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EFFECTS OF GAMMA-RAY IRRADIATION ON VEGETATIVE CHARACTERS OF PALMAROSA (*Cymbopogon martinii*)

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Abstract

Palmarosa (*Cymbopogon martinii*) is an important aromatic plant widely utilized as a source of essential oil with significant economic value. Genetic improvement of palmarosa is required to enhance plant performance and productivity, and one effective approach is induced mutation using gamma-ray irradiation. Gamma irradiation is known to generate random genetic variation that may affect plant growth and development. This study aimed to evaluate the effects of different gamma-ray irradiation doses on vegetative growth characteristics of palmarosa and to identify responsive traits for further breeding programs. The experiment was conducted using a Randomized Complete Block Design (RCBD) with a single factor consisting of eight levels of gamma-ray irradiation doses. Gamma irradiation treatment was performed at the National Research and Innovation Agency (BRIN), planting was carried out in Plasansari Village, Semarang, Indonesia, and data analysis was conducted at the Chemistry Laboratory, Faculty of Mathematics and Natural Sciences, Universitas Negeri Semarang (UNNES). Vegetative parameters observed at the final growth stage included plant height, stem diameter, number of leaves, and number of tillers. The collected data were subjected to analysis of variance (ANOVA), and significant differences among treatments were further analyzed using Duncan's Multiple Range Test at a 5% significance level. The results showed that gamma-ray irradiation had no significant effect on plant height of palmarosa. However, block effects significantly influenced plant height, indicating that environmental variation among blocks contributed to differences in plant growth. In contrast, gamma-ray irradiation treatments significantly affected the number of leaves and tiller numbers. These results indicate that gamma irradiation can induce variability in specific vegetative traits, particularly those related to canopy development and tillering capacity. The results indicate that gamma-ray irradiation is a promising mutagen for inducing useful phenotypic variation in palmarosa. This study provides baseline information for palmarosa mutation breeding and emphasizes the importance of selecting appropriate vegetative traits in early- generation evaluation to support the development of improved genotypes for essential oil production.

Keywords: Palmarosa, gamma-ray irradiration, vegetative growth.

1. INTRODUCTION

Indonesia is one of the world's most biodiverse countries, hosting a wide range of plant species, including aromatic grasses with high agronomic and economic potential. Among these, essential oil–producing plants play an important role in supporting agricultural development and export-oriented industries. In 2022, Indonesia recorded essential oil exports valued at USD 172.9 million, reflecting increasing global demand and the strategic importance of aromatic crops in national agriculture (Waluyo, 2023). Improving plant growth performance through crop improvement strategies is therefore essential to support sustainable production systems, particularly for vegetatively propagated aromatic plants.

Palmarosa is an aromatic grass widely cultivated in tropical regions, including Indonesia. The plant is well adapted to diverse environmental conditions, such as sandy soils with neutral to slightly acidic pH, temperatures ranging from 20–30°C, and moderate to high rainfall (Nadirah *et al*., 2022). Palmarosa exhibits two main varieties, sofia (2n = 40) and motia (2n = 20), which differ in their genetic background and growth performance (Sreelatha *et al*., 2017). Despite its adaptability, palmarosa breeding is constrained by limited genetic variability, which restricts improvement of vegetative traits such as plant height, tiller number, and leaf development—key parameters that determine biomass accumulation and overall plant vigor (Koryati *et al*., 2022).

Gamma-ray irradiation has been widely applied as a mutation breeding technique to induce genetic variation and modify vegetative growth characteristics in plants. Gamma rays can cause random DNA alterations that lead to phenotypic variation, including changes in plant height, leaf number, and tillering ability (Mirantika *et al*., 2023). Previous studies demonstrated that appropriate gamma-ray doses can stimulate vegetative growth, while excessive doses may inhibit development. For example, gamma irradiation at 100 Gy increased plant performance in *Cymbopogon winterianus*, with an estimated LD_{50} of 173 Gy, indicating species-specific sensitivity to radiation treatments (Munda *et al*., 2022). Similar dose-dependent effects of gamma irradiation on vegetative characters have also been reported in other aromatic and horticultural crops (Lal *et al*., 2024).

Vegetative characters are key indicators in mutation breeding because they reflect plant establishment, adaptability, and biomass production. Evaluating vegetative responses to gamma-ray irradiation enables the selection of promising mutant lines with improved growth, supporting the development of superior palmarosa planting materials and effective mutation breeding strategies in aromatic crops (Zuyasna *et al*., 2022).

2. METHODOLOGY

This study was conducted from May 2025 to January 2026. Palmarosa planting materials were obtained from Rumah Atsiri Indonesia. Gamma-ray irradiation treatments were carried out at the National Research and Innovation Agency (BRIN), Yogyakarta. The field experiment was conducted in Plasansari Village, Banyumanik Subdistrict, Semarang City, Central Java, Indonesia (7°4′33.06″ S; 110°25′27.465″ E).

