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Studies on Effect of Mutagens on Quantitative Characters in M_2 and M_3 Generation of Horsegram [*Macrotyloma uniflorum* (Lam.) Verdc]

K. Geetha[1], S. Divya[1], S. Srividya[2]

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ABSTRACT

Background: Induction of mutation plays an important role in the breeding programme among modern methods of plant breeding. Genetic variability is prerequisite for crop improvement and through induced mutation it was found to be very effective for creating variability in the quantitative and qualitative characters. Hence, the present study was aimed to induce genetic variability within short time. In the present investigation positive as well as negative impact on quantitative traits were recorded.

Methods: The experiment material comprised of seeds of horsegram [*Macrotyloma uniflorum* (Lam.) Verdc] variety Paiyur 2 which was subjected to ethyl methane sulphonate (02, 0.3, 0.4 and 0.5%), gamma radiation (100, 200, 300 and 400 Gy) and combination treatments. The mutations affecting gross morphological changes in growth and yield characters such as plant habit, flowering, pod morphology, maturity and seed yield were scored as quantitative characters. The micro mutations at population level can be easily detected in the form of increased variations for quantitative traits in the segregation of mutagen treated populations. Micro mutations can alter morpho-physiological characters hence they are of a particular interest to the plant breeders. ANOVA test was performed to determine the significant differences and Duncan's multiple range test (p=0.05) to compare the differences among treatment means.

Result: Among the twenty-five treatments studied both the mutagens, gamma radiations and Ethyl Methane Sulphonate proved to be very effective to induce variability in quantitative traits in M_2 and M_3 generations. The results of Duncan's multiple range test analysis revealed that the treatment of mutagens in combinations induced more positive effects as compared to that of using alone. The combined treatments were found positive for improvement of primary branches per plant, no. of pods per plant, pod length, no. of seeds/pod and seed yield per plant in M_2 and M_3 generations whereas the treatment of gamma irradiation imparted its significant role for improvement of plant height, days required for 50% flowering, days to maturity and 1000 seed weight in both M_2 and M_3 generations whereas the treatment with Ethyl Methane Sulphonate is useful for improving no. of pods per plant and no. of seeds/pod in M_2 generation alone.

Key words: M_2 and M_3 generations, Micro mutations, Mutagens, Quantitative traits, Variability.

INTRODUCTION

Horsegram is a hardy drought resistant, annual, traditional and tropical grain legume. It derives its name from the fact that it is important as annual feed and as such fed to horses. It is the cheapest of pulses and is hence the poor man's pulse. The bhusa of the crop after removal of pods is used as cattle feed. In Tamil Nadu the area under horsegram is about 18 per cent of the total area of 2.2 million hectares in India. The productivity of the crop is rather low with an average grain yield of 494 kg/ha as against 539 kg/ha of all the grain legumes as a whole. It ranks as the third among Indian pulses in area. The Southern States are the major horsegram growing States in the country.

Horsegram is cultivated in areas with annual rainfall of 300-600 mm and highly drought tolerant, but does not tolerate floodingor water logging. The favourable average temperature is 18-27°C. Horsegramis adapted to a wide range of well drained soils from sands and gravels to clay loams and heavy clays with neutral soils. It grows on soils having pH 5.5 to 8.

[1]Regional Research Station, Paiyur, Krishnagiri-635 112, Tamil Nadu, India.
[2]Krishi Vigyan Kendra, Papparapatty, Dharmapuri-636 809, Tamil Nadu, India.

Corresponding Author: K. Geetha, Regional Research Station, Paiyur, Krishnagiri-635 112, Tamil Nadu, India.
Email: geethakreddy@yahoo.com

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Genetic variability is the basic need in any of the crop to be improved. Mutation breeding is identified as the efficient method to enhance the genetic variability whenever the variation is not available in the natural resources. Knowing the limited variability in horsegram, present investigation has been carried out for the improvement in this crop by utilizing the variety Paiyur 2.

