

Studies on the effect of different doses of gamma radiation on seed germination of maize

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ABSTRACT

A laboratory-based experiment was conducted to see the effect of different doses of gamma radiation on maize seeds. *Zea mays* L. seeds were collected from the Grain Storage of the new Developmental Farm, The University of Agriculture Peshawar, Pakistan. Those seeds were taken to the Nuclear Institute for Food and Agriculture (NIFA), Peshawar to expose the seeds to different doses of gamma radiations i.e. 0.8, 1.0, 1.2, 1.4 KGys. The effects of different doses of gamma radiations on different genetic parameters were observed and for this purpose, those seeds were set for germination in the Laboratory of Weed Science (Botany), The University of Agriculture, Peshawar. Germination percentage was decreased under all the radiation doses applied and ranged between 51.75% and 86%. Highest doses of radiation i.e. Lowest germination percentage was recorded in the highest dose of radiation 1.4 kGy treatment (51.75%), which is significantly different from the control and 0.8, 1.0, 1.2 and 1.4 kGy treatments. Similarly, all other parameters such as root length, shoot length, fresh weight of root, fresh weight of shoot, dry weight of root, dry weight of shoot were significantly decreased with the gradual increase of radiation doses. Due to some constraints, we could not continue this experiment in the field. However, the conclusions are that gamma radiations showed the mutation effect on seeds of maize. This could possibly be used for the release of the new variety, which could be a variety with short stature and drought resistant.

Keywords: Gamma radiations, Doses, Seed germination, *Zea mays* L.

1. INTRODUCTION

Zea mays L. (Maize) is one of the most consumable crops in Pakistan, belong to family Poaceae. The application of change strategies has created a huge measure of hereditary inconstancy and has assumed a role in plant nurture and hereditary investigations all through the world [1]. Progress is also being made in deciphering the biological nature of DNA damage, repair, and mutagenesis [1]. Mutation breeding is now one of the successful techniques to induce genetic variability in the adapted crop cultivars. Induced mutations are also widely used for promoting genetic recombination, the creation of new genetic resources and breakage of unwanted linkages. As a result, mutation breeding has become an effective way for supplementing existing germplasm and improving cultivars and is recognized as one of the important accessories to the mainstream of plant breeding [2, 3]. For example, Chatterjee et al. [4] conducted an experiment on broadening the genetic base of opium poppy (*Papaver somniferum* L.); through actuated change is an advantageous instrument that can prompt the improvement of hereditary inconstancy [4].

Maize (*Zea mays* L.) and *Abelmoschus esculentus* is the most important cereal crop of Pakistan and Khyber Pakhtunkhwa grown in the irrigated and rain fed areas. It is a rich source of food and also used as a fodder crop [5, 6]. Corn yield per unit area is still far below its yield potential obtained in other corn producing countries [7]. Maize yield in Pakistan is only 3.0 t ha⁻¹ compared to USA 8.92 t ha⁻¹, Canada 7.82 t ha⁻¹, France 7.14 t ha⁻¹ and

China 4.85 t ha⁻¹ [8]. Different agricultural methods have been used to improve the quality and yield of Maize crop but in Pakistan, very little work has been done on the genetic mutation of Maize [9]. A new mutant with albino-to-zebra leaf color trait which was selected from the progeny of the indica rice maintainer line D57B by irradiation with 300 Gy Co-60 gamma rays, the mutant was then backcrossed with D57A as the recurrent parent, and finally a novel type of cytoplasmic male sterile rice line. It also had excellent combining ability with other varieties. In the mixed sowing experiment of Gaoguang A and hybrid rice combinations, the weaker competitiveness of young seedlings of Gaoguang A resulted in the natural death of its young seedlings Zhang et al. [10]. Similarly, Benslimani et al. [11] reported that *Daturas* are tropane alkaloid producing plants from the *Solanaceae* family, which have very important pharmaceutical properties. Moreover, their chemical synthesis was found to be more expensive than their direct extraction from their natural source, the plant itself. In addition, their concentration in the whole plant remains low even among the most productive bodies, both when grown in artificial (in vitro hydroponics.) or natural conditions [12]. This variety is expected to be used in an induced-mutation programmed for the sake of obtaining mutant lines that will exhibit increased tropane alkaloid concentrations [13].

