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Research Article
Response of M-1 Generation Rice Plants of the Local Aceh Cultivar Arias Putih (*Oryza sativa* L.) to Gamma Irradiation

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Abstract
Background and Objective: Rice plants have advantages and disadvantages in terms of morphological appearance and plant quality. Genetic improvement can be carried out using gamma-ray irradiation. The study aimed to determine the response of the local rice cultivar Arias Putih against administering gamma-ray irradiation. **Materials and Methods:** Arias Putih cultivar upland rice was irradiated with 3 irradiation dose intervals; G1 = 100 Gy, G2 = 200 Gy, G3 = 300 Gy and G0 = as control. Irradiation was carried out at the Isotope and Radiation Technology Application Center-National Nuclear Energy Agency PATIR BATAN, Jakarta. The planting distance used is 30×30 cm. The observed variables included plant height, number of tillers, productive tillers, flowering age and harvest age. Data analysis was conducted using means and Tukey's advanced test (HSD: Honestly significant difference) at a specified significance level (e.g., $p<0.05$). **Results:** The White Arias cultivar that was irradiated with gamma rays experienced changes in the morphology of flag leaf width, number of empty grains and filled grains, age flowering and harvest time. Changes in flowering age characteristics due to irradiation did not show significant differences compared to controls, however, plants experienced an increase in age compared to control plants. **Conclusion:** Gamma irradiation of the White Arias rice cultivar induced morphological changes in flag leaf width, grain set (empty and filled grains) and a slight increase in flowering and harvest time. Notably, a population dose of 200 Gy led to a 9.02% increase in 1000-seed weight, suggesting potential for yield improvement despite lacking statistical significance.

Key words: Mutant strain M-1, gamma irradiation, agronomic traits, rice cultivar, irradiation

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Plant breeding produces plant varieties with characteristics such as morphology, physiology, biochemistry and agronomy per existing cultivation systems and desired economic goals[1]. Mutation is one method that has been proven to increase genetic diversity that can be applied to support plant breeding programs. Mutation breeding is useful for the improvement of some plant traits without changing most of the original plant traits. Gamma irradiation treatment on rice plants resulted in several genes that can be mutated at the same time due to the mutagen treated on the network or cells hitting the target randomly. Causal factors of the low rice production is mostly local rice plants cultivated have a high stem structure, longevity long harvest, as well sensitivity to pests and plant diseases. These limiting factors can be one way to improve this is through mutation breeding[2].

Mutation is one way to obtain genetic diversity. Apart from that, mutations are also able to obtain new traits and superior traits that are not possessed by their parents with unexpected results[3]. Mutations are also able to increase the diversity of previous populations or to bring back diversity that has been lost during the evolutionary process[4]. Mutations can be induced artificially with physical mutagens via gamma-ray irradiation[5].

Gamma rays can induce mutations in upland rice plants because of their high energy that can penetrate plant tissue and cause damage to DNA. Mutations increase genetic diversity, which is essential for the internal selection of plant breeding. Induced mutations can produce new traits such as drought tolerance and increased yield, which are useful in the development of superior varieties[6,7].

Mutations can be carried out by treating mutagens, both physical and chemical. Physical mutagens use X-ray and gamma-ray radiation often used in plant breeding[8]. Mutations induced by gamma rays can produce a variety of new phenotypes, some of which may have more character profitable compared to native plants.

In the M-1 generation, which is the first generation after irradiation, genetic changes begin to appear. This mutagenesis not only creates the genetic variation necessary for the selection of plants with superior traits, but can also improve less favorable traits such as resistance to stress environments, low productivity and poor seed quality. The mutation induced by gamma rays has succeeded in increasing the resistance of rice plants gogo against drought and salinity stress. For example, research by Ana Abdelnour-Esquivel showed that gamma irradiation increased tolerance drought in rice varieties by inducing response-regulating mutation stress[9].

