Indian J. Genet., **67**(1): 41-46 (2007)

Induced mutagenesis in blackgram [*Vigna mungo* (L.) Hepper]

Arvind Kumar[1], M. N. Mishra and M. C. Kharkwal[2]

Department of Plant Breeding and Genetics, R. B. S. College, Bichpuri, Agra 283 105
[2]Division of Genetics, Indian Agricultural Research Institute, New Delhi 110 012

(Received: July 2006; Revised: December 2006; Accepted: December 2006)

Abstract

A wide range of chlorophyll and viable morphological mutations affecting almost all the parts of the plant and seed characteristics were isolated in M_2 generation of blackgram [*Vigna mungo* (L.) Hepper] variety Type 9 treated with gamma rays doses 50 to 500 Gy. The comparative study of frequency and spectrum of chlorophyll and viable mutations and the mutagenic effectiveness and efficiency of gamma rays on blackgram included two treatment conditions: 1. Irradiation of dry seeds with 50 to 500 Gy with an interval of 50 Gy, 2. Soaking of seeds in water for 6 hours before irradiation with 50 to 500 Gy again with the same interval of 50 Gy. Mutation rate indicated that 400 Gy dry treatment was most potent in inducing chlorophyll and viable mutations. The frequency and spectrum of the induced chlorophyll mutations was in the order of - Chimera, chlorina, albina and xantha. Higher doses of gamma rays treatment 500, 450 and 400 Gy showed high lethality under both the treatment conditions - dry and soaked. The treatment of 400 Gy gamma rays under dry condition showed highest mutagenic efficiency in inducing mutations in blackgram. However, lower doses of gamma rays i.e., 250 Gy under dry condition and 150 Gy under soaked condition were also quite effective and efficient.

Key words: Blackgram, induced mutations, chlorophyll and viable mutations, frequency and spectrum, mutagenic effectiveness and efficiency

Introduction

Blackgram [*Vigna mungo* (L.) Hepper] is one of the important, protein rich food legume crops of India. Being a drought and heat tolerant crop, it is normally grown during the hot and humid *kharif* season. The crop however, has very limited genetic variability and suffers from severe susceptibility to several diseases and pests which are directly related to its plant type which is not efficient for yield as well as disease resistance. Induced mutagenesis has been successfully used to generate wider variability, particularly for isolating mutants with desirable characters of economic importance such as superior plant types, synchronous maturity, disease resistance, larger seed size and desirable seed colour etc. [1]. Although studies on induced mutagenesis in blackgram have been undertaken in the past [2-4], limited reports are available on induced viable morphological mutations in blackgram.

A systematic study of induced variability for chlorophyll and viable morphological mutations in M_2 generation is the most dependable index in order to induce variability and utilize useful mutations for efficient plant breeding. Keeping in view the limited genetic variability available in the germplasm, the present investigation was undertaken to understand the nature of induced variability and response of blackgram to different doses and treatment conditions of irradiation with gamma rays. The study provides the relative sensitivity of blackgram variety Type 9 to effectiveness and efficiency of various doses of gamma rays, which are pre-requisite for induction and utilization of mutations. Effectiveness means the rate of mutations as related to dose, while efficiency usually refers to the mutation rate in relation to damage. The paper also reports frequency and spectrum of various types of viable morphological mutations (macro-mutations) identified and isolated in M_2 generation.

Materials and methods

The experimental material for the present study comprised of one prominent blackgram variety Type 9. Five hundred dry seeds with a moisture content of 10-12 % were treated with gamma rays doses of 50 to 500 Gy per dose. Equal number of seeds per dose were treated with gamma rays doses of 50 to 500 Gy after pre-soaking in distilled water for 6 hrs at room temperature. Equal number of dry and pre-soaked seeds was used as control. The M_1 crop was raised in the field in *kharif* season and all the surviving plants were selfed and harvested individually to raise the M_2 generation population along with controls. Necessary cultural practices were adopted to raise a healthy crop. Observations for recording chlorophyll mutations were noted critically right from emergence till the age of three weeks after germination when the seedlings were at two leaf stage in the field as per identification and classification recommended by Gustaffson [5]. Viable macro-mutations were scored throughout the life period

[1]Present address: Nodai Seeds India Pvt. Ltd., RB 404, Anand Vihar, Toli Chowki, Hyderabad 500 008

of the plants in the seed beds in the field as per identification and classification procedure recommended by Blixt [6]. The mutations affecting gross morphological changes in branching, stem structure, growth habit, leaf, flower, pod and seed size and maturity etc. were scored as viable macromutations. Both mutagenic effectiveness and efficiency were scored as per the following procedure recommended by Konzak *et al.* [7] and Nilan [8].

