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Mutation frequency, spectrum of induced mutagenesis in M₄ generation of soybean (Glycine max L. *Merrill*) genotypes

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Abstract

Soybean is self pollinated crop having small and fragile flowers that's why its very difficult to hybridization programme so, here Mutation breeding used here for crop improvement. The practical role of induced mutation in the improvement of crop plants can best be assessed on the basis of qualitatively inherited characters. Induced mutagenesis plays an important role in improvement of crops soybean 300 (each) Representative M4 Gamma irradiated seeds of two varieties of soybean, BSS-2 and RKS-18 procured from the Department of Genetics and Plant Breeding, BAU, Ranchi were sown in *kharif* 2017. Seeds of both the varieties viz. BSS-2 and RKS-18 were exposed to five different doses of gamma rays (50 Gy, 100 Gy, 150 Gy, 200 Gy and 400 Gy) in the year 2014 and 2015 using Cobalt 60 sources in Gamma chamber at Bhabha Atomic and Research Centre, Mumbai and their M3 progenies were used as experimental materials. Data were recorded for lethality percentage, (15 days after sowing and at flowering) injury percentage in untreated control as well as treated plants. Mutation rate, effectiveness, frequency of five different doses of gamma rays (50 Gy, 100 Gy, 150 Gy, 200 Gy and 400 Gy) and spectrum of qualitative mutations of both the variety in BSS-2 and RKS-18 was studied in the M4 generation. Mutation rate for BSS-2 (0.0596) and for RKS-18 (0.0682) mutation frequency is equal for 50 Gy (8) and 100 Gy (8) in BSS-2 and in RKS-18 in 50 Gy found highest (10). Effectiveness was highest in 50 Gy (0.16), (0.2) in both BSS-2 RKS-18.

Keywords: Induced mutagenesis, Mutation rate, Gamma rays, Mutation frequency, spectrum

Introduction

The history of mutations with early results in soybean has been well documented in the literature available. Mutation breeding has been used in recent years as a valuable supplement to other methods of plant breeding in generating new variability and development of crop varieties with new architecture, superior biochemical constitution and suitable growth and developmental rhythms. The utility of this method is evident from the fact that in several crops induced mutants have been released as new varieties. In this paper, efforts have been made to review the literature on induced mutations in soybean. The different mutagenic agents used for inducing mutations, effects of different mutagenic agents on yield, quality contributing characters and resistance to different diseases have been described. (Khan and Tyagi, 2013) ^[4]. Soybean [Glycine max (L.) Merrill] has become the miracle crop of the 211st century. It is a triple beneficiary crop, which contains about 40% proteins, possessing high level of essential amino acids except methionine and cystine, 20% oil rich in poly unsaturated fatty acids specially omega-6 and omega-3 fatty acids, 6 to 7% total minerals, 5 to 6% crude fibre and 17

to 19% carbohydrates (Chauhan *et al.*, 1988) ^[1]. Using mutation breeding, genetic improvement of any yield attributes either qualitative or quantitative trait, has been successfully achieved in soybean and also in other oil crops (Kharkwal and Shu, (2009) ^[6]. Recently Lagoda (2009) ^[9] has reported that more than 2700 mutant varieties of 170 different plant species have been produced and officially released globally using induced mutations.

The present investigation was undertaken to study role of five doses of Gamma rays in enlarging the genetic variability in two genotypes of soybean BSS-2 and RKS-18 to find out the viable mutants.

Material and methods

Representative M₄ Gamma irradiated seeds (300 each) of two varieties of soybean, BSS-2 and RKS-18 procured from the Department of Genetics and Plant Breeding, BAU, Ranchi were sown in *kharif* 2017. Seeds of both the varieties viz. BSS-2 and RKS-18 were exposed to five different doses of gamma rays (50 Gy, 100 Gy, 150 Gy, 200 Gy and 400 Gy) in the year 2014 and 2015 using Cobalt 60 sources in Gamma chamber at Bhabha Atomic and Research Centre, Mumbai and their M₄progenies were used as experimental materials. The frequency and spectrum of qualitative mutations was studied in the M4 generation.

In *Kharif* 2016, seeds of two varieties of Soybean BSS-2 & RKS-18 of five (5) different doses of γ -rays (50 Gy, 100 Gy, 150 Gy, 200 Gy& 400 Gy) were sown as progeny to row method in four row of 3.5 m length having 45 cm row to row spacing and 10cm plant to plant spacing in 2 replication in RBD design, along with control of both the varieties, BSS-2 and RKS-18. The assessments of qualitative characters were made only from selected families.

The spectrums of different types of viable mutants were scored at various developmental stages in putative mutants population of M_4 generation. The mutants were classified taking into consideration the most conspicuous characters namely stature, leaf shape, seeds per pods and seed variants etc. The frequency, effectiveness, mutation rate, injury percentage, lethality percentage, injury percentage and spectrum of viable mutants were calculated and important characters of each mutant were recorded.

Result and discussion

The spectrum of different types of viable mutants were scored at various developmental stages of M_4 plants. The mutants were classified taking into consideration the most

conspicuous characters namely stature, leaf shape, seeds per pods and seed variants etc. The frequency and spectrum of viable mutants were calculated and important characters of each mutant were recorded.

