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Assessing genetic variability induced in chickpeas by gamma rays

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Mutation breeding is a useful technique for discovering desirable traits in chickpea genomes. Therefore, the overall objective of this study was to bring out genetic variability in the genotype of chickpeas by treating the seeds with six doses (0.0, 25, 50, 100, 200, 400 and 800 rad) of gamma irradiation to generate genetic variations in morphological traits and yield components to bring out information about the magnitude of genetic parameters related to these traits. Gamma-irradiated seeds were planted in a randomized complete block design. The genotypic coefficient of variations for growth traits, except for chlorophyll a, was higher than the corresponding values of the environmental coefficient of variation. High heritability coupled with moderate genetic advance was achieved in the number of branches per plant, as well as in chlorophyll b. All yield component traits had recorded very high values of heritability ranging from 96.4% for pod number per plant to 87.21% for the weight of 50 seeds. High values of heritability coupled with high genetic advance were recorded for the number of pods, pod weight, and seed weight per plant. It can be concluded that some growth parameters, number of branches per plant, as well as all yield component traits showed small environmental effects and exhibited high additive genetic performance on the phenotypic variation of these traits which can be improved through selection programs.

Key words: Chickpea, genetic parameters, genetic advance, gamma irradiation, yield components, growth traits.

INTRODUCTION

Chickpeas a valued ranking third (accounting for 11. 67 million tons annually) in the world among pulses in productivity behind dry faba bean (25.66 million tons) and field pea (11.69 million tons), with a mean annual productivity of over 11.5 million tons. Combined production of chickpeas and peas was nearly equal to that of beans which indicated their overall importance. The land area devoted to chickpeas has increased in recent years with most of the production centered in India and now the area stands at an estimated 14.56 million hectares from which 8.9 ha in India alone (FAOSTAT, 2019). The production of the unit area has increased since 1961 with about 6 kg/ha per annum. About 2.3 million tons of chickpeas injected into the world markets yearly to support the requirements of countries are unable to meet their needs through domestic production (Merga and Haji, 2019). Grain legumes are an important nutritional value in the diet of millions of people in

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developing countries, sometimes referred to as the meat of poor people. Legumes are a significant part of the vegetarian diet because other foods they consume do not contain much protein (Latham, 1997).

Legume crops were grown in rotation with cereals because of their significant roles played in nitrogen fixation. Agricultural research in many countries all over the world has concentrated efforts on increasing cereal yields to achieve food security. Therefore, grain legume crops form systems of nutritional security, thus enabling the research will have significant impacts on nutritional security and soil fertility (Merga and Haji, 2019). Approximately 6.5 billion people currently live on our planet; this number is projected to rise by 2050 to over 9 billion people. The world will need to produce more food for more people from fewer resources to provide this growing demand. Chickpeas play a leading role in food security in the world by covering protein deficiency in the daily food ration of Indian and African populations to meet the WHO/FAO requirements (Malunga et al., 2014). Amongst pulse crops, chickpeas ranked second in area (15.3% of total) and third in productivity (15.42%) (Merga and Haji, 2019). Chickpeas contain a high level of protein that accounts for almost 40% of its weight considering it to be unique. It has potential health benefits because it reduces cardiovascular disease, diabetic disease, as well as cancer risks (Merga and Haji, 2019). Chickpeas are grown in about 57 countries all over the world. South-East Asia contributed 80% of the regional production. India is the single largest producer of chickpeas in the world accounting for 77.63% (9.075 million tonnes) of the total production (11.67 million tonnes) annually (Merga and Haji, 2019). Globally, the average global chickpea yield is about 1.8 tons/ha, but this average is about 1.46 tons/ha in West and South Asia (Merga and Haji, 2019). The outlook of chickpeas is excellent considering nutrient concentrations and food value.

Chickpea, *Cicer arietinum* L. is a self-pollinated diploid (2n = 16) plant (Varshney et al., 2013). It is the third food crop legume in the world grown in an area of about 11.2 million hectares (Qureshi et al., 2014). Different physical mutagens as gamma rays were applied to induce, as well as to increase genetic variability in chickpeas (Maluszynski et al., 2002). Few works were focused on chickpeas for mutation breeding. Gamma rays cause variation in growth at genetic, biochemical, physiological, and morphogenetic levels (Borzouei et al., 2010). Mutation breeding is highly important in chickpea improvement techniques via inserting beneficial mutations in target loci (Sangsiri et al., 2005). The available germplasm of chickpeas does not indicate enough genetic variability for the improvement of economic traits (Shah et al., 2006). Mutation breeding was used to improve the economically important traits in addition to eliminating the undesirable genes from the available lines (Lippert et al., 1964). It is a useful technique to exhibit the genetic variation spectrum in the species within a short time, which plays an important role in the development of many crop varieties (Micke, 1988).