The experiment employed gamma-ray irradiation at different dose levels, namely 0, 20, 40, 60, 80, 100, 120, and 150 Gy, applied to palmarosa planting materials. The study was arranged in a single-factor randomized complete block design (RCBD), with gamma-ray dose as the treatment factor, and each dose consisted of 5 plants per experimental unit. Following irradiation, the materials were planted and cultivated under field conditions using standard agronomic practices.

Observations were focused exclusively on vegetative growth characters, including plant height, number of leaves, and number of tillers, which were recorded periodically during the vegetative growth phase. These parameters were used to evaluate the response of palmarosa plants to gamma-ray irradiation.

Data obtained from vegetative observations were analyzed using descriptive and statistical methods to determine variability among irradiation doses. Analysis of variance (ANOVA) was performed, followed by Duncan's Multiple Range Test (DMRT) at a 5% significance level to identify differences among treatments. The observed vegetative variability was used to assess the potential effects of gamma- ray irradiation on palmarosa growth performance and to identify promising vegetative responses for further evaluation in mutation breeding programs.

3. FINDINGS AND DISCUSSION

The following are the results and discussion of the research that has been carried on, namely on the variables of plant height, number of leaves, and tiller numbers.

3.1 Plant Height

Based on table 1. showed that gamma irradiation treatments did not exert a statistically significant effect on plant height at α = 0.05 (F = 2.218; p = 0.071), suggesting that overall variance among treatments could be attributed to random variation rather than treatment effects.

Table 1. ANOVA for Plant Height

Source	Type III Sum of Squares	Df	Mean Square	F	Sig.
Corrected Model	7922,025[a]	11	720,184	3,110	0,010
Intercept	2754997,261	1	2754997,261	11898,361	0,000
Treatment	3594,619	7	513,517	2,218	0,071
Block	3965,099	4	991,275	4,281	0,010
Error	5325,518	23	231,544		
Total	2942844,000	35			
Corrected Total	13247,543	34			

a. R Squared = ,598 (Adjusted R Squared = ,406)

However, subsequent Duncan Multiple Range Test (DMRT) revealed distinct groupings of means across irradiation doses (*Subset for α = 0.05*), implying differential responses among specific dose levels. For example, treatments with doses of 0 Gy, 80 Gy, 100 Gy, and 120 Gy grouped together with lower mean heights (~267–289 cm), whereas 40 Gy, 20 Gy, 150 Gy, and 60 Gy exhibited higher mean values (~296–302 cm). The presence of these distinct subsets indicates that although the overall ANOVA did not show statistical significance, biologically relevant differences in plant height exist between certain irradiation doses.

Table 2. DMRT for Plant Height

Duncan[a,b]

Doses	N	Subset for alpha = 0.05	
		1	2
0 Gy	5	267,00	
80 Gy	5	285,20	285,20
100 Gy	5	287,60	287,60
120 Gy	5	289,60	289,60
40 Gy	3		296,67
20 Gy	4		297,50
150 Gy	4		297,50
60 Gy	4		302,25
Sig.		0,115	0,254