The achievement of flower color mutation could be founding higher doses/concentrations as compared to lower doses/conc. The seed material of *Phaseolus vulgaris* L variety

"Varun" was treated with physical mutagen with different doses of gamma rays and chemical mutagen Ethyl Methane Sulphonate (EMS) at the different concentrations. The treated M2 phenotypic progeny showed different flower color mutations like white, purple, blue, red, and yellow as mutants developed due to the effect of physical and chemical mutagens Borkar and More, [14].

Kaushik and Sastry, [15] estimate the mutagenic effects of gamma rays on yield and its attributes in M 7-generation of fenugreek cultivar RMt-1. The experiment comprised of 29 M 7 mutant lines generated through irradiation with gamma-rays along with three checks namely; RMt-1, RMt-143 and local check (desi Methi) were conducted to estimate the magnitude and pattern of diversity present in the crop using metroglyph analysis and index score values. The experiment was carried-out in completely randomized block design replicated thrice during rabi 1997-98. All the families grouped into three groups based on the frequency distribution of index score values. 11 Families of the 3rd group showed good coordination between index score values and scatter diagram. The family No. 20 (551-1) found to be the best among all families and further evaluation of this family in yield trial may prove beneficial. The sesame mutants induced through gamma ray treatment and their respective three parental sources were evaluated for five levels of drought stress using aqueous solutions of PEG-6000. Results evidence significant differences among genotypes and drought stress levels. Genotypes hsc105, lc162, mc112, ef147, and hc107 were found to be more drought tolerant than their respective parents. Although sesame is reputed to be a drought-tolerant crop, it is very susceptible to drought at the germination stage and sufficient soil moisture is required for sesame seed emergence Boureima et al. [16].

Groundnuts to be an important source of protein and other nutrients, these can be placed both in seed and in other organs of the plants. Using irradiation it can be obtained variability in plants, both morphologically and in the chemical composition of the seed. This approach for groundnut crop was applied in different doses, between 5000-10000R and there were identified variants where smaller or bigger doses presented no influence and variants with significant influence Lancu, [17]. The mutagenesis has gained popularity in plant genetics research as a means of inducing novel genetic variation. Induced mutations have been applied for the past 40 years to produce mutant cultivars in sunflower by

changing plant characteristics that significantly increase plant yield and quality. The three mutagenic agents affected seedling height, reducing it with increasing dosage Cvejic et al. [18].

The seeds of the well-adapted and popular mustard variety BARIsarisha-11 irradiated with gamma ray using ⁶⁰Co gamma cells. The selection was made from M 2 generation during 2005-06. Desirable mutants were confirmed in M 4 generation during 2007-08 and ten true breeding mutants having higher seed yield per plant with desirable morphological characters and yield attributes were selected Malek et al. [19]. The mutants also showed tolerance against Alternaria blight disease and lower aphid infestation. Results of the yield trials, as well as screening against Alternaria blight disease and aphid carried out across the country indicated that MM-10-04 and MM-08-04 were suitable for widespread cultivation. The extent of variability for yields generation of two genotypes of *Brassica napus* L. Kumar et al. [20]. The isolation of certain promising mutants signified the role of mutation breeding in enhancing the genetic variability in *Brassica napus* L. Kumar et al. [20]. The radio-sensibility of groundnuts is more emphasized in Tamburesti variety comparative with other varieties and lines, the variety has a raised specific radio-sensibility, probably determined by its physiological and biochemical peculiarities and by the fact that it was created through chemical mutagenesis (in 1983) Lancu, [21].

Barshile and Boddu, [22] studied three well-known mutagens, sodium azide (SA), ethyl methane sulphonate (EMS) and gamma radiation (GR) employed to induce genetic variability for improvement of locally popular chickpea (*Cicer arietinum* L.). Analysis of variance showed both positive and negative significant increase in the quantitative traits among the mutant lines. The higher heritability coupled with high genetic advance was observed for quantitative traits like a number of pods/plant and number of seeds/plant for the mutants in the chickpea cv. 'Vijay'. Variation was also observed for a number of bands electrophoresed in seed protein of mutants.

