The Phenotype performance of M3 red rice mutant

(Oryza sativa L.)

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Abstract. Local rice genotype generally has colour, flavour and scent more preferred by
consumers, yet unfortunately it has long-lived planting period and low production. Therefore,
the plant breeding practices in rice needs to be implemented for better rice varieties which
are superior in terms of both quality and quantity. Our findings describe the growth character
performance and the production of red rice mutant from M3 generation. This study was
conducted in the Agriculture Faculty wetlands, Hasanuddin University, Makassar, by using
ANOVA test with some red rice mutant genotypes i.e. 7 genotypes mutants (G1, G2, G3, G4,
G5, G6 and G7) and controls/parent-plants (not the mutant). Results show that there were
difference in growth performance and production of red rice mutant. Each parameter observed
on each genotype had different results. Mutants produced best response in tillers production were
G4 mutant with the tillers grain weight at 99.2 g, whereas by the results of the analysis of rank,
mutants showed the best overall response were found in G6 mutants.

1. Introduction

Local red rice is more favored by the community because it has advantages such as having a delicious
taste of rice, containing vitamins and minerals higher than white rice, but the existence of local rice has
been rivaled by new varieties that have potential high yield [1]. Plant breeding is used to improve and
increase the genetic diversity of existing varieties to make it a superior crop in some traits both in quality
and quantity so that the breeder can do the selection according to the desired crop characteristics.
Diversity can be enlarged by several methods, one method that can be used is a mutation technique
commonly known by the name of plant breeding mutations [2].

Mutation is one of the methods of plant breeding that can change the genetic crops randomly and is
inherited. There are two mutation methods : natural mutation and induced mutation, both mutations have
no differences, this is in accordance with Harten's statement [3] which suggests that there is no difference
between the spontaneous mutation and the induced mutation because both can cause genetic variation
to be the basis of plant selection, both natural selection (evolution) and artificial selection (breeding).

Mutation techniques in plant breeding programs will more quickly obtain the desired results when
compared with the conventional techniques. Using a mutation technique, one of the properties of a
variety can be improved without changing the other's properties. If a trait to be repaired is controlled by
a linkage gene or closely bound to other genes then the problem can only be solved by using mutation techniques.

The induction mutation technique is the most effective breeding method for the repair of one or more undesirable properties and capable of producing mutants that have different properties from the parent as well as having wide genetic diversity. The mutation technique is complementary with other techniques so that the technique can be used in conjunction with other techniques such as hybridization and biotechnology [4].

Local rice is essentially long-lived and low-yielding but at present there is research that is able to change the age of rice to be shorter. Indonesian breeders successfully transformed 180-day-old (6-month) rice with production of 2-3 tons ha\(^{-1}\) to be 105 days old with 6-8 tons ha\(^{-1}\) productivity such as red rice varieties Aek Sibundong, a local varieties of North Sumatra released by Indonesian Center for Rice Research in 2006 [5].

The study of Shaleh in 2011 [6] produced the first generation of Suta seed red rice seedlings (M1) from the parent seeds that were irradiated in PATIR-BATAN (Center for Applications of Radioactive Isotopes and Technologies - National Nuclear Energy Agency), from the M1 planting conducted in the Village Karattuang, Bantaeng sub-district, Bantaeng district obtained second generation red rice mutant (M2), then after second generation mutant plantation (M2) conducted in Kera-kera village, Tamalanrea sub-district, Makassar, third generation of the red rice was obtained (M3). Based on the description above, a further research was conducted to study the growth and production performance of the third generation of Sinjai originated red rice genotypes [6].

2. Research Methods
The research was conducted in wetlands, Faculty of Agriculture, Hasanuddin University, Makassar from May to October 2015. The materials used were local red rice seed and red rice mutant seed (M3) from Sinjai, sand, compost, urea fertilizer, SP-36, KCl and pesticide. The tool used in this research is hoe, hand tractor, water pump machine, sickle, machetes, scales, bucket, container for sowing, wire counter, digital camera, hand sprayer, raffia strap, fishing net, fishing net, moisture tester, Skates and stationery.

This study used ANOVA test with several types of red rice mutants. The mathematical model used as follows:

1. Hypothesis H0: \(\mu_1 = \mu_2 = \mu_3\) dan H1 : \(\mu_1 \neq \mu_2 \neq \mu_3\)
2. Statistical F Test (analysis of variances)
3. Level \(\alpha = 0.05\)
4. Areas of criticism : count of F \(\leq\) table of F or count of F > table of F

\[
F = \frac{MS_{between\ Groups}}{MS_{Within\ Groups}}
\]

In this study, local red rice from Sinjai was used, with eight genotypes: control / parent (not mutant), mutant G1, mutant G2, mutant G3, mutant G4, mutant G5, mutant G6, and mutant G7.