MATERIALS AND METHODS

The authentic seeds of horsegram [*Macrotyloma uniflorum* (Lam.) Verdc] variety Paiyur 2 released from Regional Research Station, Paiyur were used. Ethyl Methane Sulphonate (EMS), Gamma rays and their combinations were employed in present study for the treatment of seeds of horsegram. EMS was used for the seed treatment of horsegram. Various concentrations of EMS (0.2% to 0.5%) were prepared in 0.1 m phosphate buffer pH -7.0. Selected seeds were soaked in distilled water for 10 hours and the wet seeds were treated with different concentrations of EMS (such as 0.2, 0.3, 0.4 and 0.5% v/v) for four hours. Healthy, dry and uniform seeds of horsegram with moisture content of 10-12% were treated with 100, 200, 300 and 400 Gy using gamma irradiation chamber at Bhabha Atomic Research Centre, Trombay. For combination treatments the gamma irradiated seeds were treated with different concentrations of EMS. The untreated seeds served as control. The seeds treated with various concentrations of EMS were washed thoroughly with tap water for 2 hours to terminate the reaction of chemical mutagen to leach out the residual chemicals. The treated seeds (675) from each treatment were used for raising M_1 generation in field. Present investigation was carried out at Regional Research Station, Paiyur. All the experiments were carried out in triplicate following RBD design. The distance between two rows and two plants was 30 × 15 cm and the distance between two adjacent plots was one meter. The seeds of individually harvested M_1 plants were sown in the experimental field to raise M_2 generation in separate rows during *Rabi* season of the year 2018. The treated as well as control plants were screened for quantitative traits to study the induced variability. From each replication and treatment including control 20 plants were randomly selected for recording data on different quantitative characters in M_2 generations. Data on nine quantitative traits such as plant height (cm), primary branches per plant, days required for 50% flowering, days for maturity, no. of pods per plant, pod length (cm), no. of seeds/pod, 1000 seed weight (g) and seed yield per plant (g) were recorded. All the surviving M_2 plants were harvested individually and seeds of single plant from each treatment were kept separately for raising the M_3 generation. Observations on quantitative characters in M_3 generation were similar to that of M_2 generation.

ANOVA test was performed to determine the significant differences due to various treatments while DMRT test (p=0.05) was employed to compare the differences among treatment means. The DMRT analysis was performed using SPSS Software.

RESULTS AND DISCUSSION

Analysis of variance

M_2 and M_3 populations imparted significant differences for most of the traits studied.

Quantitative characters (Micro mutations) in M_2 and M_3 generations

EMS and Gamma radiations proved to be very effective to induce variability in quantitative traits in M_2 and M_3 generations (Table 1 and Table 2). Positive as well as negative impact on quantitative traits was well documented by Barshile *et al.* (2008) in chickpea, Bolbhat and Dhumal (2009), Dhumal and Bolbhat (2012) and Kanaka (2012) in horsegram.

Plant height

All the mutagens were effective for inducing variability in plant height in M_2 generation (Table 1). The DMRT represented the similar group (Group h) for minimum plant height in 200 Gy+0.3% EMS (71.47 cm), 200 Gy+0.5% EMS (70.93 cm) and 300 Gy + 0.2% EMS (60.97 cm) as compared to control (92.13 cm). Gamma treatment of 100 Gy 98.43 cm) falls under Group a showed significant increase in plant height while other treatments showed -ve influence.

Similar trend in plant height was observed in M_3 generation (Table 2). Maximum plant height 93.53 cm in M_3 was noted in 100 Gy gamma rays (Group a). The minimum plant height of 72.23 cmwas noted in 300 Gy+0.4% EMS (Group f) through DMRT. The average height of control plants was 90.33 cm. All the doses/conc.ofgamma irradiation, EMS and their combinations caused reduction in plant height with few exceptions. Similar results werereported by Dalvi (1990). Gamma irradiation exhibited a stimulatory effect of significant increase in plantheight. Findings of Yaqoob and Abdur (2001) in mungbean and Barshile *et al.* (2008) in chickpea were in agreement with above results.