Mutation breeding value could be improved by inserting different mutant genes in the same genome (Gottschalk, 1986). Mutagenesis could generate different mutant alleles different with different degrees in great modification. Gamma irradiation is an important mutagenic agent employed to increase mutation frequency in chickpeas (Borkar and More, 2010). The induction of macro morphological mutants for generating genotypic or probably phenotypic variability is of great interest to submitting additional gene biomarkers for genetic improvement of chickpeas (Laskar et al., 2015). Genetic variability is an important method for bringing out crop improvement. Mutation breeding is one of the promising approaches for inducing genetic variations in chickpea plants. This technique is used to generate new genetic variations in chickpeas to be used for improving growth and yield traits, without major changes in the total genetic structure (Khattak and Klopfenstein, 1989).

Estimates of genetic parameters such as genotypic and phenotypic coefficient of variations GCV and PCV), heritability in a broad sense, as well as genetic advances for different quantitative traits, are useful in designing an effective breeding technique (Kozgar, 2014). The genotypic coefficient of variation is measured by the level of genetic variability controlled by the traits. The genotypic coefficient of variations alone cannot estimate the amount of magnitude in the heritable variation (Wani, 2011). Heritability is an important genetic parameter for selection-based improvement because it reflects the extent of transmissibility of genetic elements related to the trait into the next generations (Kozgar, 2014). Measurement of heritability alone does not provide information about the expected genetic gain in future generations but must be considered in conjunction with the measurement of genetic advance, the alteration in mean value between generations (Wani, 2011).

In recent years, the mutagenesis technique has taken great attention as a good tool for revealing gene expression and for trait improvement. Measurements of the degree of variability in quantitative traits in the genetic resources of chickpeas are important for crop breeding techniques (Gomes et al., 2020). Characterization of morphological and growth traits is the first step in the description and classification of genetic resources (Balkaya et al., 2010). Considering the aforementioned points, the present investigation was carried out to measure the genetic variability induced by gamma rays to provide knowledge about heritability which reflected the extent of transmissibility of genetic factors to the next generations for improving economic traits in chickpeas.

MATERIALS AND METHODS

In this investigation, the effect of gamma rays as a physical mutagen was studied to increase the genetic variability in chickpea genotypic resources.

Plant

Chickpea variety Giza-195 from Kabuli type used in this study was obtained from Field Crops Research Institute, Agricultural Research Center, Giza, Egypt.

Gamma irradiation

Air-dried seeds of chickpea were irradiated with 25, 50,100, 200, 400, and 800 rad of gamma rays (dose rate 1.249 kGy h^{-1}) in addition to the control of unirradiated seeds. Where Gy is the unit of gamma rays dose in the international system of units defined as the absorption of one-Joule radiation energy per one material kilogram. The radiation dose was expressed as KR or Gray (Gy), where one Gy = 100 rad, as well as one KR = 10 Gy. Therefore, the unit of absorbed dose was rad which means irradiation absorbed dose. Irradiation was applied at the Atomic Energy Center, Nasr City, Cairo, Egypt, using irradiation equipment GSR D_1 (Germany) with a radioisotope Cobalt-60 source. The healthy and viable seeds (moisture 11%) were used to be treated with gamma rays.

Field experiment

This experiment was carried out in the Agri-field of the Genetic Department inside the campus of Mansoura University through the winter season of the academic year 2022/2023. The experiment was designed in triplicate with three rows for each treatment using a complete randomized block design. The 72 seeds were treated with each dose of gamma irradiation and then sown in three replicates in the field along with the control (24seed/replicate) to raise M_1 generation. Three seeds were sown in each hole at a distance of 40 cm between holes and 70 cm between rows. Each row contained eight holes; in each hole, three seeds were planted. The size of each replicate was 770 × 350 cm. Each experimental replicate consisted of seven rows, each row had 350 cm length and 110 cm width. The distance between replications was 1 m long × 770 cm wide. Chlorophyll concentration (mg/g fresh weight) in leaves was measured at 50 days from the day of sowing according to Mackinney's (1941) methodology. Meanwhile, the number of primary branches, plant height (cm), plant dry weight (g), total number of pods per plant, as well as 50 seed weight (g) was estimated at the harvesting stage when the plants began blooming according to Amri-Tilioiune et al. (2018).