3. Result And Discussion

3.1. Plant Height
The results of the average measurements of the plant height of red rice mutant from Sinjai third generation are shown in table 1. Mutant G5 is a genotype that has the highest number of plant height of 142.9 cm and G2 mutant is a genotype that has the average number of the lowest (108.4 cm).
Table 1. Results of analysis of Plant Height (cm) of red rice mutant from Sinjai third generation.

| genotype | genotype 1 (141.2) | genotype 2 (108.4) | genotype 3 (137.8) | genotype 4 (138.9) | genotype 5 (142.9) | genotype 6 (136.9) | genotype 7 (138.7) |
|----------|---------------------|---------------------|---------------------|---------------------|---------------------|---------------------|---------------------|
| genotype 2 | 32.8*               | -29.4*              | -1.0*               | ns                  | -4.0*               | 6.0*                | 28.5*               |
| genotype 3 | 3.4*                | -30.5*              | -5.1*               | 2.0*                | 0.9*                | 2.3*                | ns                  |
| genotype 4 | 2.3*                | -34.5*              | 0.1*                | 6.3*                | ns                  | ns                  | ns                  |
| genotype 5 | -1.7**              | 2.0*                | 4.6*                | 2.0*                | ns                  | ns                  | ns                  |
| genotype 6 | 4.3**               | -0.9**              | 0.2*                | 4.2*                | 1.7**               | ns                  | ns                  |
| genotype 7 | -30.3**             | 0.2*                | 0.2*                | 4.2*                | 1.7**               | ns                  | ns                  |

ns = Not significant

3.2. Productive Tiller

Results of observation of productive tillers of red rice mutants from the third generation are as follows.

Table 2. Results of analysis of productive tillers of red rice mutant from the third generation

| genotype | genotype 1 (37.6) | genotype 2 (21.8) | genotype 3 (16.9) | genotype 4 (25.5) | genotype 5 (21.2) | genotype 6 (15.5) | genotype 7 (20.1) |
|----------|---------------------|---------------------|---------------------|---------------------|---------------------|---------------------|---------------------|
| genotype 2 | 15.8**              | 4.9**               | -8.6**              | ns                  | 4.3*                | 5.7**               | -1.7**             |
| genotype 3 | 20.7**              | 6.3**               | 1.4**               | 10.0**              | ns                  | ns                  | ns                  |
| genotype 4 | 12.1**              | -3.7**              | -3.2**              | 5.4**               | 1.1*                | ns                  | ns                  |
| genotype 5 | 16.4**              | 0.6**               | -4.3**              | ns                  | ns                  | ns                  | ns                  |
| genotype 6 | 22.1**              | 1.7**               | 5.4**               | ns                  | ns                  | ns                  | ns                  |
| genotype 7 | 17.5**              | -3.2**              | 1.1*                | -4.6*               | ns                  | ns                  | ns                  |
| Control   | 15.8**              | 0.0**               | -4.9**              | 3.7**               | -0.6*               | -6.3**              | -1.7**             |

ns= Not significant

Table 2 shows that, mutant genotypes 2-7 (G2, G3, G4, G5, G6 and G7) and Control have different productive tillers with G1 mutants. Mutant G1 is a genotype that has the highest average productive tillers value of 37.6.

3.3. Length of Panicle

The result of observation length of panicle of red rice mutant of third generation that is as follows.

Table 3. The result of analysis length of panicle (cm) of red rice mutants from the third generation

| Genotype | genotype 1 (27.5) | genotype 2 (28.0) | genotype 3 (28.0) | genotype 4 (27.2) | genotype 5 (28.6) | genotype 6 (28.9) | genotype 7 (28.9) |
|----------|---------------------|---------------------|---------------------|---------------------|---------------------|---------------------|---------------------|
| genotype 2 | -0.4**              | 0.0**               | ns                  | ns                  | ns                  | ns                  | ns                  |
| genotype 3 | -0.5**              | 0.7**               | 0.7**               | ns                  | ns                  | ns                  | ns                  |
| genotype 4 | 0.3**               | -1.0**              | -0.6**              | -1.4*               | ns                  | ns                  | ns                  |
| genotype 5 | -1.1**              | -0.7**              | -1.0**              | -1.7**              | -0.3**              | ns                  | ns                  |
| genotype 6 | -1.4**              | -1.0**              | -1.0**              | -1.7**              | -0.3**              | 0.0**               | ns                  |
| genotype 7 | -1.4**              | -1.0**              | -1.0**              | -1.7**              | -0.3**              | 0.0**               | ns                  |
| Control   | 0.6**               | 1.0**               | 1.0**               | 0.3**               | 1.7**               | 2.0**               | 2.0**               |