Number of primary branches per plant

Data obtained in M_2 generation on number of primary branches per plant (Table 1) indicated that the mean values of this parameter showed positive and negative influence. 100 Gy and 0.4 % EMS (Group a) showed increased number of primary branches per plant. Maximum number of primary branches per plant was recorded in 100 Gy (9.23) and 0.4% EMS (8.87) over control (8.80). In combination treatments there was no definite pattern. The maximum decrease (6.63) was noted in 200 Gy + 0.5% EMS (Group j) compared to control (8.80). The highest increase (9.63) was noted in 100 Gy+0.5% EMS. In nine treatments the primary branches per plant were increased over control. Similar trend was obtained in M_3 generation (Table 2). Six treatments showed increase in number of primary branches per plant as compared to control and M_2. DMRT revealed that maximum number of primary branches was recorded in 100 Gy+0.3% EMS (9.40) over control (8.93) and categorized as Group a. The treatment 200 Gy+0.5% EMS (Group j) recorded least number of primary branches as 7.10. Almost all the treatments of gamma irradiation and EMS showed positive as well as negative impact on primary branches per plant in horsegram. Dalvi (1990) and Singh *et al.* (2000) also noted similar trend with physical as well as chemical mutagen.

Table 1: Duncan multiple range test (DMRT) of quantitative traits in M_2 generation.

Treatment	Plant height (cm)	Primary branches/plant	Days to 50% flowering	Days to first maturity	No.of pods/plant	Pod length (cm)	No.of seeds/pod	1000 seeds wt.(g)	Seed yield/plant
Control -Paiyur 2	92.13 abc	8.8 abcdef	72.77 a	103.27 a	57.00 abc	5.17 abc	5.9 a	30.70 bcdef	30.67 abcde
0.2% EMS	90.87 abc	7.7 efghij	71.50 abc	102.00 abc	57.37 a	5.23 abc	5.023 bcd	33.00 abc	29.77 abcdef
0.3% EMS	91.90 abc	7.57 ghij	73.00 a	103.50 a	51.30 def	4.60 bc	5.93 a	25 g	31.03 abcde
0.4% EMS	76.47 fgh	8.87 abcde	73.30 a	103.80 a	52.10 cdef	5.13 abc	4.67 cdefgh	28 cdefg	28.23 bcdef
0.5% EMS	82.20 cdefgh	6.77 ij	73.10 a	103.60 a	49.40 f	5.2 abc	5.07 bcdef	30 bcdefg	29.17 abcdefg
