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Assessment of Gamma rays induced variability in M_2 generation of Sesamum (*Sesamum indicum* L.)

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Abstract
A study was carried out for the assessment of magnitude of variability induced by various doses of gamma irriadiations viz., 300, 350, 400 and 450 Gy in M_2 progenies of Sesamum variety VRI 2 and TMV 7. The evaluation of 102 M_2 progenies grown in RBD for varoius yield attributing characters indicated significant differences between the progenies for plant height, Days to Maturity, number of barnches per plant, Capsule lenth, Number of seed per capsul and single plant yield. Based on Height mean and CV progenies 400 and 450 were found to have desirable character combinations and selection cane be made for desirable plant type for crop improvement programme in sesamum.

Keywords: gamma rays, mutagenic effectiveness, mutagenic efficiency, mutagens

Introduction

Sesame is probably the most ancient oilseed known and used by man. Success in any breeding programme depends on the amount of genetic variability present for the different characters in the population. The genetic variability offered by mutagenic agents is of extreme importance in plant breeding. The variability in quantitative characters increases considerably by treating the biological materials with different mutagenic agents. An estimation of the extent of variability induced in M_2 generation will be of great value to provide useful information for carrying out further selection. Mutation induction is an important complementary method of breeding crop species. Induced mutations can rapidly create variability in quantitatively and qualitatively inherited traits in crops. Mutation breeding not only creates variability in crop species, but also shortens the time taken for the development of cultivars via induced mutation compared to those via hybridizations. The average time elapsed from initial mutation treatment to the release of the mutant cultivars was approximately 9 years, while the time was more than 9 years for cultivar arising from crossing programmes. The utilization of induced mutations for the improvement of crop plants had yielded several mutants which have been used directly as new cultivars Mutation breeding is accomplished by chemical or physical treatments followed by selection for heritable changes of specific genotypes and this method has been used successfully in the genetic improvement. Hence, the objective of the present study was to induce variability in sesamum for important traits and to identify agronomical superior mutants in M_2 population.

Materials and Methods

M_1 seeds of sesamum cultivar VRI 2 and TMV 7 were used to raise M_2 population at Experimental Farm, Department of Plant Breeding and Genetics, Agriculture College and Research Institute, Killikulam. M1 seeds were produced from growing irradiated dried seeds of the sesamum (VRI 2 and TMV 7) at different doses of gamma rays (300 Gy, 350 Gy, 400 Gy and 450 Gy) in gamma chamber containing CO_{60} as a source at Department of Plant Breeding and Genetics, Tamil Nadu Agricultural University, Coimbatore. Seeds of control samples were also sown in M_2 generation. Total of 4864 plants were raised from all four doses of gamma rays. The segregating M_2 population was observed for frequency and spectrum of chlorophyll mutations (from germination to maturity). The spectrum of chlorophyll mutations was classified as per the described method [18] with modifications and also for any type of phenotypic variability. At maturity each of the putative mutant plant was harvested individually to advance for next generation.

Observations recorded and statistical procedure

The M_2 generation was screened for phenotypic variations from germination to harvesting. The frequency of the mutant plants out of the total number of individuals in M_2 generation was calculated. To identify probable mutants following phenotypic observations were recorded: Plant height, Number of primary branches per plant, Length of capsule, Number of capsules per plant, Number of seeds per capsule, Seed yield per plant, Days to maturity and 1000-seed weight. Only selected characters of the putative mutants have been presented in this paper. The recorded observations were statistically analyzed to determine the degree of significance for the variations.

Results

The M_1 population consisting of a total 4864 plants from four different doses of gamma rays 300 Gy, 350 Gy, 400 Gy and 450 Gy. A large spectrum of chlorophyll mutants were observed at seedling emergence (Table 1). Mutants were identified in whole population based on morphological traits and plants exhibiting chlorophyll variegation were observed. Among total 4864 M_1 plants, 54 plants showed different types of chlorophyll mutants were observed. Chlorophyll mutants were observed to the extent of 5.86% in M_2 generation. Among the different doses, the highest percentage (1.59%) of chlorophyll mutants were recorded in 350 Gy followed by 400 Gy (1.47%) and the lowest was recorded in 450 Gy (0.88%). Different types of chlorophyll mutants appeared in the segregating generation. Chlorina type mutants, Xanthaviridis type of chlorophyll mutants (Fig. 1c) appeared in which half of the leaves were devoid of chlorophyll and such leaves became completely white thereafter. Chlorina type mutants (Fig. 1a) were present in all the doses, whereas Xanthaviridis viridi (Fig. 1c) type of mutants also appeared in all doses except 300 Gy. In the further growth yellow leaves and irregulars leaves mutants become normal while the complete plant devoid of chlorophyll mutants were died.

