

INDUCED POLYGENIC VARIABILITY IN SESAME BY COMBINING HYBRIDIZATION AND RECURRENT MUTAGENESIS IN F₃M₃ GENERATION

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Abstract

The investigation was made to estimate the combined effects of hybridization and ethyl methane sulphonate (EMS) induced mutagenesis on the variability of seed yield and its attributes in F₃M₃ generation. A total of 15 populations were grown in randomized block design (RBD) with three replications. F₃M₃ results indicated an increase in variability for plant height, number of capsules per plant and seed yield per plant, whereas non significant variability was observed for number of primary branches per plant in mutant populations than respective controls. Increased mean value and increase in variability in the present investigation denoted the occurrence of more favorable mutations and breaking up of undesirable linkages with enhancing recombination. Correlation study in F₃M₃ generation indicated remarkable changes among seed yield per plant and its component traits and these changes might have come through independent polygenic mutations and enhanced recombination of polygenes affecting different traits. The outcome of this study will facilitate in understanding the effectiveness of combined effects of hybridization and EMS induced mutagenesis for sesame improvement.

Introduction

Sesame is one of the important ancient oilseed crops in India. Sesame is cultivated in an area of 13.9 lakh hectares in India with an annual production of 41.8 lakh tonnes and productivity of 291 kg/ha (Anon. 2018). In Odisha, sesame is cultivated in 0.23 lakh hectares with annual production of 0.55 lakh tones and a productivity of 237 kg/ha (MOA, 2015).

Recombination and mutation breeding separately was given more emphasis to create new variability for the selection of superior lines as these methods have proved effective in developing many elite lines in many crop plants (Govindarasu 1995). Recombination breeding through hybridization between selected parents followed by selection in segregating generations is a popular method for crop improvement. This helps in combining useful characters into a single genotype, however, the full potentiality of the recombination limited due to the linkage between desirable and undesirable traits in a self-pollinated crop like sesame. As a result, variability is generated to a limited extent, limiting the scope for selection. Modifications of the conventional breeding approach have been recommended by many workers to break the undesirable linkage (Singh and Paroda 1986) and create a wide range of variability (Gregory 1955 and 1956). The present investigation in sesame was done to estimate the combined effects of hybridization and EMS treatment on the variability of seed yield and its attributes. This has been done through a critical analysis of variation in F₃M₃ generation. The outcome of the investigation will help in understanding the usefulness of combined hybridization and EMS treatment for sesame improvement.

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Materials and Methods

In F_3M_3 generation, three one cycle ethyl methane sulphonate (EMS) treated populations ($F_3^1M_{3s}$), three two cycles EMS treated populations ($F_3^2M_{3s}$), three numbers of three cycles EMS treated populations ($F_3^3M_{3s}$), three F_3 populations and three parents were evaluated. Each cycle EMS treatment was done with 0.5% EMS for 3 hrs. The selection of the chemical mutagen and dose was based upon the review of Begum and Dasgupta (2010, 2015) and Kumari *et al.* (2016). Thus, there were a total of 15 treatments including controls in F_3M_3 . F_3M_3 was evaluated at EB-II section of the Department of Plant Breeding and Genetics, OUAT during summer, 2018. The designation of various treatments and controls (F_{3s}) in F_3M_3 is given in Table 1. Sowing was done in plots of 7 rows and 3 m each with 30 cm \times 10 cm spacing. Routine cultural practices like fertilizer application @ 30 kg N, 60 kg P_2O_5 and 40 kg K_2O and need-based plant protection measures were followed. In F_3M_3 generation observations were recorded on four characters, *i.e.*, plant height, number of primary branches per plant, number of capsules per plant and single plant yield. For all the characters single plant observation was taken using a sample of 30 randomly selected competitive plants per plot.

Table 1. Designation of various treatments and controls in F_3M_3 generation of sesame.

