

Studies on Induction of Mutations in Greengram (*Phaseolus aureus* Roxb.) Through Physical Mutagens¹

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Phenotypic responses of a greengram (*Phaseolus aureus* Roxb.) to X-rays and gamma irradiation were studied in the M_1 and M_2 generations. Increase in variance was noticed for all the economic traits in M_2 , offering good scope for selection. A dose of 80 kR of the mutagens has been found to be adequate for an intensive programme of improvement of the strain Co. 1 greengram through mutation breeding.

Informations regarding the possibilities of induction of mutations through physical and chemical mutagens for creating new variability in greengram are rare except for a few reports on the sensitivity studies (Santos, 1965; Murray and Newcombe, 1970; Anon., 1971). Hence the present study was undertaken to gather information on the consequences of induction of physical mutations in this crop. The observations on M_1 and M_2 generations following X-ray and gamma ray treatment in the strain Co. 1 green gram are reported herein.

MATERIALS AND METHODS

Dry seeds (8 — 10 per cent moisture) of the strain Co. 1 greengram evolved through pureline selection were treated with X-rays and gamma rays (80, 100, 120, 140 and 160 kR). X-irradiation was done from a Philips C. T. apparatus operated at 50 KV without filter at a distance of 4 cm from the source, the dose rate being 500r/second. Gamma irradiation was done

using 1175 curie ^{60}Co gamma cell available at the Sugarcane Breeding Institute, Coimbatore. One hundred seeds were treated with each of the mutagens in each dose and they were sown in four replications in the field in a simple RB design. Observations on survival, plant height, number of clusters and number of pods per plant and seed fertility were recorded in M_1 generation. Seeds were collected from M_1 plants and carried forward to M_2 . A total number of 116 M_1 plants were studied under different treatments and all of them were carried forward to M_2 generation.

Chlorophyll mutants and other macromutants were scored in M_2 . The mean, variance, coefficient of variation and heritability estimates were worked out on 50 randomly selected M_2 plants in each of the treatments. The genetic variance was arrived at by deducting the variance of control plants from the total variance of the treated population and from this heritability estimates were calculated.

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RESULTS

M₁ generation: There was a linear relationship with regard to increase in dose and reduction in survival and reduction in mean height of the plants (Table I). Fifty per cent reduction in survival was noticed in 140 to 160 kR of X-rays and there was no survival of plants in doses above 100 kR of gamma rays. With regard to mean number of clusters, there was a slight stimulatory effect in 100 kR of X-rays, but in the case of gamma ray treatments no such stimulatory effect was seen and on the contrary, a reduction to an extent of 50 per cent was seen in 100 kR dose. The mean number of pods also showed an increase in all the treatments of X-rays except at 140 and 160 kR. But in gamma ray treatments there was a decrease for this trait in both 80 and 100 kR, a decrease of 33.8 per cent being noticed in the latter. The seed fertility was reduced to 50 per cent in X-ray and gamma ray treatments.

M₂ generation: Chlorophyll mutations: The chlorophyll mutations on M₂ plant basis (Table II) ranged from 1.3 to 2.8 per cent in treatments with X-rays and 0.3 to 2.2 per cent in case of gamma rays. The maximum occurrence was noticed in 80 kR dose of both the mutagens.

Macromutations: These could be classified as plant type mutants, leaf mutants, branchless mutants and grain colour mutants and their percentage of occurrence is given in Table (III). Under plant types, there were early and dwarf ones. Maximum percentage of early types was noticed in 80 kR of X-rays. The dwarf mutants were noticed in greater frequency in 80 kR of X-rays and 100 kR of gamma rays. Plants with lobed leaflets, multifoliate (more than three leaflets), and unifoliate ones (only one leaf) and those with crinkled leaves comprise the leaf mutants. These leaf mutants occurred in populations from X-irradiated seeds. The

TABLE I. Observation on M₁ generation following X- and Gamma irradiation.

