Bot. Bull. Academia Sinica 25: 103-110 (1984)

MUTATIONS INDUCED BY GAMMA-IRRADIATION, ETHYL METHANE SULFONATE AND HYDRAZINE HYDRATE IN MUNG BEAN (*PHASEOLUS AUREUS* ROXB.)[1]

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(Received August 5, 1983; Accepted December 5, 1983)

Abstract

Mutations induced by gamma rays, ethyl methane sulfonate (EMS) and hydrazine hydrate (HZ) were evaluated on days to flowering, number of fertile branches and number of pods in the M1, M2 and M3 generations. These characters exhibited increase in variance as well as in mean values, with the few exceptions in the M1 generation. Analysis of M2 and M3 data revealed that there was a significant increase in mean values for all the characters. Coefficient of phenotypic variability increased in both the generations. The highest genotypic coefficient of variation and values of heritability were recorded in the M3 generation as compared to M2. Estimates of heritability were high for number of pods followed by days to flowering and then number of fertile branches. Days to flowering and number of pods showed the highest values of genetic advance in the M3 generation. Mutagenic treatments increased the phenotypic and genotypic coefficient of variation as well as heritability and genetic advance. This suggests that the further improvement of this variety of mung bean is possible through induced mutagenesis.

Introduction

Induced mutations can be used to generate useful variation in quantitatively inherited characters of crop plants. The classical works of Brock (1965), Gregory (1968) and Bojyo *et al.* (1969) on improvement of yield, Gustafsson (1965) and Bojyo *et al.* (1969) on adaptability, Brock (1970) on maturity time, Brock (1971) on flowering time and Sigurbjornsson and Micke (1969) on numerous other traits in

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various crop plants provide example of induction of mutations in polygenic characters. The present paper summarises the results of experiments on gamma rays, ethyl methane sulfonate (EMS) and hydrazine hydrate (HZ) induced variability for days to flowering, number of fertile branches and number of pods in M1, M2 and M3 generations in mung bean, variety PS-16.

Materials and Methods

Dry seeds (moisture content 12%) of the var. PS-16 of mung bean, *Phaseolus aureus* Roxb., were exposed to 20 kR ($1R=2.58\times10^{-4}$ C/kg) of gamma rays from ^{60}CO source. Seeds pre-soaked in distilled water for 9 hours were treated with freshly prepared 0.3% EMS and 0.04% HZ solutions for 6 hours at a constant room temperature of 27±1°C. The EMS and HZ solutions were prepared using standard buffer of pH 7. After treatment, the treated seeds were washed thoroughly in running tap water for half an hour.

The treated and control seed material was sown in a randomized complete block design with three replications. Seeds harvested from individual M1 plants were planted in 9 meter long progeny rows. The plant to plant and interrow distances were 30 cm and 60 cm, respectively. Observations were recorded on 15 normal-looking families of M2 generation for each treatment to study the three quantitative characters: days to flowering, number of fertile branches and number of pods in the M2 generation. The details of sowing in M1, M2 and M3 generations have been given elsewhere by Khan (1981, 1983a).

Data were collected on individual M2 and M3 plants and subjected to statistical analyses to assess the extent of induced variation. The different treatments were compared with their respective controls by *t*-test. The broad-sense heritability (h^2) of a character was estimated as follows:

$$h^2 = \frac{\delta_g^2}{\delta_t^2} \times 100$$

where δ_g^2 = induced genetic variance and δ_t^2 is the total phenotypic variance ($\delta_t^2 = \delta_g^2 + \delta_e^2$) calculated from the treated populations.

The genetic advance (G_s) with a 1% selection intensity was computed by the following formula:

$$G_s = k \cdot \delta_p \cdot h^2$$

where δ_p = phenotypic standard deviation of the mean performance of the treated populations; h^2 = heritability in broad-sense and $k = 2.64$ constant for 1% selection differential.