Treatment symbol	Description
V ₁	Variety 1 (Nirmala)
V ₂	Variety 2 (Prachi)
V ₃	Variety 3 (Amrit)
¹ F ₃	F ₃ of cross 1 (Nirmala \times Prachi)
² F ₃	F ₃ of cross 2 (Nirmala \times Amrit)
³ F ₃	F ₃ of cross 3 (Prachi \times Amrit)
¹ F ₃ ¹ M ₃	F ₃ M ₃ following one cycle EMS treated population of cross 1
² F ₃ ¹ M ₃	F ₃ M ₃ following one cycle EMS-treated population of cross 2
³ F ₃ ¹ M ₃	F ₃ M ₃ following one cycle EMS-treated population of cross 3
¹ F ₃ ² M ₃	F ₃ M ₃ following two cycles EMS-treated population of cross 1
² F ₂ ² M ₂	F ₂ M ₂ following two cycles EMS-treated population of cross 2
³ F ₂ ² M ₂	F ₂ M ₂ following two cycles EMS-treated population of cross 3
¹ F ₃ ³ M ₃	F ₃ M ₃ following EMS treatment (Three cycles) of $F_3^2M_3$ seeds *of cross 1
² F ₃ ³ M ₃	F ₃ M ₃ following EMS treatment (Three cycles) of $F_3^2M_3$ seeds of cross 2
³ F ₃ ³ M ₃	F ₃ M ₃ following EMS treatment (Three cycles) of $F_3^2M_3$ seeds of cross 3

*Indicates seeds of $F_2^2M_2$ plants. The superscript of F_3 in treatment symbol is not a part of the filial generation it represents the cross number, similarly superscript of M_3 indicates the mutagen treated cycles.

Variance analysis was done on plot mean values basis for each of the four quantitative traits and the “F” test was done to know the treatment differences. The significant difference between two means was tested by “t” test by estimation of CD, standard deviation and coefficient of variance (CV) for each treatment replication wise for all the four polygenic traits. ANOVA of mean, standard deviation, and CV was estimated and the significance of differences among treatments tested by “F” and “t” tests following method proposed by Bahl *et al.* (1968). The statistical significance of correlation coefficient was tested by “t” test (Snedecor and Cochran 1967).

Results and Discussion

The variability of yield and its components were analyzed in bulk F_3M_3 populations following 1 to 3 cycles of EMS treatment of F_1 hybrids of 3 intervarietal crosses (Tables 2 to 8). ANOVA of means for four characters in F_3M_3 is presented in Table 2. "F" test indicated significant differences among treatments in means of all characters, except number of primary branches per plant. ANOVA of the standard deviation and CV of four characters are presented in Tables 3 and 4. The "F" test indicated significant differences among treatments, both in the standard deviation and CV of

Table 2. ANOVA of means for four characters in F_3M_3 generation (Mean sum of squares).

Sl. No.	Characters	Source of variation (df)		
		Blocks (2)	Treatments (14)	Error (28)
1.	Plant height (cm)	22.527	39.871**	5.152
2.	Number of primary branches/plant	0.030	0.047	0.038
3.	Number of capsules/plant	3.726	121.987**	12.007
4.	Seed yield/plant (g)	0.115	3.442**	0.353

**Significant at 5 and **1% level of probability, respectively.

Table 3. ANOVA of standard deviation for four characters in F_3M_3 generation (Mean sum of squares).

Sl. No.	Characters	Source of variation (df)		
		Blocks (2)	Treatments (14)	Error (28)
1.	Plant height (cm)	0.129	19.887**	1.710
2.	Number of primary branches/plant	0.051	0.034	0.031
3.	Number of capsules/plant	5.930	77.634**	11.677
4.	Seed yield/plant (g)	0.017	3.568**	0.425

**Significant at 5 and **1% level of probability, respectively.

Table 4. ANOVA of CV for four characters in F_3M_3 generation (Mean sum of squares).

