



Effect of Gamma Radiation on M₁ Generation of Two Aromatic Rice Varieties and Determination of Their Lethal Dose

**Md. Tazul Islam ^{a,b}, Most. Shirina Akter ^{a*},
Mossammot Moriom ^a, Mohammad Hasanuzzaman Rani ^b
and Ferdaous Jahan Urmi ^c**

^a Department of Agricultural Extension (DAE), Ministry of Agriculture, Dhaka-1200, Bangladesh.

^b Bangladesh Institute of Nuclear Agriculture (BINA), Mymensingh-2202, Bangladesh.

^c Department of Plant Pathology, Sher-e-Bangla Agricultural University, Dhaka-1207, Bangladesh.

Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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Abstract

Gamma irradiation is a potent tool for inducing genetic variation in aromatic rice. This study evaluated the effects of gamma radiation on the M₁ generation of two aromatic rice varieties, Chinigura (local) and Banglamati (released), and determined their lethal dose (LD₅₀) based on plant survival. One hundred grams of seeds from each variety were irradiated at 0, 100, 200, 250, 300, and 400 Gy at the laboratory of the Bangladesh Institute of Nuclear Agriculture (BINA). The experiment followed a split-plot design with three replications and was conducted at the BINA Sub-station, Sunamganj, Bangladesh. Before irradiation, germination tests were performed under laboratory conditions (27 ± 2°C). Increasing radiation doses caused marked reductions in germination percentage, seedling height, fresh and dry biomass, tiller number, and

*Corresponding author: E-mail: aktershirin434@gmail.com;

survival. At 400 Gy, germination declined from 100% in the control to 87.0% in Chinigura and 81.3% in Banglamati. By 21 DAS, seedling height decreased by 16.7% and 19.4%, fresh weight by 51.0% and 45.6%, and dry matter by 48% in both varieties. Tiller number was reduced by nearly 50% at the highest dose. Survival declined linearly with increasing doses, and regression analysis estimated LD₅₀ values of 205.74 Gy for Chinigura and 325.92 Gy for Banglamati, indicating greater radiosensitivity in Chinigura and higher tolerance in Banglamati. These LD₅₀ values provide a key reference for mutation breeding and varietal improvement in aromatic rice.

Keywords: Mutation; gamma rays; aromatic rice; M1 generation; lethal dose (LD₅₀).

1. Introduction

Rice (*Oryza sativa* L.) is a staple cereal crop that sustains nearly half of the world's population (Golestan Hashemi et al., 2013; Fukagawa et al., 2019). Global production has steadily increased, currently reaching around 795–820 million metric tons annually (FAO, 2024). Bangladesh ranks third globally in both rice cultivation area and production (BRRI, 2025), with approximately 75% of its cropped area devoted to rice. In 2023–24, the country produced 40.69 million metric tons from 11.66 million hectares (BBS, 2025). Although aromatic rice is highly prized for its superior quality and commands a higher market price (Hasibuzzaman et al., 2025), it occupies only a small fraction of Bangladesh's rice area, with production reaching just 1.023 million tonnes in 2023–24 (The Daily Star, 2024). Its limited cultivation is attributed to low yield, lodging susceptibility, and longer growth duration compared to non-aromatic released varieties (Imam & Chakraborty, 2018). To enhance yield and shorten cultivation periods, mutation breeding has emerged as an effective approach (Solim & Rahayu, 2021).

Mutation is a sudden, heritable change in genetic material at the gene or chromosome level (Chahal & Gosal, 2002). It generates genetic diversity, enabling breeders to select desirable traits (Mohamad et al., 2005) and improve seedling height, survival, and tiller production (Mohamad et al., 2002a). Mutation breeding can also shorten crop duration by 30–35 days (Azad et al., 2012). Mutations can be induced using physical mutagens such as alpha, beta, gamma, and X-rays, or chemical mutagens like EMS, MMS, 5-bromouracil, and 5-chlorouracil. Among these, gamma rays are one of the most widely used mutagens (Bordoloi et al., 2023).

Gamma rays are electromagnetic radiation capable of interacting with cellular molecules and modifying or damaging cell structures (Minisi et al., 2013). They induce cytological, genetic, biochemical, and physiological changes in plant cells, thereby affecting development and improvement (Ashraf et al., 2003; Akshatha et al., 2013). Determining the lethal dose (LD₅₀) is essential for applying gamma rays in crop improvement, as it helps identify doses that maximize genetic variation (Harding et al., 2012). Several studies have investigated effective gamma doses for inducing desirable traits in rice (Ray et al., 2022; Solim & Rahayu, 2021; Sharma et al., 2020; Gowthami et al., 2016). Therefore, the present study aimed to evaluate the effects of gamma irradiation on two aromatic rice varieties—one local and one released—and to determine the effective gamma doses for generating genetic variation to support varietal improvement programs.

