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EMS AND GAMMA RAYS INDUCED MUTATION IN MUSTARD (*Brassica juncea*) cv. PUSA BOLD

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ABSTRACT

Pusa bold variety of Indian mustard was treated with 900, 1000, 1100, 1200 and 1300 gy doses of gamma rays. Half of the seeds of each treatments were again treated with 0.5% EMS. The treated material along with untreated control was sown in M_1 generation in the field of Agril. Botany Section, College of Agriculture, Nagpur during *rabi* 2014 and individual plants were harvested separately. Harvested seeds of individual plants from M_1 generation were sown in non replicated field trail to raise M_2 generation in *rabi* 2015. Germination percentage and mortality percentage were studied in M_1 and M_2 generation and it was found that mortality was more in M_1 generation than in M_2. Germination percentage was more in M_2 than M_1 generation. In M_2 generation days to maturity decreased in all the treatments than the control, whereas plant height, number of branches plant^{-1}, number of siliqua plant^{-1},1000 seed weight did not follow any specific trend with the increasing dose of mutagens. The mutagenic effectiveness did not follow a specific trend in any of the treatment. However, in general the treatments of gamma rays alone showed high mutagenic efficiency than the combination treatments of EMS and gamma rays in pusa bold variety of mutard.

(Key words: EMS, gamma rays, mutation, mustard)

INTRODUCTION

Indian mustard (*Brassica juncea*) called as "rai", "raya" or "laha" is one of the important oilseed crops belonging to family Cruciferae*(Syn. Brassicaceae)* and genus *Brassica.* Rapeseed-mustard is the third important oilseed crop in the world after soybean (*Glycine max*) and palm (*Elaeis guineensis* Jacq.) oil. Among the seven edible oilseeds cultivated in India, rapeseed-mustard (*Brassica* spp.) contributes 29.5% in the total production of oilseeds. In India, it is the second most important edible oilseed after groundnut sharing 27.8% in the India's oilseed economy. The concept of inducing mutation and utilising them in plant breeding was first given by Hugo de vries (1903) for generating variability and achieving the goal of generating of new strains of cultivated crop plants. Since, irradiation can to be an efficient tool in the hands of plant breeder to create a variation in a crop plant, it's use become a regular practice. India is one of the largest rapeseed mustard growing countries in the world, occupying the first position in area and second position in production after China. The world production of Rapeseed -Mustard has been increasing at a rapid rate in several countries largely in response to the continuing increase in demand for edible oils and its products. The present studies were conducted to compare the relative effectiveness of gamma rays alone and combination of gamma rays and ethyl methane sulphonate to induce genetic variability in the mustard (cv. Pusa bold).

MATERIALS AND METHODS

Dry healthy and genetically pure seeds of *Brassica juncea* cv. Pusa bold were divided into six lots of 300 seeds each for giving the gamma rays treatment separately. These 600 seeds of selected variety were subjected to irradiation with 900,1000,1100,1200 and 1300 (Co^{60} at BARC). The 300 seeds of each treatment were treated with 0.5% aqueous solution of EMS after pre-soaking with sterilized distilled water for 12 hours.

The treated seeds along with one control of dry and another of water soaked were sown in the field to raise M_1 generation in non replicated trial. M_1 generation was screened for different morphological mutants, seeds from each M_1 generation were harvested separately. M_2 generation was raised in *rabi* 2015.

Twelve treatments included different doses of gamma rays alone and combination of gamma rays and EMS along with control of mustard variety Pusa bold. 30 M_2 plants were selected at random per treatment to record the observations on Germination count in M_1 and M_2, Mortality %, Days to maturity, Plant height (cm), number of branches plant^{-1}, number of siliqua plant^{-1} and seed yield plant^{-1}(g). The efficiency and effectiveness of mutagens were estimated as suggested by Konzak *et al.* (1965). Data were analysed for mean, standard error, standard deviation and coefficient of variation.