Mutagenic effectiveness = Mf/Gy

Mutagenic efficiency = Mf/L where, Mf = Percentage of families segregating for chlorophyll mutations; Gy = Grey of gamma rays; L = Percentage of lethality in M_1

Results and discussion

The data on chlorophyll and viable mutation rate observed under various doses of gamma rays under different treatment conditions are presented in Table 1. indicates that maximum mutated progenies (12.4%) were observed under 400 Gy dry treatment followed by 250 Gy dry treatment (7.5%). Under 6 hr water soaked treatment conditions maximum mutaion rate (6.5%) was observed under 150 Gy of gamma rays followed by maximum progenies (4%) mutated under 300 Gy dose. These results are in conformity with the findings of other workers in blackgram [2-4, 9], mungbean [9, 10] and lentil [11]. There were differences in the mutation spectrum between mutagenic doses and treatment conditions - dry and 6 hours pre-irradiation water soaked treatment conditions applied. However, many similarities were also noticed between the treatment conditions in respect of the spectrum and frequency of a particular type of mutation. The frequencies of viable mutations were computed as in the case of chlorophyll mutations [12, 13]. The mutations induced and isolated in the present study are described below under five sub-categories.

Table 1. Mutation rate of various doses of gamma-rays in M_2 generation of blackgram

Mutagen & dose	Population in M_2	Chlorophyll mutants				Viable mutants				Total mutants			
		Dry		Soaked		Dry		Soaked		Dry		Soaked	
		No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
Control	300	0.0		0.0		0.0		0.0		0.0	0.0	0.0	0.0
γ-rays 50 Gy	300	4	1.33	8	2.66	8	2.66	5	1.67	12	4.00	13	4.33
γ-rays 100 Gy	310	3	0.97	8	2.58	4	1.29	4	1.29	7	2.26	12	3.87
γ-rays 150 Gy	275	9	3.27	10	3.64	4	1.45	8	2.91	13	4.07	18	6.54
γ-rays 200 Gy	300	4	1.33	8	2.66	3	1.00	5	1.66	7	2.33	13	4.33
γ-rays 250 Gy	290	10	3.45	7	2.41	12	4.14	1	0.34	22	7.59	8	2.76
γ-rays 300 Gy	300	11	3.66	4	1.33	2	0.67	8	2.66	13	4.33	12	4.00
γ-rays 350 Gy	275	9	3.27	4	1.45	2	0.73	3	1.09	11	4.00	7	2.54
γ-rays 400 Gy	250	14	5.60	2	0.80	17	6.80	7	2.80	31	12.40	9	3.60
γ-rays 450 Gy	235	3	1.28	7	2.98	3	1.28	3	1.28	6	2.55	10	4.25
γ-rays 500 Gy	230	11	4.78	10	4.35	2	0.87	1	0.43	13	5.65	11	4.78
Total	2765	**78**	**2.82**	**68**	**2.46**	**57**	**2.06**	**45**	**1.63**	**135**	**4.88**	**113**	**4.09**

Higher chlorophyll mutation rate was observed with gamma rays doses of 400, 500 and 300 Gy under dry treatment and 500, 150 and 450 Gy under soaked treatments. Frequency of chlorophyll mutations observed in the present study was very low and also fluctuating under different treatments. This finding is in conformity with the reports of earlier workers [2, 9]. For viable mutations the frequency increased randomly with the increasing doses of gamma-rays. Under soaked treatment a lower dose of 150 Gy and 400 Gy under both the treatment conditions indicated highest mutation rate. The frequency increased randomly with the increase in doses of gamma-rays. Earlier workers [2, 9] also found similar results. On the basis of total mutation rate 400 Gy followed by 50 Gy under dry treatment and 150 Gy followed by 500 Gy treatment under soaked condition showed higher mutation rate. The data