2. Materials and Methods

The experiment was conducted at the Bangladesh Institute of Nuclear Agriculture (BINA), Sub-station, Sunamganj, Bangladesh, from July to October 2020. Two aromatic rice cultivars, Chinigura (local) and Banglamati (released), were used in the study. Chinigura is a photosensitive, tall, long-duration, and low-yielding variety, whereas Banglamati is a photo-insensitive, dwarf, long-duration, and medium-yielding variety. Prior to irradiation, a germination test was performed to determine the germination percentage and seed viability. Thirty seeds from each genotype were placed in Petri dishes lined with water-soaked filter paper and kept at normal room temperature and in a growth chamber.

One hundred pure, healthy, dry seeds from each cultivar were irradiated with gamma rays at doses of 0, 100, 200, 250, 300, and 400 Gy. After irradiation, the seeds were soaked and placed on moist blotting paper in Petri dishes to assess germination under laboratory conditions (27 ± 2°C). The filter paper was kept moist with distilled water as needed. Germination percentage was recorded after seven days.

The germinated seeds were then transferred to trays containing soil. Plant height was measured at 14 and 21 days after sowing (DAS), and plant survival percentage was recorded at 21 DAS. At 21 DAS, five plants from each radiation dose were carefully uprooted, washed to remove soil, and their fresh weight recorded. The samples were then oven-dried at 90°C for more than 48 hours and weighed again to determine dry weight.

Seedlings that survived after 21 days were transplanted to the field following a randomized complete block design (RCBD) with three replications for each radiation dose. Transplanting was done in rows at a spacing of 15 × 20 cm, with one seedling per hill. At flowering, the total number of tillers per hill was counted for each replication at each dose. Data were analyzed using Microsoft Office Excel 2007. Plant survival percentage was calculated using the following formula:

$$\text{Plant survival \%} = \frac{\text{No. of survived seedlings at 21 DAS}}{\text{Total number of germinated seeds transferred to tray at 7 DAS}} \times 100$$

LD50 were calculated at 21 DAS by regression analysis.

Specific regression model used: $Y=a+bX$

Where:

- Y = plant survival percentage
- X = gamma radiation dose (Gy)
- a = intercept (survival percentage at 0 Gy)
- b = regression coefficient (rate of change in survival percentage per unit increase in dose)

3. Results and Discussion

3.1 Germination Percentage

Germination was 100% at 0 Gy for both Chinigura and Banglamati, declining progressively with higher gamma doses. In Chinigura, it decreased from 95.67% (100 Gy) to 87.00% (400 Gy), while Banglamati declined from 96.00% to 81.33%, showing sharper reductions. Mean germination was slightly higher in Chinigura (93.00%) than Banglamati (91.94%), with greater variability in Banglamati (SD 6.77 vs 4.52). Similar dose-dependent declines were reported in Kalanamak rice (Mishra et al., 2023) and prior studies (Harding et al., 2012; Cheema & Atta, 2003).

Table 1. Effect of gamma ray irradiation on germination of Chinigura and Banglamati

Dose (GY)	Chinigura		Banglamati	
	Germ. %	% over control	Germ. %	% over control
0	100.00	100.00	100.00	100.00
100	95.67	95.67	96.00	96.00
200	93.33	93.33	94.33	94.33
250	92.00	92.00	93.33	93.33
300	90.00	90.00	86.67	86.67
400	87.00	87.00	81.33	81.33
Mean	93.00		91.94	
SD	4.521553		6.77386	

3.2 Seedling Height

Higher gamma doses consistently suppressed seedling height in Chinigura and Banglamati at 14 and 21 days after sowing (DAS). At 14 DAS, Chinigura declined from 5.50 cm (0 Gy) to 4.17 cm (400 Gy, 24.12% reduction), while Banglamati dropped from 6.60 cm to 4.25 cm (35.61%), showing less consistency. At 21 DAS, Chinigura decreased from 9.67 cm to 8.06 cm, and Banglamati from 10.17 cm to 8.20 cm. Banglamati seedlings were taller but more variable. Kant et al. (2020) found plant height largely unaffected at low gamma

doses but markedly reduced at higher doses. Rani et al. (2016) found that the seedling height of both Ashfal and Binadhan-14 decreased progressively with increasing doses of gamma radiation. At 14 days after sowing, the seedling height of Ashfal ranged from 17.41 cm in the control (0 Gy) to 0.82 cm at 400 Gy, showing a reduction of over 95%. Similarly, Binadhan-14 seedlings decreased from 12.14 cm in the control to 3.70 cm at 400 Gy, representing a reduction of approximately 70%. The mean seedling height across all gamma ray treatments was 11.34 cm for Ashfal and 9.66 cm for Binadhan-14, indicating that higher doses of gamma radiation significantly inhibited early seedling growth. Similarly, Rani et al. (2016), Harding et al. (2012), and Faustino et al. (2024) reported gradual declines in rice seedling height with increasing radiation.

