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Effects of gamma irradiation on growth and productivity in dose-dependent response of Indian mustard (*Brassica juncea* L.) genotypes

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Abstract

Gamma irradiation is a widely used mutagenic technique in plant breeding, employed to induce genetic variations for crop improvement. In this study, the effects of different doses of gamma irradiation (0, 5, 10, 15, 20 and 25 kR) on the morphological, phenological, yield, and quality traits of two Indian mustard (*Brassica juncea* L.) genotypes, P13RGN-303 and RH-761, were investigated. The study focused on key parameters such as field emergence, survival rate, plant height, days to 50% flowering, siliqua length, siliqua per plant, number of seeds per siliqua, seed yield, test weight and oil content. Significant variations were observed across the different radiation treatments, with the highest performance noted at 20 kR for both genotypes. The optimal doses of gamma irradiation (20 kR) resulted in increased field emergence and survival rates, indicating improved seed vigour. Furthermore, plant height and days to 50% flowering were favorably affected, suggesting enhanced growth and early maturity. Siliqua traits, including siliqua length and seed number, as well as seed yield (per plant, plot, and hectare), were all significantly improved, reflecting increased reproductive efficiency. The highest test weight and oil content were recorded at 20 kR, signaling enhanced seed quality. Conversely, higher doses (25 kR) led to a reduction in growth and yield, likely due to excessive radiation stress. Overall, the study demonstrates the potential of gamma irradiation at 20 kR for improving mustard traits, providing valuable insights for crop improvement initiatives.

Keywords: *Brassica juncea, gamma irradiation, growth, Indian mustard, mutagenesis, yield*

Introduction

Brassica species, including *Brassica napus*, *B. rapa*, and *B. juncea*, are essential oilseed crops cultivated across 11 million hectares worldwide under diverse climatic conditions. Among these, *Brassica juncea* is extensively grown in the Indian subcontinent, where it serves multiple purposes, including as a source of edible oil, a condiment, a lubricant, and a component of cattle feed and fertilizers. Given its agronomic and economic significance, *B. juncea* breeding programs focus on enhancing yield potential, disease resistance, and quality traits. Conventional breeding methods have played a crucial role in genetic improvement; however, recent advancements in nonconventional approaches such as mutation induction, tissue culture, and molecular genetics have significantly contributed to trait enhancement (Tiliouine *et al.,* 2018; Gupta 2019; Lal *et al.,* 2020).

One of the most effective nonconventional breeding techniques is gamma radiation, a form of ionizing radiation that induces cytological, biochemical, physiological, and morphological changes in plant cells and tissues. Gamma irradiation has been widely used in plant breeding programs to generate novel genetic variations, leading to improved stress tolerance, enhanced growth, and increased yield potential (Moghaddam *et al*., 2011; Celik and Atak, 2017). The interaction of gamma rays with plant cells triggers the production of reactive oxygen species (ROS) such as superoxide radicals, hydroxyl radicals, and hydrogen peroxide, which alter cellular structures and metabolic functions (Esfandiari *et al*., 2007). While high doses of gamma radiation can cause severe physiological damage, including chromosomal aberrations and impaired photosynthesis, low doses have been shown to stimulate growth, enhance secondary metabolite production, and improve stress resilience by activating specific physiological pathways (Aly, 2010; Vardhan and Shukla, 2017).

In mustard (*B. juncea*), gamma radiation has been explored as a means to address key challenges such as low genetic diversity, vulnerability to abiotic stresses, and limited yield potential. Studies have demonstrated that low doses of gamma irradiation can improve photosynthetic efficiency, increase chlorophyll content, and enhance stress tolerance mechanisms (Kulandaivelu and Noorudeen, 1983; Wi *et al*., 2005). Additionally, chlorophyll fluorescence analysis has emerged as a reliable tool for assessing plant health and photosynthetic performance under stress conditions (Sousaraei *et al*., 2021; Esmaeili *et al*., 2022). Given the high nutritional and economic value of mustard oil, which is rich in omega-3 and omega-6 fatty acids with significant health benefits, the development of improved mustard varieties through gamma irradiation is of paramount importance (Gupta *et al*., 2014 and Bhatia *et al*., 2021).

This study aims to evaluate the mutagenic effectiveness and efficiency of different doses of gamma radiation on in vitro regeneration and physiological traits of *B. juncea*. By examining dose-dependent responses, the research seeks to determine the optimal radiation levels that induce beneficial genetic variations while minimizing deleterious effects. The findings will contribute to the development of superior mustard genotypes with enhanced agronomic performance, thereby supporting sustainable agricultural practices and global food security.