I. Mutations affecting chlorophyll apparatus:

Chlorophyll mutations: The frequency and spectrum of various types of chlorophyll mutations i.e., albina, xantha, chlorina and chimera observed in the present study (Table 2) indicated that dry treatments showed higher percentage of chlorophyll mutations than the pre-soaked treatments. The chlorina mutant plants were very light green or pale green which persisted for throughout the growth period. The albina mutants, which were white and without chlorophyll in leaves, branches or stem did not survive after few days. The highest frequency of chlorophyll mutations recorded was for chimera type followed by chlorina type under dry as well as soaked treatment condition. High frequency and spectrum of chlorophyll mutations was recorded under higher doses of gamma rays i.e., 400 and 500 Gy treatment under dry condition and 500 Gy and 450 Gy under soaked

Table 2. Spectrum and frequency of induced chlorophyll mutations in M_2 generation of blackgram

Mutagen & dose	Treatment condition	Number and % of chlorophyll mutants scored										No. of M_2 plants	Mutants/ 1000 M_2 plants
		Albina		Chlorina		Xantha		Chimera		Total			
	Dry/soaked	No.	%	No.	%	No.	%	No.	%	No.	%		
Control	Dry	0		0		0		0		0		300	
γ-rays 50 Gy	Dry	1	0.333	2	0.666	1	0.333	0		4	1.333	300	13.333
γ-rays 100 Gy	Dry	2	0.645	1	0.322	0		0		3	0.968	310	9.677
γ-rays 150 Gy	Dry	0		3	1.090	2	0.727	4	1.454	9	3.273	275	32.727
γ-rays 200 Gy	Dry	0		0		1	0.333	3	1.000	4	1.333	300	13.333
γ-rays 250 Gy	Dry	3	1.034	2	0.689	3	1.034	2	0.689	10	3.448	290	34.483
γ-rays 300 Gy	Dry	1	0.333	3	1.000	3	1.000	4	1.333	11	3.666	300	36.666
γ-rays 350 Gy	Dry	4	1.454	0		-		5	1.818	9	3.273	275	32.727
γ-rays 400 Gy	Dry	0		6	2.400	3	1.200	5	2.000	14	5.600	250	56.000
γ-rays 450 Gy	Dry	1	0.425	0		2	0.851	0		3	1.276	235	12.766
γ-rays 500 Gy	Dry	5	2.174	1	0.435	2	0.870	3	1.304	11	4.783	230	47.826
Total	Dry	**17**	**0.615**	**18**	**0.651**	**17**	**0.615**	**26**	**0.940**	**78**	**2.821**	**2765**	**28.210**
Control	Soaked	0		0		0		0		0		300	
γ-rays 50 Gy	Soaked	1	0.333	7	2.333	0		0		8	2.666	300	26.666
γ-rays 100 Gy	Soaked	2	0.645	2	0.645	1	0.322	3	0.968	8	2.581	310	25.806
γ-rays 150 Gy	Soaked	0		4	1.454	2	0.727	4	1.454	10	3.636	275	36.364
γ-rays 200 Gy	Soaked	2	0.666	1	0.333	2	0.666	3	1.000	8	2.666	300	26.666
γ-rays 250 Gy	Soaked	4	1.379	2	0.690	1	0.345	0		7	2.414	290	24.138
γ-rays 300 Gy	Soaked	2	0.666	0		1	0.333	1	0.333	4	1.333	300	13.333
γ-rays 350 Gy	Soaked	0		2	0.727	0		2	0.727	4	1.454	275	14.545
γ-rays 400 Gy	Soaked	1	0.400	0		1	0.400	0		2	0.800	250	8.000
γ-rays 450 Gy	Soaked	3	1.276	0		4	1.702	0		7	2.979	235	29.787
γ-rays 500 Gy	Soaked	2	0.869	3	1.304	3	1.304	2	0.869	10	4.348	230	43.478
Total	Soaked	**17**	**0.615**	**21**	**0.759**	**15**	**0.542**	**15**	**0.522**	**68**	**2.459**	**2765**	**24.593**
Grand Total		34	**0.615**	**39**	**0.705**	**32**	**0.579**	**41**	**0.741**	**146**	**2.640**	**5530**	**26.401**

treatment and the lowest response for chlorophyll mutations was recorded under 100 Gy.

II. Mutations affecting leaf morphology: The frequency of various leaf type mutations observed under various treatments (Table 3) showed that lower doses of gamma rays were more effective in inducing more leaf type mutations. Highest frequency of leaf type mutations was recorded under 250 Gy dry treatment and 100 Gy under soaked treatment. Among the various types of viable mutations induced, leaf type mutants showed highest percentage of 0.54 in M_2 generation. Some of the distinct leaf type mutants isolated (Fig. 1) are described below:

1. *Small leaf*: Observed under 250 Gy dose of gamma rays under dry treatment, this mutant had very small leaflets of light green colour and was associated with dwarfness.