The plants were thinned after complete emergence (which reached 14 days after planting) to two plants per hill. The plants were grown without any irrigation throughout the season, except for drain water. The recommended dose of phosphorus chemical fertilizer was applied by hand on one side of the row at 6 cm depth, without application of chemical nitrogen fertilizer to the plants throughout the growing season. The recommended nature of the Egyptian Ministry of Agriculture concerning chickpea production was applied at the proper time. Natural selfing takes place between germ cells, therefore, mutation takes place in the germ part cells of seeds. The M_1 generation faced lethality at various stages of growth and development. The significance of M_1 is as a source for M_2 generation, as well as ensuring maximum viability in their plants which is beneficial. M_1 plants may carry some seeds resulting from the fusion of male and female gametes which may carry the same mutation and thus are homozygous mutation. These seeds generated individual plants that carried the mutant phenotype which can be observed in M_2 generation (Calicius, 2006). Most mutations are recessive and cannot be observed in heterozygous states. Meanwhile, dominant mutants were observed in heterozygous condition, therefore it can appear in the M_1 generation, but recessive mutation was observed in a homozygous state in the M_2 generation after the seeds were formed by fusion between male and female gametes that carry the same mutation. These mutant phenotypes appeared in the M_2 generation (Kumar et al., 2019). In this study, growth and yield traits were assessed in M_1 individual plants that may carry dominant mutant phenotype.

Seedlings emerged

The germination ratio of M_1 seeds was recorded every day from the sowing till the last seedling emerged. The germination ratio was expressed by the following formula according to Qureshi et al. (2014).

$$\text{Seedling germination ratio} = \frac{\text{Number of seedling emerged}}{\text{Total seed sown}}$$

where survival ratio = number of M1 seedlings emerged / number of seedlings emerged in unirradiated seeds.

Statistical analysis

Results are the mean values of three biological replicates. The data collected were subjected to the analysis of variance to test the significance of differences between means using the F-test. A least significant difference (LSD) was used to test the significance of differences between two means at 0.05 and 0.01 levels of probability according to Duncan (1955). PCV and GCV, broad sense heritability, as well as genetic advance, were assessed according to Amri-Tilioiune et al. (2018). The PCV and GCV were assessed according to Singh and Choudhary (1985). GCV and PCV were classified as low (0-10%), moderate (10-20%), and high (above 20%) according to Sivasubramanian and Madhava Menon (1973). Meanwhile, heritability percentage was classified as low (0 - 30%), moderate (30 - 60%) and high (above 60%) according to Robinson et al. (1949).

The estimates of genetic advance (GA) and genetic advance as a percentage of the mean (GAM) were calculated according to the method of Assefa et al. (1999).

$$\text{GAM (\%)} = \frac{\text{GA}}{\bar{\text{X}}} \times 100$$

Genetic advance assumed as a percentage of the mean was classified as low (0-10%), moderate (10-20%), as well as high (above 20%) according to Johnson et al. (1955).

RESULTS AND DISCUSSION

Growth traits

The estimates of genetic parameters (Table 1) revealed that the PCV was higher than the corresponding values of the GCV. The higher values of GCV (> 20%) were recorded for the number of branches per plant (35.13%) followed by plant length (33.78%), plant dry weight (27.78%), and chlorophyll b (23.16%). Nevertheless, the smallest GCV was recorded for chlorophyll (5.24%) and total chlorophyll (5.83%). Whereas the higher values in

Table 1. Genetic components of growth traits in M_1 generation of chickpea treated with gamma rays.

Genetic parameter	Number of branches per plant	Plant dry weight	Plant height	Chlorophylls concentration		
				Chl a	Chl b	Total Chl
Mean±Sd	24.57± 5.28	43.43±6.82	70.57±5.27	1.35±0.26	0.45±0.45	1.81±0.24
DMS	71.55	98.31	69.96	0.165	0.526	0.15
EM	7.98	27.88	11.06	0.107	0.267	0.02
GV	21.19	23.47	19.63	000.03	000.17	00.04
PV	29.17	51.35	30.69	000.69	000.18	00.06
EV	07.98	27.88	11.06	000.67	000.01	00.02
H%	72.64	45.71	63.96	003.73	094.40	68.25
GCV%	35.13	27.78	33.78	005.24	023.16	05.83
PCV%	41.21	41.09	42.24	027.13	023.83	07.06
ECV%	06.08	13.31	08.46	021.89	000.67	01.23
EGA	08.08	06.74	07.29	002.92	000.83	00.35
GAM%	32.88	15.52	10.34	210.07	185.03	02.78

Sd, Standard deviation; DMS, doses of gamma rays mean squares; EM, error mean squares; GV, genotypic variance; PV, phenotypic variance; EV, environmental variance; H (%), heritability in a broad sense in percent; GCV (%), genotypic coefficient of percent variation; PCV (%); phenotypic coefficient of percent variation; ECV (%), environment coefficient of percent variation; EGA, expected genetic advance; GAM, genetic advance as percent of mean at 5% selection intensity.

PCV (> 20%) were recorded for plant length (42.24%) followed by the number of branches per plant (41.21%), plant dry weight (41.09%), chlorophyll a (27.13%), and chlorophyll b (23.83%). Meanwhile, the smallest PCV was recorded for total chlorophyll (7.06%).