Gamma rays can mimic spontaneous or natural mutations in rice plants' gogo because of their high energy can penetrate plant tissue and induce DNA damage. This damage includes strand breaks single and double as well as nitrogen base modification, which is similar to mutase spontaneously happening naturally. This DNA damage is not always repaired completely by cell repair mechanisms, resulting in mutations permanent. Gamma-ray-induced mutations increase genetic diversity, which is important for selection in plant breeding, allowing the identification and development of genotypes with superior traits such as drought tolerance or increased yield[10,11].

Some of the resulting M-1 mutants showed an increase in p number of seeds per panicle and seed weight. Research by Viana shows that rice varieties irradiated with gamma rays have increased yields significant yield compared to non-irradiated control varieties[4]. Use of induced mutation techniques to improve characters which is less profitable in rice plants has produced a lot of varieties which has an advantage. Results of research over the last five decades show that the character has been improved through induced mutation techniques namely early age, tolerance of low-temperature stress and inundation, increasing number of tillers, resistance to blast and leaf blight and increased quality grain and yield[12].

The use of gamma rays as a mutagen agent has resulted in higher genetic diversity in the M-1 population. This provides the base for wider genetics for the selection and development of more new good varieties. Gamma ray-induced mutations can also improve characteristics of seed quality, such as seed size, nutritional content and seed texture, for example in Katiyar *et al.*[13] research found that gamma irradiation can increase protein and amylose content in rice seeds. The objective of the study was to evaluate the response of the local rice cultivar Arias Putih to gamma-ray irradiation.

MATERIALS AND METHODS

Study area: The research was conducted from January to July, 2023 on Jalan TM Bahrum, Langsa City with a height of $\pm$10 m above sea level. The local upland rice cultivar Arias Putih was irradiated with 3 irradiation dose intervals; G0 = control, G1 = 100 Gy, G2 = 200 Gy and G3 = 300 Gy. Irradiation is carried out in Isotope Technology Application Center and Radiation-National Nuclear Energy Agency PATIR-BATAN, Jakarta.

Study design: A total of 972 rice seeds Local rice cultivar Arias Putih cultivar were irradiated (3 doses of irradiation G1, G2 and G3) and sent to PATIR-BATAN. Planting plus control (M-0) was 324 Rice seeding is done by planting seeds directly using a pointed wooden stick to make holes in the ground where the seeds will be planted. The planting distance used is 30×30 cm. Every planting hole planted 1 seed. Observations were made on morpho agronomic characters in all populations including plant height, number of tillers, number of productive tillers, age at flowering, age at harvest, number of grains, percentage of grain containing and empty, as well as grain production per hill.

Statistical analysis: Data analysis was conducted using means and Tukey's advanced test (HSD: Honestly significant difference) at a specified significance level (e.g., $p<0.05$).

RESULTS AND DISCUSSION

Irradiated White Arias cultivar with gamma rays at plant height range 151.23 up to 139.34 cm. Table 1 shows that there is no real difference in characters on plant height and flag leaf length. The research results do not yet provide any diversity of mutants obtained so the plant's height is high. Tall plants are vulnerable and experience a fall. Varieties with posture tall are susceptible to lodging, while varieties that are too short (<80) often make harvesting difficult[14].

A decrease in leaf width occurs as a result of gamma-ray irradiation. The highest decrease in leaf width was found in the 100 Gy irradiation treatment it was 1.98 cm. A wide decrease in Leaves has a positive impact on plants to be more tolerant of drought due to a decrease in leaf surface area (Table 1).

There is no real difference in flag leaf length characters compared with control plants (Table 1). These results did not produce the expected diversity for improving the morphology of the flag leaf length of the Arias Putih rice cultivar. Radiation mutation plant breeding hopefully, it won't cause any changes in real physiology but changes occur in genetics in large quantities to improve genetic diversity.

The differences are not significant in all characteristics of the number of tillers, productive tillers and panicle length (Table 2). These results show that there is no difference significant impact on the generative character of rice plants gogo irradiated cultivar White Arias compared with control. The number of tillers, number of productive tillers and panicle length due to irradiation treatment have not resulted in physical damage, namely the deterministic effect of irradiation treatment so it has not shown significant results. This can also be seen in the percentage of control that occurred in the decrease in the number of offspring in the G3 treatment, around -1.88%; the decrease in the number of productive tillers in the G2 treatment was around -2.34% and the reduction in panicle length in the G1 treatment was around -4.66%. In this case, if there is a decrease in the number of tillers, the number of productive tillers and panicle length in the irradiation treatment will affect decreasing production.