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RESULTS AND DISCUSSION

Induced mutations in polygenes governing characters can best be inferred by the estimation of mean, range, coefficient of variation, variance and standard deviation in the successive generations of mutagen treated populations. Nature and amount of genetic variability generated in different quantitative traits after exposure to gamma rays and EMS alone and in combination.

Data regarding effect of various treatments of EMS and gamma rays on germination in M_1 and M_2 generations are presented in table 1. Reduced germination per cent in all the treatment of variety was observed in M_1 generation as compared to control. Treatments T_7 (900gy+0.5% EMS) and T_{11} (1300gy+ 0.5% EMS) recorded the highest reduction in germination per cent (57.49%) and (55.75%) respectively, while it was the lowest in treatment T_1 (900gy, 70.00%) and T_7 (900gy+0.5% EMS, 57.49%). The germination per cent in control was (82.23%) and (80.50%) respectively. Similar results for decrease in percentage of germination with high dose of gamma rays were also reported by Sangsiri *et al.* (2005).

Data regarding survival of seedling as affected due to different treatments in M_1 and M_2 are presented in table 1.The effect was recorded as mortality in percentage. It increases in M_2 generation as compared to their control in all the treatments.Voice *et al.* (2004) reported greater mortality in EMS, DES, gamma rays treated mustard.

Data regarding the effect of different treatments of EMS and gamma rays on days to maturity in M_2 generation are presented in table 2. The mean value for days to maturity decreased in all the treatments than control T_6 (control, 119.50) and T_{12} (control, 118.00). The minimum mean value for the character was observed in T_2 (1000gy, 109.40) of Pusa bold. The coefficient of variation increased in all the treatments as compared to control. Maximum mean variation was found to be in treatment T_4 (1200gy, 51.24%) while the minimum in T_{12} (control, 5.49%). The range of coefficient of variation was 5.49% to 51.24%. Barve *et al.* (2009) reported early maturity with increase in dose of EMS and gamma rays. The mutants were early to mature in 90-105 days.

The observations regarding the effect of different combination of treatments of EMS and gamma rays and gamma rays alone on plant height in M_2 generation are presented in table 3. Maximum plant height was observed in treatment T_4 (1200gy, 151.80cm), while the minimum was in treatment T_{10} (1200gy+ 0.5% EMS,102.50cm) of Pusa Bold as compared to control T_6 (control, 126.70cm) and T_{12} (control, 121.90cm).The coefficient of variation for plant height increased in all the treatments except T_2 (1000gy), T_8 (1000gy+0.5% EMS), T_{10} (1200+0.5% EMS) and T_{11} (1300+0.5% EMS). The maximum variation was in treatment T_4 (1200gy, 51.80%) and T_5 (1300gy, 51.15%) as compared to control T_6 (control, 18.37%) and T_{12} (control, 18.58%) respectively. The range for the coefficient of variation was 12.37 to 51.80. The mean height in general, reduced significantly in combination treatment of EMS and gamma rays, but the height was increased in treatment gamma rays alone as compared to control. Jambulkar (2007) also reported the similar result of increase in height of plant with decrease in gamma rays dose.

The data on variation caused due to the different treatments of EMS and gamma rays on number of branch plant^{-1} are presented in table 4. The highest mean value for the number of branches plant^{-1} was in treatment T_{10} (1200+0.5% EMS, 3.30) and the lowest in treatment T_8 (1000gy+0.5% EMS, 1.90). In general the number of branches decreased in all the treatments except T_{10} (1200+0.5% EMS) and T_{11} (1300+0.5% EMS) as compared to their control T_6 (control, 2.7) and T_{12} (control, 2.17). The variability studies showed that the coefficient of variation increased against their control except in treatments T_7 (900gy+0.5% EMS), T_{10} (1200+0.5% EMS) and T_{11} (1300+0.5% EMS). The variation for the character ranges from 34.82 to 60.80% as compared to control T_6 (control, 42.94%) and T_{12} (control, 42.94%). The highest variation was recorded in T_8 (1000gy+0.5% EMS, 60.80%) and T_{10} (1100gy+0.5% EMS, 34.82%). Malek *et al.* (2012) also reported significant differences for number of branches plant^{-1} of plants.