Results

The variability induced by gamma rays, EMS and HZ in different quantitative characters was studied in M1, M2 and M3 generations. The data presented in Table 1 clearly show that the variability induced was more in the treated populations as compared with their respective controls in the M1 generation. However, mean days to flowering reduced only at 20 kR of gamma rays. The EMS treatment increased the mean number of fertile branches and pods. But the number of pods decreased at 20 kR and 0.04% HZ.

Table 1. *Estimates of mean values* ($\bar{X}$), *standard error of mean* ($S_{\bar{x}}$) *and variances* (δ^2) *for different quantitative characters in the* M_1 *generation*

Treatments	N*	Days to Flowering		Number of Fertile Branches		Number of Pods	
		$\bar{X}\pm S_{\bar{x}}$	δ^2	$\bar{X}\pm S_{\bar{x}}$	δ^2	$\bar{X}\pm S_{\bar{x}}$	δ^2
Control	84	42.22±0.44	5.78	3.97±0.14	0.69	15.00±1.27	33.79
20 kR	76	40.62±0.68	9.59	4.00±0.22	1.25	13.67±1.53	58.17
0.3% EMS	61	43.83±0.59	8.31	4.07±0.18	0.99	18.87±1.32	61.70
0.04% HZ	60	44.04±0.55	7.81	3.79±0.19	0.88	14.06±1.29	40.36

N*: Number of plants survived.

The data recorded for days to flowering in M2 and M3 generations are presented in Tables 2 and 3. The results, in general, showed that the range of variability shifted only in the positive direction in both the generations. The mean flowering time increased significantly ($P\geq0.01$) from the control. Highest phenotypic coefficient of variation (27.08%) was shown in the M2 generation with the treatment of 0.04% HZ. However, the genotypic coefficient of variability, heritability and expected genetic advance were recorded more in the M3 generation.

The mean number of fertile branches and number of pods increased significantly in M2 and M3 generations (Tables 2 and 3). The range of variability increased in both the directions, but it was more in the positive direction. The highest coefficient of phenotypic variability (102.19%) and (36.98%) was recorded with the treatment of 0.04% HZ in the M2 and M3 generations, respectively. The same trend was noticed for the other character, number of pods. But the genotypic coefficient of variability was highest with the treatment of HZ followed by EMS and then gamma rays in M2 and M3 generations for both the characters. The total broad-sense heritability was highest (186.72%) in the M3 generation whereas it was 117.23% in the M2 generation for the character, number of fertile branches. However, mean number of pods showed highest values of heritability in the M3 generation. The expected genetic advance was recorded more in the M2 generation with all these

mutagens whereas it was quite high with the treatment of gamma rays only (Tables 2 and 3).

Discussion

Mutations affecting quantitative characters can best be inferred by the estimation of mean and the genetic parameters in the successive generations of the mutagen-treated populations. The effects of ionizing radiation and chemical mutagens were measured on the polygenic traits for the further improvement of the genetic architecture of this pulse.

Direction of Shift in Mean

The data presented in Table 1 show that the mean values increased in the treated populations of M1 generation as compared to the control. The exceptions were noticed for days to flowering and number of pods at 20 kR and for fertile branches, it was with the treatment of 0.04% HZ. However, the mean values shifted significantly in the M2 and M3 generations (Tables 2, 3 and 4). There has

Table 2. *Estimates of range (R), mean values ($\bar{X}$), standard error of mean ($S_{\bar{x}}$), coefficient of variation, heritability (h^2) and expected genetic advance (Gs) for three quantitative characters in the M_2 generation*