There is a real difference in the parameters of the number of filled grains and the number of empty grains. The higher the irradiation value, the smaller the amount of grain contained. The highest reduction in the number of containing grains compared to the control was -22.69% with G3 irradiation (300 Gy). The number of empty grains reached 75.64% in G3 plants (300 Gy) compared to controls (Table 3). An increase in the amount of grain Vacancy in irradiation treatment is a form of physical damage due to the deterministic effects of irradiation. In line with research by Tuhina-Khatun *et al.*[15] which reports that irradiation doses above 300 Gy cause the percentage of grain void per panicle (sterility) to increase significantly so that research in general still has potential for continued in the next generation for the breeding program. Mutagens will cause a reduction reproductive ability of plants and can increase the number of empty grains more large compared to environmental influences. Reduced amount of fertile grain caused by one of the causes of chromosomal aberrations. Increasing the dose of irradiation causes a higher number of empty grains in this study.

There is no significant difference in the b character of planting seed density and weight of 1000 seeds (Table 4). These results showed that there is no significant impact on the generative character of rice plants gogo irradiated cultivar White Arias compared with the control. Reduction in weight of planted seeds at irradiation of 100 Gy -22.47%, 200 Gy -17.28% and 300 Gy -16.65% compared to control. In the weight parameter of 1000 seeds, there was a decrease in weight at 100 Gy irradiation of -2.25% and -0.74% at 300 Gy irradiation, while at 200 Gy irradiation, there was an increase in the weight of 1000 seeds by 9.02%.

There are significant differences in the parameters of flowering time and harvest time. The higher the irradiation value, the longer the flowering period. The highest increase in flowering age compared to the control was 10.39% with G3 irradiation (300 Gy) (Table 5). The Arias Putih cultivar is upland rice which has a long harvest period. Gamma irradiation can

Table 1: Character performance plant height, flag leaf length, flag leaf in several gamma-ray treatments

Gamma ray treatment	Plant height (cm)	Flag leaf width (cm)	Flag leaf length (cm)
G0 (Control	151.23	2.18[a]	31.45
G1 (100 Gy)	139.34	1.98[b]	30.30
G2 (200 Gy)	148.34	1.92[b]	30.87
G3 (300 Gy)	149.28	1.90[b]	31.17
Average	147.05	2.00	30.95
	ns	0.22	ns

ns: Non significant and $p<0.05$

Table 2: Character performance of the number of tillers, productive tillers, panicle length compared to controls of several gamma-ray treatments

Gamma ray treatment	Number of tillers	Percentage against control	Number of productive tillers	Percentage against control	Panicle length (cm)	Percentage against control
G0 (Control	12.78	-	11.13	-	21.89	-
G1 (100 Gy)	12.84	0.47	11.32	1.71	20.87	-4.66
G2 (200 Gy)	13.54	5.95	10.87	-2.34	22.08	0.87
G3 (300 Gy)	12.54	-1.88	11.93	7.19	21.46	-1.96
	ns	-	ns	-	ns	-

ns: Non significant and $p<0.05$

Table 3: Character performance of filled grains and empty grains compared to controls in several gamma-ray treatments

Gamma ray treatment	Number of filled grains	Percentage against control	Number of empty grains	Percentage against control
G0 (Control	144.49[a]	-	55.73[b]	-
G1 (100 Gy)	143.97[a]	-0.36	60.39[b]	8.37
G2 (200 Gy)	122.80[b]	-15.01	85.33[a]	53.13
G3 (300 Gy)	111.70[c]	-22.69	97.88[a]	75.64
	8.74	-	13.86	-

Numbers followed by the same letter indicate no significant difference was found in the Tukey's test in the same column at the 5% level of significance

Table 4: Character performance of planting seed weight and 1000 seed weight compared to controls in several gamma-ray treatments