| Mutagen and dose | Survival per cent (transformed) | Plant height (cm) | Number of clusters per plant | Number of pods per plant | Seed fertility per cent (transformed) |
|------------------|---------------------------------|-------------------|------------------------------|--------------------------|---------------------------------------|
| Control | 90.0 | 73.6 | 29.4 | 76.9 | 90.0 |
| X-rays | | | | | |
| 80 kR | 78.7 | 60.2 | 27.2 | 87.6 | 45.2 |
| 100 kR | 57.2 | 68.8 | 30.4 | 109.0 | 44.6 |
| 120 kR | 52.0 | 67.0 | 28.0 | 83.0 | 43.2 |
| 140 kR | 37.5 | 59.2 | 24.7 | 67.7 | 39.0 |
| 160 kR | 24.0 | 58.4 | 27.0 | 71.5 | 39.7 |
| Gamma rays | | | | | |
| 80 kR | 49.6 | 64.2 | 26.0 | 84.3 | 45.9 |
| 100 kR | 48.5 | 48.8 | 15.7 | 50.9 | 44.6 |
| CD. (P=0.05) | 10.7 | 9.5 | 8.3 | 33.0 | 11.0 |

TABLE II. Frequency of chlorophyll mutants in the M_2 .

| Treatment | No. of M_1 families studied | No. of M_1 families segregating | No. of M_2 plants scored | No. of chlorophyll mutants | Frequency of mutation for 100 M_2 plants |
|-------------------|-------------------------------|-----------------------------------|----------------------------|----------------------------|--|
| Control | 30 | — | 1542 | — | — |
| X-rays | | | | | |
| 80 kR | 39 | 15 | 1626 | 45 | 2.8 |
| 100 kR | 22 | 9 | 854 | 24 | 2.8 |
| 120 kR | 16 | 4 | 585 | 8 | 1.4 |
| 140 kR | 9 | 1 | 253 | 4 | 1.6 |
| 160 kR | 3 | 3 | 543 | 7 | 1.3 |
| Gamma rays | | | | | |
| 80 kR | 15 | 2 | 846 | 19 | 2.2 |
| 100 kR | 12 | 1 | 378 | 1 | 0.3 |

mutants with lobed leaves were observed in progenies receiving 80 and 120 kR doses. The multifoliate mutants were observed in 80 kR dose and the unifoliate mutants in 80 and 140 kR doses. The unifoliate mutants were completely sterile. In these, the ped-

uncle is elongated and the buds formed in them do not contain either stamens or pistil and dropped away. All the other leaf mutants were fertile. The branchless mutants were noticed in all the doses of X-rays except at 160 kR. The grain colour mutants were noticed in gamma ray treatment (80 kR) only. The grains in them turned yellow from green. These mutants could be distinguished easily from a distance as the leaves, stem, petiole and pods in them were yellowish green in appearance.

Micromutations: The mean, variance, C. V., heritability estimates and genetic advance as percentage of mean were worked out for different treatments (Table IV) for plant height, number of branches, number of clusters and number of pods.

(a) X-rays: The mean plant height showed a decrease in different treatments compared to control, the maximum reduction being at 160 kR

TABLE III. Characteristics of the Macromutants observed in the M_2 generation

| Treatment | Family size | Early | | Dwarf | | Lobed | | Multifoliate | | Unifoliate | | Crinkled | | Branchless | | Yellow grain | |
|-------------------|-------------|-------|-----|-------|-----|-------|-----|--------------|-----|------------|-----|----------|-----|------------|-----|--------------|-----|
| | | F | % | F | % | F | % | F | % | F | % | F | % | F | % | F | % |
| Control | 1542 | — | — | — | — | — | — | — | — | — | — | — | — | — | — | — | — |
| X-rays | | | | | | | | | | | | | | | | | |
| 80 kR | 1177 | 62 | 5.3 | 76 | 6.5 | 2 | 0.2 | 53 | 4.5 | 5 | 0.4 | 7 | 0.6 | 13 | 1.1 | — | — |
| 100 kR | 578 | 15 | 2.6 | 13 | 2.2 | — | — | — | — | — | — | — | — | 3 | 0.5 | — | — |
| 120 kR | 336 | — | — | 13 | 3.8 | 1 | 0.3 | — | — | — | — | — | — | 2 | 0.6 | — | — |
| 140 kR | 204 | 2 | 1.0 | 2 | 1.0 | — | — | — | — | — | — | — | — | — | — | — | — |
| 160 kR | 243 | 5 | 2.0 | 9 | 3.6 | — | — | — | — | — | — | — | — | — | — | — | — |
| Gamma rays | | | | | | | | | | | | | | | | | |
| 80 kR | 295 | — | — | 7 | 2.4 | — | — | — | — | — | — | — | — | — | — | 6 | 2.0 |
| 100 kR | 86 | — | — | 4 | 4.6 | — | — | — | — | — | — | — | — | — | — | — | — |