Sl. No.	Characters	Source of variation (df)		
		Blocks (2)	Treatments (14)	Error (28)
1.	Plant height (cm)	0.470	16.042**	1.449
2.	Number of primary branches/plant	65.717	22.397	24.743
3.	Number of capsules/plant	12.897	83.675**	18.755
4.	Seed yield/plant (g)	0.676	167.441**	26.696

**Significant at 5 and **1% level of probability, respectively.

all the characters except number of primary branches per plant. Significant treatment differences in mean, standard deviation and CV were quite expected. However, a non-significant difference for number of primary branches per plant was observed. The test of significant differences among

treatments was observed, however a prerequisite for critical comparison of treatments with regard to different parameters.

The range of variation, mean and standard deviation of four characters in F_3M_3 generation are presented in Tables 5 to 8. Among the crosses and mutant populations, population ${}^2F_3{}^3M_3$ (three cycles, EMS treated population of Nirmala \times Amrit) showed the highest range of variation *i.e.* 83.59 - 159.66 for plant height (Table 5). One and two cycles EMS treated populations of Nirmala \times Prachi had lower mean, whereas three cycles EMS treated population ${}^1F_3{}^3M_3$ had similar mean than the control F_3 (Nirmala \times Prachi). Similarly, one and three cycles EMS treated populations of Nirmala \times Amrit had significantly lower mean and two cycles EMS treated population ${}^1F_3{}^2M_3$ had a similar mean than the control 2F_3 . Among the mutant populations of Prachi \times Amrit, populations ${}^3F_3{}^1M_3$ and ${}^3F_3{}^3M_3$ had similar mean and ${}^3F_3{}^2M_3$ had significant increased mean than control 3F_3 . All the mutant populations of Nirmala \times Prachi had significant increased standard deviation than the control for plant height. Among the mutant populations of Nirmala \times Amrit, populations ${}^2F_3{}^1M_3$ and ${}^2F_3{}^2M_3$ had similar standard deviation and population ${}^2F_3{}^3M_3$ had significant higher standard deviation than control (2F_3). Population ${}^3F_3{}^3M_3$ had significant positive, ${}^3F_3{}^2M_3$ reduced and ${}^3F_3{}^1M_3$ with similar standard deviation than control F_3 (Prachi \times Amrit).

Table 5. Range, mean and standard deviation of plant height (cm) in F_3M_3 generation.

Treatment	Range	Mean	Sd.
V_1	93.83 - 130.85	115.60	7.506
V_2	99.46 - 130.96	117.36	6.319
V_3	99.26 - 129.26	114.74	6.762
1F_3	99.00 - 140.88	117.92	8.005
${}^1F_3{}^1M_3$	91.74 - 139.06	111.31*	10.246*
${}^1F_3{}^2M_3$	87.54 - 141.72	108.76*	12.555*
${}^1F_3{}^3M_3$	91.15 - 144.28	115.35	14.864*
2F_3	101.35 - 144.20	121.38	11.814
${}^2F_3{}^1M_3$	86.76 - 140.22	114.72*	11.637
${}^2F_3{}^2M_3$	91.02 - 141.02	121.25	9.968
${}^2F_3{}^3M_3$	83.59 - 159.66	115.31*	15.005*
3F_3	86.54 - 144.43	111.78	11.875
${}^3F_3{}^1M_3$	93.41 - 145.91	113.32	10.611
${}^3F_3{}^2M_3$	97.14 - 147.07	120.52*	8.941*
${}^3F_3{}^3M_3$	81.72 - 152.72	114.41	14.482*
CD at 5%	-	3.80	2.19
SE(m)	-	1.31	0.75
CVe	-	1.96	12.53

*Significantly different from control.

The range of variation, mean and standard deviation of the number of primary branches per plant in F_3M_3 are presented in Table 6. All the mutant populations had similar mean and standard deviation for primary branches per plant.

Table 6. Range, mean and standard deviation of number of primary branches per plant in F₃M₃ generation.

