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The Pharma Innovation

ISSN (E): 2277-7695
ISSN (P): 2349-8242
NAAS Rating: 5.23
TPI 2022; 11(11): 2497-2502

www.thepharmajournal.com
Received: 07-09-2022
Accepted: 10-10-2022

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Influence of gamma rays on quantitative traits in M_2 generation of lathyrus (*Lathyrus sativus* L.)

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Abstract
The present study was carried out at experimental farm of Agricultural Botany Section, College of Agriculture, Nagpur. An experiment on induction of mutation in lathyrus by gamma rays was performed by using treated seeds of lathyrus cv. NLK-73 with 250, 300 and 350 Gy doses of gamma rays in non-replicated trial along with control. The treated material along with untreated control were sown in M_1 generation and the seeds collected from individual plant of M_1 generation were used to raise M_2 generation during *rabi* 2020 and 2021 respectively. Observations on morphological traits and biometrical traits were recorded on each treatment and data were subjected to statistical analysis. Germination and mortality percentage were recorded in M_1 and M_2 generation and it was found that there was reduction in germination percentage and increase in mortality as compared to control. In M_2 generation number of branches plant^{-1}, number of pods plant^{-1}, 100 seed weight and seed yield plant^{-1} increased in all the three treatments of gamma radiation *i.e*., 250, 300 and 350 Gy as compared to control. Days to first flower, days to maturity and plant height increased in some treatments and decreased in some treatments.

Keywords: Lathyrus, gamma rays, mutation, M_2 generation

Introduction
The *Lathyrus sativus* L. (2n=14), also known as grass pea, khesari dal, peavine, or chanamatra, is an annual plant and a significant pulse crop rich in protein with (28%) next to soybean. *Lathyrus sativus* is a herbaceous annual plant with many branches that is capable of straggling or climbing. It has a well-developed taproot system, and its rootlets are covered in tiny, cylindric, branched nodules that are typically clustered together in dense groups. It belongs to the Leguminoceae family, subfamily Papilionoideae, and genus Lathyrus, with 130 species spread over the temperate zone of the Northern Hemisphere and the higher altitudes of tropical Africa. In India, besides the ornamental *Lathyrus odoratus*, the only other species cultivated is *Lathyrus sativus* which yield the khesari dal. The edible *Lathyrus sativus* originated in the West Central Asia Mediterranean region and North India was its center of domestication. The plant is regarded as an excellent fighter against many pests, drought, floods, and hail. The plant has a high tolerance for drought. Due to its status as a legume, it fixes atmospheric nitrogen through its root nodules, part of which could be available to succeeding crop. During the rabi season, it is typically sown as a "Utera" or "Paira" crop in standing crops of paddy. Since the primary pulse crop, Tur (pigeon pea), is exclusively planted on bunds, the production of Tur is insufficient to meet the needs of these regions hence Lathyrus can be sown as the alternative pulse. The lathyrus plant type is thought to be highly resistant to drought (Tripathy *et al*., 2011) [16] and grows luxuriantly without any cultivation input. This pulse is consumed in a number of ways, including chapaties, wadas, and curries, and it has been fed to cattle as green fodder and stover (dried chaff) since ancient times.

Since ancient times, this pulse has been used to make a variety of dishes, including chapaties, wadas, and curries, as well as to feed cattle as stover (dry chaff) and green fodder. It fulfils major pulse need in our country. However, a ban has been imposed on it because of its association to Neurolathyrism, a neurological illness that is irreversible in both humans and animals, which is caused by the neurotoxin, (β-N-oxalyl-L-α, β-diaminopropionic acid) β-ODAP, that is present in its seedlings and seeds. Even though research experts have created a number of improved lathyrus cultivars, India's average production and land area are fairly poor when compared to other nations. Due of its high protein content (28–32%), grasspea is the second-most significant pulse crop after soybean (42%). Thus, in order to increase economic output and satisfy the large population's need for protein, a variety of breeding techniques are being used on this crop. Mutation is a sudden heritable change in the characteristics of an organism other than those due to mendelian segregation and recombination. Mutations are the rare events mostly recessive to wild type, often associated with deleterious or lethal effects and are inducible by a variety of external agents called mutagen. Naturally occurring spontaneous mutations are extremely uncommon and have a very low frequency. As a result, they cannot resolve the issues faced by breeders. By inducing artificial mutations in organisms and plants, several genetic and breeding issues can be resolved. Therefore the technique of mutation breeding has been adopted as a valuable supplement to conventional breeding to create additional genetic variability. The variability produced through these mutations is utilized for further crop improvement. Therefore, mutation breeding is more desirable to create variability in grasspea.