Table 2. Effect of gamma ray irradiation on seedling height at 14 and 21 days after sowing on Chinigura and Banglamati

Dose (GY)	Chinigura		Banglamati		Chinigura		Banglamati	
	Seedling ht. at 14 days (cm)	% over control	Seedling ht. at 14 days (cm)	% over control	Seedling ht. at 21 days (cm)	% over control	Seedling ht. at 21 days (cm)	% over control
0	5.50	100.00	6.60	100.00	9.67	100.00	10.17	100.00
100	5.08	92.42	6.34	96.11	9.56	98.83	9.73	95.71
200	4.66	84.79	6.25	94.70	9.33	96.52	9.50	93.41
250	4.50	81.82	5.03	76.26	9.17	94.79	9.23	90.79
300	4.37	79.39	5.00	75.76	9.11	94.21	9.17	90.13
400	4.17	75.88	4.25	64.39	8.06	83.32	8.20	80.63
Mean	4.71		5.58		9.15		9.33	
SD	0.493057		0.946163		0.576745		0.663995	

3.3 Fresh Weight

Fresh weight of Chinigura and Banglamati declined progressively with increasing gamma radiation doses (Table 3). Chinigura exhibited a more pronounced reduction, losing 51.01% of fresh weight at 400 Gy, whereas Banglamati showed a 45.56% decrease compared with the control (0 Gy). The reduction trend was approximately dose-dependent and closely paralleled the decrease observed in seedling height. Across all irradiation levels, Chinigura consistently recorded lower fresh weight than Banglamati, indicating greater sensitivity to gamma exposure. Similarly, Gupta et al. (2021) reported that the shoot fresh weight was highest in Binadhan-17 at 100 Gy and in Galon at 200 Gy; however, it decreased with increasing levels of gamma radiation.

Table 3. Effect of gamma ray irradiation on fresh weight of Chinigura and Banglamati at 21 DAS

Dose (GY)	Chinigura		Banglamati	
	Fresh wt. (g)	% over control	Fresh wt. (g)	% over control
0	2.81	100.00	3.00	100.00
100	2.30	81.85	2.85	95.00
200	2.01	71.53	2.53	84.22
250	1.50	53.26	2.10	70.00
300	1.43	51.01	1.99	66.44
400	1.38	48.99	1.63	54.44
Mean	1.90		2.35	
SD	0.575224		0.530477	

3.4 Dry Matter Accumulation

Dry matter accumulation decreased proportionally with increasing gamma radiation doses in both Chinigura and Banglamati (Table 4). At 400 Gy, both varieties exhibited a 48% reduction in dry weight compared to the control. Throughout all treatments, Chinigura consistently recorded lower dry matter values (0.07–0.13 g) than Banglamati (0.08–0.15 g), reflecting similar declines observed in seedling height and fresh weight. The reduction pattern suggests a dose-dependent inhibitory effect of gamma irradiation on biomass production.

Lasmana et al. (2025) reported that lower doses, particularly 50 Gy and 100 Gy, induced a hormetic effect by enhancing germination, plant height, and tillering, whereas higher doses (>150 Gy) significantly reduced growth due to physiological and genetic damage.

Table 4. Effect of gamma ray irradiation on Dry matter accumulation of Chinigura and Banglamati at 21 DAS

Dose (GY)	Chinigura		Banglamati	
	Dry wt. (g)	% over control	Dry wt. (g)	% over control
0	0.13	0.13	0.15	0.15
100	0.11	82.50	0.14	94.67
200	0.10	75.00	0.13	87.78
250	0.08	61.25	0.11	70.67
300	0.08	60.00	0.10	66.67
400	0.07	52.00	0.08	52.00
Mean	0.10		0.12	
SD	0.023571		0.027766	

3.5 Tiller Production

Tiller production decreased with increasing gamma doses, showing a non-proportional, zigzag trend (Fig. 1). In Chinigura, tillers dropped significantly at 100 Gy and reached about 50% of control at 400 Gy, while in Banglamati, significant reduction occurred at 200 Gy, also declining to 50% at 400 Gy. This aligns with Harding et al. (2012), who reported reduced tillering with higher radiation.