Treatment	Range	Mean	Sd
V ₁	1.82 - 5.06	3.32	1.039
V ₂	1.74 - 4.81	3.01	0.762
V ₃	1.96 - 6.06	3.49	1.091
¹ F ₃	1.27 - 5.73	3.20	0.927
¹ F ₃ ¹ M ₃	1.86 - 5.86	3.29	0.877
¹ F ₃ ² M ₃	0.78 - 5.78	3.28	0.980
¹ F ₃ ³ M ₃	1.83 - 5.83	3.27	0.917
² F ₃	0.94 - 6.21	3.31	1.118
² F ₃ ¹ M ₃	1.90 - 6.00	3.47	1.091
² F ₃ ² M ₃	0.99 - 5.99	3.32	1.100
² F ₃ ³ M ₃	0.84 - 5.98	3.34	1.020
³ F ₃	1.81-6.14	3.24	1.077
³ F ₃ ¹ M ₃	1.99-4.02	3.12	0.860
³ F ₃ ² M ₃	1.79-6.12	3.32	1.005
³ F ₃ ³ M ₃	1.89-6.09	3.46	0.922
CD at 5%	-	0.33	0.29
SE(m)	-	0.11	0.10
CVe	-	5.92	17.88

*Significantly different from control.

Table 7. Range, mean and standard deviation of number of capsules per plant in F₃M₃ generation.

Treatment	Range	Mean	Sd
V ₁	26.53 - 120.53	64.83	18.521
V ₂	40.72 - 101.89	61.62	10.658
V ₃	39.86 - 124.09	70.02	20.434
¹ F ₃	39.00 - 102.07	65.67	14.685
¹ F ₃ ¹ M ₃	44.26 - 122.29	69.86	13.864
¹ F ₃ ² M ₃	38.53 - 124.43	74.53*	20.547*
¹ F ₃ ³ M ₃	40.20 - 133.07	76.13*	26.345*
² F ₃	42.10 - 129.80	64.27	16.116
² F ₃ ¹ M ₃	39.18 - 121.18	65.78	16.180
² F ₃ ² M ₃	39.50 - 131.33	78.80*	22.170*
² F ₃ ³ M ₃	41.23 - 133.43	78.37*	26.150*
³ F ₃	42.18 - 103.08	64.78	13.252
³ F ₃ ¹ M ₃	51.07 - 135.70	68.30	14.959
³ F ₃ ² M ₃	40.37 - 127.83	74.27*	21.286*
³ F ₃ ³ M ₃	48.30 - 133.40	82.50*	27.059*
CD at 5%	-	5.80	5.72
SE(m)	-	2.00	1.97
CVe	-	4.90	17.82

*Significantly different from control.

The range of variation, mean and standard deviation of number of capsules per plant in F_3M_3 generation are presented in Table 7. Among the crosses and mutant populations, population ${}^1F_3{}^3M_3$ had the highest range of variation with an estimated range of 40.20 - 133.07. All the $F_3{}^2M_{3s}$ and $F_3{}^3M_{3s}$ populations had significant greater mean as compared to their respective controls (F_{3s}) for number of capsules per plant. All the three $F_3{}^1M_{3s}$ had similar mean as compared to their respective controls (F_{3s}). Two and three cycles EMS treated populations of Nirmala \times Prachi and Nirmala \times Amrit had similar mean, whereas three cycles EMS treated population of Prachi \times Amrit had significant greater mean than its two cycles EMS treated population for number of capsules per plant. All the $F_3{}^2M_{3s}$ and $F_3{}^3M_{3s}$ populations had significant greater standard deviation as compared to their respective controls (F_{3s}) for number of capsules per plant. All the three $F_3{}^1M_{3s}$ had similar standard deviation as compared to their respective controls (F_{3s}).

Among the crosses and mutant populations, three cycles EMS treated population of Nirmala \times Amrit (${}^2F_3{}^3M_3$) had the highest range of 5.90 - 25.83 (Table 8). All the mutant populations of Nirmala \times Prachi had slightly increased mean value for seed yield per plant than control (1F_3), however, only populations ${}^1F_3{}^2M_3$ and ${}^1F_3{}^3M_3$ had a higher standard deviation for seed yield per plant as compared to control (1F_3). Single cycle EMS treated mutant population ${}^1F_3{}^1M_3$ had a similar standard deviation as compared to control 1F_3 . Among the mutant populations of Nirmala \times Prachi, three cycles, EMS treated population had the highest standard deviation followed by two

Table 8. Range, mean and standard deviation of seed yield per plant (g) in F_3M_3 generation.