Materials and Methods

This study aimed to evaluate the effect of gamma irradiation on growth, yield, and oil content in two mustard (*Brassica juncea*) genotypes, P13RGN-303 and RH-761, under field conditions. The research was conducted during the Rabi season of 2023–24 at the Field Experimentation Centre, Department of Genetics and Plant Breeding, Naini Agricultural Institute, Sam Higginbottom University of Agriculture, Technology and Sciences (SHUATS), Prayagraj, Uttar Pradesh, India. The mustard seeds were procured from ICAR-Directorate of Rapeseed-Mustard Research (ICAR-DRMR), Bharatpur, Rajasthan, India. The seeds were irradiated with gamma rays at the National Botanical Research Institute (CSIR-NBRI), Lucknow, using a GIC-1200 model gamma irradiator equipped with a Cobalt-60 (^60Co) radioactive source. The treatments included a control (T_0 - unirradiated seeds) and five gamma irradiation doses: 5 kR (T_1), 10 kR (T_2), 15 kR (T_3), 20 kR (T_4), and 25 kR (T_5).

The experiment was laid out in a Randomized Block Design (RBD) with six treatments, each replicated three times. Each plot had a uniform size of 2 × 2 m² with a seed rate of 5 kg per hectare and a row-to-row spacing of 30 cm, while the plant-to-plant spacing was 10 cm to ensure optimal growth conditions. The experimental soil was sandy loam in texture, with a pH of 7.1, electrical conductivity of 0.37 dS/m, organic carbon content of 0.50%, available nitrogen of 189.6 kg/ha, available phosphorus of 12.96 kg/ha, and available potassium of 225.7 kg/ha. Standard agronomic practices, including irrigation, thinning, weeding, and pest management, were uniformly followed across all plots.

The survival rate of mustard plants under different gamma irradiation doses was assessed using the exponential survival model (FAO, 2013):

$$S(D) = S_0 * e^{-kD}$$

where S(D) represents the survival rate at a given radiation dose D, S0 is the survival rate in the control treatment, and k is the survival decline constant. Oil content was estimated using the AOAC method, where oil was extracted using a Soxhlet apparatus with petroleum ether as the solvent. The oil percentage was calculated using the formula:

Oil Content (%) = (Weight of extracted oil / weight of seed sample) * 100

Data for survival rate, growth parameters, yield, and oil content were statistically analyzed using analysis of variance (ANOVA) for a randomized block design (RBD), with significance determined at a 5% probability level (P = 0.05) using the Least Significant Difference (LSD) test. Statistical software was used for precision in data analysis. Observations were systematically recorded at critical growth stages, including germination, seedling establishment, vegetative growth, flowering, pod formation, and physiological maturity. The recorded parameters included germination percentage, plant survival rate, seedling vigor, plant height, number of branches per plant, days to 50% flowering, pod formation percentage, seed yield per plant and per plot, and oil content percentage. The study provides insights into the impact of gamma irradiation on mustard genotypes and its potential application in crop improvement through induced mutagenesis.

Results and Discussion

Gamma irradiation is widely utilized in mutation breeding to induce genetic variability, enhance stress tolerance, and improve agronomic traits in crops. Its impact on plant growth and yield is mediated through complex physiological, biochemical, and molecular changes rather than a direct effect on seed yield. The extent of these alterations depends on the radiation dose, with moderate levels often stimulating beneficial mutations and metabolic processes, while excessive doses may induce oxidative stress, DNA damage, and metabolic dysfunctions. This study evaluated the response of two mustard genotypes, P13RGN-303 and RH-761, to varying doses of gamma irradiation, with a focus on growth attributes, reproductive traits, and seed yield potential (Tables 1 to 4).

Table 1 : Analysis of variance on effect of different doses of Gamma irradiation on Indian mustard (*Brassica juncea* L.) Genotype P13RGN-303

Characters	Mean Sum of Square		
	Treatment (d.f=5)	Replication (d.f=2)	Error (d.f=10)
Field emergence	23.3*	2.00	4.40
Survival rate	26.93*	2.16	7.50
Plant height	22.04*	34.26	4.61
Days to 50% flowering	48.35*	34.72	6.79
Siliqua length	0.27*	0.00	0.01
Siliqua/plant	363.68*	338.74	1,088.61
Number of seeds/ Siliqua	0.27*	0.00	0.01
Seed yield/Plant	363.68*	338.74	108.86
Seed yield/Plot	3484.06*	504.39	211.79
Seed yield/Hectare	3.87*	0.56	0.24
Test weight	0.07*	0.34	0.02
Days to maturity	33.55*	7.39	7.92
Oil content	8.29*	4.66	2.07

*Indicates significant at 5% level of significance.

Table 2 : Analysis of variance on effect of different doses of Gamma irradiation on Indian mustard (*Brassica juncea* L.) Genotype RH-761.