2. *Long leaf*: The mutants observed in 400 Gy of gamma rays under soaked treatment and 500 Gy dry treatment, were having arrow like very long leaflets tapering towards the tip (Fig. 1-b).

3. *Long apex leaf*: Isolated from 400 Gy of gamma rays under soaked treatment this mutant had broad and long apex leaves (Fig. 1-c).

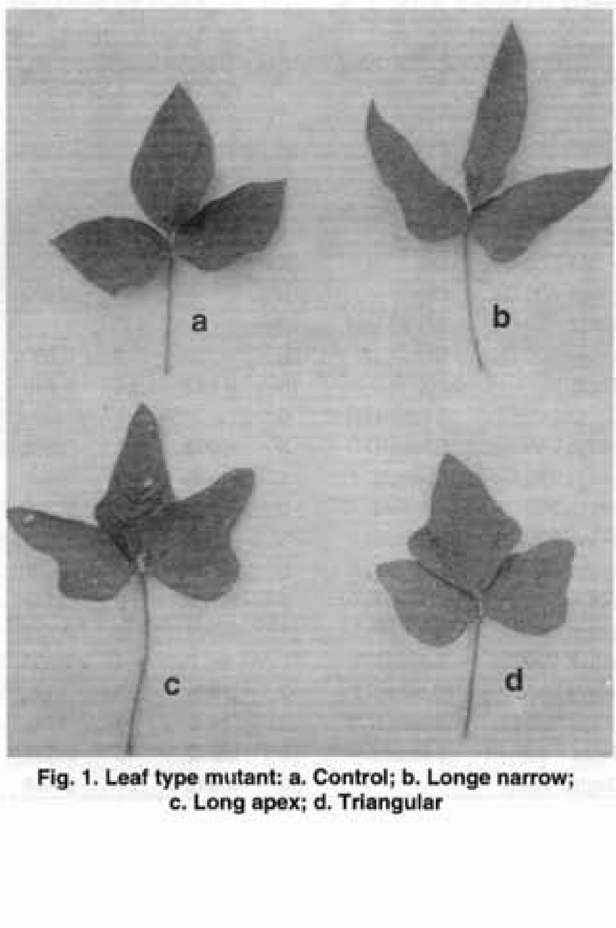

Fig. 1. Leaf type mutant: a. Control; b. Longe narrow; c. Long apex; d. Triangular

4. *Triangular leaf*: Observed under 150 Gy of gamma rays under soaked treatment this mutant had wide based tri-lobed leaflets (Fig. 1-d).

5. *Broad leaf*: The leaves of this mutant isolated from under 250 Gy dose of gamma rays under soaked treatment were thicker and succulent with broad lamina.

III. Mutations affecting plant type: The frequency of various plant type mutations observed under various treatments (Table 3) showed that higher doses of gamma rays were more effective in inducing more plant type mutations. Highest frequency of plant type mutations was recorded under 400 Gy dry treatment both under dry as well as soaked treatments. Some of the important and distinct plant type mutants isolated are described below:

1. *Tall mutant*: Isolated under 250 Gy dry treatment, tall mutants were vigourous with larger leaves, few branches and attained height ranging from 150 to 175 cm compared to 60 cm of the control plant.

2. *Dwarf mutant*: Isolated from 450 Gy dose of gamma rays of soaked condition has a height of 15 cm only.

3. *Erect mutant*: Erect and tall mutant with shy branching were isolated from 50 Gy and 250 Gy doses of gamma rays under dry treatments.

4. *Unbranched mutant*: Observed under 450 Gy treatment of gamma rays under dry treatment.

5. *Single pod mutant*: Isolated from 250 Gy dose of soaked treatment this mutant with spreading habit had only one pod on each node (Fig. 2).