100 Gy	98.43 a	9.23 abc	72.50 ab	103.00 ab	57.93 a	5.13 abc	4.47 fgh	36.33 a	25.70 efg
200 Gy	87.10 bcde	7.09 hij	66.87 c	97.37 c	57.90 a	5.17 abc	4.73 cdefgh	28.67 cdefg	28.90 abcdefg
300 Gy	76.47 fgh	7.63 fghij	72.33 ab	102.83 ab	52.40 cdef	4.53 c	4.60 defgh	28.67 cdefg	32.67 ab
400 Gy	71.80 h	7.6 fghij	73.77 a	104.27 a	54.8 abcde	5.27 abc	4.63 defgh	32.33 abcd	25.23 fg
100 Gy+0.2% EMS	94.07 ab	9.3 abc	73.57 a	104.07 a	54.63 abcde	4.87 abc	5.9 a	25.67 fg	27.97 bcdefg
100 Gy+0.3% EMS	75.57 fgh	7.31 ghij	73.27 a	103.77 a	59.23 a	4.97 abc	4.67 cdefgh	26.67 efg	23.83 g
100 Gy+0.4% EMS	88.57 abcd	8.2 cdefgh	72.90 a	103.40 a	52.1 cdef	4.53 c	4.91 bcdefg	28 cdefg	27.27 cdefg
100 Gy+0.5% EMS	90.57 abc	9.63 a	73.60 a	104.10 a	51.60 def	5.10 abc	4.90 bcdefg	29.13 bcdefg	28.17 bcdefg
200 Gy+0.2% EMS	94.37 ab	9.53 ab	73.53 a	104.30 a	58.66 a	5.20 abc	4.73 cdefgh	25.30 fg	31.27 abcd
200 Gy+0.3% EMS	71.47 h	7.77 efghij	71.60 a	102.10 abc	52.20 cdef	4.73 bc	5.20 bcde	28.63 cdefg	32.13 abc
200 Gy+0.4% EMS	88.10 bcd	7.03 hij	74.23 a	104.73 a	48.40 f	4.57 bc	4.43 fgh	34.33 ab	28.97 abcdefg
200 Gy+0.5% EMS	70.93 h	6.63 j	73 a	103.50 a	49.00 f	4.90 abc	4.53 fgh	26.83 defg	27.30 cdefg
300 Gy+0.2% EMS	68.97 h	7.63 fghij	67.63 bc	98.13 bc	50.03 ef	5.63 a	4.40 gh	28.20 cdefg	33.63 a
300 Gy+0.3% EMS	75.03 fgh	7.87 defghi	70.33 abc	100.83 abc	59.13 a	4.73 bc	5.97 a	32 abcde	26.53 defg
300 Gy+0.4% EMS	72.23 gh	9.03 abcd	75.17 a	105.67 a	51.70 def	4.77 abc	4.20 h	26.63 efg	28.83 abcdefg
300 Gy+0.5% EMS	79.13 defgh	7.9 defghi	73.43 a	103.93 a	55.80 abcd	5.00 abc	5.30 bc	29.23 bcdefg	26.83 cdefg
400 Gy+0.2% EMS	77.13 fgh	7.67 efghij	71.11 abc	101.60 abc	58.97 a	5.45 ab	6 a	27 bcdef	26.17 defg
400 Gy+0.3% EMS	78.28 efgh	8.33 cdefg	73.33a	103.83 a	55.20 abcd	4.50 c	5.4 ab	27 defg	24.7 fg
400 Gy+0.4% EMS	83.43 cdef	7.53 ghij	73.53 a	104.03 a	52.63 bcdef	5.10 abc	4.8 bcdefgh	29.33 bcdefg	26.83 cdefg
400 Gy+0.5% EMS	76.67 hgf	8.4 bcdefg	74.37 a	104.87 a	48.50 f	4.85 abc	4.57 efgh	28.70 cdefg	27.07 cdefg