Treatment	Range	Mean	Sd
V ₁	4.86-19.19	11.29	3.622
V ₂	5.07-15.99	8.69	2.047
V ₃	4.18-18.54	11.03	3.451
1F_3	5.19-15.85	9.45	2.454
${}^1F_3{}^1M_3$	5.52-21.34	10.70*	2.642
${}^1F_3{}^2M_3$	4.94-23.21	11.54*	4.010*
${}^1F_3{}^3M_3$	5.52-24.63	11.87*	5.113*
2F_3	6.54-23.34	11.03	3.755
${}^2F_3{}^1M_3$	5.48-21.97	10.30	3.332
${}^2F_3{}^2M_3$	5.29-23.90	12.48*	4.922*
${}^2F_3{}^3M_3$	5.90-25.83	12.60*	6.029*
3F_3	6.46-20.52	10.19	2.563
${}^3F_3{}^1M_3$	5.87-19.38	10.75	2.718
${}^3F_3{}^2M_3$	4.50-23.73	10.89	4.210*
${}^3F_3{}^3M_3$	6.10-24.09	12.12*	5.446*
CD at 5%	-	0.99	1.09
SE(m)	-	0.34	0.38
CVe	-	5.40	17.65

*Significantly different from control.

cycles EMS treated population. Among the mutant populations of Nirmala \times Prachi, one and two cycles EMS treated populations (${}^1F_3{}^1M_3$ and ${}^1F_3{}^2M_3$) and two and three cycles EMS treated populations (${}^1F_3{}^2M_3$ and ${}^1F_3{}^3M_3$) had similar mean, whereas one and three cycles EMS treated populations had significant different mean for seed yield per plant. Two and three cycles EMS treated populations of Nirmala \times Amrit had slightly higher mean and standard deviation than

control 2F_3 , while population ${}^2F_3{}^1M_3$ had similar mean and standard deviation than 2F_3 for seed yield per plant. Two and three cycles EMS treated populations of Nirmala \times Amrit had similar mean, but three cycles EMS treated population had a significant higher standard deviation for seed yield per plant. Population ${}^3F_3{}^3M_3$ had a higher mean, whereas population ${}^3F_3{}^1M_3$ and ${}^3F_3{}^2M_3$ had a similar mean than control 3F_3 . Populations ${}^3F_3{}^2M_3$ and ${}^3F_3{}^3M_3$ had significant higher standard deviation for seed yield per plant than controls (3F_3).

Overall, F_3M_3 results indicated positive or no shift in mean for plant height, while a positive shift in mean for number of capsules per plant and seed yield per plant in mutant populations than respective controls. Increased mean values in the present investigation, denoted the occurrence of more favorable mutations and breaking up of undesirable linkages with enhancing recombination. Similar results of an increase in mean value have been reported by Begum and Dasgupta (2015) and Ravichandran and Jayakumar (2018) for number of capsules per plant and seed yield per plant.

In general, with a few exceptions, variability increased with increase in number of EMS treatment cycles and maximum variability was observed at three cycles EMS treated populations for all the characters except number of primary branches per plant. The amount of variability created also varies from genotype to genotype. All the mutant populations had similar mean and standard deviation for primary branches per plant. The results indicated no scope of improvement for primary branches per plant by induced mutagenesis. Among the two and three cycles EMS treated populations, three cycles EMS treated populations had the highest variability for plant height, number of capsules per plant and seed yield per plant.

Table 9. Correlations among four characters in F_3M_3 generation.

