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DEVELOPMENT OF NEW MUTANTS USING GAMMA RADIATION IN GLADIOLUS CV. PUNJAB DAWN

Yograj Kushwaha^{1*}, Bhagwan Das Bhuj¹, Ranjan Srivastava¹, N.K. Singh² and Satish Chand¹

¹Department of Horticulture, Govind Ballabh Pant University of Agriculture and Technology, Pantnagar-263 145, Uttarakhand, India.

²Department of Genetics & Plant Breeding, Govind Ballabh Pant University of Agriculture and Technology, Pantnagar-263 145, Uttarakhand, India.

*Corresponding author E-mail : 53999_yograjkushwaha@gbpuat-tech.ac.in
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ABSTRACT

This study presents the development of two new gladiolus mutants derived from the original cultivar Punjab Dawn (PD) through mutation breeding using gamma radiation. The experiment was conducted at the Model Floriculture Centre, Govind Ballabh Pant University of Agriculture and Technology, Pantnagar (Uttarakhand), using a randomized block design. *Gladiolus grandiflorus* var. Punjab Dawn was chosen as the experimental material. Gladiolus corms of the Punjab Dawn (PD) variety were exposed to controlled doses of gamma rays *i.e.* 4.0 kR (T₁) and 5.0 kR (T₂) and Control (non-radiated T₃) to induce random mutations in the genetic material. Following irradiation, the mutagenized populations were evaluated for novel phenotypic traits including flower color, size, shape and overall plant vigor. Two distinct mutant lines showing desirable characteristics were selected and further propagated to establish stable new varieties. The newly developed gladiolus mutants exhibit unique ornamental features and represent successful outcomes of mutation breeding using gamma radiation. This abstract highlight the application of mutation breeding to expand the genetic diversity of gladiolus cultivars and introduce innovative mutants with enhanced ornamental qualities derived from the popular Punjab Dawn cultivar.

Key words : Gamma radiation, Mutation breeding, Gladiolus, Mutants, Cultivar, Punjab Dawn.

Introduction

Gladiolus, a cut flower well-regarded for its tall spikes and effervescent blooms, is globally cultivated and often used in splendid floral arrangements. The name “gladiolus” was coined by Pliny the Elder, derived from the Latin word “gladiolus,” meaning sword, owing to its sword-shaped foliage. Due to its unique leaves, it is commonly known as the ‘sword lily’ and ‘corn flag’ in Europe. In India, gladiolus holds noteworthy importance among commercial flowers due to its remarkable spikes festooned with striking and elusive florets in innumerable shades. This tender plant exhibits a sequential blooming of flowers, offering a long-lasting display and excellent durability as cut flowers (Singh, 2006). The increasing demand for gladiolus in the floriculture market underscores the need for genetic improvements that can

be achieved within shorter time frames. Induced mutagenesis serves as a valuable method for genetic enhancement in crops, providing a range of variants for effective selection (Dhumal and Bolbhat, 2012). Exposing genetic materials to mutagenic agents induces changes in nuclear DNA and/or cytoplasmic organelles, resulting in genomic or chromosomal mutations. This allows plant breeders to identify and select beneficial mutants. In mutation breeding strategies aimed at producing a high frequency of desirable mutations, the selection of an effective and efficient mutagen is crucial (Roychowdhury and Tah, 2011). Gamma rays are high-frequency electromagnetic radiations with short wavelengths. Their characteristics, such as high energy per photon, precise dosimetry, reproducibility and uniform penetration in multicellular systems, make gamma rays suitable as a physical mutagen. Given these attributes, this experiment

was conducted to explore the spectrum and frequency of color variants in *Gladiolus* varieties treated with gamma rays.