Identification and selection of putative mutants in M_2 under field conditions

The whole surviving M_2 generation were screened for any type of variability and putative mutants were selected from these segregating population. A total of 114 mutants were identified based on different morphological characters (Fig. 2). Plants carrying putative mutation affect Plant height, Number of primary branches per plant, Length of capsule, Number of capsules per plant, Number of seeds per capsule, Seed yield per plant, Days to maturity, 1000-seed weight were identified. The maximum number of mutants were selected from the gamma rays dose of 400 Gy (35) followed by 350 Gy (29) and 300 Gy (19). The identified and selected mutants were grouped into different groups to know the extent of variability created through gamma rays. In M_2 population, many plants showed unique phenotypic traits (Fig. 2a). There were some early flowering mutants (Fig. 2c), early muturity mutant sterile type (Fig. 2a) mutants with higher number of capsule (Fig. 2h). Bushy mutants. Tall mutant (Fig. 2d). Multi capsule (Fig. 2j). Close arragnment of capsule (Fig. 2i) and five loculed per capsule (Fig. 2i) were also noticed in the M_2 population (Fig. 2i).

The mutants were grouped based on phenotypic characters as enlisted in Table 2. By grouping based on plant height revealed that mutants could be classified into six different groups. The maximum number (26.85%) of mutants was of short type (75-105 cm) followed by dwarf type (25-40 cm) accounting for 20.30% of the selected mutants. Tall mutant is a desirable trait in sesamum and 14.80% of selected mutants were of medium height (150-220 cm) with good number of primary and secondary branches. Ultra dwarf mutant having plant height less than 32 cm with no branching and early flowering formed 11.96% of total identified mutants. Long type mutants (5.33%) with plant height more than 285 cm were also isolated. The time taken for appearance of flowering is presented in Table 2. The flowering time in 7.48% of the mutants identified was less than 48 DAS and classified as very early flowering mutants. Some mutants (15.89%) flowered between 50-60 DAS, while 60.00% flowered between 63-90 DAS and some (17.59%) of the mutants flowered very late (after 90 DAS). Mutation affecting number of branches per plant (Table 2), 4.63% of mutants did not produce primary branches and only main stem were emerged. About 15.59% of mutants showed relatively less number (1-5) of branches, while 32.41% of mutants were recorded medium number of branches (6-10). Nearby 45.37% of mutants produced more than 11 branches. The number of capsule per plant is one of the important yield attributing traits in sesamum crop and it was improved in some selected mutants (Table 2). Spectrum of mutation affecting maturity period were isolated in M_2 generation. Early mutants maturing less than 48 DAS accounted for 8.33% of mutants, while 62.96% of the mutants matured between 80 and 90 DAS. About 28.70% of mutants matured late taking more than 100 days. For almost all growth and yield attributing characters variation was observed in both directions i.e. positive and negative in comparison to parent variety ascertaining the efficiency of gamma rays to induced genetic variability in sesamum crop.

Discussion

Frequency and spectrum of chlorophyll mutants

Chlorophyll mutation frequency is an indication in assessing effectiveness of a mutagen, genetic effects of a mutagen and in estimation of mutational events in crop plants. The appearance of chlorophyll mutants during seedling stage was indicator for gamma rays efficiency in sesamum crop. The scoring of chlorophyll mutation frequency in M_2 generation is one of the most reliable measures for evaluating the mutagenic efficiency and effectiveness in the plants. From a breeder's point of view, the frequency of chlorophyll mutants expressed as per cent of M_2 population is more realistic and helpful. Hence, results were explained on M_2 plant basis. The reports of chlorophyll mutants in large number of crops could be ascribed to various causes such as impaired chlorophyll biosynthesis, further degradation of chlorophyll and bleaching taking place due to deficiency of carotenoids. Our results are in confirm with previous findings where mutation affecting chlorophyll, growth period and maturity period have been identified in M_2 population which was the sign of efficiency of gamma rays for the induction of mutation. Chlorophyll deficient chimeras in M_1 generation and their segregation in M_2 generation are often observed in an induced population. Several authors have reported the occurrence of different types of chlorophyll mutations such as, xantha, chlorina, and xantha viridis.