The main objective for choosing a pulse crop for mutation breeding is that they are rich in protein, which is the building block of life. Protein makes up the majority of the human brain, blood, bones, muscles, and skin. Thus, there is no need to elaborate on the role that protein plays in human nutrition. Therefore, the breeding strategy for this crop's enhancement focuses on creating cultivars with high protein content, high yield, and low neurotoxic levels. There are numerous ways to accomplish these goals, with mutation breeding being one of the most effective method. Hence, the purpose of this experiment was to examine the effect of gamma rays on quantitative traits in the M_2 generation of lathyrus.

Material and Methods

In the present study dry, healthy and genetically pure seeds of *Lathyrus sativus* cv. NLK-73 (high yielding cultivar) were obtained from Agril. Botany section, College of Agriculture, Nagpur. Four different lots of 5000 seeds of lathyrus seed cultivar NLK-73 were made. Out of these, three lots of seeds were sent to Bhabha Atomic Research Centre, Trombay, for irradiation with three different doses of gamma rays *i.e*. 250 Gy, 300 Gy and 350 Gy (Co^{60} at BARC Trombay, Mumbai) and used for raising M_1 during rabi 2020 and individual plant in each treatment were harvested separately. The harvested seed were used to raise M_2 generation in *rabi* 2021 and mutants were identified.

M_2 generation was raised in *rabi* 2021. The seeds of individual plants harvested from each of the treatment were sown to raise M_2 population. The sowing was undertaken on the fertile and well levelled piece of land in the research farm of Agril. Botany section, College of Agriculture, Nagpur. The M_2 population was observed for different parameters besides scoring of different mutants. The treated populations were carefully screened for all morphological characters. The observations on the following traits were recorded on normal vigorous and healthy plant in each treatment for germination per cent, mortality per cent (in M_1 and M_2), days to first flower, days to maturity, plant height (cm), number of branches plant^{-1}, number of pods plant^{-1}, 100 seed weight (g) and grain yield plant^{-1} (g) (in M_2) on each and every plant in each treatment except for germination per cent and mortality per cent on which plot wise observations were recorded. Germination count was taken 12 days after sowing and was reported in percentage.

$$\text{Germination \%} = \frac{\text{Total number of seeds germinated}}{\text{Total number of seeds sown}} \text{ X } 100$$

The number of plants which failed to survive upto flowering from the date of germination were counted and mortality percentage was calculated as per the formula given below:

$$\text{Mortality \%} = \frac{\text{Number of plants failed to survive upto flowering}}{\text{Number of seeds germinated}} \text{ X } 100$$

The data recorded were analysed statistically as described by Panse and Sukhatme (1954) [13].

Results and Discussion

The effect of various treatments of gamma rays on germination in M_1 and M_2 generations are presented in table 1. Reduced germination per cent in all the treatments was observed in M_1 generation as compared to control. The germination percentage in M_1 generation ranged from 41.12 (T_3 – 350 Gy) to 71.50 (T_4-control) per cent. Reduction of germination percentage was highest in T_3 – 350 Gy (41.12%) followed by T_2 – 300 Gy (47.26%) of M_1 generation while, lowest reduction in T_1-250 Gy (51.30%) as compared to control. In M_2 generation, similar pattern of reduced germination was observed as in M_1 generation. The germination percentage in M_2 generation ranged from 56.96 (T_3 – 350 Gy) to 84.84 (T_4-control) per cent. Reduction of germination percentage was highest in T_3 – 350 Gy (56.96%) followed by T_2 – 300 Gy (67.81%) of M_2 generation while, lowest reduction in T_1-150 Gy (68.34%) as compared to control. The results on germination per cent revealed that gamma rays reduced the germination in M_1 and M_2 generation with increasing dose of gamma rays clearly indicating that gamma rays as mutagen have induced an inhibitory effect on seed germination. The germination percentage increased in M_2 as compared to M_1. The reduction in germination may be either due to genetic cause or inhibition of physiological process in cell by mutagen. In accordance to this result Kusmiyati *et al*. (2018) [12] and Kankal *et al.* (2018) [10] also observed that higher doses of gamma rays significantly reduced germination percentage of the first and final count in soybean. Chavan *et al.* (2019) [7] also reported decrease in germination percentage compared to control in both M_1 and M_2 generation in lathyrus.