The viable mutants isolated in the present study included mutants with agronomically desirable features which could possibly be utilized in future for breeding programmes. In soybean variety, BSS-2 the spectrum of viable mutation was maximum at 50 Gy (8), 150Gy (8). (Table 2) In the variety RKS-18, the spectrum of viable mutants was maximum in 50 Gy (10) followed by 100 Gy (8) and 150 Gy (4).

The possible cause of these macro mutations may be chromosomal aberrations, small deficiencies or duplications and most probably gene mutations. Maximum number of mutants was obtained for leaf related traits followed by seed related traits like Hilum colour and seed coat colour.

Short plant height is an very important traits because it having lodging resistant characters. The plant mutants with mean height of plant of more than 20 cm but less than 30 cm were considered as dwarfism in this present study. The mean height of control were 48.58cm in BSS-2 and 48.09 in RKS-18, whereas in dwarf mutant it was in BSS-2 24.46 in 400 Gy, 24.70 in 200 Gy while in RKS-18 28.22 in 400 Gy, 28.76 in 150 Gy,29.22 in 100 Gy some dwarf mutant showed early maturity observed in BSS-2 in 50 Gy (1), 100 Gy (1), 200 Gy (1) but it does not give any variation with number of pods per plant as well as for yield per plant.

In both the variety BSS-2 and RKS-18 maximum number of Leaf mutant observed in M4 generation included changes in leaflet number, leaf shape, leaf size, sessile leaves, lanceolate leaves, bifoliate leaves, tetrafoliate leaves, heart shape leaves, hexafoliate.

The variation in leaflet number did not give any variation in seed yield as compared to control.

In variety BSS-2 mutant for leaf shape like Bifoliate, Bifoliate and sessile, Trifoliate with one leaf sessile, Quadrifoliate, Pentafoliate, Small leaf, Heart shaped leaf were observed while in RKS-18 bifoliate, bifoliate and sessile, brown venation leaves were observed in M4 generation.

In variety, BSS-2 light dark brown hilum, dark brown seed coat, Dark yellow seed coat, Large seed, Brown patch on seed were observed while in RKS-18 Small seeds, elongated seeds, dark brown seed coat, broad hilum, small hilum were observed in M4 generation.

In variety, RKS-18, light violet flower color observed in dose 100 Gy (1) and 200 Gy (2).

 Table 1: Frequency, mutation rate, injury, efficiency and effectiveness of macro mutation induced by gamma rays in putative mutant population of two varieties (BSS-2 and RKS-18) of soybean M4 generation.

Variety	Dose	Total plants studied in M ₄	Total plants segregated in M ₄	Mutation Rate	Mutation Frequency	Effectiveness
BSS - 2	Control	30				
	50 Gy	50	4		8	0.16
	100 Gy	50	3	0.0596	6	0.06
	150 Gy	50	4	0.0390	8	0.053
	200 Gy	50	2		4	0.02
	400 Gy	50	1		2	0.005
	Control	30				
	50 Gy	50	5		10	0.2
RKS - 18	100 Gy	50	4	0.0682	8	0.08
	150 Gy	50	2	0.0082	4	0.026
	200 Gy	50	3		6	0.03
	400 Gy	50	1		2	0.005

Table 2: Spectrum of viable mutants induced by gamma rays of putative mutant population in the variety BSS-2 of soybean in M4 (2017-18)

S. No	Mutant Character/Dose	50 Gy	100 Gy	150 Gy	200 Gy	400 Gy
	Plant					
1	Short height plant			1		
2	Short height plant and early flowering	1	1		1	
	Leaf					
1	Bifoliate	2	1	-	1	1
2	Bifoliate and sessile	1	2	-	1	-
3	Trifoliate with one leaf sessile					
4	Quadrifoliate	2	2	-	1	-
5	Sesilequadrifoliate leaf					
6	Pentafoliate					
7	Hexafoliate	-		2	-	-
8	Yellow leaf					
9	Small leaf					
10	Heart shaped leaf					
11	Obovate leaf shape			1		
12	Yellow patch on leaf					
13	Brown venation	1	-	-		-
	Stem			1		
1	Twining stem					
2	Tricotyledon					
3	Tetracotyledon					
4	Branched cotyledon					
5	Brown hypocotyl (Pigmentation present)					
	Seed					
1	Light Brown Hilum					
2	Black Hilum					
3	Dark brown Hilum	-		1		
4	Medium Brown seed coat					
5	Dark brown seed coat					1
6	Light brown seed coat					
7	Dark yellow seed coat	1				
8	Small seed					
9	Large seed	-	-	1		
10	Elongated seed					
11	Brown patch on seed			1		
•	Total	8	6	8	4	2

Table 3: Spectrum of viable mutants induced by gamma rays of putative mutant population in the variety RKS-18 of soybean in M4 generation(2017-18).

S. No	Mutant Character/Dose	50 Gy	100 Gy	150 Gy	200 Gy	400 Gy
	Plant Height					
1	Short height plant	2	1		2	1
	Leaf					
1	Bifoliate	3	2	1		
2	Bifoliate and sessile				1	
3	Brown venation					
	Flower Colour					
1	Light violet flower colour		1		2	
	Seed					
1	Small seeds	1		1	1	1
2	Elongated seeds	2	1	1		
3	Yellow seed coat					
4	Dark brown seed coat					
5	Black seed coat					
6	Broad Hilum	1	1	1		
7	Small Hilum		2			
8	Black Hilum					
9	Light Brown Hilum					
10	Bold seed	1				
	Total	10	8	4	6	2

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