Higher estimates of genetic advance (> 30%) as a percentage of the mean were recorded for chlorophyll a (210.07%), followed by chlorophyll b (185.03) and number of branches per plant (32.88). The lowest values of genetic advance were recorded for total chlorophyll (2.78%), followed by plant length (10.34%), as well as plant dry weight (15.52%). The highest value of heritability was recorded for chlorophyll b (94.4%), followed by the number of branches per plant (72.64%), total chlorophyll (68.25%), plant length (63.96%), and plant dry weight (45.71%). The lowest value of heritability (3.73%) was recorded for chlorophyll a. Higher GCV (> 20%) with a higher corresponding PCV (> 40%) coupled with high genetic advance (> 30%) was recorded for the number of branches per plant and chlorophyll b. Genetic variability increased in these traits providing great attention for further selection. This is because genetic variations in the population are prerequisites to selecting superior genotypes in any crop. Increasing genetic diversity within any crop population could improve the efficiency of the breeding technique process. The success in breeding mutation technique based on the irradiation doses applied produced sufficient diversity in addition to maintaining fertility and viability of seeds.

High heritability coupled with a moderate genetic gain was shown in the number of branches per plant and chlorophyll b, indicating that these traits were governed by additive gene interactions. This is considered an important trait in breeding and must be controlled by more than one locus. This study highlights the efficacy of applying both phenotypic and genotypic characterization approaches. The new variations induced in chickpeas via irradiated seeds were seen through phenotypic observation, suggesting that the population is suitable for breeding. For further evaluations, light must be shed on the relationship between genotypic variations induced and phenotypes expressed observed. This will provide important information about gene function and gene biomarkers that facilitate plant breeding techniques (Wani, 2011).

The diversity in crops was classified as genetic and non-genetic variability, including GCV and PCV. The GCV for growth characteristics varied from 35.13% (number of branches per plant) to 5.24% (chlorophyll a). Meanwhile, the PCV ranged from 42.24% (plant length) to 7.06% (total chlorophyll). The environmental coefficient of variation for chlorophyll a (21.89%) was higher than the GCV (5.24%) in the same trait. This indicated that chlorophyll was more influenced by environmental factors than genetic factors, leading to this trait being complicated in genetic improvement, through a selection of mutant genotypes. Meanwhile, the GCV was more than the environmental coefficient of variation for the number of branches per plant, plant dry weight, plant length, chlorophyll b, and total chlorophyll. This finding indicated that the genetic factors played a significant impact on these traits, which are not mainly susceptible to environmental factors. Thus, the heritability of these traits was increased above 40%. These results are in line with those reported before by Chen et al. (2020), who reported that heritability in a broad sense revealed the reliability of phenotypic value expressed as a breeding guide including additive and non-additive gene action.

Therefore, traits with high heritability estimates could be improved more quickly by selection than those with low heritability measurements because the latter are mainly affected by environmental factors. Thus, traits less affected by environmental factors can be easily selected because of high additive gene effects. These results are consistent with those reported earlier in peanuts by Kavera and Nadaf (2017).

All growth characteristics studied herein recorded different values of genetic gain as a percent of mean ranging from 210.07% (chlorophyll a) to 2.78% (total chlorophyll). This may be attributed to pleiotropic effects of mutagenic agents or newly mutated genes. This finding was agreed and supported by Rania et al. (2022) in cowpea. According to Qureshi et al. (2014), the dose of 100 Gy gamma rays induces the best increase in chlorophyll pigments (a and b) in addition to the total chlorophyll in chickpeas. On the other hand, Borzouei et al. (2010) stated that irradiated wheat genotypes with 100 Gy induced high concentrations in chlorophyll a, b, and total chlorophyll. They further demonstrated that chlorophyll pigment concentration was decreased with the doses of gamma rays increased. In addition, Qureshi et al. (2014) observed that total leaf chlorophyll had a maximum increase at 250 Gy of chickpea gamma irradiated seeds. Deshmukh et al. (1986) categorized phenotypic and GCV values > 20% referred to as high, values between 10 and 20% medium, and values < 10% low.

Depending on the aforementioned classification, the GCV obtained in this study was coupled with a PCV which produced high values exceeding 20% for the following traits; plant length, number of branches per plant, plant dry weight, and chlorophyll b. The GCV values were greater than ECV although the differences were small, indicating that environmental factors have lower effects on the gene expression of these traits. This agreed with Hailu et al. (2016), who found that PCV and GCV exceeding 20% were shown for the number of tillers/m^2 and number of kernels/spikes occurring in locations of barley cultivation. Meanwhile, the same authors found that lower values (< 10%) of GCV and PCV were recorded for days to heading and maturity in all locations, offering less scope of selection, because these traits were subjected to the influence of environmental factors (Panse, 1957).