Treatments	N*	R	$\bar{X}\pm S_{\bar{x}}$	*t*-test	CV(*p*) (%)	CV(*g*) (%)	h^2 (%)	Gs
			Days to Flowering					
Control	273	28-43	35.70±0.32	—	18.68	—	—	—
20 kR	264	29-43	38.15±0.26	**	11.99	0.64	0.60	0.06
0.3% EMS	262	29-43	37.11±0.30	**	16.13	2.28	5.38	0.68
0.04% HZ	273	29-45	37.12±0.27	**	27.08	4.09	4.75	0.54
			Number of Fertile Branches					
Control	213	3-9	5.07±0.18	—	43.80	—	—	—
20 kR	211	3-13	6.22±0.30	**	85.88	23.80	40.61	4.73
0.3% EMS	201	4-13	6.86±0.21	**	62.01	16.55	36.66	2.82
0.04% HZ	207	3-10	5.28±0.16	a	102.19	27.53	39.96	2.42
			Number of Pods					
Control	213	17-73	42.81±1.62	—	42.14	—	—	—
20 kR	211	13-98	43.18±2.41	a	88.15	24.49	41.17	37.99
0.3% EMS	201	18-89	48.93±1.58	**	73.45	18.87	31.20	18.46
0.04% HZ	207	15-87	46.60±1.40	**	88.89	24.74	33.98	18.08

N*: Number of plants survived.
** Significant at 1% level, a=Non-significant.
CV(*p*)=Phenotypic coefficient of variation, CV(*g*)=Genotypic coefficient of variation.

Table 3. *Estimates of range (R), mean values ($\bar{X}$), standard error of mean ($S_{\bar{x}}$), coefficient of variation, heritability (h^2) and expected genetic advance (Gs) for three quantitative characters in the M_3 generation*

Treatments	N*	R	$\bar{X}\pm S_{\bar{x}}$	*t*-test	CV(*p*) (%)	CV(*g*) (%)	h^2 (%)	Gs
			Days to Flowering					
Control	306	33-43	38.11±0.66	—	15.57	—	—	—
20 kR	301	33-44	39.21±0.21	**	20.92	18.25	76.06	7.35
0.3% EMS	300	33-46	40.12±0.20	**	21.03	18.29	75.63	6.92
0.04% HZ	306	33-45	40.11±0.18	**	22.32	19.46	76.02	6.39
			Number of Fertile Branches					
Control	200	3-6	4.27±0.07	—	29.94	—	—	—
20 kR	200	3-7	5.15±0.07	**	29.45	23.21	62.13	1.54
0.3% EMS	200	3-8	5.33±0.07	**	24.10	16.78	48.48	1.29
0.04% HZ	200	3-5	4.92±0.07	**	36.98	32.26	76.11	1.88
			Number of Pods					
Control	200	13-70	37.98±0.29	—	41.10	—	—	—
20 kR	200	16-76	39.88±0.90	*	65.99	60.02	80.33	26.93
0.3% EMS	200	15-90	45.41±0.99	**	59.93	53.00	79.34	29.36
0.04% HZ	200	16-90	41.59±0.99	**	71.15	61.85	80.54	29.86

N*: Number of plants survived.
*,** Significant at 5% and 1% level, respectively.
CV(*p*)=Phenotypic coefficient of variation, CV(*g*)=Genotypic coefficient of variation.

Table 4. *Shift in mean in different treatments against control in M_1, M_2 and M_3 generations*

Treatments	Days to Flowering			Number of Fertile Branches			Number of Pods		
	M_1	M_2	M_3	M_1	M_2	M_3	M_1	M_2	M_3
Control	—	—	—	—	—	—	—	—	—
20 kR	−1.60	+2.44	+1.10	+0.03	+1.15	+0.88	−1.33	+0.38	+1.90
0.3% EMS	+1.61	+1.41	+2.01	+0.10	+1.79	+1.07	+3.87	+6.12	+7.43
0.04% HZ	+1.82	+1.42	+2.00	−0.18	+0.21	+0.66	+0.94	+3.79	+3.61

been an increase in the mean values in both the generations. This can be explained as a recovery effect in the successive generations. Borojevic (1965) studied the effect of thermal-neutrons and gamma rays in *Triticum vulgare*, observed an increase in the M3 means, might have resulted from the purposeful elimination of all mutants which produce abnormal-spike morphology and fertility prior to M3 generation. Khan (1981) exercised selection for normal-looking plants in the M1 generation, there was an increase in the mean values of the treated populations in