Fig. 2. Plant type mutant — Single poded and spreading

Table 3. Spectrum and frequency of induced viable mutations in M_2 generation of blackgram

Mutagen & dose	Treatment condition	Number and % of viable mutants scored										No. of M_2 plants	Mutants/ 1000 M_2 plants
		Leaf type		Plant type		Pod type		Seed type		Total			
	Dry/soaked	No.	%	No.	%	No.	%	No.	%	No.	%		
Control	Dry	0		0		0		0		0		300	
γ-rays 50 Gy	Dry	3	1.000	2	0.666	1	0.333	2	0.666	8	2.666	300	26.666
γ-rays 100 Gy	Dry	1	0.322	2	0.645	0		1	0.322	4	1.290	310	12.903
γ-rays 150 Gy	Dry	2	0.727	0		2	0.727	0		4	1.454	275	14.545
γ-rays 200 Gy	Dry	0		0		3	1.000	0		3	1.000	300	10.000
γ-rays 250 Gy	Dry	6	2.069	3	1.034	0		3	1.034	12	4.138	290	41.379
γ-rays 300 Gy	Dry	1	0.333	0		1	0.333	0		2	0.666	300	6.666
γ-rays 350 Gy	Dry	2	0.727	0		0		0		2	0.727	275	7.273
γ-rays 400 Gy	Dry	0		9	3.600	8	3.200	0		17	6.800	250	68.000
γ-rays 450 Gy	Dry	0		0		0		3	1.276	3	1.276	235	12.766
γ-rays 500 Gy	Dry	0		2	0.689	0		0		2	0.869	230	8.696
Total	Dry	**15**	**0.542**	**18**	**0.650**	**15**	**0.542**	**9**	**0.325**	**57**	**2.061**	**2765**	**20.615**
Control	Soaked	0		0		0		0		0		300	
γ-rays 50 Gy	Soaked	3	1.000	1	0.333	0		1	0.333	5	1.666	300	16.666
γ-rays 100 Gy	Soaked	4	1.290	0		0		2	0.645	4	1.290	310	12.903
γ-rays 150 Gy	Soaked	0		2	0.727	4	1.454	0		8	2.909	275	29.091
γ-rays 200 Gy	Soaked	3	1.000	0		0		2	0.666	5	1.666	300	16.666
γ-rays 250 Gy	Soaked	0		0		0		1	0.345	1	0.345	290	3.448
γ-rays 300 Gy	Soaked	2	0.666	0		3	1.000	3	1.000	8	2.666	300	26.666
γ-rays 350 Gy	Soaked	1	0.364	2	0.727	0		0		3	1.091	275	10.909
γ-rays 400 Gy	Soaked	0		4	1.600	0		3	1.200	7	2.800	250	28.000
γ-rays 450 Gy	Soaked	2	0.851	0		1	0.425	0		3	1.277	235	12.766
γ-rays 500 Gy	Soaked	0		0		1	0.435	0		1	0.435	230	4.348
Total	Soaked	**15**	**0.542**	**9**	**0.325**	**9**	**0.325**	**12**	**0.434**	**45**	**1.627**	**2765**	**16.275**
Grand Total		30	**0.542**	**27**	**0.488**	**24**	**0.434**	**21**	**0.380**	**102**	**1.844**	**5530**	**18.445**

6. *Sterile mutant*: Observed in 200 Gy dose of gamma rays under soaked treatment, the mutant having full green leaves had no flowers or pods till maturity.

IV. Mutations affecting pod characters:

1. *Bunchy pod mutant*: This mutant isolated from 200 Gy dose of soaked treatment had a bunch of nine pods with normal shape and size on every branch (Fig. 3).

Fig. 3. Bunchy pod type mutant

2. *Smooth pod mutant*: Recovered under 400 Gy dose gamma rays under dry treatment, this mutant did not have any hair on its dark black coloured pods (Fig. 4 & 5).

V. Mutations affecting seed colour, shape and size:

1. *Brown seeded mutant*: Observed under 450 Gy dose of gamma rays of soaked treatment, this mutant had brown seeds with normal shape and size.

2. *Dark black seeded mutant*: Isolated from 400 Gy dose of gamma rays, the seeds of this mutant were dark black.

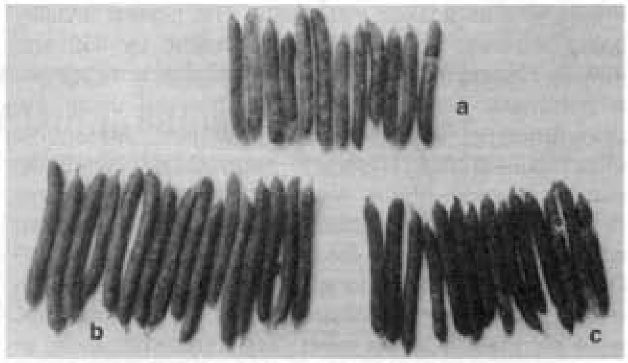

Fig. 4. Pod type mutants: (a) Small hairy pods; (b) Large pods with normal hairs; (c) Small pods without hairs