The data on mortality of seedling as affected due to different doses of gamma rays in M_1 and M_2 generation are presented in table 1. The effect was recorded as mortality in percentage. Increasing the dose of gamma rays caused increase in mortality of seedling in both M_1 and M_2 generation**.** Data in table 1 reveals that the maximum mortality was recorded in T_3-350 Gy (77.96%) while minimum in T_1-250 Gy (58.28%) as compared to their respective control (25.37%) in M_1 generation. In M_2 generation the maximum mortality was recorded in T_3-350 Gy (54.38%) while minimum in T_1-250 Gy (49.31%) as compared to their respective control (21.42%). Increased doses of treatment of gamma rays had exerted increasing effect in this character. The results on mortality per cent revealed that mortality per cent was more in gamma rays treatments as compared to control. It was also found that mortality increased in M_1 and M_2 generation with increase in doses of gamma rays treatment. The prime cause of mortality was physiological imbalance or different types of chromosomal aberrations as reported by Aditya *et al*. (2017) [1]. In accordance to the above result, Ahire *et al*. (2005) [2] in soybean reported that high lethality may be attributed to the injuries caused by mutagenic treatments of gamma rays. Similar to this result Beltagi *et al*. (2006) [6] also observed that under highest dose of gamma irradiation the seedling immerged but it did not continue growth and indicated 100 per cent lethality and the low dose significantly reduced the shoot length and leaf area in common bean. Aditya *et al*. (2017) [1] in soybean also observed and reported increase in mortality per cent with increase in the dose of gamma rays treatment. Chavan *et al*. (2019) [7] also reported that mortality percentage increased as compared to control in both M_1 and M_2 generation in lathyrus. Vijaykumar *et al*. (2021) [18] also observed that rate of reduction in biological damage on quantitative characters was directly proportional to the dose of mutagen irrespective of the varieties and mutagens used in lathyrus. Physical mutagen showed the highest biological damage than chemical mutagen.

Data regarding effect of different gamma rays treatments on days to first flower in M_2 generation are as presented in table 1. It is observed from the table that the maximum mean value for days to first flower initiation was in T_3-350 Gy (49.33) while the minimum days to first flower was recorded in T_1-250 Gy (47.89 days) as compared to their respective controls. Maximum range for days to first flower initiation was observed in T_3-350 Gy and minimum range in T_4-Control. The coefficient of variation increased in T_2 and T_3 while in T_1 it was lower than control for days to first flower. The maximum coefficient of variation was observed in T_3-350 Gy (11.37%) followed by T_2-300 Gy (10.16%) while the minimum in T_1-250 Gy (8.93%) as compared to control (9.15%). The results on days to first flower revealed that time taken for flowering was more in control as compared to gamma rays treatments. The results showed that gamma irradiation affected days to first flower. In contrary to this result Khan *et al*. (2018) [11] reported that gamma irradiation non-significantly affected days to flower initiation in pea. In accordance to this result Aditya *et al*. (2017) [1] reported gradual decrease in the mean value for 50 per cent flowering with increase in dose of gamma rays in soybean. Chavan *et al.* (2019) [7] also reported that days to first flower increased in 250 Gy, 200 Gy and 300 Gy while decreased in 150 Gy and 350 Gy over the control in lathyrus.