Materials and Methods

The experiment was conducted at the Model Floriculture Centre, Govind Ballabh Pant University of Agriculture and Technology (GBPUA&T), Pantnagar (Uttarakhand), using a randomized block design. *Gladiolus grandiflorus* var. Punjab Dawn was chosen as the experimental material. Medium-sized corms pretreated with 0.2% carbendazim were exposed to varying doses of physical mutagen for inducing mutations, specifically gamma irradiation, based on Lethal Dose (LD_{50}) values. The gladiolus corms underwent gamma irradiation individually with two doses: 4.0 kR and 5.0 kR, at the Gamma Chamber Facility of the Radiation & Isotopic Tracers Laboratory (RITL), College of Basic Science and Humanities, GBPUA&T, Pantnagar, Uttarakhand. The crop was cultivated under uniform cultural practices with corms of uniform diameter (4-6 cm) selected for planting at spacings of 30 cm between rows and 20 cm between plants. Weeding was performed manually at intervals to maintain a weed-free field, and irrigation was carried out periodically using flooding method. The final irrigation was applied two weeks before harvesting the corms and cormels from the experimental field. Harvested corms from the M_1 generation were used as the base population for subsequent M_2 to M_7 generations. Variations resulting from mutagenic treatments were meticulously observed, focusing on differences in vegetative growth and flowering characteristics between treated and untreated corms. Detailed observations were recorded for each treatment to evaluate the impact on growth, flowering, and corm attributes. Approximately 60 days after spike cutting, corms and cormels were lifted and any infected or injured specimens were removed. Detail of the treatments showed in Table 1. The statistical analysis was done by using OPSTAT developed by O.P. Sheoran, Computer Section, CCS HAU, Hisar.

Table 1 : Details of treatment for the experiment.

Variety – Punjab Dawn (PD)		
Mutagen – Physical Mutagen <i>i.e.</i> gamma irradiation		
Treatments	Dose	R.H.S Colour Chart Reading
T ₁	4.0kR	White Group 155B (Mutant 1)
T ₂	5.0kR	Purple Red 72 C (Mutant 2)
T ₃	Control	Red Group 49A (Parent)

Results and Discussion

Impact of gamma rays on vegetative growth of the plants

The results pertaining to vegetative growth is depicted in Table 2 and Fig. 1. The average days to 50% sprouting for the Control treatment is 6.525 days, which is higher compared to the 4.0 kR and 5.0 kR treatments, indicating a slower sprouting rate. The 4.0 kR treatment has the lowest average days to 50% sprouting at 6.160 days, followed closely by the 5.0 kR treatment at 6.055 days. It could be due to the food reserve in the corms and delayed growth regulator activity due to environmental condition. It could also be hampered by the intercultural operation while planting of corms such as the depth of the corm planted in the soil. But the most important is its gene expression which is obstructed by giving the dose of gamma irradiation. The 4.0 kR treatment results in the highest average number of tillers (2.388), suggesting that this dose promotes vegetative growth more effectively than the other treatments. The 5.0 kR treatment averages 1.838 tillers, while the Control treatment has the lowest average number of tillers at 1.705. The changes in number of tillers may be attributed to its genetic makeup of the plant and changed gene expression, which may lead to the increase or decrease in the number of tillers. The 4.0 kR treatment results in the tallest plants with an average height of 64.030 cm. In contrast, the Control and 5.0 kR treatments have similar plant heights, with averages of 54.143 cm and 53.850 cm, respectively. Leaf count provides insights into the foliar development of the plants. The 4.0 kR treatment results in the highest average leaf count at 10.328 leaves, followed by the 5.0 kR treatment at 9.155 leaves, and the Control treatment at 8.588 leaves. The 4.0 kR treatment results in the longest leaves with an average length of 33.270 cm. The Control treatment has an average leaf length of 30.560 cm, and the 5.0 kR treatment has the shortest leaves with an average length of 29.873 cm. Vegetative characters discussed above may be attributed to the action of plant growth regulator such auxin, cytokinin and gibberellin etc. and also the intercultural/agronomical operation during the cropping time. The growth of these character may also be hampered due the slower and faster mitotic cell division in the plant which helps to increase the palisade cell areas where chloroplast reside. Application of different dose of gamma irradiation might impact the synthesis of auxin or proteins which helps in increase the plant stature in these mutants which ultimately resulted the changes in vegetative character such as plant height, leaf length and leave width. Aligning with the observations of Patil and Dhadhuk (2009), it was found that in *gladiolus*, lower

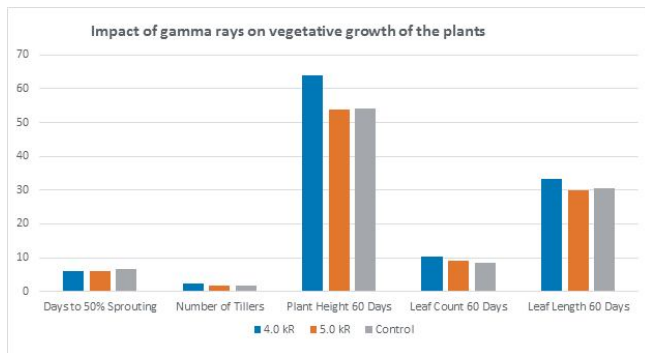


Fig. 1 : Impact of gamma rays on vegetative growth of the plants.