Characters	Mean Sum of Square		
	Treatment(d.f=5)	Replication(d.f=2)	Error(d.f=10)
Field emergence	10.23*	0.67	2.60
Survival rate	6.22*	8.22	1.49
Plant height	22.58*	102.13	2.31
Days to 50% flowering	6.89*	9.06	1.46
Siliqua length	0.10*	0.01	0.56
Siliqua/plant	159.45*	1,474.46	47.16
No. of seeds/ Siliqua	1.58*	0.11	0.47
Seed yield/Plant	16.05*	22.07	3.46
Seed yield/Plot	34,491.97*	52.67	99.13
Seed yield/Hectare	3.88*	0.06	0.11
Test weight	0.28*	0.05	0.09
Days to maturity	13.69*	5.06	2.86
Oil content	9.83*	1.50	2.83

*Indicates significant at 5% level of significance.

Field emergence (FE) and survival rate (SR) showed significant variation among treatments, reflecting the effect of gamma irradiation on seed viability and seedling vigor (Tables 3 and 4). In P13RGN-303, the highest FE (85.0%) and SR (82.7%) were recorded at 20 kR, whereas the lowest values (76.67% and 74.00%, respectively) occurred at 25 kR. A similar pattern was observed in RH-761, where 20 kR resulted in the highest FE (78.33%) and SR (75.67%). The reduction at 25 kR can be attributed to excessive radiation-induced oxidative stress and impaired cellular function, as described by Alikamanoglu *et al.* (2007) and Ali *et al.* (2019). In contrast, moderate doses may enhance germination and seedling establishment by activating DNA repair pathways and antioxidant defense mechanisms (Khurana *et al.,* 2020; Hamideldin and Eliwa, 2015).

Plant height and days to 50% flowering were significantly affected by gamma irradiation. The tallest plants (177 cm) in P13RGN-303 were observed at 15 kR, while the shortest plants (169 cm) occurred at 25 kR. Similarly, RH-761 exhibited maximum height (158 cm) at 20 kR, with a significant reduction at 25 kR (150 cm). These findings

Table 3: Mean performance on effect of different doses of Gamma irradiation on morphological and yield components of mustard (*Brassica junceae*) Genotype P13RGN-303

Treatment	FE (%)	SR (%)	PH (cm)	50% DOF	SL (cm)	SPP	NSPS	SYPP (g)	SYPP (kg)	SY (ha)	TW (g)	DM	OC (%)
T_0	81.67	79.33	175.04	58.67	5.45	271.28	12.17	23.74	560.00	18.67	4.13	138	38.48
T_1	79.67	77.00	174.29	55.67	5.23	267.17	11.83	21.75	550.67	18.36	3.63	139	37.65
T_2	80.00	80.00	174.07	60.00	5.25	262.55	12.47	24.31	566.67	18.89	3.93	141	39.32
T_3	82.00	77.00	177.81	53.33	5.33	271.47	12.72	23.42	576.67	19.22	4.17	138	39.69
T_4	85.00	82.67	177.51	51.33	5.80	272.08	12.85	24.66	614.00	20.47	4.33	136	41.29
T_5	76.67	74.00	169.82	61.67	4.88	243.44	11.34	21.66	509.67	16.99	3.57	146	36.54
CD (5%)	5.07	5.45	5.28	7.31	0.55	20.03	1.04	2.32	62.00	2.07	0.56	6.37	3.02
CV (%)	3.45	3.83	1.66	7.08	5.66	4.16	4.68	5.49	6.05	6.05	7.76	2.51	4.28

Legends: FE-Field emergenc e, SR-Survival rate, PH-Plant height, DOF-Days of flowering, SL-Siliqua length, SPP-Siliqua per plant, NSPS=Number of seeds per siliqua, SYPP-Seed yield per plant, SYPP-Seed yield per plot, SY-Seed yield, TW-Test weight, OC-Oil content, DM-days to maturity,T_0-Control, T_1-5kR, T_2-10kR, T_3-15kR, T_4-20kR, T_5-25kR treatment of Gamma rays

Table 4 : Mean performance on effect of different doses of Gamma irradiation on morphological and yield components of mustard (*Brassica junceae*) Genotype RH-761

Treatment	FE (%)	SR (%)	PH (cm)	50% DOF	SL (cm)	SPP	NSPS	SYPP (g)	SYPP (kg)	SYPH	TW (g)	DM	OC (%)
T_0	76.00	73.33	155.40	77.33	5.26	131.22	12.17	22.92	493.66	16.46	3.03	143	34.33
T_1	74.67	72.67	154.11	76.67	5.20	135.53	12.17	24.38	475.33	15.84	3.00	143	32.33
T_2	76.00	72.33	155.50	75.33	5.30	131.08	11.59	22.67	481.00	16.03	3.27	142	33.00
T_3	77.00	74.00	152.00	76.00	5.32	136.78	12.50	24.20	492.33	16.41	3.17	141	35.33
T_4	78.33	75.67	158.39	73.00	5.34	140.33	13.50	26.00	561.66	18.72	3.17	140	36.33
T_5	73.00	70.33	150.70	76.33	5.13	119.33	11.50	19.18	467.00	15.57	2.83	146	31.67
CD (5%)	4.87	5.22	5.10	6.95	0.48	11.36	1.02	2.15	56.70	1.94	0.41	5.23	2.97
CV (%)	3.25	3.67	2.14	6.89	4.92	4.08	4.52	5.12	5.87	5.79	6.72	2.27	4.11

