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Effect Of Gamma Ray Irradiation And Harvesting Age On Viability And Vigor Of M2 Mutant Rice (*Oryza Sativa* L)Seeds

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Abstract:
This study aims to obtain genotypes and the accurate harvesting age of rice crops to increase seed viability and vigor. This research was conducted at the Experimental Garden and Seed Science and Technology Laboratory, Faculty of Agriculture, Syiah Kuala University started from June to December 2019. This study used a Factorial Completely Randomized Design (CRD). The observed variables were viability and vigor of rice seeds, namely: potential growth rate, germination capacity, vigor index, relative growth rate, seed emergence uniformity, dry weight of normal germination. The results showed that the genotypes that were irradiated by gamma ray radiation at a dose of 200 Gy were able to increase the potential growth rate, germination capacity, vigor index, relative growth rate and seed emergence uniformity. The harvesting age at a week after physiological maturity was able to increase the potential growth rate, germination capacity, vigor index, relative growth rate and seed emergence uniformity. There was an interaction between genotypes that already irradiated with gamma rays at a dose of 200 Gy and harvesting time of a week after physiological maturity on the potential growth rate, germination capacity, vigor index, relative growth rate and seed emergence uniformity.

Keywords: *M2 mutant, rice, gamma rays, harvest age*

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I. Introduction

Rice (Oryza sativa L) is one of the staple foods of the Indonesian population. The increasing population of Indonesia is also related with increasing the need for rice[1]. Rice production in the last three years has fluctuated, according to[2]states that Indonesia's rice production in 2018 was 59.2 million tons of milled dried grain, which decreased in 2019 by 54.6 million tons, and in 2020 by 55.1 million tons.

One of the main causes of low rice production is the limited good quality of the seeds. According to[3]seeds are planting materials that determine the initial success of a production process of a plant. [4]Stated that seed quality has 95% growth rating will provide that (a) have viability or can maintain their continued growth into a healthy good plant, (b) Pure seeds that are free from dirt, seeds of other plants. [5]in responding to this problem, one of the efforts that can be done is to create the superior varieties that have high production. New superior varieties can be obtained from rice genetic sources originating from local rice varieties which have natural characteristics of high adaptability. BadanTenaga Atom Nasional (BATAN) or The National Atomic Energy Agency is an institution that produces superior rice varieties using non-conventional breeding techniques, namely[6]

Mutation is a process of changing the structure of genes or chromosomes that results in phenotype changes that are inherited from one generation to another[7]. One of the methods in mutation breeding techniques is by using gamma rays. [8]Suggested that gamma rays are the most energetic form of electromagnetic radiation, which can be useful for changes in physiological characters. Furthermore, [9]explained that improving local rice varieties by mutation techniques using gamma ray irradiation is an alternative way to create new genetic sources. According to [10] gamma ray irradiation at a dose of 250 Gy can increase germination rate, seed emergence uniformity, plant height, number of leaves and number of tillers of rice plants. In addition, [1]effective dose of gamma ray irradiation at a dose of 200 Gy was able to increase the percentage of germination and seedling height.

In addition, the harvesting time is an important factor in determining seed quality[11]. Also, [12]further explained that the germination percentage is more sensitive to harvesting time. [13]explained that testing of quality seeds is rapidly constrained by seed dormancy. Rice seeds have various of “after ripening” periods from 0 - 11 weeks, while the after-ripening period is the length (weeks or months) of seeds in a dormant period since harvest, this period is necessary to allow seed embryos to overcome dormancy during the development process[14]

This study aims to obtain the best genotype and the accurate harvesting time to increase the viability and vigor of rice seeds, as well as to examine the interaction between genotype and harvesting age on the viability and vigor of rice seeds.