Table 2: Duncan multiple range test (DMRT) of quantitative traits in M_3 generation.

Treatment	Plant height (cm)	Primary branches/plant	Days to 50% flowering	Days to first maturity	No.of pods/plant	Pod length (cm)	No.of seeds/pod	1000 seeds wt. (g)	Seed yield/plant
Control -Paiyur 2	90.33 abc	8.93 abcde	72.4 ab	103.40 abc	58.63 ab	5.60 abcd	5.70 abc	32.33 abcde	32.37 abcd
0.2% EMS	91.07 ab	9.07 abc	73.6 ab	104.60 abc	56.37 abcdef	5.20 abcde	5.13 abcdef	33.07 abcd	33.20 abcd
0.3% EMS	89.23 abcde	8.23 bcdefghi	71.17 ab	102.17 abc	58.77 ab	5.67 abc	5.73 abc	33.80 abc	28.23 abcde
0.4% EMS	78.67 def	8.60 abcdef	73.30 ab	104.30 abc	58.53 abc	5.13 bcdef	4.57 f	28.23 defg	27.73 abcde
0.5% EMS	82.20 bcdef	8.03 cdefghij	72.70 ab	103.70 abc	50.43 ghi	5 def	4.93 def	30.03 abcdefg	25.87 e
100 Gy	93.53 a	9.00 abcd	72.7 ab	106.27 ab	59.60 ab	4.53 f	4.80 ef	34.53 a	30.43 abcd
200 Gy	86.23 abcde	9.10 abc	69.23 b	100.23 bc	57.90 ab	5.27 abcde	5.57 abcd	27.33 efg	32.40 abcd
300 Gy	81.30 bcdef	7.97 defghij	68.03 b	99.03 c	50.40 ghi	4.90 ef	4.80 ef	28.90 cdefg	30.73 abcde
400 Gy	77.87 ef	8.07 cdefghij	74.6 ab	105.60 abc	53.83 cdefgh	5.30 abcde	4.97 def	30.10 abcdefg	26.87 de
100 Gy+0.2% EMS	82.43 bcdef	8.33 abcdefgh	71.90 ab	102.90 abc	58.90 ab	5.60 abcd	5.73 abc	32.60 abcde	31.27 abcde
100 Gy+0.3% EMS	91.03 ab	9.40 a	73.33 ab	104.33 abc	57.17 abcd	5.10 cdef	5.10 bcdef	27.73efg	33.47 ab
100 Gy+0.4% EMS	81.97 bcdef	8.17 bcdefghij	73.77 ab	104.77 abc	52.43 defghi	4.97 def	4.97 def	27.80 defg	30.50 abcde
100 Gy+0.5% EMS	79.67 cdef	9.17 ab	72.90 ab	103.90 abc	52.43 defghi	4.83 ef	5.23 abcdef	31.40 abcdefg	30.07 abcde
200 Gy+0.2% EMS	81.47 bcdef	8.47 abcdefg	73.77 ab	104.67 abc	58.90 ab	5.80 a	4.90 def	30.63 abcdefg	29.23 abcde
200 Gy+0.3% EMS	90.77 abc	\7.67 fghij	70.87 ab	101.87 abc	52.33 efghi	4.83 ef	5.77 ab	32.63 abcde	33.40 abc
200 Gy+0.4% EMS	79.97 bcdef	7.63 fghij	71.20 ab	102.20 abc	48.57 i	5.00 def	5.00 def	34.33 ab	27.50 bcde
200 Gy+0.5% EMS	77 ef	7.10 j	74.97 ab	105.97 abc	49.23 hi	5.13 bcdef	4.97 def	26.83 fg	28.60 abcde
300 Gy+0.2% EMS	84.87 abcde	8.97 abcde	69.03 b	100.03 bc	48.43 i	5.67 abc	5.80 a	28.20 defg	33.90 a
300 Gy+0.3% EMS	81.23 bcdef	7.33 hij	70.13 b	101.13 bc	59 ab	5.00 def	5.50 abcd	32 abcdefg	28.80 abcde
300 Gy+0.4% EMS	72.23 f	7.43 ghij	74.10 ab	105.10 abc	59.67 a	5.20 abcde	5.07 cdef	26.63 f	30.83 abcde
300 Gy+0.5% EMS	80.63 bcdef	7.50 ghij	73.27 ab	104.27 abc	57.07 abcde	5.73 ab	4.93 def	29.23 bcdefg	29.13 abcde
400 Gy+0.2% EMS	78 def	7.60 fghij	77.70 a	108.70 a	55.73 abcdef	5.07 cdef	5.27 abcde	34.93 a	33.60 ab
400 Gy+0.3% EMS	77.20 ef	8.17 bcdefghij	73.10 ab	104.10 abc	52.07 fghi	5.13 bcdef	5.73 abc	28.93 cdefg	29.70 abcde
400 Gy+0.4% EMS	84.23 abcde	7.20 ij	73.53 ab	104.53 abc	56.77 abcdef	4.77 ef	5.20 abcdef	28.83 cdefg	27.07 cde
400 Gy+0.5% EMS	83.20 abcdef	7.90 efghij	73.80 ab	104.80 abc	54.73 bcdefg	5.33 abcde	5.23abcdef	27.67 efg	28.50 abcde