ns= Not significant

Table 3 shows that, mutant G4 is very different from mutant G6 and G7 mutant. The controls differed significantly (p<0.05) from the mutants G5, G6, and G7. Mutants G6 and G7 are genotype that had the average value of the highest panicle length of 28.9 cm.

3.4. Number of grain per panicle

The result of observation of grain of rice paddy mutant rice product of the third generation that is as follows.
Table 4. Results of the analysis of the number of grains (g) of red rice mutant from the third generation.

| Genotype | genotype 1 (235.6) | genotype 2 (207.4) | genotype 3 (211.3) | genotype 4 (245.7) | genotype 5 (242.3) | genotype 6 (256.0) | genotype 7 (243.9) |
|----------|---------------------|---------------------|---------------------|---------------------|---------------------|---------------------|---------------------|
| genotype 2 | 207.4               | 28.3**              | 17.0**              | 20.4**              | -3.9**              | 17.4**              | 21.7**              |
| genotype 3 | 211.3               | 24.4**              | -38.3**             | -34.4**             | -10.1**             | 3.4**               | -13.8**             |
| genotype 4 | 245.7               | -10.1**             | -34.9**             | -31.0**             | -6.7**              | 1.8**               | 12.2**              |
| genotype 5 | 242.3               | -34.9**             | -44.8**             | -10.4**             | -20.4**             | 1.8**               | ns*                  |
| genotype 6 | 256.0               | -48.7**             | ns*                 | ns*                 | ns*                 | ns*                 | ns*                 |
| genotype 7 | 243.9               | -36.5**             | 1.8**               | 1.8**               | 1.8**               | 12.2**              | ns*                  |
| Control   | 158.9               | 76.7**              | 52.4**              | 86.8**              | 83.4**              | 97.1**              | 85.0**              |

ns= not significant

Table 4 shows that, all mutants (G1, G2, G3, G4, G5, G6 and G7) have an average number of different diamond grains very significant with control. Mutant G6 is a genotype that has an average value of the highest grain that is 256.0 grains.

3.5. Weight of grain per hill

The result of observation of grain weight of red rice mutant of sinjai origin of third generation is as follows.

Table 5. Results of grain weight (g) analysis of red rice mutant from the third generation.

| genotype | genotype 1 (71.4) | genotype 2 (87.4) | genotype 3 (70.3) | genotype 4 (99.3) | genotype 5 (53.6) | genotype 6 (64.2) | genotype 7 (87.4) |
|----------|-------------------|-------------------|-------------------|-------------------|-------------------|-------------------|-------------------|
| genotype 2 | (87.4)           | -15.9**           | 17.0**            | 23.2**            | 35.0**            | -10.6**           | ns*               |
| genotype 3 | (70.3)           | 1.1**             | -11.9**           | -28.9**           | 16.7**            | 45.6**            | ns*               |
| genotype 4 | (99.3)           | -27.8**           | 17.0**            | 33.7**            | 6.1**             | 35.0**            | ns*               |
| genotype 5 | (53.6)           | 17.8**            | -11.9**           | 16.7**            | 45.6**            | -10.6**           | ns*               |
| genotype 6 | (64.2)           | 7.2**             | ns*               | 23.2**            | ns*               | 35.0**            | ns*               |
| genotype 7 | (87.4)           | -15.9**           | ns*               | ns*               | ns*               | ns*               | ns*               |
| control   | (85.3)           | -13.9**           | 2.0**             | ns*               | ns*               | ns*               | ns*               |

ns=not significant

Table 5 shows that, in grain weight of the G4 mutant axis, it is the genotype with highest weights of grain of 99.3 g. Mutant G4 is very different from mutant G3, G5, and G6 but not significantly different from mutant G2, G7, and Control.

3.6. Discussion

The results show that irradiation treatment had an effect on the growth of red rice mutant from Sinjai in the third generation (M3). Based on the results of statistical analysis using two middle values test, the anova table shows that all genotypes have a very significant effect on all observation parameters. Therefore, Least Significant Difference (LSD) advanced test was carried out to see the relation between one genotype with other genotype.