II. Material And Methods

This research was conducted at the Experimental Garden and Seed Science and Technology Laboratory, Faculty of Agriculture, Syiah Kuala University and IRRI (Aceh Rice Research Institute) started from June to December 2019. This research material used seeds from the Aceh Rice Research Institute which consisted of the five genotypes of the M2 generation mutant of Sigupai, consisting of G0 (Sigupai local rice variety from Southwest of Aceh), G1 (200 Gy), G2 (250 Gy), G3 (300 Gy), G4 (350 Gy), and G5 (400 Gy), water, tissue paper, fertilizers, stencil paper, and other research tools, namely: tractors, nursery containers, tweezers, petridists, analytical scales, hand sprayers, germinators, and cameras.

This research used a Factorial Completely Randomized Design (CRD). In this experiment consisted of 2 factors and 3 replications. The first factor is the genotype which consists of 6 levels, namely: G0 (Sigupai local rice variety from Southwest of Aceh), G1 = 200 Gy, G2 = 250 Gy, G3 = 300 Gy, G4 = 350 Gy, and G5 = 400 Gy. The second factor is harvesting age which consists of 3 levels, namely: U1 = a week before physiological maturity, U2 = Physiological maturity, U3 = a week after physiological maturity. Thus, there are 18 treatment combinations, each treatment combination was repeated 3 times, so that there are 54 experimental units. The research data were analyzed by using Analysis of Variance (ANOVA). If the F test results show a significant effect, it will be followed by an Honestly Significant Difference test (HSD) at the 5% level. The observed variables were viability and vigor of the rice seed, namely: potential growth rate, germination capacity, vigor index, relative growth rate, seed emergence uniformity, and dry weight of normal germination.

III. Result and Discussion

Genotype against Viability and Vigor

Table 1. Average of the potential growth rate, germination capacity, vigor index, relative growth rate, seed emergence uniformity, dry weight of normal germination due to the M2 genotype of Sigupai local rice from Southwest of Aceh

Genotype	Potential Growth Rate (%)	Germination Capacity (%)	Vigor Index (%)	Relative Growth Rate (%/etmal)	Seed Emergence Uniformity (%)	Dry Weight of Normal Germination (gram)
G_0 (Kontrol)	18,00 ^{c}	17,11 ^{c}	8,17 ^{b}	2,29 ^{c}	14,44 ^{c}	0,04
G_1 (200Gy)	37,56 ^{d}	36,67 ^{d}	17,78 ^{c}	4,76 ^{d}	20,44 ^{d}	0.03
G_2 (250Gy)	12,44 ^{bc}	12,00 ^{bc}	4,61 ^{b}	1,66 ^{bc}	9,11 ^{bc}	0,05
G_3 (300Gy)	4,11 ^{a}	4,11 ^{a}	1,39 ^{a}	0,50 ^{a}	3,00 ^{a}	0,07
G_4 (350Gy)	8,67 ^{ab}	8,67 ^{ab}	3,67 ^{b}	1,00 ^{ab}	4,89 ^{ab}	0,02
G_5 (400Gy)	13,39 ^{bc}	13,39 ^{bc}	6,56 ^{b}	1,63 ^{bc}	11,00 ^{c}	0,02
HSD 0.05	6,05	5,58	10,94	1,78	4,96	-

Note: Numbers followed by the same letter in the same column are not significantly different at the 5% level based on HSD test

Table 1 shows that the highest of potential growth rate, germination capacity, vigor index, relative growth rate, seed emergence uniformity were found in the genotype that was irradiated by gamma rays at the dose 200 Gy (G1). It is suspected that the irradiation of various doses of gamma ray radiation has been able to affect the germination capacity of seeds. According to gamma ray irradiation can affect electrons in the nucleus[12]. The effect of gamma rays is the ionization of water which can form highly reactive hydrochloric radicals. These radical reactions can affect chemical changes and immediately react with all organic molecules both structurally and functionally. Furthermore, the radical reactions also decrease the metabolism and germination rate. In addition, higher radiation dose can negatively affect the strength of the seeds, due to gamma ray irradiation works is to produce an ionization process [13]This process differs for every plant depending on the morphology and physiology of the plant. This phenomenon is called hormesis which has a positive impact on plants. [14]Added that high seed vigor causes tolerant seeds and develops in sub-optimum land conditions, in the form of an environment that is not suitable for seed growth and germination. In the results of [15]research increasing the dose of gamma ray irradiation in SiGadis rice seeds reduced the germination capacity and seed emergence uniformity, further explained that at a dose of 100 Gy produced 80% of germination capacity while at doses of 200 Gy and 300 Gy the germination capacity was 70% and 60 % respectively.