Data regarding effect of different gamma rays treatments on days to maturity in M_2 generation are presented in table 1. It is observed from the table that the maximum mean value for days to maturity was in T_2-300 Gy (109.71) while the minimum days to maturity was recorded in T_3-350 Gy (108.40). Maximum range for days to maturity was observed in T_3-350 Gy (25 days) and minimum range T_1-250 Gy (15 days) as compared to control (5 days). The extent of coefficient of variation is low for this trait. The results on days to maturity revealed that the mean value of days to maturity increased in some treatments and decreased in some treatments. Similar to this Patil *et al.* (2011) [14] reported in soybean that in higher doses of mutagen and their combinations, days required to maturity were found to be significantly increased. Khan *et al.* (2018) [11] also observed and reported that increase in the radiation dose of gamma rays delayed maturity in pea. Chavan *et al.* (2019) [7] also reported that days to maturity increased in 250 Gy and 300 Gy and decreased in 150 Gy, 200 Gy and 350 Gy over the control in lathyrus.

Plant height is an important measure of growth. It is one of the visible measurement and is a function of the internode and leaf emergence, since leaves are born on stem. Leaf area development and biomass production shows a close relationship with plant height. The data regarding the effect of gamma rays on plant height are presented in table 1. Mean plant height increased significantly in all the treatments except T_1-250 Gy as compared to control. Maximum plant height was observed in T_2-300 Gy (105 cm) followed by T_3-350 Gy (97 cm) while the minimum in T_1-250 Gy (88 cm) as compared to control (72 cm). The maximum range for plant height was recorded in T_2-300 Gy (81 cm) and minimum range was recorded in T_1-250 Gy (63 cm) as compared to control (30 cm).The coefficient of variation for the plant height increased in all the treatments as compared to the control. The maximum variation was in T_2-300 Gy (24.08%) followed by T_3-350 Gy (21.87%) and minimum in T_1-250 Gy (20.85%) as compared to control (13.32%) respectively. The range for the coefficient of variation was 20.85% to 24.08%. The data on plant height indicated that mean height reduced in lower doses and increased in higher doses of gamma rays. Gamma rays induced increase in plant height. Similar to this result Gopinath and Pavadai (2015) [9] reported the positive shift and the maximum mean value were recorded at 50 kR gamma rays in M_2 generation of soybean. In contrary to this result Rybinski *et al.* (2006) [15] reported that in M_2 progeny of lathyrus, plant height for cultivar Krab ranged from 104.2 to 111.5 cm, and that of the mutant from 89.9 to 129.4 cm. Alikamanoglu *et al*. (2011) [3] also reported decrease in plant height in inverse proportion of dose in soybean. Chavan *et al.* (2019) [7] also recorded that plant height increased in 150 Gy and 250 Gy while decreased in 200 Gy, 300 Gy and 350 Gy over control in lathyrus.

The data on number of branches plant^{-1} was recorded at harvest stage and are presented in table 1. Significant variation was observed for this trait. The highest mean value for the character was recorded in T_2-300 Gy (3.86) and the lowest in T_1-250 Gy (3.70). In general the number of branches increased in all the treatments as compared to their control (3.66). Maximum range for number of branches plant^{-1} was noticed in T_1-250 Gy and T_2-300 Gy (7) as compared to control (3). The variability studies showed that the coefficient of variation increased against their control in all the treatments. The coefficient of variation for the character ranged from 27.40% to 32.69% as compared to control (23.35%). The highest variation was recorded in T_1-250 Gy (32.69%) followed by T_2-300 Gy (29.02%) and the lowest in T_3-350 Gy (27.40%). The data revealed that increase in dose of gamma rays lead to increase in number of branches plant^{-1} with increase in dose of gamma rays except for T_3-350 Gy. In contrary to this result Usharani and Kumar (2015) [17] reported that the mean values for number of primary branches in blackgram decreased below the control in most of the treatment. Aditya *et al*. (2017) [1] observed that the coefficient of variance for number of branches plant was highest in 50 Gy followed by 150 Gy, 200 Gy, 100 Gy, 400 Gy and control in decreasing order in soybean. Chavan *et al.* (2019) [7] observed that number of branches plant^{-1} increased significantly in all the treatments as compared to control in lathyrus.

