Performance and estimation genetic variability of M3 pearl millet (*Pennisetum glaucum*) populations

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Abstract. Improvement of agronomic characteristics of pearl millet plants such as short plants and high production can be done through plant breeding techniques. The success of plant breeding programs is largely determined by the availability of genetic variability. The application of gamma-ray irradiation is used to increase the genetic variability of pearl millet plants. This study aims to determine the genetic variability and performance in the population and to predict genetic parameters in several characters of pearl millet plants in M3 generation. The research material used was M2 seed from gamma-ray irradiation at doses of 0, 100, 200 and 300 Gy. The observed characters were plant height, the number of nodes per plant, stem diameter, panicle length, panicle diameter, panicle weight of seed weight per panicle and weight of 1000 seeds. The study was conducted from February to April 2019 at Center for Application of Isotope and Radiation, National Nuclear Energy Agency. Through estimating variance, characters that have high heritability and GVC values are not obtained in the four populations (0, 100, 200 and 300 Gy) so that selection cannot be done. The pearl millet phenotype in the M3 generation is more influenced by environmental factors compared to genetic factors, and, therefore, selection is not effective in the M3 generation. Purification in the next generation needs to be done. However, in the populations of 200 and 300 Gy start look potentially to be selected in the next generation.

1. Introduction

Pear millet (*Pennisetum glaucum*) is C4 annual cereal for food and fodder. It was grown well in Africa, India, Australia and also in the United States. It is commonly grown in the hottest and driest regions where other cereals are likely to fail because of drought, high-temperature stress, and poor soil conditions [1]. It is the most drought tolerant of all domesticated cereal [2]. Pearl millet grain is more nutritious than wheat, rice, maize, and sorghum [3] [4] [1]. Its nutritional superiority comes from high levels of protein, vitamins, essential amino acids, antioxidants, and essential micronutrients, such as iron and zinc [3].

The induction of mutations as a technique for crop improvement occupies a significant place among modern methods of plant breeding. The induced mutations are of considerable value for comprehension, evaluation and accelerating the process of plant improvement [5]. Based on the results of doses orientation, the highest diversity is found at doses of 100-300 Gy. Tall plants cause more photosynthate allocation to stems than seeds and lodging risk. Therefore, improving the agronomic traits of short pearl millet plants, producing high seeds and large stem diameters needs to be done through plant breeding techniques. In many cereal crops, reduced height is
a key trait for enhancing yield, and dwarf mutants have been extensively used in breeding to reduce yield loss due to lodging under intense management [6][7]. This study aims to study the performance and genetic variability of the M3 pearl millet mutant population to obtain information on selection criteria for the next generation. Improvement of plant genetic and agronomic traits can be done through breeding, analyzing genetic variability and estimating the heritability of some characters in pearl millet plants.

2. Material and methods
The experiments were conducted from February - April 2019 at wire house of Center for Application of Isotopes and Radiation, National Nuclear Energy Agency (CAIR-NNEA), Jakarta. The genetic material used in this study was the introduction from Institute of Agricultural Research, Mozambique Institute of Agricultural Research (IIAM). The seeds irradiated with different doses (0, 100, 200 and 300 Gy) of gamma-rays from 60Co at CAIR-NNEA, Jakarta-Indonesia. The seedling were planted by population without an experimental design with a spacing of 15 cm x 70 cm. Each population of each dose consists of 200 individual plants. Morphological characters observed consist: the number of nodes per plant, plant height (cm), the diameter of stem (cm), panicle length (cm), the diameter of panicle (cm), panicle weight (g), seed weight per panicle (g), and weight of 1000 seeds (g). The data were analyzed using a quantitative estimation of the value of broad-sense heritability and coefficient of genetic variability. Estimation of heritability broad sense (H2BS) use the following formula:

$$h^2_{bs} = \frac{\sigma^2_g}{\sigma^2_p} \times 100$$

The estimated value of heritability is classified according to Stanfield [8]:  
- $h^2_{bs} > 50$ (high), 20 ≤ $h^2_{bs} ≤ 50$ (medium), and $h^2_{bs} < 20$ (low). Genetic Variability Coefficient (GVC) use the following formula Singh and Chaudary [9]:  
  $$GVC = \frac{\sqrt{\frac{\sigma^2_g}{\bar{x}}}}{\bar{x}} \times 100\%.$$  
Exp : $\sigma^2_g$ = genetic varians $\bar{x}$ = mean value of population. GVC criteria base on Knight [10]: low (0–10%), medium (12–20%) and high (> 20%).

3. Results and discussion
A reference to Table 1 showed that each character of 100 Gy, 200 Gy and 300 Gy populations have a high genetic variability in terms of the mean and standard deviations. Almost all the characters showing that the highest average value showed from 300 Gy population compared with 100 and 200 Gy populations. While the lowest average value is found in a population of 100 Gy.