Number of days required for 50% flowering

The results of the gamma radiation in M_2 generation in different treatments indicated that there was significant change in the number of days required for 50% flowering while in EMS and combination treatments there was no definite pattern. In all the gamma treatments, days to 50% flowering were less than control except 400 Gy. The minimum numbers of days required for 50% flowering were 66.87 DAS in 200 Gy (Group c) as compared to control (72.77 DAS). In the combination treatments, days to 50% flowering were less than control in 300 Gy with different concentrations of EMS *viz.,* 0.2 and 0.3% and 400 Gy + 0.2% EMS. Similarpattern was noted for M_3 generation (Table 2). The minimum days required for 50% flowering were 68.03 DAS in 300 Gy (Group b), 69.23 DAS in 200 Gy (Group b), 69.03 in 300 Gy+0.2% EMS (Group b), 70.13 in 300 Gy+0.3% EMS (Group b). The number of days required for 50% flowering was not muchas compared to control except few treatments. However, gamma radiation and combination with EMS treatments caused early flowering. Dalvi (1990) also noted similar results in horsegram with different mutagens. The results recorded by Gaikawad *et al.* (2005); Rudraswami *et al.* (2006); Manjaya and Nandavar (2007) and Ahire (2008) in different legumes were supportive to the present findings.

Number of days required for pod maturity

The data recorded in Table 1 revealed that GR had succeeded in reducing the number of days required for pod maturity as compared to control. The results of gamma irradiation were highly significant for reducing the number of days required for pod maturity. The minimum number of days (97.37 DAS) required for pod maturity was noted in 200 Gy (Group c). The data obtained for M_3 generation was on par with M_2 generation (Table 2) where the minimum days required for pod maturity were 99.03 DAS in 300 Gy. Gamma radiation (200 Gy) was successful to induce earlier pod maturity by about 3-4 days as compared to control. Singh *et al.* (2000) reported contradictory findings with reference to this parameter.

Number of pods per plant

Gamma radiation and EMS single and combination had induced variability in number of pods per plant in M_2 generation. The data recorded in Table 1 revealed that the treatments had stimulatory as well as inhibitory effect. In M_2 generation maximum number of pods per plant (59.23) was noted in 100 Gy+0.2% EMS (Group a), 59.13 in 300 Gy +0.3% EMS (Group a), 58.97 in 400 Gy+0.2% EMS (Group a), 58.66 in 200 Gy+0.2% EMS (Group a), 57.93 in 100 Gy (Group a) and 57.90 in 100 Gy (Group a) than control (57.00). The minimum number of pods per plant (48.40) were recorded at 200 Gy+0.4% EMS (Group f), (48.50) at 400 Gy+0.5% EMS (Group f), (49.40) at 0.5% EMS (Group f) and (49) 200 Gy+0.5% EMS (Group f) as compared to control. However, all the combination treatments have caused reduction in number of pods per plant except 100 Gy + 0.2% EMS, 200 Gy + 0.2% EMS, 300 Gy + 0.3% EMS and 400 Gy + 0.2% EMS.

The trend in variation of pod number observed in M_3 generation was similar to that of M_2 generation. M_3 generation had shown slight increase in pod number as compared to M_2 generation (Table 2). Maximum pods were recorded in 300 Gy + 0.4% EMS (59.67- Group a) and minimum pods was recorded in 200 Gy+0.4% EMS (48.57- Group i) and 300 Gy + 0.2% EMS (48.43- Group i). There was increase as well as decrease in number of pods per plant with different doses/concentrations used. The results of Dalvi (1990) for horsegram were in agreement with present study. Gaikawad *et al.* (2005) in lentil noted similar results. However, decrease in pod number was also recorded by Barshile *et al.* (2008) in chickpea.

Pod length

All the mutagenic treatments showed inhibitory effect on pod length except 300 Gy+0.2% EMS (5.67 cm - Group a) than control (5.2 cm) in M_2 generation. The treatments of GR, EMS and their combinations in M_2 as well as in M_3 did not show any significant change in pod length (Table 1). The results reported by Singh *et al*. (2000) in *Vigna* and Dalvi (1990) in horsegram were in conformity with present findings. M_3 generation had shown slight increase in pod length as compared to M_2 generation (Table 2). In M_3 all the mutagenic treatments showed inhibitory effect on pod length except 200 Gy+0.2% EMS (5.80 cm- Group a) than control (5.60 cm).