F = Frequency

TABLE IV. Mean, Variance, Coefficient of Variability, heritability (%) and genetic advance as percentage of mean of various quantitative characters in the M₂ generation

| Mutagen and dose | Mean | Variance | C. V. | h ² % | G. A. as % of mean |
|------------------------------|---------------|----------|--------|------------------|--------------------|
| PLANT HEIGHT (CM) | | | | | |
| Control | 78.1 ± 1.782 | 158.7 | 16.13 | — | — |
| X-rays | | | | | |
| 80 kR | 53.6 ± 5.710 | 367.1 | 35.75 | 56.76 | 41.80 |
| 100 kR | 55.2 ± 2.211 | 244.4 | 28.32 | 35.06 | 20.46 |
| 120 kR | 61.2 ± 2.048 | 209.8 | 23.67 | 24.36 | 11.88 |
| 140 kR | 48.2 ± 1.622 | 131.6 | 23.80 | — | — |
| 160 kR | 46.0 ± 2.090 | 218.4 | 32.13 | 27.33 | 18.09 |
| Gamma ray | | | | | |
| 80 kR | 62.3 ± 2.261 | 255.7 | 25.57 | 45.39 | 24.00 |
| 100 kR | 52.0 ± 2.067 | 213.7 | 28.11 | 25.74 | 14.91 |
| NUMBER OF BRANCHES | | | | | |
| Control | 4.6 ± 0.173 | 1.5 | 26.62 | — | — |
| X-rays | | | | | |
| 80 kR | 4.5 ± 0.307 | 4.7 | 48.18 | 68.09 | 65.58 |
| 100 kR | 3.6 ± 0.290 | 4.2 | 56.93 | 64.29 | 75.39 |
| 120 kR | 2.6 ± 0.219 | 2.4 | 59.58 | 37.50 | 46.08 |
| 140 kR | 3.3 ± 0.257 | 3.3 | 55.05 | 54.55 | 61.85 |
| 160 kR | 3.0 ± 0.205 | 2.1 | 48.30 | 28.57 | 28.43 |
| Gamma rays | | | | | |
| 80 kR | 3.6 ± 0.184 | 1.7 | 36.22 | 11.76 | 8.78 |
| 100 kR | 3.5 ± 0.228 | 2.6 | 46.07 | 42.31 | 40.14 |
| NUMBER OF CLUSTERS PER PLANT | | | | | |
| Control | 28.4 ± 1.143 | 65.3 | 28.45 | — | — |
| X-rays | | | | | |
| 80 kR | 20.1 ± 2.074 | 215.0 | 72.95 | 69.63 | 104.64 |
| 100 kR | 12.3 ± 1.446 | 104.6 | 83.15 | 37.57 | 64.35 |
| 120 kR | 14.7 ± 1.521 | 115.7 | 73.17 | 43.56 | 65.66 |
| 140 kR | 14.4 ± 1.388 | 96.3 | 68.15 | 32.19 | 45.19 |
| 160 kR | 15.1 ± 1.921 | 184.5 | 89.95 | 64.60 | 119.71 |
| Gamma rays | | | | | |
| 80 kR | 19.8 ± 1.269 | 254.3 | 80.57 | 74.34 | 123.39 |
| 100 kR | 19.9 ± 1.165 | 183.7 | 68.11 | 64.45 | 90.43 |
| NUMBER OF PODS PER PLANT | | | | | |
| Control | 105.6 ± 5.698 | 1623.5 | 38.16 | — | — |
| X-rays | | | | | |
| 80 kR | 74.6 ± 8.869 | 3933.6 | 84.07 | 58.73 | 101.71 |
| 100 kR | 56.5 ± 8.759 | 3835.7 | 109.62 | 57.67 | 130.22 |
| 120 kR | 63.1 ± 7.358 | 2706.7 | 82.45 | 40.02 | 67.97 |
| 140 kR | 44.5 ± 4.306 | 927.2 | 68.43 | — | — |
| 160 kR | 54.7 ± 8.605 | 3702.5 | 111.24 | 56.15 | 128.67 |
| Gamma rays | | | | | |
| 80 kR | 62.8 ± 10.848 | 5883.6 | 122.14 | 72.41 | 182.19 |
| 100 kR | 86.1 ± 10.642 | 5662.5 | 87.40 | 71.30 | 82.82 |