Isolation of suitable mutants under field conditions

Mutation breeding provides a powerful means of creating new and useful variability in crop plants both in qualitative and quantitative characters. Morphological characterizations of germplasm/mutants are necessary for the efficient use of the material through conventional methods or modern techniques. The success of induced mutation also depends on the selection of putative mutants based on phenotypic characters. The findings on plant morphological and reproductive parameters showed that gamma rays irradiated plants can alter the flowering and maturity in either positive or negative direction resulting in sufficient variability in the treated population that can be utilized for selection of early or late flowering plants for further improvement of this versatile crop. The flowering and maturity in the case of induced populations were consistently shifted in the direction of earliness. It is valuable in obtaining varieties associated with escape from pests, drought and other stress injuries that occur in standing crop during cropping season. The results obtained in M_2 generation revealed the possibility of increasing seed yield and yields components through irradiation with gamma rays in sesamum. Improvement of agronomic characteristics of sesamum by using gamma radiation has been reported and the gamma irradiation is important for inducing genetic variability and mutants can be selected in segregating population.

Plant height is one of the important parameters which need to be addressed in sesamum crop. In segregating population a number of the variants identified showed tall mutant. In a report, induced genetic variability in wild type pepper and numbers of mutant selected in segregating population (M_2 generation) showed tall mutant. Tall mutant is an enviable character for breeding programs to increase the nodal segments directly or indirectly correlated to single plant yield in sesamum. In the present investigation, the mutation created tall character and we recorded a plant height more than 285 cm with short internodes and more capsules. Dwarf mutants were also reported earlier in rice, which might be due to the inhibition of the elongation of epidermal cells, or a defect in gibberellic acid biosynthesis. Discovery of dominent, monogenic mutations causing tall characters is very important to allow for easier identification of the genes that control plant growth.

Days to flowering and maturity in case of irradiated populations were consistently shifted towards earliness. The M_2 generation was analysed for variability generated for early flowering and maturity as compared to parent plant. The results revealed that M_2 progenies of fennel significantly differed among themselves and also as a group from the control plant (parent variety) in respect to flowering. The early flowering was recorded in several mutants as compared to control (42 DAS). In our results, it was found that nine mutants reached maturity before 94 DAS. Similar results were reported by other worker [28] and they observed significant variation for days to flowering and maturity in coriander through induced mutation. The number of primary branches between the selected mutants varied in both, positive and negative direction. Maximum number of primary branches (>5) mutants were obtained whereas no primary branches were recorded in four mutants. These characters significantly contributed in yield of crop. A wide range of variability for number of primary branches was observed in several lines of blackgram. One line showed high yield/plant than the check variety, indicating the possibility of further yield improvement by mutation breeding. In M_2 generation of sesamum, 1230 macromutants identified namely, mono stem, prostrate stem, pigmented stem, dwarf, pigmented flowers, narrow leaf and early flowering etc. These lines were advanced by selfing and evaluated in M_3 generation along with control. The eight phenotypic traits including Plant height, Number of primary branches per plant, Length of capsule, Number of capsules per plant, Number of seeds per capsule, Seed yield per plant, Days to maturity and 1000-seed weight were improved.

In case of number of capsule per plants improved significantly from the parent plant. The mutants recorded maximum number of capsule per plants as compared to parent variety. It was also seen that a number of progenies exhibiting superiority in many characters as compared to control. This allowed for possible identification of even better progenies through selection. Yield is a complex character, which depends on it main components, *viz.*, plant height, number of capsule per plant and number of seed per capsule and Length of capsulel etc. These components are further dependent for the expression or reveal morphological and developmental traits, which are interrelated with each other. Therefore, the parents selected in the breeding program, aimed at increasing seed yield should possess wide range of genetic variation for the above said morphological and developmental characters. Besides, it would be of interest to know the magnitude of variation due to heritable component, which in turn would be a guide post in selection for the improvement of a population. In other words, for the improvement of any crop species, the knowledge of genetic variability for characters of economic importance and their heritability and genetic advance is of utmost importance in further breeding programmes. The high yielding progenies resulting from mutation breeding have been earlier reported in urd bean and black cumin.

Table 1: Effect of gamma irradiation on the frequency of the chlorophyll mutation in M_2 generation of VRI 2.