Data on variation caused due to different gamma rays treatment on number of pods plant^{-1} are presented in table1. The highest mean value for number of pods plant^{-1} was observed in T_2-300 Gy (79.60) followed by T_3-350 Gy (76.59). In general number of pods plant^{-1} increased in all the treatment as compared to control (59.71). Maximum range was recorded in T_2-300 Gy (332), followed by T_1-250 (280) and T_3-350 (259) all more than the range of the control (83).The coefficient of variation increased in all the treatments for number of pods plant^{-1} as compared to control. The maximum coefficient of variation was observed in T_2-300 Gy (65.83%) followed by T_1 – 250 Gy (65.20%) while the minimum in T_3-350 Gy (52.80%). The coefficient of variation was observed to be higher in all the treatment as compared to control (44.16%). The result on number of pods plant^{-1} revealed that the mean number of pods plant^{-1} increased in all the treatments as compared to control. But the wide range of variation observed in all the treatments indicated the presence of high within treatment variation. In accordance to this result Rybinski *et al.* (2006) [15] reported a wide variation for number of pods plant^{-1} in selected grasspea mutants. The number of pods plant^{-1} ranged from 56.2 to 69.8 for cv. Krab and 37.1 to 95.4 in their mutants. Badr *et al*. (2014) [5] also noticed that the number of pods plant^{-1} were significantly increased in cowpea varieties treated with gamma radiation as compared to control. Aditya *et al*. (2017) [1] reported in soybean that the standard deviation values and coefficient of variation increased in radiation doses in comparison to control. Khan *et al*. (2018) [11] reported that number of pods plant^{-1} decreased significantly in some treatment of gamma irradiation in pea while increased significantly in some treatment. Chavan *et al.* (2019) [7] also reported that number of pods plant^{-1} were increased in 250 Gy and 300 Gy and decreased in 150 Gy, 200 Gy and 350 Gy over the control in lathyrus.

Data regarding effect of different doses of gamma rays on 100 seed weight in M_2 generation are presented in table 1**.** The mean value for 100 seed weight showed significant difference. The maximum mean value for the character was observed in T_3-350 Gy (6.91 g) and minimum in T_1-250 Gy (6.30 g) and control showed a mean value of 4.26 g. Highest range for this trait was recorded in T_2-300 Gy (9.29 g) followed by T_1-250 Gy (8.96 g) and lowest range in T_3-350 Gy (5.66 g) as compared to control (2.20 g). The coefficient of variation increased in T_1-250Gy for 100 seed weight and reduced in T_2-300 Gy and T_3-350 Gy as compared to control. The maximum coefficient of variation was observed in T_1-250 Gy (17.14%) while the minimum in T_3-350 Gy (11.40%). The range of coefficient of variation in treated population was 11.40% to 17.14% as compared to control (14.67%). The results on 100 seed weight revealed that gamma rays treatment resulted in increase in mean 100 seed weight in some treatments and decreased in some treatments as compared to control. In accordance to this result Waghmare and Mehara (2000) [19] reported significant reduction in 100 grain weight in grasspea irradiated with gamma rays. Similar to this results Usharani and Kumar (2015) [17] also reported that the mean value for 100 seed weight decreased below the control in most of the treatments in blackgram. Chavan *et al.* (2019) [7] reported that 100 seed weight increased in 250 Gy, 300 Gy and 350 Gy while decreased in 150 Gy and 200 Gy over control in lathyrus.