Harvesting Age on Rice Seed Viability and Vigor

Table 2. Average of seed moisture content, potential growth rate, germination capacity, vigor index, relative growth rate, seed emergence uniformity, dry weight of normal germination due to harvesting age

Harvesting Age	Maximum Growth Potential (%)	Germination Capacity (%)	Vigor Index (%)	Relative Growth Rate (%)	Seed Emergence Uniformity (%)
U_1 (1 week before physiological cooking)	8,08^{a}	7,86^{a}	3,82^{a}	0,98^{a}	4,11^{a}
U_2 (Physiological cooking)	12,33^{b}	12,22^{a}	7,36^{ab}	1,58^{b}	10,67^{b}
U_3 (1 week after physiological cooking)	26,44^{c}	25,89^{b}	10,50^{b}	3,37^{c}	21,67^{c}
HSD 0.05	6,05	5,58	10,94	1,78	4,96

Note: Numbers followed by the same letter in the same column are not significantly different at the 5% level based on HSD test

Table 2 shows that the best of potential growth rate, germination rate, vigor index, relative growth rate and seed emergence uniformity were found at the harvesting age which was carried out at a week after physiological maturity (U3). The condition of the seed when it reaches physiological maturity is important, the seeds that have physiological maturity will have a condition to express their maximum vigor, the seeds that are used before physiological maturity have low physiological qualities. Also, the seeds that are harvested too long decrease the physical and physiological qualities of rice seed, due to the harm of environment, humidity and high temperature. The determination of physiological maturity cannot always be determined by its morphological appearance, because the physiological maturity of seeds can be achieved before the fruit looks morphologically ripe[15]. Furthermore , it is thought that the seed dormancy occurs since the seeds are still in the plant, when the embryo develops it is called primary dormancy or innate dormancy. The cause of primary dormancy occurring in rice seeds is after ripening which has a role in the low germination value of rice seeds. The seeds that have undergone after ripening period will germinate if stored for a certain period of time[16.]

Interaction of M2 Mutant Rice and Harvest on Viability and Seed Vigor

Table 3. The value of the interaction between genotypes of generation M2 and harvesting age on potential growth rate, germination capacity, relative growth rate, seed emergence uniformity, vigor index and dry weight of normal germination

Gynotype	Maximum Growth Potential (%)		
	U_1 (1 week before physiological cooking)	U_2 (Physiological cooking)	U_3 (1 week after physiological cooking)
G_0 (Control)	20,15 Ba	14,33 ABa	36,46 Bb
G_1 (200Gy)	18,81 Ba	37,94 Ca	53,30 Cb
G_2 (250Gy)	14,80 ABa	9,27 Aa	31,89 Bb
G_3 (300Gy)	5,41 Aa	10,15 Aa	16,35 Aa
G_4 (350Gy)	21,09 Ba	10,15 Aa	15,68 Aa
G_5 (400Gy)	10,67 ABa	26,55 BCb	23,47 ABab
HSD 0.05	13.06		
Gynotype	Germination Capacity (%)		
G_0 (Kontrol)	18,95 Ba	12,91 Aa	36,46 Cb
G_1 (200Gy)	18,28 Ba	37,56 Bb	52,48 Bc
G_2 (250Gy)	14,80 ABa	9,27 Aa	31,07 Bb
G_3 (300Gy)	5,41 Aa	10,15 Aa	16,35 Aa
G_4 (350Gy)	21,84 Ba	12,16 Aa	14,93 Aa
G_5 (400Gy)	10,67 ABa	26,55 Aa	23,47 ABb
HSD 0.05	12.03		
Gynotype	Vigor Index (%)		
G_0 (Kontrol)	5,41 Aa	6,77 Aa	27,91 Cb
G_1 (200Gy)	11,54 Aa	19,91 Ba	37,66 Db
G_2 (250Gy)	9,27 Aa	4,05 Aa	18,99 Bb
G_3 (300Gy)	4,05 Aa	5,41 Aa	9,27 Aa
G_4 (350Gy)	12,42 Aa	6,55 Aa	12,16 ABa
G_5 (400Gy)	6,55 Aa	19,56 Bb	14,93 ABa