The concept of heritability demonstrated whether variations observed in the population arose as a result of differences in their genotypes or due to environmental factors. Heritability was categorized according to Singh (2001) as follows: very high 80% or above, moderately high 60-79%, medium 40-59%, low heritability < 40%. Based on this classification, chlorophyll b exhibited very high heritability (94.4%), whereas the number of branches per plant, plant length, and total chlorophyll exhibited moderately high heritability (72.64, 63.96, and 68.25%, respectively). Meanwhile, medium heritability was recorded for plant dry weight (45.71%) and the lower heritability value was recorded for chlorophyll a (3.73%). The results indicated that traits exhibited very high heritability values reflecting the minimum effect of environmental factors on the phenotypic expression of these traits leading selection to be effective in their improvement (Singh, 2001). Therefore, selection may be difficult or virtually with low heritability traits due to the masking effect of the environmental factors. This is in line with Luzi-Kihupi (1998), who estimated heritability values for plant height, panicle length, number of filled grains per panicle, and 1000-grain weight in rice. Further explanation by Sardana et al. (2007), stated that high heritability values might not necessarily lead to an increase in the gain of genetic advancement unless sufficient genetic diversity in the germplasm was available. In this respect, Chand et al. (2008) detected high heritability values coupled with low genetic gain for the number of days to ear emergence in barley.

Yield components

The assessment of genetic parameters (Table 2) showed that the PCV in all traits of yield components was higher than the corresponding values in the GCV. The higher values in GCV (> 20%) were obtained in pod number per plant (77.42%) followed by pod weight per plant (67.18%) and seeds weight per plant (64.57%). Nevertheless, the smallest GCV was obtained for the weight of 50 seeds (12.82%). Whereas the higher PCV values (> 20%) were recorded in pods number per plant (78.84%) followed by pods weight per plant (69.16%) and seeds weight per plant (67.15%). The smallest PCV was recorded in 50 seeds weight. All yield traits recorded very high values of heritability ranging from 96.4% (pod number per plant) to 87.21% (weight of 50 seeds). In addition, all the parameters of yield components recorded higher values in GCV than that corresponding in ECV. Higher estimates of genetic advance (> 30%) were recorded for seed weight per plant (41.41%) followed by pod weight per plant (36.50%). The lower value for genetic advance as a percentage of the mean was recorded for 50-seed weight (19.11%). Higher values of heritability were coupled with high genetic advance for the number of pods per plant, pod weight per plant, and seed weight per plant.

In addition, high values of PCV were coupled with high estimates of GCV, indicating lower effects of environmental factors on these traits. The increase in GCV for these traits provides a great possibility for improving them through a selection program. The results obtained herein agreed with Amri-Tiliouine et al. (2018), who obtained higher values in genetic parameters for the number of seeds yielded per plant and yield of chickpea, as well as, total yield which was highly positively correlated with the number of pods and seeds yield per plant. However, the relative difference between the PCV

Table 2. Genetic parameters of yield components in M_1 generation of chickpea treated with gamma rays.

Genetic parameter	Weight of 50 seeds	Pods weight per plant	Pods number per plant	Seed weight per plant
Mean±Sd	11.56± 1.37	94.88± 16.97	255.11±31.70	66.79±7.75
DMS	4.185	917.28	3250.29	600.76
EM	0.195	17.96	39.94	15.89
GV	01.33	299.70	1070.11	194.96
PV	01.52	317.33	1110.05	210.85
EV	00.19	017.96	0039.94	015.89
H%	87.21	094.34	0096.40	092.46
GCV%	12.82	067.18	0077.42	064.57
PCV%	13.72	069.16	0078.84	067.15
ECV%	00.90	001.98	0001.42	002.58
EGA	02.21	034.63	0066.15	027.65
GAM%	19.11	036.50	0025.93	041.41

Sd, Standard deviation; DMS, Doses of gamma rays mean squares; EM, Error mean squares; GV, genotypic variance; PV, phenotypic variance; EV, environmental variance; H (%), heritability in a broad sense in percent; GCV (%), genotypic coefficient of percent variation; PCV (%); phenotypic coefficient of percent variation; ECV (%), environment coefficient of percent variation; EGA, expected genetic advance; GAM, genetic advance as percent of mean at 5% selection intensity.

and GCV was small for the weight of 50 seeds, pod weight per plant, pod number per plant, and seed weight per plant. This suggested that improving these traits through selection has a high chance. This indicated the greater influence of genetic factors on the gene expression of these traits. The results obtained in this study agreed with Yeremko et al. (2024), who increased the productivity of seed chickpeas through the application of mineral and organic mineral fertilizers in addition to microbial preparations for nitrogen fixation.