Data regarding effect of different gamma rays doses on seed yield plant^{-1} in M_2 generation are presented in table 1. The mean value for seed yield plant^{-1} showed significant differences among the treatment. The maximum mean value for this characters was observed in T_3-350 Gy (24.56 g) and minimum in T_2-300 Gy (20.20 g) and control showed a mean value of 20.25 g. Highest range for this trait was recorded in T_2-300 Gy (79.31 g) and lowest range in T_3-350 Gy (63.6 g) as compared to control (26.92 g). The coefficient of variation increased in all the treatments for seed yield plant^{-1} as compared to control. The maximum coefficient of variation was observed in T_2-300 Gy (53.31%) followed by T_1-250 Gy (45.66%) while the minimum in T_3-350 Gy (40.51%). The range of coefficient of variation in treated population was 40.51% to 53.31% as compared to control (34.17%). The result on seed yield plant^{-1} revealed that the mean seed yield plant^{-1} increased in most of treatments *viz.*, T_3-350 Gy and T_1-250 Gy and decreased in some treatments *i.e*., T_2-300 Gy as compared to control. But the wide range of variation observed in all the treatments as compared to control indicated the presence of high within treatment variation. In accordance to this result Aney (2013) [4] reported significant variability in yield in gamma irradiated population of pea. In contrary to this Usharani and Kumar (2015) [17] observed decrease in mean values for single plant yield below control in most of the treatments and more than control in some treatment. Aditya *et al*. (2017) [1] also reported that seed yield plant^{-1} in variety BSS-2 of soybean showed maximum mean in control closely followed by 50 Gy, 100 Gy and 150 Gy, while it decreased in higher doses. Chavan *et al.* (2019) [7] also reported that grain yield plant^{-1} was found to increase in 150 Gy, 250 Gy and 300 Gy and decreased in 200 Gy and 350 Gy treatments as compared to control.

The results obtained from this investigation revealed that the germination per cent reduced in both M_1 and M_2 generation as compared to control. The reduction in germination was more in M_1 than M_2 generation. Mortality percentage was more in M_1 than M_2 generation. In M_2 generation number of branches plant^{-1} increased significantly in all the treatments as compared to control. Days to first flower increased in 350 Gy and decreased in 250 Gy and 300 Gy and 300 Gy as compared to control. Days to maturity decreased in 350 Gy and increased in 250 Gy and 300 Gy over the control. Plant height increased in 350 Gy and 300 Gy while decreased in 250 Gy over control. Number of pods plant^{-1} was observed to increase in all treatments *i.e*. 250 Gy, 300 Gy and 350 Gy over the control. 100 seed weight also increased in all the three treatment of gamma irradiation as compared to control. Seed yield plant^{-1} was found to increase in 250 Gy, 300 Gy and 350 Gy as compared to control.

Table 1: Effect of gamma ray on quantitative traits in M_2 generation

Treatments	Parameters	Doses of gamma rays			
		T_1 (250 Gy)	T_2 (300 Gy)	T_3 (350 Gy)	T_4 (Control)
Germination%	M_1	51.30	47.26	41.12	71.50
	M_2	68.34	67.81	56.96	84.84
Mortality%	M_1	58.28	58.69	77.96	25.37
	M_2	49.31	50.01	54.38	21.42
Days to first flower (Days)	Range	15	20	22	4
	Mean	47.89	48.02	49.33	59.62
	SD	4.27	4.88	5.61	1.16
	CV (%)	8.93	10.16	11.37	1.94
Days to maturity (Days)	Range	15	19	25	5
	Mean	109.04	109.71	108.40	115.52
	SD	4.62	5.61	7.60	7.01
	CV (%)	4.23	5.12	7.01	1.49
Plant height (cm)	Range	63	81	70	30
	Mean	48.21	53.09	56.35	53.04
	SD	10.54	12.78	11.75	7.06
	CV (%)	20.85	24.08	21.87	13.32
Number of branches plant^{-1}	Range	7	7	6	3
	Mean	3.70	3.86	3.74	3.66
	SD	1.21	1.12	1.02	0.85
	CV (%)	32.69	29.02	27.40	23.35
Number of pods plant^{-1}	Range	280	332	259	83
	Mean	62.71	79.60	76.59	59.71
	SD	40.89	52.41	40.44	26.37
	CV (%)	65.20	65.83	52.80	44.16
100 seed weight (g)	Range	8.96	9.29	5.66	2.20
	Mean	6.30	6.84	6.91	4.26
	SD	1.08	0.84	0.78	0.62
	CV (%)	17.14	12.32	11.40	14.67
Seed yield plant^{-1} (g)	Range	76.54	79.31	63.66	26.92
	Mean	23.77	20.20	24.56	20.25
	SD	10.85	11.17	9.95	6.92
	CV (%)	45.66	53.31	40.51	34.17

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