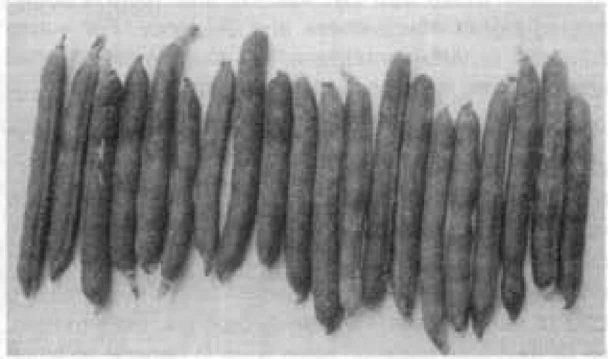

Fig. 5. Bold pods without hairs

Mutagenic effectiveness and mutagenic efficiency: Mutagenic effectiveness usually means the rate of mutation as related to dose. Mutagenic efficiency refers to the mutation rate in relation to damage Table 4. The results obtained for mutagenic effectiveness (Mf/Gy) and efficiency (Mf/L) of various doses of gamma rays on blackgram (Table 4) showed differential behaviour depending upon doses and treatment conditions. Increasing trend in lethality percentage was observed with increasing doses of gamma rays both in case of

Table 4. Mutagenic effectiveness and efficiency of gamma-rays in M_2 generation of blackgram

Treatment & dose	Population in M_2	Lethality		Mutagenic effectiveness and efficiency			
		Dry	Soaked	Dry		Soaked	
				Effectiveness	Efficiency	Effectiveness	Efficiency
Control	300	0.00	0.00	0.00	0.00	0.00	0.00
γ-rays 50 Gy	300	7.50	6.24	0.72	0.84	1.20	1.02
γ-rays 100 Gy	310	7.96	6.57	0.70	0.88	1.20	1.02
γ-rays 150 Gy	275	8.12	9.23	0.86	1.60	1.20	1.95
γ-rays 200 Gy	300	8.17	11.34	0.35	0.85	0.65	1.46
γ-rays 250 Gy	290	10.25	12.26	0.88	2.14	0.32	0.65
γ-rays 300 Gy	300	8.26	12.76	0.43	1.57	0.40	0.53
γ-rays 350 Gy	275	11.73	13.24	0.31	1.93	0.20	0.55
γ-rays 400 Gy	250	11.87	16.70	0.77	2.61	0.22	0.54
γ-rays 450 Gy	235	12.50	17.29	0.13	0.48	0.22	0.58
γ-rays 500 Gy	230	14.43	19.24	0.26	0.96	0.22	0.57
Total	2765						

dry as well as soaked treatments. The highest lethality being observed in 500 Gy was followed by 450 and 400 Gy respectively. A noticeable reduction in mutagenic effectiveness and efficiency was observed under the pre-irradiation water soaked treatment. Mutagenic effectiveness and efficiency showed an increasing pattern in case of the lower doses of gamma rays (from 50 to 250 Gy). However, an inverse relationship was observed in case of higher doses (300 to 500 Gy). Similar observations of general decrease in effectiveness with increasing doses of gamma rays were reported in foxtail millet [14]. The lower doses of gamma rays i.e., 50, 100 and 150 Gy were found more effective under soaked treatments than dry treatments where 250 Gy, 400 Gy and 150 Gy doses showed higher effectiveness and efficiency. For macro mutations or viable mutations, dry treatments exhibited more mutants than soaked treatments.

The usefulness of a mutagen depends both on its mutagenic effectiveness and efficiency, efficient mutagenesis being the production of maximum desirable changes accompanied by the least possible undesirable changes. Although both the mutagenic effectiveness and efficiency generally decreased with the increasing dose of gamma rays with few exceptions, nevertheless, the highest total mutation rates were in general obtained with higher doses of gamma rays (e.g. 400 Gy). It would thus be seen that higher mutagenic effectiveness and efficiency does not reflect the *per se* mutation frequency and they can not be used as an index for maximization of mutation rates [15, 16]. In effect, these factors modify the effectiveness (mutation per unit dose) and efficiency (proportion of mutations as against associated undesirable biological effects such as gross chromosomal aberrations, lethality and sterility) of the mutagen in question [7, 8].