The variance showed an increase in all treatments except in 140 kR and the maximum increase was in 80 kR. The C. V. ranged from 23.67 to 35.75 per cent and the maximum heritability and genetic advance was noticed in 80 kR. With regard to mean number of branches also there was a reduction in treatments compared to control. The variance for this trait was maximum in 80 kR followed by 100 kR. The heritability was also maximum in 80 kR. In respect of number of clusters and number of pods also, a similar trend was noticed as compared to control. The maximum variance and heritability were seen in 80 kR.

(b) **Gamma rays:** The mean plant height showed a decrease in treatments compared to control, the maximum reduction being at 100 kR. The variance showed an increase in treatments. The maximum variance, heritability and genetic advance were observed in 80 kR. The mean number of branches also showed a decrease in treatments and the values for C. V., heritability and genetic advance were the greatest in 100 kR. In respect of mean number of clusters and pods also there was a decrease in treatments. The maximum variance, heritability and genetic advance for those two traits were noticed in 80 kR.

DISCUSSION

The study of M_1 generation receiving the same doses of X-rays and gamma rays has indicated that a 50 per cent reduction in survival was between 140 to 160 kR of X-rays and

around 100 kR of gamma rays. The treatment with gamma rays was found to be more drastic with regard to reduction in survival and other characters as compared to X-rays. Stimulation effects were observed in 100, 80 and 120 kR of X-rays and 80 kR of gamma rays for number of pods per plant.

The chlorophyll mutation frequency which is taken as a reliable index to determine the mutagenic efficiency was found to be maximum in 80 kR of both X-rays and gamma rays. The spectrum of macromutants was found to be wider in 80 kR of gamma rays. Of the leaf mutants reported in the present study, the occurrence of unifoliate and multifoliate mutants have already been observed in this crop by Santos (1969). Rajput (1971) has also reported the occurrence of morphological abnormalities such as stunted growth with formation of rough leathery leaves, plants with hexafoliate leaves and formation of leaf clusters in the place of flowers in green gram following gamma irradiation.

The mean plant height showed a reduction in higher doses of X-rays and gamma rays as compared to the control. High variance and maximum heritability and genetic advance for plant height was noticed in 80 kR dose of both X-rays and gamma rays and the variance increased in all treatments except of 140 kR of X-rays, compared to the control. The same trend was observed in mean and variance for number of branches for X-ray treatments. In gamma ray treatments,

however, maximum increase in variance was noticed in 100 kR dose. With regard to number of clusters and number of pods also maximum values for variance, C.V. and heritability were noticed in 80 kR dose of both X-rays and gamma rays. It has been reported by Brock (1965) that in species previously subjected to breeding and selection, random mutation results in the increase of variance and shift in the mean away from the direction of selection for quantitative traits. The present findings also confirm this view. The increase in variance for all the traits in M_2 generation is due to the presence of some plants exceeding the expression of parental mean indicating good scope for selection in respect of economic traits. In this context, application of a dose of 80 kR of both the mutagens is recommended for a programme of improvement of the Co. 1 strain of greengram.

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