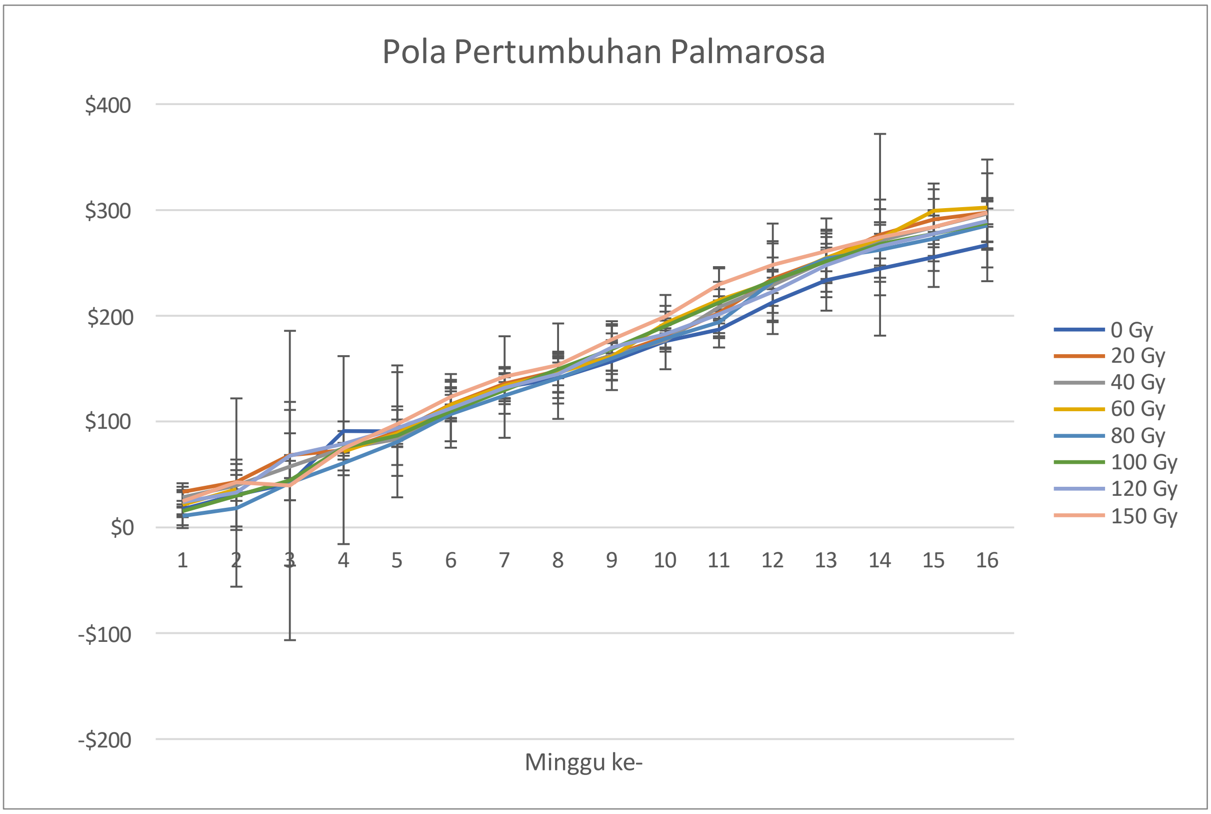
Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 4,248.
b. The group sizes are unequal. The harmonic mean of the group sizes is used. Type I error levels are not guaranteed.

These observations are consistent with known mutation induction mechanisms triggered by gamma radiation. Gamma rays cause ionization events that lead to DNA strand breaks and the formation of free radicals. If the DNA damage is not lethal and undergoes imperfect repair, mutations can arise, potentially altering gene expression involved in cell division and growth regulation. Cytogenetic studies have shown that gamma irradiation can induce chromosomal aberrations in meristematic cells, which may modify growth patterns in subsequent plant development (Sayed *et al*., 2025). Similar patterns have been reported in other studies, where low to moderate irradiation doses stimulated morphological traits while high doses inhibited growth, depending on genotype radiosensitivity and cellular repair capacity (Jeong *et al*., 2024). Therefore, while the overall ANOVA confirms a lack of global significance, the DMRT results provide insight into specific dose-dependent effects that may be biologically meaningful in the context of mutation breeding and genetic variation.

3.1.1 The Growth Pattern of Palmarosa

The height of palmarosa plants exhibited a linear growth pattern over 16 weeks across all gamma-ray irradiation doses. Intermediate doses showed a greater stimulatory effect on vegetative growth, whereas higher doses resulted in lower or control-like responses, indicating a dose-dependent hormetic effect. Similar trends have been reported in other crops, such as quinoa, where moderate gamma irradiation enhanced plant height compared to higher doses (Song *et al*., 2021).

Figure 1. The growth pattern of Palmarosa



3.2 Number of Leaves

Based on table 3. revealed that gamma-ray irradiation significantly affected the number of leaves in palmarosa plants at the 5% significance level (F = 4.282; p = 0.004). This indicates that irradiation dose plays an important role in altering vegetative development, particularly leaf formation. The block effect was also significant (p = 0.007), suggesting acceptable experimental variability among replications.

Table 3. ANOVA for Number of Leaves

Source	Type III Sum of Squares	Df	Mean Square	F	Sig.
Corrected Model	8949,891[a]	11	813,626	4,818	0,001
Intercept	4395423,042	1	4395423,042	26026,632	0,000
Treatments	5062,357	7	723,194	4,282	0,004
Block	3107,170	4	776,792	4,600	0,007
Error	3884,280	23	168,882		
Total	4681551,000	35			
Corrected Total	12834,171	34			

a. R Squared = ,697 (Adjusted R Squared = ,553)

The DMRT further classified the irradiation treatments into two homogeneous subsets. Lower mean leaf numbers were observed in the control and 80 Gy treatments (approximately 346–348 leaves), whereas higher doses ranging from 60 Gy to 150 Gy formed a second subset with increased leaf numbers (approximately 365–386 leaves). The highest leaf number was recorded at 150 Gy, followed by 120 Gy and 40 Gy, indicating a stimulatory effect of moderate to relatively high irradiation doses on leaf production.