This study will be conducted with the following objectives: To see the effect of different doses of Gamma radiation on seed germination and growth of Maize, and also aims to identify the most effective dose of gamma radiations in reducing or improving the seed germination and growth, and to see a potential mutation in Maize Seeds.

2. EXPERIMENTAL SECTION

Maize (*Zea mays* L.) seeds were collected from the Grain Storage of the New Developmental Farm, the University of Agriculture, Peshawar. Those seeds were taken to the Nuclear Institute for Food and Agriculture (NIFA), Peshawar to expose the seeds to different doses of Gamma i.e. 0.8, 1.0, 1.2, 1.4 KGys. The effects of different doses of gamma radiations on different genetic parameters were observed and for this purpose, those seeds were set for germination in the Laboratory of Weed Science (Botany), The University of Agriculture, Peshawar.

Under laboratory conditions, the following parameters were studied by placing 10 seeds in each Petri dish with four replications for each dose in a randomized complete block design.

Those seeds were kept on filter paper with distilled water (5 ml/Petri dish) in sterilized Petri dishes which were kept at room temperature in the Weed Science Laboratory, The University of Agriculture, Peshawar for four weeks. The seeds were kept moist until the termination of the experiment.

There were four radiation doses i.e. 0.8, 1.0, 1.2, 1.4 KGys and one control treatment, each treatment was replicated four times. The seeds were observed daily and whenever water required, the same amount of water was added.

After each 48 hours, data for the following genetic parameters were recorded.

- Length of the stem

- Length of roots
- Number of roots
- Germination rate
- Time of germination
- Germination percentage

Length of the stem and roots were measured on a calibrated scale in cm after 48 hours. The number of roots was obtained by counting the roots of each grain. Germination percentage was calculated by multiplying the total number of grains germinated

3. RESULTS SECTION

Germination percentage. When the effect of gamma radiation on seed germination of maize seed was compared with the controlled one at Least Significant Test at (LSD = 4.0999) a significant difference was observed (Table 1). Germination percentage decreased under all the radiation doses applied and ranged between 51.75% and 86% (Table 1). Lowest germination percentage was recorded in the 1.4 kGy treatment (51.75%), which is significantly different from the control and 0.8, 1.0, and 1.2 kGy treatment (P = 0.0001). However, germination percentage in 0.8 kGy (Table 1).

Root length. When the effect of gamma radiation on the root length of maize seed was compared with the controlled one at Least Significant Test at (LSD = 2.6977) a significant difference was observed (Table 2). Root length increased under all the radiation doses applied and ranged between 1.310% and 11.6% (Table 2). Lowest root length was recorded in the 1.0 kGy treatment (1.310%), which is significantly different from the control and 0.8, 1.2, and 1.4 kGy treatment (P = 0.0002). However, root length in 0.8 kGy (Table 2).

Table 1. Effect of gamma radiation on seed germination of seeds of maize.

Treatments/doses gamma radiation	Means
T1 (0.8 Kgys)	86.000 A
T2 (1.0 Kgys)	73.250 B
T3 (1.2 Kgys)	56.750 C
T4 (1.4 Kgys)	51.750 D
T5 (Control)	87.750 A
(LSD)0.05	4.0999

Table 2. Effect of different doses of gamma radiation on the effect of root length of maize.

Treatments/doses gamma radiation	Means
T1 (0.8 Kgys)	11.623 A
T2 (1.0 Kgys)	1.3100 D
T3 (1.2 Kgys)	2.7500 CD
T4 (1.4 Kgys)	4.2500 BC
T5 (Control)	6.8750 B
(LSD)0.05	2.6977

Shoot length. When the effect of gamma radiation on shoot length of maize seed was compared with the controlled one at Least Significant Test at (LSD = 0.6528) a significant difference was observed (Table 3). shoot length decreased under all the radiation doses applied and ranged between 1.470% and 12.78% (Table 3). Lowest shoot length was recorded in the 1.0 kGy

with 100 and divided by the total number of grains planted. Time of germination was calculated by daily observation of each Petri dish.

Statistical analysis. Data on the above parameters were recorded and was statistically analyzed through computer software Statistix 8.1 using the Analysis of Variance (ANOVA). The significant means were separated through Least Significant Different test (LSD).

treatment (1.470%), which is significantly different from the control and 0.8, 1.2, and 1.4 kGy treatment (P = 0.000). However, shoot length in control kGy (Table 3).