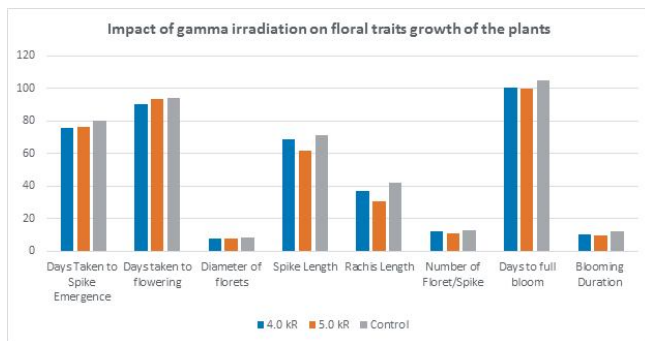


Fig. 2 : Impact of gamma irradiation on floral traits growth of the plants.

doses of radiation lead to an increase in the number of leaves due to the activation of physiological substances present in corms. Correspondingly, higher doses inhibit cell division by arresting mitotic processes, negatively impacting auxin levels and resulting in reduced plant growth characteristics. Minor enhancements in leaf number with lower doses of gamma irradiation may occur due to the synthesis and release of substances like biochemicals and enzymes, which play crucial roles in plant metabolism and growth, ultimately leading to increased plant growth (Misra and Bajpai, 1981; Cesarett, 1968). Additionally, these effects could be attributed to disruptions in mitosis resulting in frequent mitotic aberrations and alterations in nutrient levels due to disturbances in assimilation rates (Sparrow, 1961). The differential impacts of gamma radiation doses on plants may be linked to modifications in cell anatomy, components, morphology, and ultimately plant physiology

(Wi *et al.*, 2007). Reductions in leaf length after exposure to gamma rays might be caused by interference in chromosomal aberrations and mitotic activities, leading to physiological damage (Gunckal, 1957). The decrease in leaf dimensions, both width and length, in “Dutch Iris” with increasing gamma irradiation doses was also observed by Rather and John (2000). Observable changes in nutrient assimilation rates altering nutrient levels in plants (Ehrenberg, 1955), or anatomical, physiological, and cytological variations in plants, could contribute to reductions in plant growth. The stimulatory effects of gamma doses in certain varieties may be associated with the release of enzymes induced by irradiation, which play pivotal roles in metabolism, consequently increasing metabolic activities and stimulating plant growth (Misra and Bajpai, 1983). Reductions in the width of the longest leaf when corms are exposed to higher doses of irradiation were also observed by Dhara and Bhattacharya (1972). The results were also confirmed with the finding of Belwal *et al.* (2023) and Rawat *et al.* (2021). The data from the experiment indicates that the 4.0 kR treatment generally promotes better vegetative growth and development compared to the 5.0 kR and Control treatments. The 4.0 kR treatment results in the fastest sprouting, the highest number of tillers, the tallest plants, the highest leaf count, and the longest leaves, making it the most effective treatment among the ones studied.

Impact of gamma rays on floral growth of the plants

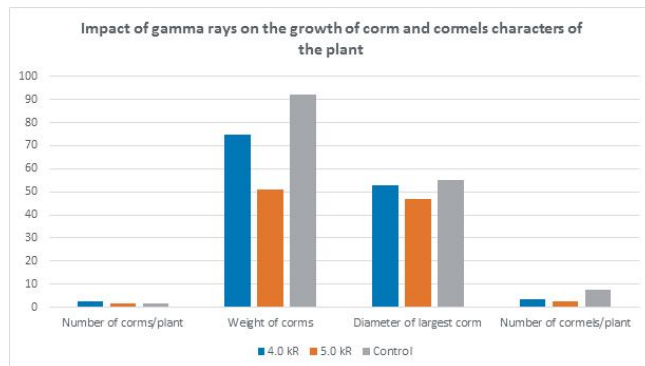
The observation related to floral growth is illustrated in Table 3 and Fig. 2. The 4.0 kR treatment results in the shortest time to spike emergence with an average of 75.840 days. The 5.0 kR treatment follows with an average of 76.308 days, while the Control treatment has the longest time to spike emergence at 79.833 days. The 4.0 kR treatment also results in the shortest time to flowering with an average of 90.268 days, followed by the 5.0 kR treatment at 93.210 days. The Control treatment takes the longest time to flowering with an average of 94.318 days. The Control treatment has the largest average diameter of florets at 8.270 mm. The 5.0 kR treatment has an average diameter of 7.768 mm, while the 4.0 kR treatment has the smallest diameter at 7.490