The results are also in line with Nechifor et al. (2011), who found a large difference between PCV and GCV for seed yielding in common beans attributed to environmental conditions. Based on the classification of Johnson et al. (1955), genetic advance as a percent of the mean can be categorized as low (0-10%), moderate (10-20%), or high (above 20%). In this study, all estimates of genetic advance for seed weight per plant, the weight of pods per plant, and pod number per plant exceed 20%, classified as high genetic advance. Moderate genetic advance was recorded for the weight of 50 seeds (19.11%). This is in harmony with Alemayehu (2010), who found moderate values of heritability and genetic advance as a percentage of the mean for seeds number per pod in common bean. The estimated GCV and PCV exceeded 60% for most of the yield components classified as high values if compared with the low value obtained in the weight of 50 seeds. Therefore, selection based on the phenotypic performance of variant genotypes leads to an increase in the performance of selected offspring. This indicated the involvement of additive gene action in the inheritance of yield components traits leading to selection will be effective.

The results are in close harmony with the findings of Chand et al. (2008), who stated that high heritability values in barley were associated with high genetic gain for the number of grains/spikes, grain yield/plant, and biological yield per plant. Therefore, the information released from this study required to be highlighted by chickpea breeders who are interested in generating high-yielding genotypes. Besides, the mutant genotypes need to be tested for their genetic stability in different agro-ecologies. Genetic variability induced in this study by the different doses of gamma irradiation was defined as the occurrence of diversity among individuals in the population due to variations in their genetic composition and the environment in which they are raised (Falconer and Mackay, 1996). A reverse genetic technique named Targeting Induced Local Lesions In Genomes (TILLING) can also help to increase genetic variability because it is an efficient tool for generating point mutations in genes of interest leading to increased genetic variations in the population, which is usually applied in combination with physical or chemical mutagenic agents to induce spectra magnitude of mutations at high density to overcome the recovery of single nucleotide mutants.

Therefore, measuring genetic parameters is useful in designing effective programs in chickpea breeding techniques (Kozgar, 2014). The GCV measures the level of genetic variability shown in the traits; it alone cannot measure the amount of heritable variations (Wani, 2011). Information about heritability was important for selection-dependency improvement because it indicates the extent of genetic factors' transmissibility of the trait into the next generations (Kozgar, 2014). Assessment of heritability alone does not bring out information about the expected genetic advance in the future generation but must be in conjunction with the measuring of genetic advance as the change in mean performance between generations

Table 3. Effect of gamma irradiation on the germination and viability ratio of M_1 seeds.

Doses of gamma rays (rad)	Germination	Viability
00	0.88	1.00
25	1.00	1.13
50	0.94	1.06
100	0.91	1.02
200	1.00	1.13
400	0.98	1.11
800	1.00	1.13
F-test	NS	NS
LSD		
0.05	0.13	0.129
0.01	0.18	0.314

NS = Non significant differences.

(Wani and Khan, 2006). Notably, all yield components were more affected by genetic factors indicating the minimum effect of environmental factors on the gene expression of these traits leading selection to be effective in their improvement. This reflected the involvement of additive gene action in the inheritance of yield components. Meanwhile, the traits possessing low genetic advances as the weight of 50 seeds (19.11%) with high heritability (87.21%) indicating the presence of non-additive gene action, leading selection technique will not be effective in the improvement of the desirable trait, because slow progress will be obtained through selection of these traits. The results reflected that the different doses of gamma irradiation had different effects on quantitative and vegetative traits. This will provide important information on gene expression, as well as provide genetic biomarkers to facilitate breeding techniques. Therefore, selection based on the phenotypic performance of mutant genotypes leads to an increase in the performance of selected progenies.

Viability of seeds

The results in Table 3 demonstrate that the germination ratio ranged between 0.88 in the control and 1.0 at some doses of gamma irradiation. The highest ratio of germination (1.0) was recorded in M_1 seeds released from the doses of 25, 200, and 800 rad followed by 400 rad (0.98), 50 rad (0.94), and 100 rad (0.91). The lowest germination ratio was recorded in unirradiated seeds (0.88). The differences between doses of gamma rays affecting germination and viability ratio were insignificant.

The viability of M_1 seeds ranged between 1.0 (00 rad) and 1.13 (at doses of 25, 200, and 800 rad). The highest value in seeds viability (1.13) was recorded by the doses of 25, 200, and 300 rad followed by 400 rad (1.11), 50 rad (1.06), and 100 rad (1.02). The decrease in germination ratio, as well as in seed viability will cause a great loss in seed yielding per unit of cultivated area. These results agreed with El-Batal et al. (2012), who found an increase in the viability of *Gliocladium virens* and *Gliocladium deliquescens,* as a chickpea root rot caused fungi, by increasing the dose of gamma irradiation. The increase in germination rate and viability percentage at some doses of irradiation may be due to enhanced enzyme activity induced by gamma irradiation which is related to activating the mitotic cell cycle. Similar results were also obtained by Melki et al. (2011), who found that the dose of 20 Gy-gamma rays enhanced plant growth by 146.35% compared with the plants released from unirradiated seeds. In contrast, Shah et al. (2008) found that germination percentage in chickpeas irradiated with gamma rays decreased gradually with increasing the doses of gamma rays from 400 to 1200 Gy.