Treatment	r_{12}	r_{13}	r_{14}	r_{23}	r_{24}	r_{34}
V_1	0.146	0.023	0.048	0.666**	0.625**	0.930**
V_2	0.002	0.074	0.124	0.511**	0.351**	0.726**
V_3	-0.030	0.120	0.004	0.765**	0.729**	0.906**
1F_3	-0.011	0.072	0.039	0.802**	0.655**	0.871**
${}^1F_3{}^1M_3$	-0.077	-0.023	-0.063	0.655**	0.511**	0.803**
${}^1F_3{}^2M_3$	0.132	0.230*	0.286**	0.735**	0.700**	0.921**
${}^1F_3{}^3M_3$	0.152	0.092	0.112	0.622**	0.629**	0.945**
2F_3	0.334**	0.342**	0.367**	0.643**	0.605**	0.912**
${}^2F_3{}^1M_3$	0.450**	0.606**	0.567**	0.791**	0.659**	0.847**
${}^2F_3{}^2M_3$	0.175	0.193	0.168	0.782**	0.688**	0.927**
${}^2F_3{}^3M_3$	0.378**	0.513**	0.545**	0.760**	0.730**	0.943**
3F_3	0.015	-0.046	-0.006	0.741**	0.723**	0.844**
${}^3F_3{}^1M_3$	0.427**	0.509**	0.354**	0.639**	0.551**	0.813**
${}^3F_3{}^2M_3$	0.223*	0.329**	0.258*	0.763**	0.730**	0.930**
${}^3F_3{}^3M_3$	0.547**	0.556**	0.601**	0.685**	0.662**	0.918**

*,**Significant at 5 and 1% level of probability, respectively.

¹Plant height, ²number of primary branches/plant, ³number of capsules/plant, ⁴seed yield/plant.

The increase in variability for these characters had arisen from induced micro mutations and that resulting from segregation and recombination of polygenes in hybrid populations are cumulative. Similar results have been reported by Ravichandran and Jayakumar (2018) for number of capsules per plant and single plant yield and for plant height, number of branches per

plant, number of capsules per plant and seed yield per plant by Sarwar *et al.* (2010) and Kumar (2017).

Selection for yield will be effective when it is considered along with yield attributes rather than relying on yield only. Phenotypic correlations among seed yield per plant and three of its components in F_3M_3 of the different populations are presented in Table 9. In all F_{3s} and F_3M_{3s} populations characters like number of primary branches per plant and number of capsules per plant showed significant positive correlations with seed yield per plant but the magnitude of association varies from population to population. Similar positive associations have been reported by Ismail *et al.* (2013), Shabana *et al.* (2015) and Kumar (2017).

Plant height showed significant positive correlation with seed yield per plant in population 2F_3 (Nirmala \times Amrit), but in populations 1F_3 (Nirmala \times Prachi) and 3F_3 (Prachi \times Amrit) the association was non significant. Among mutant populations $F_3{}^3M_3$ Nirmala \times Amrit (${}^2F_3{}^3M_3$) and $F_3{}^3M_3$ Prachi \times Amrit (${}^3F_3{}^3M_3$), plant height had a significant positive association with seed yield per plant. Among mutant populations ${}^1F_3{}^2M_3$ and ${}^3F_3{}^2M_3$, plant height had a significant positive relationship with seed yield per plant. Plant height had significant positive association with seed yield per plant among mutants ${}^2F_3{}^1M_3$ and ${}^3F_3{}^1M_3$.

Considering all the possible correlations of three categories (*i.e.* one, two and three cycles mutated populations of a particular cross) of F_3M_{3s} together, seven of the 18 correlations in the F_3M_{3s} of Nirmala \times Prachi were greater, four correlations smaller and seven similar to those in 1F_3 . In F_3M_{3s} of Nirmala \times Amrit, nine correlations were greater, four smaller and five similar to those in F_3 (Nirmala \times Amrit). Similarly, among F_3M_{3s} of Prachi \times Amrit, nine correlations were greater, three were smaller and six similar to those in F_3 (Prachi \times Amrit). In general, considering all the possible correlations three cycles EMS treated populations had increased in magnitude than corresponding controls with few exceptions. Also, three cycles EMS treated mutants had increased associated magnitude as corresponding two cycles EMS treated mutants of sesame.

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