Table 1. Mean Value and Standard Deviation of Genetic Variability of Morphological Character of M3 Pearl Millet population.

| Character                   | 0 Gy   | 100 Gy  | 200 Gy  | 300 Gy  |
|-----------------------------|--------|---------|---------|---------|
| Number of nodes per plant   | 8.0±0.8| 7.0±1.5 | 6.0±1.5 | 8.0±1.3 |
| Plant height (cm)           | 185.0±32.5 | 175.5±37.5 | 153.0±36.5 | 185.0±37.7 |
| Diameter of stem (cm)       | 1.4±0.5 | 1.9±1.16 | 1.7±1.1  | 2.4±1.1 |
| Panicle Length (cm)         | 27.0±6.2 | 23.0±6.3 | 24.0±7.8 | 29.0±6.4 |
| Panicle weight (g)          | 22.9±10.1 | 16.5±10.8 | 11.7±10.2 | 21.8±11.6 |
| Diameter of panicle (cm)    | 2.3±0.4 | 2.0±1.1  | 2.1±1.0  | 2.6±1.0 |
| Seed weight per panicle (g) | 13.7±8.2 | 9.7±8.4  | 11.6±9.2 | 16.1±9.5 |
| Weight of 1000 seeds (g)    | 9.5±2.4 | 7.7±2.4  | 7.8±2.4  | 10.1±2.5 |

The results in Table 1 showed that the standard deviation values in all of the populations are still diverse and higher than the standard deviation values of control. A low standard deviation means that
most of the numbers are close to the average and a high standard deviation means that the numbers are more spread out. Genetic variability of 100, 200 and 300 Gy populations are caused by the influence of gamma-ray irradiation. Then the selection of mutant lines is expected to be done.

The expected characteristics of pearl millet plants are short plants with a height of about 100-140 cm, high production and large panicle. Short plants can simplify the process of maintaining, harvesting and making plants resistant to lodging. A population of 300 Gy produced plants that were the same height as the parents among those observed, which were 185 cm, while the population of 200 Gy produced the lowest plants, which were 153 cm (Table 1). A large diameter of the stem can produce a greater weight of the seed because the stem is a place for photosynthesis and storage of food reserves. The largest stem diameter is in the population of 300 Gy (2.36 cm) and the smallest stem diameter is in the population of 200 Gy (1.68 cm) (Table 1). Sungkono [11] states that there is a positive correlation between plant height and stem diameter of sorghum plants. In Table 1 it is proven that the largest seed weight is produced by a population of 300 Gy (16.1 g) and the smallest seed weight produced by 100 Gy population (9.7 g). A population that have tall plants (> 180 cm) is population of 300 Gy, as well as the largest stem diameter (2.4 cm). Populations classified as having short-sized plants (<180 cm) are populations of 100 and 200 Gy with stem diameters in both populations not as large as stem diameter in population of 300 Gy (1.9 and 1.7 cm).

Panicle is the main part of the pearl millet plant which produces seeds and influences the yield and components of seed yield. Phenotypically the panicle form of each population tested differs depending on the radiation dose. That is caused by the effects of gamma rays irradiation which can cause genetic changes in somatic cells (somatic mutations). Besides being able to be derived, the effects of radiation can also cause phenotypic changes. The performance of pearl millet panicle in M3 generation is found in Figure 1.

Figure 1. Performance and variation in panicle morphology, panicle size, grain size, and grain color of pearl millet populations of 100 Gy, 200 Gy and 300 Gy.

The estimates of genotypic variation ($\sigma^2_g$), phenotypic variation ($\sigma^2_p$), and genetic variation coefficient (GVC) for different characters have been presented in (Table 2). Estimates of genetic variance are used to evaluate the number of planted populations that have high or low variability. The population used in this study is in the M3 generation so that this population still has high variability.
This experiment used a parent/control as a variety of environments because each individual plant in the parent has genetic similarities, so the phenotype variations that arise are not caused by genetic variations but are influenced by the environment. Components of environmental variability can be calculated using populations that do not have genetic diversity or populations with low genetic diversity such as pure lines.