Meanwhile, Brahmi et al. (2014) stated that the dose of 150 Gy caused a 50% reduction in seed survival of the local chickpea variety, but high doses of more than 250 Gy caused a slow decline in germination percent. The low doses of gamma irradiation may affect plant cells to increase the anti-oxidative activity of the cells via induced reactive oxygen species (ROS) which mediate cell cycle acceleration entry to G_0/G_1 inducing a positive effect on plant cell cycle machinery (Sharma et al., 2012). Meanwhile, the high doses of gamma rays may induce cell cycle arrest at the G_2/M phase during mitosis and/or damage induced in the genome (Preuss and Britt, 2003). The retarded germination of unirradiated seeds was stimulated by different doses of gamma irradiation. Retarded germination may be associated with a low rate of cell division at the early emergency of seminal root and shoot. This agreed with Girija et al. (2013), who stated that low doses of gamma irradiation increased the

Table 4. Coefficient of variation for seeds viability and their germination rate affected by gamma irradiation.

Doses of gamma irradiation (rad)	Germination rate	Viability of seeds
00	0.11	0.000
25	0.00	0.000
50	0.11	0.110
100	0.17	0.120
200	0.00	0.000
400	0.03	0.037
800	0.00	0.000

proportion of divided cells, meanwhile, higher doses induced a reduction in the mitotic index in cowpeas following irradiation with the doses ranging from 10 to 300 Gy. Similar findings were also demonstrated in soybean and cowpea by De Veylder et al. (2003), who found in plant root tip cells that arrest in cell cycle progression induced by checkpoints that mediate entry of the cells into S-phase and mitosis. Thus, stimulation shown in this study for germination and viability of M_1 seeds may be due to broking the arrest in cell cycle progression leading the cells to enter into S-phase and mitosis to increase cell division at the early emergency of seminal root and shoot. The mitotic cells often spontaneously continue in cell cycle progression, which may be followed by genome instability leading to cell survival including chromosomal abnormalities (Harting and Beck, 2006). Therefore, Abdoun et al. (2022) found that the higher doses of gamma rays ranging from 300 to 600 Gy induced degradation in nuclear cell membranes in most root meristematic cells of all chickpea genotypes. Similar results were also obtained by Wani (2009) in chickpeas treated with ethyl methane sulphonate and gamma irradiation, as well as their combination treatments.

Ionizing radiation generated reactive oxygen species. Plant enzymes are involved in cell protection against oxidative stress (Zaka et al., 2002). Antioxidants and peroxidase are involved in the mechanisms of free radicals' inhibition upon seed irradiation. Activation of these enzymes in irradiated seeds may lead to an increase in germination rate because these enzymes are involved in the protection of cells against oxidative stress (Rogozhin et al., 2000). The results obtained herein are in harmony with Al-Enezi et al. (2012), who found that root growth of palm (*Phoenix dactylifera* L.) was stimulated in response to the doses increased of gamma rays. In addition, Bonde et al. (2020) found that seed germination percentage increased progressively with increasing doses of gamma irradiation at specific doses and then decreased thereafter. Meanwhile, the reduction in the percentage of seed germination may be due to injury in the seed's tissue, chromosomal abnormalities, the severity of chromosome damage, and subsequent mitotic retardation (Datta, 2009).

Homogeneity

The degree of homogeneity was assessed based on the coefficient of variability (Table 4), which was used to determine the magnitude of variation in germination rate due to gamma-irradiated chickpeas. Concerning germination rate the coefficient of variance values ranged between 00 at 25 rad and 0.17 at 100 rad, compared with the check value (0.11). All the doses of gamma rays, except for 100 rad showed coefficient of variance values lower or close to the chick value. This indicated that the doses of 25, 50, 200, 400, and 800 rad showed a lower degree in coefficient of variance ranging between 0.0 and 0.11 which is lower or close to the check value (0.11), reflecting homogeneity in germination rate. Meanwhile, the dose of 100 rad recorded the highest value in coefficient of variance (0.17) higher than the check (0.11), indicating high heterogeneity towards decreasing germination rate, shown before in Table 3.

The estimated coefficient of variance for seed viability ranged between 0.0 and 0.12 compared with the check value (0.0). The doses of 25, 200, and 800 rad recorded a 0.0 coefficient of variance as that in check value (0.0), indicating high homogeneity in seeds viability, because they recorded their values close to the check. Meanwhile, the doses of 50, 100, and 400 rad recorded high values in coefficient of variation ranging between 0.037 (400 rad) and 0.120 (100 rad) higher than the check (0.00), indicating high heterogeneity in seed viability. Generally, the degree of homogeneity for germination and viability states differed between the doses of gamma irradiation. The lowest homogeneity was obtained if the coefficient of variance recorded the highest values exceeding the check genotype. However, high homogeneity was shown if the coefficient of variance recorded lower or close values to the check genotype. High homogeneity obtained in seed viability by the doses of 25, 200, and 800 rad indicated that the seeds exhibited high uniformity in their viability. These results agreed with El-Eslamboly et al. (2020), who found that the coefficient of variance

Table 5. Genetic parameters of seeds germination and their viability in M_1 generation released from gamma irradiation.