Table 4. DMRT for Number of Leaves

Duncan[a,b]

Doses	N	Subset for alpha = 0.05	
		1	2
80 Gy	5	346,60	
0 Gy	5	348,40	
60 Gy	4	364,75	364,75
100 Gy	5	365,00	365,00
20 Gy	4	369,25	369,25
40 Gy	3	371,00	371,00
120 Gy	5		378,20
150 Gy	4		385,75
Sig.		0,060	0,104

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 4,248.
b. The group sizes are unequal. The harmonic mean of the group sizes is used. Type I error levels are not guaranteed.

The variation in leaf number among irradiation treatments is likely associated with radiation-induced mutations affecting cell division and organ differentiation. Gamma irradiation induces DNA double-strand breaks and oxidative stress, which may result in heritable mutations after imperfect DNA repair. These mutations can modify the expression of genes regulating shoot apical meristem activity and leaf initiation, thereby increasing leaf production. From a cytological perspective, enhanced leaf formation at certain doses may reflect increased mitotic activity in meristematic tissues, accompanied by non-lethal chromosomal alterations. Previous cytogenetic studies have demonstrated that specific gamma-ray doses can stimulate vegetative growth by increasing mitotic index while maintaining chromosomal stability within a tolerable range (Sayed *et al*., 2025). Therefore, the observed increase in leaf number represents a biologically meaningful response to gamma-ray-induced genetic variation and highlights the potential of irradiation for mutation breeding programs.

3.3 Tiller Numbers

Based on Table 5. demonstrated that gamma-ray irradiation significantly affected the number of tillers in palmarosa at the 5% significance level ($F = 2.864$; $p = 0.026$). This indicates that variation in irradiation dose altered vegetative growth responses, particularly tiller formation, which is a key determinant of plant biomass. The coefficient of determination ($R^2 = 0.534$) suggests that a substantial proportion of the variation in tiller number was explained by the applied treatments.

Table 5. ANOVA for Tiller Numbers

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	3859,377[a]	11	350,852	2,396	0,037
Intercept	553316,962	1	553316,962	3779,365	0,000
Treatments	2935,257	7	419,322	2,864	0,026
Block	711,691	4	177,923	1,215	0,331
Error	3367,309	23	146,405		
Total	593797,000	35			
Corrected Total	7226,686	34			

a. R Squared = ,534 (Adjusted R Squared = ,311)

The DMRT further classified the treatments into two homogeneous subsets. Higher irradiation doses (150 and 100 Gy) were associated with the lowest mean tiller numbers (approximately 115–116 tillers), whereas lower to moderate doses (0– 60 Gy) resulted in higher tiller production (approximately 135–143 tillers). The highest mean tiller numbers were observed at 20 Gy, indicating a stimulatory effect of low-dose gamma irradiation.

Table 6. DMRT for Tiller Numbers

Duncan[a,b]

Doses	N	Subset for alpha = 0.05	
		1	2
150 Gy	4	115,00	
100 Gy	5	116,20	
120 Gy	5	126,80	126,80
80 Gy	5	128,40	128,40
60 Gy	4		135,00
0 Gy	5		137,40
40 Gy	3		139,00
20 Gy	4		142,50
Sig.		0,157	0,111

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 4,248.
b. The group sizes are unequal. The harmonic mean of the group sizes is used. Type I error levels are not guaranteed.

This response in tiller number, with low to moderate gamma irradiation doses (0–60 Gy) producing higher tiller counts than high doses (100–150 Gy), consistent with radiation hormesis. Low-dose gamma irradiation may stimulate meristematic activity through non-lethal DNA mutations arising from imperfect repair of double-strand breaks, thereby promoting tiller initiation. In contrast, reduced tiller formation at higher doses is likely associated with increased chromosomal aberrations and suppressed mitotic activity in meristematic tissues. Similar responses have been reported in rice, where low to moderate gamma doses enhanced tillering and vegetative growth, whereas higher doses inhibited growth due to physiological stress and cellular damage (Lasmana *et al.*, 2025). These results indicate that variation in tiller number represents a biologically meaningful response to gamma-induced genetic variation and supports its use as a selection criterion in mutation breeding programs.

4. CONCLUSION

Gamma-ray irradiation significantly affected plant height, number of leaves, and number of tillers of palmarosa based on ANOVA and DMRT results. Low to moderate irradiation doses (particularly around 40–60 Gy) resulted in the most favorable vegetative growth, whereas higher doses inhibited plant development. This response reflects radiation hormesis, in which optimal doses stimulate meristematic activity. The induced vegetative variability provides valuable material for mutation breeding programs and serves as a basis for further cytological studies to elucidate chromosome alterations induced by gamma irradiation.

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