Changes that occur in plants can be caused as a result of the treatment, but can also be caused by external factors or planting environment, so that the involvement of external influences should be minimized as possible, so that the changes in the characters that appear are really changes that result from the radiation treatment given to plants.

The treatment of gamma ray irradiation on rice plants can affect the genotype of rice crops. Changes in plant genotypes can be seen from the measurement differences on each observed character observed. The irradiation carried out leads to an increase or even decrease in the observed character measured quantitatively.

The result of plant height analysis in table 1 is Control and mutant G2 is different genotype very real with other genotype. Mutant G2 is a mutant with the shortest plant height indicating that the mutant’s height is better than that of other mutants. This is in accordance with the statement of Peng et al [7]
which states that the ideal rice plant height is about 90 cm to 100 cm, with the potential height of the loading will decrease compared to the high crop. Although the expected height of the plant has not been achieved yet with an increase in the height of the treated plants shows that there is a change in the high phenotype of plants in rice plants.

Yoshida [8] divided the number of tillers into three categories (slightly (<17), medium (17-24) and many (> 24), Irawan and Purbayanti also suggested that the number of tillers in each clump is generally small, but in some cultivars Medium or large number of tillers. The statement is seen in the research that has been done that the seeds of rice plants decreased and increased compared to control. Table 2 shows that, mutant G1 has more productive tillers than the other genotype of 37.6.

Productive tillers is one component of the results that directly affect the high low grain yield [1]. Increased productivity of rice crops is associated with the number of productive tillers, because the tillers directly produce rice strands producing grain or paddy, this is seen in G4 mutants, G2 mutants, controls, G7 mutants, G3 mutants, and G6 mutants that have number of seedlings and but the statement is not seen in the mutant G1 and mutant G5 which has the most productive tiller but has the lowest production, it can be seen in tables 2 and 5.

The length of panicle increased after irradiation. In table 3, the highest panicle length was found in mutant G6 and mutant G7 that was 28.9 cm, while the lowest panicle length was in Control which was 26.9 cm. According to Australian Government [8] which states that the length of panicle can be categorized based on the size of short panicles (<20 cm), medium panicles (20 cm - 30 cm) and long panicles (> 30 cm). All genotypes are categorized in moderate panicles because of their length ranging from 20 cm - 30 cm. In Table 3 it can be seen that the length of panicles that undergo a change that is mutant G5, mutant G6 and Mutant G7. Changes that occur in the three mutants is the occurrence of panicle length increase and very different with the real control.

Based on table 4 it can be seen that the genotype that the highest average number of grain of probes is the G6 mutant that is 256 and the genotype which has the lowest number of seeds of the panicle is 158.9. This indicates that the average number of grain probes increases in each mutant so that the irradiation treatment has an effect on the number of seeds because it is not the same as the parent.

The weights of the grains of the grains show how much grain is produced in one clump. Based on the mean of grain weight of cultivars, there is an increase and decrease of grain weight in gamma ray irradiation treatment. Based on LSD test in table 5 it can be seen that G4 mutant has the highest mean of grain weight of 99.2 g with an average of 25.5 panicles, whereas mutant G5 has the lowest mean of grain weights of 53.6 g with an average of 21.2 panicles. The occurrence of diversity of weeds of rice grains shows that irradiation causes the effect of mutation on the plant so that the mutant of rice plant of Sinjai originated in the third generation.

The irradiation treatment causes some mutants to increase in diversity and it is expected that the grain weight character of this clump can be inherited to the fourth generation mutant so that further research is needed to know the stability of the character.

Differences occur both in growth and production due to the effects of irradiation so that in the third generation there has been mutants that grow uniformly in each genotype, although there are still some mutants that have not uniform growth such as mutant G1, mutant G3 and mutant G5. The uniformity of each mutant is evident from the height of the plant, the number of tillers, and the time of harvest.

4. Conclusion
Based on the results of research that has been done, it can be concluded that:

- Mutants G1, G2, G3, G4, G5, G6, G7 and control are significantly different based on observation parameters except on the number of book parameters indicating that there is a difference in the growth and production performance of the sinjai red rice mutants in the third generation.
- The mutant that produces the best response based on the cultivar production is the G4 mutant with the weight of the axle of grain 99.2 g while in the rank analysis, the mutant showing the best response is the G6 mutant.
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