Mutagen dose	Number of M_1 Plants		Number of M_2 seedlings		Mutation frequency	
	Studied	Segregating	Studied	Chlorophyll mutants	M_1 Plants basis	M_2 seedling basis
Gamma rays						
Control	100	-				
300 Gy	100	9	1324	22	0.09	0.016
350 Gy	100	5	1213	14	0.05	0.011
400 Gy	100	7	1127	9	0.09	0.007
450 Gy	100	3	1063	7	0.07	0.006

Table 2: Effect of gamma irradiation on the frequency of the chlorophyll mutations in M_2 generation of TMV 7.

Mutagen dose	Number of M_1 Plants		Number of M_2 seedlings		Mutation frequency	
	Studied	Segregating	Studied	Chlorophyll mutants	M_1 Plants basis	M_2 seedling basis
Gamma rays						
Control	100	-	900			
300 Gy	100	7	1386	15	0.07	0.010
350 Gy	100	4	1200	10	0.04	0.008
400 Gy	100	2	1164	6	0.02	0.005
450 Gy	100	3	1083	7	0.03	0.006

Table 3: Phenotypic characters of the selected mutants in M_2 generation of sesamum.

Group	Number of mutants in M_2 generation	Percent
Plant height (cm)		
Short (51-100 cm)	29	26.85
Medium (101-150 cm)	15	13.89
Normal (151-175 cm)	18	16.67
Tall (>175 cm)	9	8.33
Flowering time (days)		
Early (<38)	7	6.48
Medium (40-45)	15	13.89
Normal (50-55)	67	62.04
Late (>60)	19	17.59
Number of primary branch per plant		
No branching (0)	5	4.63
Less branch (1-5)	19	17.59
Medium branch (6- 10)	35	32.41
Number of capsule per plant		
Less (50-85)	13	12.04
Medium (150-200)	28	25.93
High (300-400)	44	40.74
Number of seeds per capsule		
Less (30-40)	21	19.44
Medium (45-50)	45	41.67
High (>55-70)	19	17.59
Days to maturity		
Early (<38)	9	8.33
Medium (40-45)	68	62.96
Late (>50)	31	28.70

A b c

Fig 1: Appearance of different types of chlorophyll mutants during the seedling stage in segregating population (M_2). a. chlorina mutant (light green colour), b. xantha type (completely white plant), c; viridi type

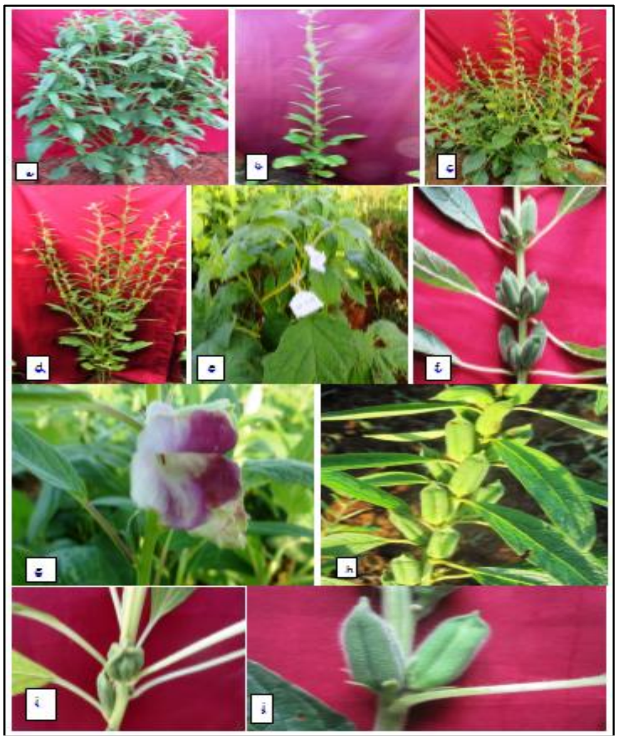

Fig 2: Phenotypic appearance mutant in M_2 generation of sesamum: a; sterile mutant, b; monostem mutant, c; branching mutant, d; tall plants, e; early typemutant, f; multicapsule mutant, g; flower colour pigmentation, h;flower colour mutant, I; multilocule mutant, j; two capsule per node

The present study revealed that the utility of gamma rays in inducing genetic variability in sesamum crop. The irradiation had significant effect on induction of chlorophyll mutants in M_2 generation. Maximum number of mutants were identified at 350 Gy followed by 400 Gy. The mutants varied in different morphological characters which are generally absent in available germplasm. The gene which is responsible for tall character and early maturity in sesamum can be identified and used to develop monostemp of sesamum varieties.