Number of seeds per pod

All the treatments showed decreased number of seeds per pod except (0.3% EMS (5.93- Group a), 100 Gy+ 0.2% EMS (5.90- Group a), 300 Gy + 0.3% EMS and 400 Gy+0.2% EMS (6.00- Group a) compared to control (5.90). Data on total number of seeds per pod (Table 1) in M_2 progeny showed non-significant changes as compared to control. M_3 generation showed similar trend (Table 2). The results recorded in table indicated that all the treatments of mutagens of GR, EMS and GR+EMS exerted inhibitory effects on number of seeds per pod except 300 Gy + 0.2% EMS. Decrease in number of seeds per pod was recorded by Dalvi (1990), Dubey (1990), Gaikwad *et al.* (2005) and Barshile and Apparao (2006).

1000 seed weight

Results recorded on 1000 seed weight (Table 1) indicated that Gamma ray treatment of 100 Gy had exercised +ve effect on this parameter and showed increased 1000 seed weight (36.33 g-Group a) compared to control (30.70 g). The results of M_3 generation were on par with M_2 generation (Table 2). The results with GR, EMS and GR+ EMS showed negative as well as positive impact in horsegram. Similar observations were also made by Singh *et al.* (2000) in urdbean, Gaikawad *et al.* (2005) in lentil Sagade (2008).

Increase in 1000 seed weight was observed in 100 Gy (34.53- Group a) and 400 Gy + 0.2% EMS (34.93- Group a) compared to control (32.23 g) in M_3 generation.

Seed yield per plant

Mean values for seed yield per plant decreased in all treatments as compared to control (Table 1). All the mutagenic treatments of gamma radiation showed -ve effect except 300 Gy + 0.2% EMS (33.63 g- Group a). EMS (0.3%) (Group- abcde) caused maximum increase (31.03 g), while all other treatments showed reduction in seed yield per plant over control. The combination treatment 300 Gy+0.2% EMS had induced maximum increase (33.63 g) over control (30.67 g). But all other treatments had caused reduction as compared to control except 200 Gy+0.2% EMS (31.27 g). In M_3 generation seed yield per plant was increased in 300 Gy+0.2% EMS (33.90 g- Group a) as compared to control (32.37 g). The results are in line with the findings of Priyanka *et al.* (2019). All the mutagenic treatments except few treatments showed inhibitory effect on seed yield per plant. Patil *et al.* (2004) in soybean, Auti (2005) in mung bean, Banu *et al*. (2005) in cowpea, Barshile *et al*. (2008) in chickpea and Senapati *et al.* (2008) in black gram also recorded adverse effect on seed yield per plant due to various types of mutagenic treatments. Hakande (1992) reported wider variability in yield due to mutagenic treatments in winged bean which was attributed to pollen sterility and genetical as well as physiological alterations caused by mutagens.

Yield is an important trait, as it governs the economic benefit. Its expression is inherited by many genes, which control the production, transport and storage of assimilates. Previous studies indicated that both additive and non-additive genes contribute to yield. The variability in yield was induced by mutagenic treatments. In the present study increased seed yield was attributed to increase in number of pods per plant, length of pods and 1000 seed weight per plant.

CONCLUSION

Both the mutagens proved to be very effective to induce variability in quantitative traits like plant height, primary branches per plant, number of days required for 50% flowering, days required for first pod maturity, number of pods per plant, pod length, number of seeds per pod, 1000 seed weight and yield per plant in M_2 and M_3 generations. Combination of EMS and gamma irradiation treatments showed better positive effects as compared to that of separate treatment of EMS and Gamma irradiation. The traits *viz*., primary branches per plant, no. of pods per plant, pod length, no. of seeds/pod and seed yield per plant in M_2 and M_3 generations can be improved through combined treatments. Gamma irradiation exerted its significant role for improvement of plant height, days required for 50% flowering, days to maturity and 1000 seed weight in both M_2 and M_3 generations. Within separate treatments gamma irradiation showed more positive effects than EMS.

Conflict of interest

All authors declared that there is no conflict of interest.

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