The results of the present study in terms of induction of useful variability particularly for a number of economically important characters which can contribute in development of high yielding genotypes having improved plant type clearly indicate to the vital role of mutation breeding in crop improvement. Systematic and serious efforts in persuing the methodology of mutation breeding has already been successfully demonstrated in development and release of large number of improved high yielding varieties in several crops [1]. TAU-1 and TU 94-2 in blackgram [1, 3] and Pusa 408, Pusa 413, Pusa 417 and Pusa 547 (BGM 547) in chickpea [1, 17] are some of the recent examples of high yielding mutant varieties of pulses released in India.

References

1. **Kharkwal M. C., Pandey R. N. and Pawar S. E.** 2004. Mutation Breeding for Crop Improvement, 601-645. *In*: Plant Breeding — Mendelian to Molecular Approaches (H. K. Jain and M. C. Kharkwal, eds). Narosa Publishing House P. Ltd. New Delhi.
2. **Singh V. P. and Lal J. P.** 1998. Mutagenic effects of gamma rays and EMS on frequency of chlorophyll and macromutations in urdbean [*Vigna mungo* (L.) Hepper]. Indian J. Genet., **59**: 203-210.
3. **Pawar S. E., Manjaya J. G. Souframanien J. and Bhatkar S. M.** 2000. Genetic improvement of blackgram using induced mutations. *In*: DAE/BRNS Symposium on the use of nuclear and molecular techniques in crop improvement. December 6-8, 200. BARC, Mumbai. 170-174.
4. **Sharma S. K. Ritu Sood and Pandey D. P.** 2005. Studies on mutagen sensitivity, effectiveness and efficiency in urdbean [*Vigna mungo* (L.) Hepper]. Indian J. Genet., **65**: 20-22.
5. **Gustaffson A.** 1940. The mutation system of chlorophyll apparatus. Lund Iniv. Arskr., **36**: 1-40.
6. **Blixt S.** 1972. Mutation in *Pisum*. Agri. Hort. Genet., **30**: 1-293.
7. **Konzak C. P., Nilan R. A., Wagner J. and Foster R. J.** 1965. "Efficient chemical mutagenesis", Radiat. Bot., (Suppl.), **5**: 49-70.
8. **Nilan R. A.** 1967. Nature of induced mutations in higher plants. *In*: Induced mutations and their utilization. Proc. Symp. Erwin-Bauer-Gedachtnisvorlegungan IV. Gatersleben, 1966. Akademie-Verlag. Berlin. Pp. 5-20.
9. **Ignacimuthu S. and Babu C. R.** 1993. Induced quantitative variations in pod and seed traits of wild and cultivated mung and urdbean. J. of Nuclear Agriculture and Biology, **22**: 133-137.
10. **Dahiya B. S.** 1973. Improvement of mungbean throough induced mutations. Indian J. Genet., **33**: 460-468.
11. **Solanki I. S. and Sharma B.** 2001. Frequency and spectrum of chlorophyll mutations in macrosperma lentil (*Lens culinaris* Medik.). Indian J. Genet., **61**: 283-286.
12. **Kharkwal M. C.** 1999. Induced mutations in chickpea (*Cicer arietinum* L.) III. Frequency and spectrum of viable mutations. Indian J. Genet., **59**: 451-464.
13. **Kharkwal M. C.** 2000. Induced mutations in chickpea (*Cicer arietinum* L.) IV. Types of macromutations induced. Indian J. Genet., **60**: 305-320.
14. **Gupta P. K. and Yashvir.** 1975. Induced mutations in foxtail millet (*Sataria italica* Beauv.). I. Chlorophyll mutations induced by gamma rays, EMS and DES. TAG., **45**: 242-249.
15. **Prasad M. V. R.** 1972. A comparision of mutagenic effectiveness and efficiency of gamma rays, EMS, NMU and NG. Indian J. Genet., **32**: 360-367.
16. **Kharkwal M. C.** 1998. Induced mutations in chickpea (*Cicer arietinum* L.) I. Comparative effectiveness and efficiency of physical and chemical mutagens. Indian J. Genet., **58**: 159-168.
17. **Kharkwal M. C., Nagar J. P. and Kala Y. K.** 2005. BGM 547 — A high yielding chickpea (*Cicer arietinum* L.) mutant variety for late sown condition in north western plain zone of India. Indian J. Genet., **65**: 229-230.