Table 2 : Impact of gamma rays on vegetative growth of the plants.

Dose/Traits	Days to 50% Sprouting	Number of tillers	Plant height 60 Days	Leaf Count 60 Days	Leaf Length 60 Days
4.0 kR	6.160	2.388	64.030	10.328	33.270
5.0 kR	6.055	1.838	53.850	9.155	29.873
Control	6.525	1.705	54.143	8.588	30.560
C.D.	N/A	0.379	2.130	0.907	1.130
SE(m)	0.165	0.108	0.604	0.257	0.320

Table 3 : Impact of gamma irradiation on floral traits growth of the plants.

Dose/Traits	Days taken to Spike Emergence	Days taken to flowering	Diameter of florets	Spike length	Rachis length	Number of Floret/ Spike	Days to full bloom	Blooming Duration
4.0 kR	75.840	90.268	7.490	68.953	36.735	12.000	100.345	10.350
5.0 kR	76.308	93.210	7.768	61.883	30.300	10.790	99.748	9.575
Control	79.833	94.318	8.270	71.285	42.238	12.615	104.645	11.880
C.D.	2.055	1.512	0.362	2.229	4.027	0.610	N/A	0.679
SE(m)	0.583	0.429	0.102	0.632	1.142	0.173	1.748	0.193

**Fig. 3 :** Impact of gamma rays on the growth of the corms and cormels characters of the plant.

mm. The control treatment showed that the longest spikes with an average length of 71.285 cm. The 4.0 kR treatment follows with an average spike length of 68.953 cm, while the 5.0 kR treatment has the shortest spikes at 61.883 cm. In control the results observed that longest rachis with an average length of 42.238 cm. The 4.0 kR treatment has an average rachis length of 36.735 cm, while the 5.0 kR treatment has the shortest rachis at 30.300 cm. The control treatment reflects the highest number of florets per spike with an average of 12.615 florets. The 4.0 kR treatment follows closely with an average of 12.000 florets, while the 5.0 kR treatment has the fewest florets per spike at 10.790. Longest time to reach full bloom with an average of 104.645 days was noticed in control treatment. While, the 4.0 kR treatment follows with an average of 100.345 days, while the 5.0 kR treatment has the shortest time to full bloom at 99.748 days. The control treatment gives longest blooming duration with an average of 11.880 days. The 4.0 kR treatment follows with an average blooming duration of 10.350 days, while the 5.0 kR treatment has the shortest blooming duration at 9.575 days. The findings of the experiment found contradictory to the findings of Sisodia and Singh (2014) and Tiwari *et al.* (2018), which observed that spike emergence changes with increasing doses of gamma rays. Conversely, all other gamma ray doses resulted in early spike emergence. The delay in spike emergence may be attributed to disruptions in biochemical

pathways that support the flower induction process, as suggested by Bagnall *et al.* (1995), their study focused on how altered phytochrome expression in mutants and transgenic lines of *Arabidopsis* affected flowering responses. Variations in spike length caused by gamma irradiation in certain varieties could be attributed to changes in auxin levels. It is likely that the decrease in auxin content with higher irradiation doses is responsible for this effect, as suggested by Sisodiya and Singh (2014). Exposure to ionizing radiation may cause slight disruptions in pigment synthesis, potentially explaining the varying shades of floret colors in gladiolus varieties. However, stable and consistent mutants likely result from DNA-level changes (Kumari and Kumar, 2020). The alterations in spike and rachis length may be linked to the hindered growth of irradiated plants resulting from radiation-induced damage. Previously, Kainthura and Srivastava (2015) similarly observed shorter rachis length in Tuberosa and attributed it to physiological, morphological, and cytological disruptions caused by gamma radiation. Dobanda (2004) and Patil (2014) noted that lower gamma ray doses promoted earlier opening of the first floret, whereas higher doses resulted in delayed opening, increase diameter of florets and number of florets. Whereas, decrease of diameter and number of florets with increase in irradiation doses. The initiation of flowering may be influenced by mutagenic treatments due to alterations in several biosynthetic pathways directly and indirectly linked to flowering physiology (Mahure *et al.*, 2010; Ismael and Mohmoud, 2015). Misra and Bajpai (1983) subjected various gladiolus varieties to gamma ray doses of 3, 4, 5, 7 and 10 kR. They identified stable mutations in Picardy, Sans Sauci, Himprabha, and Ratna's Butterfly in the vM₁ and vM₂ generations. According to Cantor *et al.* (2002), exposure to gamma radiation enhanced the growth of roots and shoots, likely leading to improved nutrient absorption and higher rates of photosynthesis. This, in turn, caused earlier spike emergence and better-quality flowers.