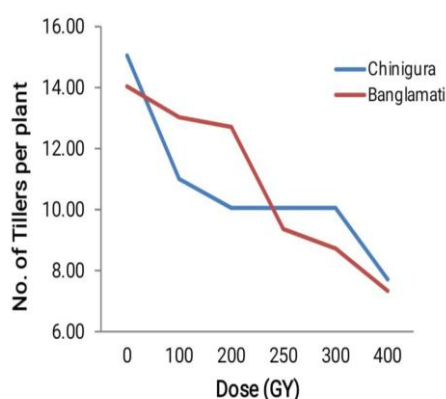


Fig. 1. Effect of gamma ray irradiation on tiller production

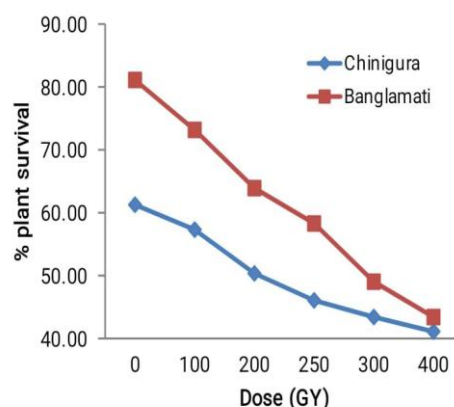


Fig. 2. Effect of gamma ray irradiation on survival percentage

3.6 Survival Percentage

Survival percentage of both varieties decreases with increasing gamma radiation doses (Fig. 2). Banglamati consistently shows higher survival than Chinigura across all doses. At 400 Gy, survival drops to about 43% in Banglamati and 41% in Chinigura, indicating greater tolerance in Banglamati. Increasing gamma radiation reduced survival percentage, a trend observed by Wijesena et al. (2019) and similarly reported in aromatic rice by Harding et al. (2012).

3.7 Determination of Lethal Dose and Relative Biological Value (RBE)

The LD₅₀ value, representing the gamma dose at which 50% of plants fail to survive, was determined based on plant survival percentages (Table 5). Chinigura required a lower gamma dose (LD₅₀ = 205.74 Gy) than the released variety Banglamati (LD₅₀ = 325.92 Gy) to achieve the relative biological effect (RBE) 1.58 and 1.00, respectively (Table 5), indicating higher sensitivity. Similarly, Rani et al. (2016) reported that the local variety

Ashfal had lower LD₅₀ and LD₃₀ values (241.68 Gy and 153.19 Gy) than the improved variety Binadhan-14 (353.29 Gy and 254.33 Gy), with correspondingly higher RBE values (1.12–1.29 vs. 1.02–1.05). These observations consistently show that local varieties are more radiosensitive due to limited genetic tolerance, whereas improved or released varieties are more resilient, requiring higher doses for the same biological effect. This pattern highlights the influence of genetic background and breeding history on varietal radiosensitivity. Harding et al. (2012) reported that LD₅₀ doses for rice range from 345 to 423 Gy, while Sharma et al. (2020) suggested an optimal gamma dose range of 280 to 350 Gy. Ramchander et al. (2015) observed that LD₅₀ values varied among rice varieties, with 354.80 Gy for White Ponni and 288.40 Gy for BPT 5204, highlighting varietal differences in sensitivity to gamma irradiation; similarly, Gowthami et al. (2017) reported that for ADT-37 and ADT(R) 45, the LD₅₀ doses were 300.03 Gy and 300.00 Gy, respectively.

Table 5. lethal dose (LD50) of the two varieties Chinigura and Banglamati

Variety	Regression Equation	LD50	RBE
Chinigura	$Y = 61.11 - 0.054x$	205.74	1.58
Banglamati	$Y = 81.94 - 0.098x$	325.92	1.00

4. Conclusion

The study demonstrated that increasing gamma irradiation doses negatively affected the growth and development of both Chinigura and Banglamati rice varieties. The LD₅₀ values, calculated from survival percentage, were 205.74 Gy for Chinigura and 325.92 Gy for Banglamati, confirming higher radiosensitivity in the local variety. Overall, the results highlight a clear dose-dependent suppression of growth parameters, with Chinigura being more sensitive. These findings provide critical information for determining optimal gamma doses for mutation breeding and varietal improvement programs in aromatic rice.

Disclaimer (Artificial Intelligence)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc.) and text-to-image generators have been used during the writing or editing of this manuscript.

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Competing Interests

Authors have declared that no competing interests exist.

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