Table 3. Effect of different doses of gamma radiation on the effect of shoot length of maize.

Treatments/doses gamma radiation	Root length
T1 (0.8 Kgys)	12.358 A
T2 (1.0 Kgys)	1.4700 C
T3 (1.2 Kgys)	2.0925 C
T4 (1.4 Kgys)	3.4500 B
T5 (Control)	12.780 A
(LSD)0.05	0.6528

Number of roots. When the effect of gamma radiation on No of roots of maize seed was compared with the controlled one at Least Significant Test at (LSD = 1.5422) a significant difference was observed (Table 4). No of roots increased under all the radiation doses applied and ranged between 1.4750% and 11.55% (Table 4). Lowest No of roots was recorded in the 1.0 kGy treatment (1.4750%), which is significantly different from the control and 0.8, 1.2, and 1.4 kGy treatment (P = 0.0002). However, a number of roots in 0.8 kGy (Table 4).

Table 4. Effect of different doses of gamma radiation on the number of root of maize after 15 days of germination.

Treatments/doses gamma radiation	Means
T1 (0.8 Kgys)	11.550 A
T2 (1.0 Kgys)	1.4750 C
T3 (1.2 Kgys)	1.5750 C
T4 (1.4 Kgys)	1.8500 C
T5 (Control)	7.9500 B
(LSD)0.05	1.5422

Fresh weight of root. When the effect of gamma radiation on fresh weight of root of maize seed was compared with the controlled one at Least Significant Test at (LSD = 0.1585) a significant difference was observed (Table 5).).

Table 5. Effect of different doses of gamma radiation on fresh weight of root after 15 days of germination.

Treatments/doses gamma radiation	Means
T1 (0.8 Kgys)	2.4715 A
T2 (1.0 Kgys)	0.3378 BC
T3 (1.2 Kgys)	0.2073 C
T4 (1.4 Kgys)	0.4530 B
T5 (Control)	2.3150 A
(LSD)0.05	0.1585

Fresh weight of root increased under all the radiation doses applied and ranged between 0.2073% and 2.47% (Table 5). The lowest fresh weight of root was recorded in the 1.2 kGy treatment (0.2073%), which is significantly different from the control and 0.8, 1.0, and 1.4 kGy treatment ($P = 0.0002$). However, the fresh weight of root in 0.8 kGy (Table 5).

Dry weight of root. When the effect of gamma radiation on the dry weight of root of maize seed was compared with the controlled one at Least Significant Test at ($LSD = 0.0281$) a significant difference was observed (Table 6). The dry weight of root decreased under all the radiation doses applied and ranged between 0.0350% and 0.1578% (Table 6). Lowest dry weight of root was recorded in the 1.0 kGy treatment (0.0350%), which is significantly different from the control and 0.8, 1.2, and 1.4 kGy treatment ($P = 0.0001$). However, the dry weight of root in control (Table 6).

Table 6. Effect of different doses of gamma radiation on dry weight of root after 15 days of germination.

Treatments/doses gamma radiation	Means
T1 (0.8 Kgys)	0.0683 B
T2 (1.0 Kgys)	0.0350 C
T3 (1.2 Kgys)	0.0538 BC
T4 (1.4 Kgys)	0.0528 BC
T5 (Control)	0.1578 A
(LSD)0.05	0.0281

Fresh weight of shoot. When the effect of gamma radiation on fresh weight of shoot of maize seed was compared with the controlled one at Least Significant Test at ($LSD = 0.4505$) a significant difference was observed (Table 7).). Fresh weight of shoot decreased under all the radiation doses applied and ranged between 0.480% and 4.867% (Table 7). The lowest fresh weight of shoot was recorded in the 1.0 kGy treatment (0.480%), which is significantly different from the control and 0.8, 1.2, and 1.4 kGy treatment ($P = 0.0002$). However, the fresh weight of shoot in control (Table 7).

Table 7. Effect of different doses of gamma radiation on fresh weight of shoot after 15 days of germination.