Data on variation caused due to different concentrations of gamma rays and EMS in combinations on number of siliqua plant^{-1} are presented in table 5. The mean value of number of siliqua plant^{-1} ranged from in treatments T_9 (1100gy+0.5% EMS, 69.30%) to T_{10} (1100gy+0.5% EMS, 153.00%). The coefficient variation for the character increased in all the treatments except T_2 (1000gy) and T_3 (1100gy) as compared to the control. The highest variation was noticed in T_9 (1100gy+0.5% EMS, 66.79%) and the lowest in T_2 (1000gy, 29.67%). Both mutagens proved effective in increasing the number of siliqua plant^{-1}. The variations were induced by both the mutagens. Malek *et al.* (2012) and Yassein and Ali (2014) also exhibited similar results of significant difference for number of siliqua plant^{-1} in mustard treated with gamma rays.

The data regarding effect of different treatments of EMS and gamma rays on the seed yield plant^{-1} in M_2 generation are presented in table 6.The maximum mean seed yield plant^{-1} was observed in treatment T_3 (1100gy, 16.80g) and minimum in treatment T_9 (1100gy+0.5% EMS, 10.65g). The variability studies showed that coefficient of variation increased in all the treatments except treatment T_7 (900gy+0.5% EMS) and treatment T_{11} (1300gy+0.5% EMS). Maximum coefficient of variation was noticed in treatment T_5 (1300gy, 54.62%) followed by treatment T_4 (1200gy, 52.00%) and the minimum was in treatment T_2 (1000gy, 24.21%). The variation for this parameter ranged between 24.21 to 54.62%. Javed *et. al.* (2000) reported that five mutants produced significantly higher yield than parents. Similar result with radiation were also reported by Jambulkar and Shitre (2009), who reported mutant lines with significant superioity in seed yield.

The efficiency and effectiveness of mutagens were estimated as suggested by Konzak *et al.* (1965) and are

Table 1. Effect of different treatments of EMS and gamma rays on germination per cent of variety Pusa bold in M_1 and M_2 generation

Treatments	Germination (%)		Mortality	
	M_1	M_2	M_1	M_2
T_1 (900gy)	70.00	90.15	2.24	0.95
T_2(1000gy)	76.00	95.65	4.55	1.04
T_3(1100gy)	68.56	85.71	6.88	3.25
T_4(1200gy)	59.25	84.00	8.21	2.56
T_5(1300gy)	62.50	76.56	9.45	4.65
T_6(control)	82.23	86.50	1.34	0.83
T7(900gy+ 0.5% EMS)	57.49	75.43	6.45	2.70
T_8(1000gy+ 0.5% EMS)	62.00	78.41	8.34	1.20
T_9(1100gy+ 0.5% EMS)	60.12	73.85	7.33	4.21
T_{10}1200gy+ 0.5% EMS)	59.00	72.55	9.95	4.45
T_{11} (1300gy+ 0.5% EMS)	55.75	73.45	10.15	5.20
T_{12} (control)	80.50	85.36	2.35	0.93

Table 2. Effect of different treatments of EMS and gamma rays on days to maturity of variety Pusa bold in M_2 generation

Treatments	Range	Mean	Variance	S D	CV %
T_1 (900gy)	28.00	115.80	1759.49	41.95	36.22
T_2(1000gy)	28.00	109.40	77.83	8.82	8.06
T_3(1100gy)	28.00	115.23	1186.00	34.44	29.89
T_4(1200gy)	26.00	110.53	3208.20	56.64	51.24
T_5(1300gy)	25.00	112.53	3253.33	57.04	50.69
T_6(control)	20.00	119.50	32.94	5.74	4.80
T7(900gy+ 0.5% EMS)	28.00	116.70	66.29	8.14	6.98
T_8(1000gy+ 0.5% EMS)	29.00	116.17	53.32	7.30	6.29
T_9(1100gy+ 0.5% EMS)	31.00	113.23	90.81	9.53	8.42
T_{10}1200gy+ 0.5% EMS)	31.00	112.10	109.75	10.48	9.35
T_{11} (1300gy+ 0.5% EMS)	31.00	110.42	1989.89	44.61	40.40
T_{12} (control)	20.00	118.00	42.00	6.28	5.49