Genetic parameter	Seeds germination	Viability of seeds
Mean± Sd	14.61± 5.00	0.79±0.24
DMS	4.185	0.195
EM	917.28	17.96
GV	00.00086	00.001
PV	00.00580	00.006
EV	00.00500	00.005
GCV%	01.13000	00.770
PCV%	02.94000	01.890
ECV%	01.81000	01.110
H%	14.82000	16.660
EGA	00.02000	00.026
GAM%	02.40000	02.400

Sd, Standard deviation; DMS, doses of gamma rays mean squares; EM, error mean squares; GV, genotypic variance; PV, phenotypic variance; EV, environmental variance; GCV (%), genotypic coefficient of percent variation; PCV (%); phenotypic coefficient of percent variation; ECV (%), environment coefficient of variation in percent; H (%), heritability in a broad sense in percent; EGA, expected genetic advance; GAM, genetic advance as percent of mean at 5% selection intensity.

values for plant length, number of female flowers, and average fruit weight in summer squash were highly homogeneous. In addition, El-Morsy et al. (2021) applied selection on sixteen new lines of tomatoes based upon exhibited high homogeneity for plant height, shape index, vitamin C, number of leaves/plants, and total soluble sold (TSS). The genotypes exhibited the lowest values of variance coefficient than the check cultivar, indicating that their plants were more phenotypically uniform than other plant genotypes.

Phenotypic and genotypic variations

Estimates of genetic parameters (Table 5) showed that the GCV for the germination and viability of seeds were lower than the environmental coefficient of variation. On the other hand, environmental variance was a larger magnitude of difference than genotypic variance. Meanwhile, in all variance analyses, the PCV was larger than the GCV. This indicates higher effects of environmental factors than genetic elements in the expression of seed germination and viability. Therefore, selection depending on the phenotypic performance may be not appropriate for these traits. The PCV and GCV for germination and viability of seeds were regarded as low values because they were lower than 10% based on Deshmukh et al. (1986). Similarly, relatively lower genetic variances share of the total phenotypic variance were observed for seed germination and their viability, indicating a greater share of environmental variance in the total variations, leading to little opportunity for improving these traits through selection, because of non-additive gene effects.

The results obtained herein agreed with Ejara et al. (2018), who found a relatively large difference between the GCV and PCV for grain yield/hectare, reflecting a greater influence of environmental factors on this trait. Estimates of heritability in a broad sense ranged from 14.82% for seed germination to 16.66% for the viability of seeds. These heritability values are categorized as low based on Robinson et al. (1949). Heritability in a broad sense describes the reliability of phenotypic variance as a breeding guide that includes additive and non-additive gene action. Therefore, seed germination and viability with low heritability values cannot be improved through selection than the traits with high heritability values, because these traits were more affected by environmental factors than genetic background. The germination of seeds recorded genetic advance as a percent of mean equal to 2.4%, classified as low genetic advance based upon the characterization of Johnson et al. (1955). In this study, low heritability was coupled with low genotypic variance. Low GCV and low genetic advance suggest that selection based upon phenotypic expression of genotypes for germination rate and viability of seeds will not improve the performance of offspring in the next generations. The low heritability obtained herein reflected the influence of environmental factors that limit improvement by selection, because of non-additive gene effects on these traits. These findings were detected earlier in peanuts by Kavera and Nadaf (2017). In addition, Hailu et al. (2016) found that plant height and biological yield in barley showed a greater share of environmental variance in the total phenotypic variability.

This is because there is a close correspondence between the environmental factors and the phenotype due to the small relative contribution of the genotype on the phenotype performance (Singh, 2001). The results obtained in this study agreed with Hailu et al. (2016), who found low heritability was coupled with low genetic gain for plant height in barely. Further study by Sardana et al. (2007) stated that high heritability may not necessarily be coupled with increased genetic gain unless sufficient genetic variations existed in the population.

In conclusion, yield components exhibited relatively high values of heritability which possibly is linked to its genetic advance as a percentage of the mean for each trait. Mutation progenies can be used as patents for mutational breeding techniques in chickpeas. The differences between the PCV and GCV were relatively few in magnitude of yield components, indicating the higher influence of genetic factors than environmental conditions in the gene expression of these traits. This indicated that the involvement of additive gene action on gene expression of these traits leading to selection will be effective.

CONFLICT OF INTERESTS

The author has not declared any conflict of interests.

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