**Table 2.** Heritability Broad Sense of Morphological Character of M3 Pearl Millet populations.

| Character                           | $\sigma^2_g$ | $\sigma^2_p$ | $\sigma^2_e$ | $h^2_{bs}$ (%) | GVC (%) |
|------------------------------------|--------------|--------------|--------------|----------------|---------|
| **100 Gy**                         |              |              |              |                |         |
| Number of nodes per plant          | 1.67         | 2.39         | 0.72         | 69.83          | 0.46    |
| Plant height (cm)                  | 345.10       | 1419.19      | 1074.08      | 24.31          | 0.16    |
| Diameter of stem (cm)              | 1.11         | 1.36         | 0.24         | 81.76          | 0.66    |
| Panicle Length (cm)                | 1.28         | 40.57        | 39.28        | 3.17           | 0.23    |
| Panicle weight (g)                 | 14.3         | 117.61       | 103.30       | 12.16          | 0.85    |
| Diameter of panicle (cm)           | 1.02         | 1.2          | 0.18         | 84.98          | 0.67    |
| Seed weight per panicle (g)        | 12.43        | 71.52        | 68.23        | 4.59           | 0.51    |
| Weight of 1000 seeds (g)           | 0.05         | 5.94         | 5.88         | 0.99           | 0.08    |
| **200 Gy**                         |              |              |              |                |         |
| Number of nodes per plant          | 1.39         | 2.11         | 0.72         | 65.95          | 0.46    |
| Plant height (cm)                  | 268.92       | 1343.00      | 1074.08      | 20.02          | 1.33    |
| Diameter of stem (cm)              | 1.04         | 1.29         | 0.24         | 80.73          | 0.71    |
| Panicle Length (cm)                | 22.61        | 61.89        | 39.28        | 36.53          | 0.95    |
| Panicle weight (g)                 | 2.51         | 105.82       | 103.30       | 2.37           | 0.39    |
| Diameter of panicle (cm)           | 0.9          | 1.08         | 0.18         | 83.37          | 0.55    |
| Seed weight per panicle (g)        | 18.13        | 86.37        | 68.23        | 20.99          | 1.16    |
| Weight of 1000 seeds (g)           | 0.15         | 6.04         | 5.88         | 2.52           | 0.13    |
| **300 Gy**                         |              |              |              |                |         |
| Number of nodes per plant          | 1.21         | 1.93         | 0.72         | 62.61          | 0.39    |
| Plant height (cm)                  | 356.44       | 1430.62      | 1074.08      | 24.92          | 1.37    |
| Diameter of stem (cm)              | 1.0          | 1.25         | 0.24         | 80.16          | 0.61    |
| Panicle Length (cm)                | 2.82         | 42.11        | 39.28        | 6.71           | 0.31    |
| Panicle weight (g)                 | 33.9         | 137.21       | 103.30       | 24.71          | 1.18    |
| Diameter of panicle (cm)           | 0.85         | 1.03         | 0.18         | 82.58          | 0.54    |
| Seed weight per panicle (g)        | 23.19        | 91.43        | 68.23        | 25.37          | 1.14    |
| Weight of 1000 seeds (g)           | 0.78         | 6.67         | 5.88         | 11.76          | 0.27    |

$\sigma^2_g$ (genetic variance); $\sigma^2_p$ (phenotype variance); $\sigma^2_e$ (environments variance); $h^2_{bs}$ (broadsense heritability); GVC (Genetic Variation Coefficient)

Based on the estimated phenotypic variance, environmental variance and genetic variance indicate that the environmental influence is still quite high on the diversity of the pearl millet genotype population except for the character of the number of nodes per plant, stem diameter, and panicle diameter where genetic factors are more dominantly controlling the character as indicated by the heritability value of broad-sense which is high (Table 2).

Table 3. showed that the coefficient of genetic variability (GVC) for each character in the three populations is low. GVC is used to measure the amount of genetic diversity of a character. Broad
criteria on GVC values can indicate a wide level of genetic diversity so that the selection process can be done more easily and efficiently. Based on the results of data analysis, the GVC obtained in this experiment is low in all characters and each population so that selection cannot be done. However, according to Boer [12] characters with GVC values <0.10 are not recommended as character selection. It means that all characters in populations of doses 100, 200 and 300 Gy can be recommended as selection characters because they have a GVC value >0.10 except for the 1000 seed weight character in 100 Gy population that have a GVC value <0.10 (0.08).

The graph in Figure 2. illustrates that each M3 population in the characters of plant height, the diameter of stem, and seed per panicle are spread normally. Environmental influences play a role in controlling M3 populations variability. The distribution of values in the observed characters of genotypes in M3 populations, extending to the right or left, showed transgressive segregations whose value exceeds or decreases the value of the control. This is consistent with the opinions of Roy [13] and Jayaramachandran et al [14] which say that quantitative characters in plants whose spread extends to the left or right show the influence of the environment, genotype and environmental interactions.

![Graphs showing distribution of M3 population based on plant height, diameter of stem, and seed weight per panicle.](image)

**Figure 2.** Distribution of M3 pearl millet population-based on character of plant height, the diameter of stem, and seed weight per panicle.

Variance and quartil distribution of the populations (100, 200 and 300 Gy) on the characters of plant height, the diameter of stem and seed per panicle are shown in Figure 3. The upper, median and lower quartiles of boxes represent the 75th, 50th and 25th percentiles of the populations, respectively. The horizontal lines represent the highest and lowest variation of each trait in the population. Among the characters studied, plant height character showed that 200 Gy populations are the best variation for short plant, followed by 100 and 300 Gy. However, the best variation in stem diameter is in the
population of 100 Gy, followed by 300 and 200 Gy. While in the character of seed per panicle, the best variation is in the population of 300 Gy followed by 100 and 200 Gy.

![Boxplot quartile distribution of different M3 populations of pearl millet-based on the characters of plant height, the diameter of stem and seed weight per panicle.](image)

4. Conclusion

Genetic variability through estimation of variance obtained that all characters in populations of 100, 200, and 300 Gy have low heritability and, except for the characters of the number of nodes per plant, diameter of stem and diameter of panicle that have high heritability values, but the values of genetic variance coefficient are narrow. Then the selection is not effective in the M3 generation. Purification in the next generation needs to be done. However, populations of 200 and 300 Gy start looks potentially to be selected in the next generation.

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