Table 3. Effect of different treatments of EMS and gamma rays on plant height (cm) of variety Pusa bold in M_2 generation.

Treatment	Range	Mean	Variance	S D	CV %
T_1 (900gy)	70	128.70	2375.22	48.74	37.87
T_2(1000gy)	63	137.70	290.15	17.03	12.37
T_3(1100gy)	100	124.37	2014.68	44.78	36.03
T_4(1200gy)	88	151.80	6183.75	78.64	51.80
T_5(1300gy)	73	149.60	5854.79	76.52	51.15
T_6(control)	62	126.70	541.79	23.28	18.37
T_7(900gy+ 0.5% EMS)	110	104.93	481.31	21.94	20.91
T_8(1000gy+ 0.5% EMS)	64	111.20	266.44	16.32	14.68
T_9(1100gy+ 0.5% EMS)	98	102.70	435.94	20.88.	20.33
T_{10}1200gy+ 0.5% EMS)	67	102.50	245.71	15.68	15.29
T_{11}(1300gy+ 0.5%EMS)	80	119.20	465.48	21.57	18.10
T_{12} (control)	63	121.90	512.77	22.64	18.58

Table 4. Effect of different treatments of EMS and gamma rays on number of branches plant^{-1}of variety Pusa bold in M_2 generation

Treatments	Range	Mean	Variance	S D	CV %
T_1 (900gy)	4	2.53	2.21	1.48	58.62
T_2(1000gy)	5	2.6	1.28	1.13	43.56
T_3(1100gy)	4	2.57	1.48	1.22	47.38
T_4(1200gy)	4	2.67	2.30	1.52	56.38
T_5(1300gy)	4	2.00	1.44	1.20	60.01
T_6(control)	3	2.70	1.34	1.16	42.94
T_7(900gy+ 0.5% EMS)	2	1.73	0.41	0.64	36.90
T_8(1000gy+ 0.5% EMS)	5	1.90	1.33	1.16	60.80
T_9(1100gy+ 0.5% EMS)	4	2.47	1.22	1.11	44.83
T_{10}1200gy+ 0.5% EMS)	4	3.30	1.32	1.15	34.82
T_{11}(1300gy+ 0.5%EMS)	4	2.93	1.24	1.11	37.91
T_{12} (control)	4	2.70	1.34	1.16	42.94

Table 5. Effect of different treatments of EMS and gamma rays on number siliqua plant^{-1} of variety Pusa bold in M_2 generation

Treatments	Range	Mean	Variance	S D	CV %
T_1 (900gy)	195	93.20	2830.22	53.20	57.08
T_2(1000gy)	125	116.07	1186.20	34.44	29.67
T_3(1100gy)	115	108.50	1649.74	40.62	37.43
T_4(1200gy)	140	102.40	3471.41	58.92	57.54
T_5(1300gy)	85	83.33	2106.37	45.90	55.07
T_6(control)	85	72.00	807.56	28.42	39.47
T_7(900gy+ 0.5% EMS)	221	93.03	2523.00	50.23	53.99
T_8(1000gy+ 0.5% EMS)	155	91.38	2784.30	52.77	57.74
T_9(1100gy+ 0.5% EMS)	229	69.13	2132.33	46.18	66.79
T_{10}1200gy+ 0.5% EMS)	398	153.00	9313.79	96.51	63.08
T_{11} (1300gy+ 0.5% EMS)	158	133.60	1871.83	43.26	32.38
T_{12} (control)	63	85.20	395.20	19.88	23.34