HSD 0.05	8,00		
Gynotype	Relative Growth Rate (%)		
G_0 (Kontrol)	6,24 Aa	4,80 Aa	12,81 Cb
G_1 (200Gy)	6,87 Aa	12,27 Cb	16,64 Dc
G_2 (250Gy)	4,81 Aa	5,43 ABa	10,45 BCb
G_3 (300Gy)	3,60 Aa	3,26 Aa	4,82 Aa
G_4 (350Gy)	6,63 Aa	4,15 Aa	5,96 Aa
G_5 (400Gy)	4,26 Aa	8,94 BCb	7,79 ABa
HSD 0.05	3.84		
Gynotype	Seed Emergence Uniformity(%)		
G_0 (Kontrol)	13,84 Aa	12,16 ABa	34,84 CDb
G_1 (200Gy)	15,02 Aa	35,13 Cb	45,38 Db
G_2 (250Gy)	11,28 Aa	9,27 Aa	26,93 BCb
G_3 (300Gy)	5,41 Aa	9,27 ABa	12,70 Aa
G_4 (350Gy)	12,16 Aa	11,28 ABa	14,18 Aa
G_5 (400Gy)	6,55 Aa	24,09 Bb	22,37 ABb
	10,70		
Genotipe	Dry Weight of Normal Germination (gram)		
G_0 (Kontrol)	0,72	0,76	0,72
G_1 (200Gy)	0,74	0,72	0,72
G_2 (250Gy)	0,72	0,74	0,77
G_3 (300Gy)	0,75	0,74	0,77
G_4 (350Gy)	0,71	0,73	0,72
G_5 (400Gy)	0,72	0,72	0,73

Note : The numbers followed by the same letter (horizontal lowercase, vertical uppercase) show insignificant differences at the 5% level (LSD 0.05).

Table 3 shows that the potential growth rate, germination capacity, vigor index, relative growth rate and seed emergence uniformity, the best treatment was found in G1U3, which is irradiated with 200 Gy of gamma ray irradiation dose and a week of harvest after physiological maturity. According to[16]200 Gy gamma rays have been able to increase the viability and vigor of M2 strain of rice seeds. Furthermore, it explained that the way gamma rays produce the reactive radicals that will be reacted with molecules in the physiological system so that they can increase biochemical processes in cells. Mutations in plants after irradiation with gamma ray change the plant parts and plant characteristics. Gamma radiation changes the genetics which causes addition or loss of one or more bases content of DNA molecule. In addition, another factor that affects vigor and viability of rice seeds is the harvesting time, because the difference of seed maturity level is based on the position of the seeds in the panicles. Additionally, healthy seeds that germinate is when the seeds have formed a food reserve

IV. Conclusion

Conclusion: The genotypes of rice seeds that have been irradiated with 200Gy of gamma ray radiation were able to increase the potential growth rate, germination capacity, vigor index, relative growth rate, and seed emergence uniformity. Harvesting age in a week after physiological maturity can increase the potential growth, germination capacity, vigor index, relative growth rate, and seed emergence uniformity. There was an interaction between genotype and harvesting age on potential growth rate, germination capacity, vigor index, relative growth rate, and seed emergence uniformity.

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