the M2 and M3 generations, might be due to the elimination of aberrant plants and also due to genetic nature of the changes induced after mutagenic treatments. But in the present study, the change was a consequence of the elimination of undesirable genes and partly due to the recovery effect in the successive generations. The mean number of fertile branches and pods increased concurrently with the increase in days to flowering. Increase in pods has also been reported by Rajput (1974) and Khan (1981, 1982, 1983a). Thus, it appears from the present study that there is scope for further improvement of this variety of mung bean with regard to the polygenic characters like fertile branches and pods by using micro-mutation technique.

Extent of Induced Variation

The data recorded in the M1 generation revealed that the induced variability was more in the treated populations as compared to control (Table 1). The range of variability was on either side of the control but it was more on the positive side in the M2 and M3 generation (Tables 2 and 3). The estimates of genetic parameters showed higer values for phenotypic and genotypic coefficient of variation in all the characters in both the generations, i. e. mutations of polygenes governing the quantitative characters. Increase in variability after mutagenic treatments was reported by Yamaguchi (1964), Sharma and Saini (1970), Daly (1973), Rajput (1974), Rao and Siddiq (1976), Shakoor *et al.* (1978), Larik (1979), Chaturvedi and Singh (1980) and Khan (1982, 1983a, b). The genotypic coefficient of variability was highest in the M3 generation as compared to M2. The maximum genetic variability was recorded for number of pods followed by number of fertile branches and then days to flowering.

Induced maximum variability in polygenes might be due to increased mutations and recombination induced by gamma rays, EMS and HZ. These mutagens could successfully be used for creating genetic variability and ultimately the selection of mutants with more number of pods and increased number of fertile branches be made. From practical point of view, increased variation assumes greater significance; thus, induced genetic variability can effectively be exploited for improving yield of this variety of mung bean through more number of pods and branches.

Heritability and Genetic Advance

Estimates of heritability and genetic advance increased many fold in the treated populations and differed trait to trait in both M2 and M3 generations (Tables 2 and 3). The combination of higher values of heritability and genetic advance was noticed in the M3 generation for the characters, days to flowering and number of pods. However, genetic advance was slightly more in the M2 for number of fertile

branches. Higher values of heritability and genetic advance were reported by Larik *et al.* (1980), Rao (1982) and Khan (1983). All the three characters; days to flowering, number of fertile branches and number of pods showed considerable increase in the values of heritability and genetic advance, indicating that these characters can be transmitted to future generations and a potential gain could possibly be achieved through selection in early generations. The success of selection, however, will be greater in subsequent generations when there will be increased recombination and elimination of cytological variants (Kumar, 1972). Higher values of heritability coupled with higher values of genetic advance met with in the present investigation suggests that the further improvement is likely to be very effective in this variety through induced mutagenesis.

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以珈瑪射線、磺酸乙基甲烷和水化聯氨誘變綠豆

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以珈瑪射線、磺酸乙基甲烷（EMS）和水化聯氨（HZ）誘發綠豆突變，評估 M_1, M_2, M_3 子代之開花日數、結果枝數和果莢數。上述性狀之變方與平均值都增加，僅突變第一代（M_1）有少數例外。M_1、M_2 之資料分析顯示所有性狀之平均值均顯著增加。此二突變代之表現型變異係數都增加。與 M_2 比較，M_3 有最高之基因型變異係數以及遺傳率。遺傳率估值大小依序爲果莢數、開花日數、結果枝數。突變第三代（M_3）之開花日數及果莢數表現出最高之遺傳進階值（genetic advance）。誘變處理不僅增高了表現型和基因型之變異係數，同時也增高了遺傳率與遺傳進階值。此項結果提示未來綠豆品種之改良或可採取誘變方法。