Gamma ray treatment	Planting seed weight (g)	Percentage against control	Weight of 1000 seeds (g)	Percentage against control
G0 (Control	25.11	-	20.40	-
G1 (100 Gy)	19.40	-22.74	19.94	-2.25
G2 (200 Gy)	20.77	-17.28	22.24	9.02
G3 (300 Gy)	20.93	-16.65	20.25	-0.74
	ns	-	ns	-

ns: Non significant and $p<0.05$

Table 5: Character performance of age of flowering and age of harvest compared to controls in several gamma ray treatments

Gamma ray treatment	Flowering age (DAP)	Percentage against control	Harvest age (DAP)	Percentage against control
G0 (Control	91.53[c]	-	122.55[d]	-
G1 (100 Gy)	96.79[b]	5.74	127.09[c]	3.70
G2 (200 Gy)	100.60[a]	9.90	129.02[b]	5.28
G3 (300 Gy)	101.04[a]	10.39	131.39[a]	7.21
Average	97.49	-	127.51	-
	1.57	-	1.56	-

Numbers followed by the same letter indicate no significant difference was found in the Tukey's test in the same column at the 5% level of significance

cause changes in chromosomes, but in this experiment, it was not able to reduce the length of flowering life. White Arias cultivar irradiated with light gamma at doses of 100, 200 and 300 Gy experienced an increase in flowering time and lifespan harvest. The age of flowering of control plants (without irradiation) is 91.35 HST while irradiated plants are 96.79 to 101.04 HST.

The results of this study did not decrease the harvest age but increased it, as in the study of Purwanto *et al.*[11], black rice variety Compo Ireng experienced an increase in flowering age to obtain yield index >2, the criteria for medium harvest age are very mature (105-124 days after sowing), medium (125-150 days after sowing) and immature (>151 days after sowing). The data from this study in the form of days after planting can be directly compared with data from the Center for Rice Research in the form of days after sowing because the study did not carry out sowing, but planting was carried out by planting seeds directly.

The results showed that the characteristics of flag leaf width, the number of filled grains and the number of empty grains decreased as the irradiation dose increased to 300 Gy. This suggests that higher radiation doses are likely to have detrimental effects on some aspects of rice plant productivity. This decline is most likely caused by cellular and molecular damage caused by radiation, which affects the plant's ability to develop optimally[10].

On the other hand, the characteristics of flowering age and harvest time showed an increase along with increasing irradiation dose. This shows that plants take longer to reach the flowering and harvest phases when exposed to higher radiation doses. This increase in flowering time and harvest time can be considered as a form of adaptive response to radiation-induced stress, where the plant lengthens its life cycle perhaps to overcome the damage[16].

However, the characteristics of plant height, flag leaf length, panicle length, number of tillers and number of productive tillers did not show significant changes due to gamma irradiation treatment in the M-1 generation. The stability of this character indicates that some morphological aspects of the Arias Putih rice cultivar are less sensitive to changes induced by gamma ray radiation or may require more than one generation to show significant variation.

CONCLUSION

Gamma irradiation induces morphological and physiological changes in the rice cultivar Arias Putih. These changes included a decrease in flag leaf width, a reduction in the number of filled grains, an increase in the number of hollow grains and an increase in flowering and harvesting time. Higher irradiation doses (300 Gy) generally produced more pronounced negative effects on grain yield and plant development compared to lower doses (100 and 200 Gy). Some traits, such as plant height, number of tillers and panicle length, were largely unaffected by gamma irradiation in the M-1 generation. This suggests that these traits may be less sensitive to radiation-induced changes in this cultivar. The research focused on analyzing the effects of gamma-ray treatment on this cultivar's growth and development.

SIGNIFICANCE STATEMENT

This research provides a foundation for the development of improved varieties of Arias Putih local rice through radiation mutation techniques. The findings offer initial insights for selecting mutant plants with enhanced traits, such as shorter harvest periods, higher productivity and increased resistance to environmental stress. Further studies are essential to assess the next generations (M2, M3, etc.) to explore the emergence of broader genetic variations and identify mutant plants with desirable characteristics. Additionally, evaluating subsequent generations will help determine the long-term effects of gamma irradiation and identify potential beneficial mutations.

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