] showed a significant decrease in RGR during severe

drought stress compared to RGR values for plants under normal water supply conditions. The observation of RGR at 7 DAT or 42 DAP and 21 DAT or 56 DAP. The Rice plant has reached the maximum vegetative phase, so the accumulation of assimilation was focused on the filling seeds.

Table 3. Relative Growth Rates (g/day) and Net Assimilation Rate ($\mu\text{g}/\text{m}^2/\text{day}$) of local black and red rice cultivars at various levels of drought stress (g/day)

| Cultivars | RGR | | | NAR | | |
|-----------|---------|--------|--------|---------|--------|--------|
| | 100% FC | 70% FC | 40% FC | 100% FC | 70% FC | 40% FC |
| V1 | 0.00 | -0.05 | 0.07 | 0.25 | -10.41 | 14.38 |
| V2 | 0.04 | 0.19 | 0.12 | 8.92 | 25.96 | 22.31 |
| V3 | 0.04 | 0.15 | 0.03 | 9.23 | 20.13 | 4.19 |
| V4 | 0.09 | 0.09 | 0.07 | 14.35 | 11.15 | 6.63 |
| V5 | 0.01 | 0.01 | 0.05 | 1.61 | 0.63 | 6.86 |
| V6 | 0.10 | 0.04 | 0.02 | 15.08 | 8.21 | 3.31 |

Note: V1= Wonogiri black rice, V2= Jelitheng Karanganyar, V3= Salatiga black rice, V4= Sragen black rice, Wonogiri red rice= V5, and V6= Karanganyar red rice.

3.5 Net Assimilation Rate (NAR)

Based on Table 3, cultivars V4 and V6 experienced a decrease in NAR as the soil field capacity decreased. In line with [23], NAR of all soybean varieties tested at 80, 60, and 40% moisture content decreased gradually with age. The decrease in NAR value is thought to be because the water demand decreases as the age of the plant increases. It causes the decline of photosynthesis. According to [24], drought stress during the flowering and pod filling phases significantly decreased NAR. When plants enter the reproductive stage, plant growth will be active and abiotic stress can cause a downward trend in NAR. Jelitheng Karanganyar showed the highest NAR value at 70% FC (25.96 $\text{g}/\text{m}^2/\text{day}$) and 40% FC (22.31 $\text{g}/\text{m}^2/\text{day}$). It means that Jelitheng Karanganyar has a low-stress sensitivity under drought conditions.

3.6 Shoot-root ratio

The root system size affects the absorption of water and minerals from the soil and is related to the size of the plant part above the soil surface [25]. The statistical tests showed that rice cultivars significantly affected the shoot-root ratio. However, stress treatment and the interaction between cultivars and drought treatment did not affect the shoot-root ratio. It showed that the allocation of organic matter between shoots and roots is almost the same. Table 4 showed a significant difference in the root crown ratio of Wonogiri black rice (16.19) and Karanganyar red rice (7.40). According to Fenta et al. [26], drought resulted in changes in biomass partitioning between root and shoot by decreasing the shoot-root ratio

Table 4. Shoot-root ratio of Central Java local black and red rice cultivars under drought stress

| Cultivars | Shoot-root ratio |
|-----------------------|------------------|
| Wonogiri black rice | 16.19 a |
| Salatiga black rice | 14.30 ab |
| Sragen black rice | 12.16 ab |
| Jelitheng Karanganyar | 11.80 ab |
| Wonogiri red rice | 9.31 ab |
| Karanganyar red rice | 7.40 b |

Note: Numbers followed by the same letter in the column and the same treatment indicate that there is no significant difference between treatments in the HSD test at a 95% confidence level

3.7 Flowering Age

Flowering age is one of the effective selection criteria for obtaining drought-tolerant plants (Table 5). The analysis of variance showed that the cultivar had a significant effect on the flowering age. Jelitheng Karanganyar shown a faster flowering period (64.89 DAP) than other local rice cultivars, but it was not significantly different from Wonogiri red rice (66.22 DAP) and Karanganyar (66.78 DAP). Although the drought stress treatment did not show a significant response, the 40% FC drought stress inhibited the flowering time of 76.01 days after planting. Karanganyar red rice accelerated flowering age at 40% FC drought stress, but the same stress treatment delayed the flowering of Salatiga black rice to 94.67 days after planting. According to Alghabari dan Ihsan [27], genotypes with early flowering when experiencing drought stress will produce a higher yield due to a more extended photosynthetic period. Monggesang et al. [28] suggested that drought during the initiation of flowering will affect the reproductive system, so flower sterility increases and triggers flowering.

Table 5. The flowering age of Central Java local black and red rice cultivars under drought stress

| Cultivars | Drought stress | | | Mean |
|-----------------------|----------------|--------|--------|---------|
| | 100% FC | 70% FC | 40% FC | |
| Wonogiri black rice | 85.00 | 90.30 | 88.67 | 88.00 a |
| Jelitheng Karanganyar | 64.00 | 66.00 | 64.67 | 64.89 b |
| Salatiga black rice | 75.00 | 77.30 | 94.67 | 82.33 a |
| Sragen black rice | 85.00 | 83.67 | 78.00 | 82.22 a |
| Wonogiri red rice | 65.00 | 67.30 | 66.30 | 66.22 b |
| Karanganyar red rice | 69.00 | 67.30 | 64.00 | 66.78 b |

Note: Numbers followed by the same letter in the column and the same treatment indicate that there is no significant difference between treatments in the HSD test at a 95% confidence level

4 Conclusions

Based on the study results, Sragen black rice decreased plant dry weight, leaf area, and relative growth rate as drought stress increased. Jelitheng Karanganyar showed a high relative growth rate, net assimilation rate, and the fastest flowering age response during drought. Wonogiri black rice gave a growth response by reducing specific leaf areas during drought. The highest shoot-root ratio on Wonogiri black rice. Each cultivar gives different growth responses to different stress levels.

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