The data indicates that the Control treatment generally promotes longer and more extensive flowering and floret



Fig. 4 : Comparison of changes in flowering characteristics of the plant due to gamma rays.

Table 4 : Impact of gamma rays on the growth of corm and cormels characters of the plant.

Dose/Traits	Number of corms/plant	Weight of corms	Diameter of largest corm	Number of cormels/ plant
4.0 kR	2.388	74.808	52.655	3.325
5.0 kR	1.838	51.035	46.733	2.545
Control	1.705	92.040	55.040	7.500
C.D.	0.379	21.877	4.352	0.987
SE(m)	0.108	6.201	1.234	0.280

development compared to the 4.0 kR and 5.0 kR treatments. The 4.0 kR treatment results in the shortest time to spike emergence and flowering, making it effective for early flowering.

Impact of gamma rays on the growth of corm and cormels characters of the plant

The data regarding corms and cormels growth is depicted in Table 4 and Fig. 3. The average number of corms per plant is highest in the 4.0 kR treatment with 2.388 corms per plant. The 5.0 kR treatment averages 1.838 corms per plant, while the Control treatment has the lowest average number of corms at 1.705 per plant. The critical difference suggests that the 4.0 kR treatment is significantly better at promoting the formation of corms compared to the 5.0 kR and Control treatments. The Control treatment results in the heaviest corms with an average weight of 92.040 grams. The 4.0 kR treatment follows with an average corm weight of 74.808 grams, while the 5.0 kR treatment has the lightest corms at 51.035 grams. The average diameter of the largest corm is highest in the Control treatment at 55.040 mm. The 4.0 kR treatment has an average diameter of 52.655 mm, and the 5.0 kR treatment has the smallest corm diameter at 46.733 mm. The Control treatment results in the highest average number of cormels per plant at 7.500. The 4.0 kR treatment averages 3.325 cormels per plant, while

the 5.0 kR treatment has the lowest average number of cormels at 2.545. The variation in the number of corms per plant is closely linked to vegetative growth and the number of shoots per plant, which impacts the overall corm and cormel production. These findings are consistent with the research conducted by Kaur and Bajpay (2019), emphasizing the significance of these growth parameters in determining corm and cormel yields. It is important to recognize that different cultivars may demonstrate varied responses to different doses of gamma radiation, resulting in both inhibitory and stimulatory effects. This variability can be attributed to the inherently random and non-directional nature of mutational processes occurring in the natural environment, making it challenging to predict

their specific impact on plant traits. This phenomenon has been observed in other studies as well. For example, Jun et al. (2007) documented a reduction in corm count in saffron plants exposed to higher doses of gamma radiation, aligning with the findings of Sisodia and Singh (2014).

Conclusion

On the basis of experiment finding the 4.0 kR treatment generally promotes better vegetative growth and development compared to the 5.0 kR and Control treatments. The 4.0 kR treatment results in the fastest sprouting, the highest number of tillers, the tallest plants, the highest leaf count, and the longest leaves, making it the most effective treatment among the ones studied. Also, the Control treatment generally promotes longer and more extensive flowering and floret development compared to the 4.0 kR and 5.0 kR treatments. The 4.0 kR treatment results in the shortest time to spike emergence and flowering, making it effective for early flowering. Overall, while the 4.0 kR treatment shows positive effects on the number and weight of corms, the Control treatment excels in all measured traits, especially in promoting the formation and growth of cormels.

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