Treatments/doses gamma radiation	Means
T1 (0.8 Kgys)	2.4343 B
T2 (1.0 Kgys)	0.4800 C
T3 (1.2 Kgys)	0.7058 C
T4 (1.4 Kgys)	0.7165 C
T5 (Control)	4.8670 A
(LSD)0.05	0.4505

Dry weight of shoot. When the effect of gamma radiation on the dry weight of shoot of maize seed was compared with the controlled one at Least Significant Test at ($LSD = 0.0671$) a significant difference was observed (Table 8).).

Table 8. Effect of different doses of gamma radiation on dry weight of shoot after 15 days of germination.

Treatments/doses gamma radiation	Means
T1 (0.8 Kgys)	0.2148 B
T2 (1.0 Kgys)	0.0560 C
T3 (1.2 Kgys)	0.0878 C
T4 (1.4 Kgys)	0.0738 C
T5 (Control)	0.3273 A
(LSD)0.05	0.0671

Dry weight of shoot decreased under all the radiation doses applied and ranged between 0.056% and 0.3273% (Table 8). The lowest dry weight of shoot was recorded in the 1.0 kGy treatment (0.056%), which is significantly different from the control and 0.8, 1.2, and 1.4 kGy treatment ($P = 0.0001$). However, dry weight of shoot in control (Table 8).

Discussion. Application of different doses of gamma radiations significantly changed different parameters of maize while growing in Petri dishes. Our results are in line with the results obtained by Borkar and More, [14] and Barshile and Boddu, [22]. However, they germinated seedlings to flowering, where the effects of radiations have been shown in flowers, while we tested seeds in Petri dishes. Therefore, possibilities are the colour of flowers, plant height and number of flowers is also expected to be increased if seeds of maize are grown in the field. Moreover, based on the decreased in the shoot length, the effect of that gamma radiation could lead us to release a short-statured variety of maize [8].

In this experiment, with the increase of doses from 0.8 to 1.4 kGy the germination percentage was decreased and root length, shoot length was decreased gradually and a maximum number of seeds germination were inhibited and roots and shoot length were decreased significantly at 1.4 kGy. Our results are also similar to Borkar and More, [14] and [23], who concluded that the percentage of flower color mutation increased according to the doses/concentrations of the mutagens.

However, our results are in contradiction to those of Arulbalachandran *et al.* [24], who concluded that genetic variation has led to an increase in the quantitative traits of crops. The variability on the genome is induced by mutation, which enhances the productivity. They evaluated variability on quantitative characters such as, plant height, number of branches/plant, number of leaves/plant, number of fruit clusters/plant, number of pods/plant, number of seeds/pod, yield/plant and 100 seed weight of black gram in M(2) generation by the effect of mutation by gamma rays. The results were shown high genetic variability, heritability and genetic advance with significant enhancement ($P 0.05$ and $P 0.01$) in growth and yield traits. Our results of various parameters show similar results to [25], he explains the effects of gamma radiation on aflatoxin B1 levels in soybean and the properties of soybean

In our experiment, all parameters were significantly affected by the different doses of gamma radiations, which are different to those of Kaushik and Sastry, [15], who stated that some of the parameters could not be affected such as branches per plant, pod length, days to maturity and harvest index. We could have seen those effects as well if we planted Maize plant in the field. But due to unavoidable circumstance and time limit, we could not continue our experiment in the field [26].

Another positive use of those gamma radiations could be used to protect crops from pathogens such as Sharma *et al.* [27] used those mutagens for the control of *Fusarium oxysporum* f. sp. Pisi, which is a serious production constraint for peas worldwide. Applications of different doses of mutagens have a positive and negative effect on both qualitative and quantitative characteristics of plants (Barshile and Boddu, [22]). However, this effect could be different on different plant species and different growth conditions. As our experiment was limited to the growth of maize

plant for 15 days only, in which are parameter was significantly affected both positively and negatively. Such as germination

percentage was decreased and root and shoot length were inhibited and consequently fresh and dry weight was gradually decreased.

4. CONCLUSIONS

On the basis of above work, it can be concluded that the effects of all the doses of Gamma Irradiations applied were highly effective against all parameters studied where the growth parameter is

shown a significant decrease in growth, which could possibly be used to introduce a new short statured variety with less water requirement.

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