Legends: FE-Field emergence, SR-Survival rate, PH-Plant height, DOF-Days of flowering, SL-Siliqua length, SPP-Siliqua per plant, NSPS-Number of seeds per siliqua, SYPP-Seed yield per plant, SYPP-Seed yield per plot, SY-Seed yield, TW-Test weight, OC-Oil content, DM-days to maturity, T_0-Control, T_1-5kR, T_2-10kR, T_3-15kR, T_4-20kR, T_5-25kR treatment of Gamma rays.

align with earlier reports by Sharma et al. (2017), indicating that moderate radiation doses enhance auxin biosynthesis and promote cell elongation, whereas excessive exposure disrupts mitotic activity and reduces plant height. Early flowering was recorded at 20 kR for both genotypes (51 days in P13RGN-303; 73 days in RH-761), while 25 kR delayed flowering. Gamma irradiation at optimal levels may modulate hormonal homeostasis, particularly gibberellins and cytokinins, accelerating floral initiation and shortening the vegetative phase (Guangyaol *et al.*, 2007 and Ahmed *et al.*, 2018).

Siliqua-related traits, including siliqua length (SL), siliqua per plant (SPP), and the number of seeds per siliqua (NSPS), were significantly influenced by irradiation. In P13RGN-303, the highest SL (5.8 cm), SPP (272.08), and NSPS (12.85) were recorded at 20 kR, while similar improvements were observed in RH-761. However, 25 kR caused a decline in these traits, likely due to radiation-induced disruptions in reproductive development, such as ovule abortion and reduced pollen viability, as reported by Garg *et al.* (2022). Moderate doses, however, may induce beneficial genetic variations that enhance seed set and assimilate partitioning.

Seed yield per plant (SYPP), seed yield per plot (SYPPl), and seed yield per hectare (SY) exhibited significant improvements at 20 kR. In P13RGN-303, the highest SYPP (24.66 g), SYPPl (614.00 kg), and SY (20.47 q/ha) were recorded at 20 kR, whereas 25 kR resulted in the lowest values. A similar pattern was observed in RH-761, with peak yield recorded at 20 kR. These findings corroborate earlier studies by Yassein and Amina, 2014 and Singh *et al*. (2020), which demonstrated that moderate gamma irradiation enhances photosynthetic efficiency, nutrient uptake, and carbon assimilation, leading to improved seed productivity. However, excessive radiation exposure negatively impacts chloroplast function and disrupts source-sink relationships, ultimately reducing yield potential.

Test weight (TW), oil content (OC), and days to maturity (DM) were also significantly affected by irradiation. The highest TW (4.33 g) and OC (41.29%) in P13RGN-303 were observed at 20 kR, while the lowest values (3.57 g and 36.54%, respectively) were recorded at 25 kR. Similarly, RH-761 exhibited the highest TW (3.17 g) and OC (36.33%) at 20 kR, with a significant decline at 25 kR. The improvement in oil content at moderate doses suggests a positive influence on lipid biosynthesis, possibly through the up-regulation of genes involved in fatty acid metabolism (Mohurle *et. al.,* 2017 and Fatima *et al.*, 2016). In contrast, excessive irradiation may impair metabolic pathways, leading to reduced oil accumulation and seed deterioration.

Days to maturity were slightly reduced at 20 kR for both genotypes, indicating an accelerated reproductive phase. This effect may be attributed to radiation-induced modulation of flowering genes and hormonal signals, promoting early transition to reproductive growth. These findings highlight the potential of moderate gamma irradiation as a strategic tool for improving mustard yield and seed quality while emphasizing the need to avoid excessive doses that may induce deleterious effects.

Conclusion

The results of this study highlight the potential of gamma irradiation as an effective tool for improving the morphological and yield characteristics of mustard genotypes P13RGN-303 and RH-761. Moderate doses, particularly 20 kR, showed significant positive effects on field emergence, plant height, seed yield, and oil content, suggesting the suitability of gamma radiation for enhancing these traits. However, higher doses (25 kR) were observed to have adverse effects, underscoring the importance of optimizing irradiation doses for maximum benefit. These findings contribute valuable insights into the use of gamma irradiation for mustard breeding programs aimed at improving productivity and quality.

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