References

1. Choudhary S, Pereira A, Basu S, Verma AK. Differential antioxidant composition and potential of some commonly used Indian spices. Journal of Agrisearch. 2017; 4:160-166.
2. Rather MA, Dar BA, Sofi SN, Bhat BA, Qurishi MA. *Foeniculum vulgare*: A comprehensive review of its traditional use, phytochemistry, pharmacology, and safety. Arabian Journal of Chemistry. 2012; 9:1574-1583.
3. Spice Board India, 2015-16. Database. http://www.indianspices.com
4. Verma AK, Sharma S, Kakani RK, Meena RD, Choudhary S. Gamma radiation effects seed germination, plant growth and yield attributing characters of fennel (*Foeniculum vulgare* Mill.). International Journal of Current Microbiology and Applied Sciences. 2017a; 6:2448-2458.
5. Tanaka A, Shikazono N, Hase Y. Studies on biological effects of ion beams on lethality, molecular nature of mutation, mutation rate, and spectrum of mutation phenotype for mutation breeding in higher plants. Journal of Radiation Research. 2010; 51(3):223-233.
6. International Atomic Energy Agency (IAEA) Mutant variety database. https://mvd.iaea.org, 2017.
7. Arisha MH, Shah SNM, Gong ZH, Jing H, Li C, Zhang HX. Ethyl methane sulfonate induced mutations in M2 generation and physiological variations in M1 generation of peppers (*Capsicum annuum* L.). Frontiers in Plant Science. 2015; 6:399.
8. Roychowdhury R, Alam MJF, Bishnu S, Dalal T, Tah J. Comparative study for effects of chemical mutagenesis on seed germination, survivability and pollen sterility in M1 and M2 generations of *Dianthus*. Plant Breeding and Seed Science. 2012; 65:29-38. 16.
9. Swaminathan MS. A comparison of mutation induction in diploids and polyploids. Radiation Botany. The use of induced mutation in plant breeding. 1964; 5:619-641.
10. Shah TM, Mirza JI, Haq MA, Atta BM. Induced genetic variability in chick pea (*Cicer arietinum* L.). I. Frequency and spectrum of chlorophyll mutations. Pakistan Journal of Botany. 2006; 38(4):1217-1226.
11. PawarN, Pai S, Nimbalkar M, Kolar F, Dixit G. Induction of chlorophyll mutants in *Zingiber officinale* Roscoe by gamma rays and EMS. Emirates Journal of Food and Agriculture. 2010; 22:406-411.
12. Kolar F, Pawar N, Dixit G. Induced chlorophyll mutations in *Delphinium malabaricum* (Huth) Munz. Journal of Applied Horticulture. 2011; 13(1):18-24.
13. Sarada C, Jyothi KUV, Rao S, Reddy PV. Mutagenic sensitivity of gamma rays, EMS and their combinations on germination and seedling vigour in coriander (*Coriandrum sativum* L). International Journal of Advances in Pharmacy, Biology and Chemistry. 2015; 4(2):430-438.
14. Jabeen N, Mirza B. Ethyl methane sulfonate induces morphological mutations in *Capsicum annuum*. International Journal of Agriculture and Biology. 2004; 6:340-345.
15. Fridborg I, Kuusk S, Moritz T, Sundberg E. The Arabidopsis dwarf mutant shi exhibits reduced gibberellin responses conferred by overexpression of a new putative zinc finger protein. Plant Cell. 1999; 11:1019-1031.
16. Wang D, Bosland PW. The genes of *Capsicum*. Hort Science. 2006; 41:1169-1187.
17. Datta AK, Sengupta K. Induced viable macromutants in coriander (*Coriandrum sativum* L.). Indian Journal of Genetics and Plant Breeding. 2002; 62(3):273-274.
18. Verma AK, Kakani RK, Solanki RK, Meena RD. Improvement in yield attributing traits of cumin (*Cuminum cyminum*) through acute exposure of gamma ray. International Journal of Pure & Applied Bioscience. 2017b; 5(2):312-318.
19. Paul R, Datta KA Quantitative evaluation of induced macromutants in fennel (*Foeniculum vulgare* Mill.). International Journal of Research in Ayurveda and Pharmacy. 2016; 7(6):107-109.