Table 6. Effect of different treatments of EMS and gamma rays on seed yield plant^{-1} (g) of variety Pusa bold in M_2 generation

Treatments	Range	Mean	Variance	S D	CV %
T_1 (900gy)	20.57	15.43	51.37	7.17	46.44
T_2(1000gy)	11.60	15.29	13.70	3.70	24.21
T_3(1100gy)	17.79	16.80	43.89	6.63	39.45
T_4(1200gy)	7.29	14.15	54.11	7.36	52.00
T_5(1300gy)	12.94	12.28	44.97	6.71	54.62
T_6(control)	12.90	17.46	15.91	3.99	22.85
T_7(900gy+ 0.5% EMS)	20.04	14.80	18.22	4.27	28.85
T_8(1000gy+ 0.5% EMS)	25.00	14.09	32.49	5.70	40.44
T_9(1100gy+ 0.5% EMS)	24.01	10.65	26.71	5.17	48.53
T_{10}1200gy+ 0.5% EMS)	21.75	16.63	32.89	5.73	34.48
T_{11} (1300gy+ 0.5% EMS)	15.24	14.60	12.52	3.54	24.24
T_{12} (control)	15.10	17.83	33.62	5.80	32.52

presented in table 7. Treatment T_1 (900gy) exhibited the highest mutagenic efficiency (6.78), while treatment T_9 (1100gy+0.5% EMS, 0.23) showed the lowest. The treatments of gamma rays alone in general, showed the higher efficiency than combination treatment of gamma rays and EMS in Pusa bold. The mutagenic effectiveness was highest in treatment T_1 (900gy, 0.28), while treatment T_9 (1100gy+0.5% EMS, 0.040) showed lowest. Mohamed and Abd El Haleem (2014) indicated significantly differential response of cultivars to radiation treatments. The treatments of gamma rays alone in general, showed the higher effectiveness than combination treatment of EMS and gamma rays in pusa bold.

Reduction in germination percentage was more in M_1 than in M_2 generation and hence the mortality was also more in M_1 than in M_2 generation as compared to control. There was decrease in days to maturity and number of branches plant^{-1} as compared to control. Plant height reduced in combination treatments of EMS and gamma rays but increased in treatment of gamma rays alone. Number of siliqua plant^{-1} and seed yield plant^{-1} did not follow any specific trend in any of the treatments. Treatment T_4 (1200 gy gamma rays) had maximum coefficient variance for most of the characters viz., days to maturity , plant height, and higher coefficient variance for number of siliqua plant^{-1} and seed yield plant. Hence, irradiation of 1200gy gamma rays induced maximum variability for days to maturity, plant height , no of siliqua plant^{-1} and seed yield plant^{-1}, but mutagenic efficiency and effectiveness was highest in treatment T_1 (900gy gamma rays).

Table 7. Mutagenic and effectiveness of different treatments of EMS and gamma rays in *Brassica junceae (*L.)

Sr.No.	Treatments	% Lethality (L)	% Mutation 30 M_2 Plants (MP)	Mutagenic efficiency (MP/L)	Mutagenic effectiveness (MP/tc)
1	T_1 (900gy)	0.95	6.45	6.78	0.280
2	T_2(1000gy)	1.04	5.61	5.39	0.230
3	T_3(1100gy)	3.25	2.05	0.63	0.085
4	T_4(1200gy)	2.56	2.01	0.78	0.083
5	T_5(1300gy)	4.65	2.84	0.61	0.110
6	T_7(900gy+ 0.5% EMS)	2.70	2.07	0.76	0.086
7	T_8(1000gy+ 0.5% EMS)	1.20	5.26	4.38	0.210
8	T_9(1100gy+ 0.5% EMS)	4.21	0.98	0.23	0.040
9	T_{10}1200gy+ 0.5% EMS)	4.45	4.08	0.91	0.170
10	T_{11} (1300gy+ 0.5% EMS)	5.20	3.40	0.65	0.140

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