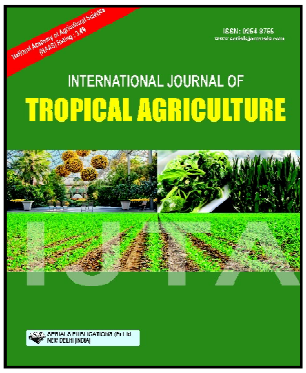

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Mutagenesis for Oligogenic Traits with Gamma Rays and Ems in Soybean (*Glycine max* L.)

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Abstract: Induced qualitative mutation in JS-335 Soybean cultivar consisted of chlorophyll and viable mutants. The combination treatments exhibits wide spectrum and frequency on both M_1 family and M_2 plant basis for both. Increased in doses and concentrations of mutagens showed increased in spectrum and frequency. Chlorophyll mutants viz. albino, albo-viridis, xantha, xantha- viridis and viridis and viable mutants viz; plant type, leaf, growth habit, flowering, pod, kernel and economic mutation were observed. Desirable mutants like bold poded, early maturing, and high yielding were also isolated.

Key Words: JS-335, Spectrum, Frequency, Gamma Rays and EMS)

INTRODUCTION

Mutation breeding has contributed to increase genetic resources and has a valuable tool for plant breeding. Genetic variations induced by mutation represent a more efficient source of genetic variability than gene pools conserved by nature (Brock, 1977). The advantage of mutation breeding is that it can be applied to altering specific characters in other wise good varieties by incorporating some useful changes such as earliness, high oil content and high yielding ability in a comparatively shorter time than the conventions breeding methods. So, the induced mutations supplement plant breeding method and confer specific improvements on a genotype without significantly altering its otherwise acceptable phenotype. Since the induction of mutation has been accepted as a useful tool in plant breeding, a systematic study of the induced mutagenesis in crop like soybean appears to be

essential specially in the light of the growing need for food feed, fuel and fertilizer. Both radiations and chemical mutagens have been employed to generate the desired variability in various crop species. Though their effect on quantitative characters, investigations involving chemical mutagens are mere in soybean. Even these investigations were directed to assess the physical sensitivity of varieties in groundnut (Gregory,1956; Ashri and Goldin,1965) Intensive studies on the effect of physical and chemical mutagens and their combinations inducing viable mutations in soybean cultivars are required to derive the maximum benefits from these tools. Therefore, the present investigation was undertaken to study the effect of physical and chemical mutagens on induction of viable mutation.

MATERIALS AND METHODS

Two mutagens viz; gamma rays, ethyl methane sulphonate and their combination treatments were administered. The breeder seeds of JS-335 variety of soybean were procured from Oilseed Research Unit, Dr.PDKV, Akola (MS) India and seeds of uniform size were treated with the details given in Table 1. Hundred seeds of each treatment were sowed in augmented block design with spacing of 40 X 15cm. The M_1 plants were harvested on single plant basis. The seeds harvested from randomly selected 50 M_1 plants were sown to raise M_2generation. It was expected that in each treatment should consisted 50 plants but due to lethality in some higher doses the number of plant progenies were obtained less than fifty. These all plant progenies were raised on progeny row basis along with two controls with 45 X 15 spacing. The M_2 generation was examined up to 15th day after germination for chlorophyll mutation mutations. The spectrum and frequency was estimated and expressed as percentage on both M_1 family and M_2 plant basis. The mutant and normal seedlings were counted separately to determine the segregation ratio i.e., percentage of mutants to total progenies. The chlorophyll mutations were classified according to the system proposed by Gustafsson (1940) and Blixt (1961).The viable mutants were observed periodically from seedling stage to maturity in M_2 generation and its frequency and spectrum was also estimated as like chlorophyll mutants.

Table 1
Treatment details

Treatment code	*Treatment details*
Gamma rays	
T_1 – 150gy	Irradiation of seeds with 150 Gy Gamma-rays
T_2 – 250gy	Irradiation of seeds with 250 Gy Gamma-rays
T_3 – 300gy	Irradiation of seeds with 300 Gy Gamma-rays
EMS	
T_4 – 0.05% EMS	Presoaking of seeds for 6 hrs. followed by 6hr.soaking in 0.05% EMS
T_5 – 0.1% EMS	Presoaking of seeds for 6 hrs. followed by 6hr. soaking in 0.1% EMS
T_6 – 0.2% EMS	Presoaking of seeds for 6 hrs. followed by 6 hrs. soaking in 0.2%EMS
Combinations	
T_7 – 150 Gy + 0.2% EMS	Irradiation of seeds with 150 Gy, presoaking for 6 hrs. followed by 6 hrs. soaking in 0.2% EMS
T_8 – 250 Gy + 0.1% EMS	Irradiation of seeds with 250 Gy, presoaking for 6 hrs. followed by 6 hrs. soaking in 0.1% EMS
T_9 – 300 Gy + 0.05% EMS	Irradiation of seeds with 300 Gy, presoaking for 6 hrs. followed by 6hr. soaking in 0.05% EMS
Control	
T_{10} – Dry	Control (Dry seeds)
T_{11} – Presoaked	Control (Seeds presoaked in distilled water only)

RESULT AND DISCUSSION

Chlorophyll Mutations: The spectrum of chlorophyll mutation induced by mutagenic

treatment was found to vary according to the mutagen doses or concentrations. The xantha-viridis and viridis mutations were of common occurrence in most of the mutagenic treatments. The different mutagenic treatments used, differed significantly from each other for inducing chlorophyll mutations. However, combination treatments of gamma rays with ethyl methane sulphonate were found most effective in inducing chlorophyll mutations and sole treatments of ethyl methane sulphonate were found least effective. Generally all the mutagenic treatment induced maximum chlorophyll mutations namely albina, albo-viridis, xantha, xantha-viridis, viridis and chlorina. Similar spectrum of chlorophyll mutations was also reported by Rajput and sarwar (1996) in lentil, Das and Kundagrami (2000) in grass pea, Geeta and Vaidyanathan (2000) in Soybean.

(Table 2) The mutation frequency on both M_1 family basis and M_2 plant basis was found maximum (26.09 percent on M_1 family basis and 1.98 percent on M_2 plant basis) in 300 Gy gamma rays + 0.05percent EMS. It was indicated from the results that increased doses or concentrations of the mutagens showed increased both frequency and spectrum of the chlorophyll mutations. The differential response obtained in the present investigation to various mutagenic treatments could be due to the reaction of specific genes to mutagens, which could be responding differentially to physical, chemical mutagens or their combinations. It is also believed that these mutations seemed to be brought about by genes located on different chromosomes. Swaminathan (1965) has suggested that the genes controlling the chlorophyll characters may be located near the centromere and proximal system is responsible for the high incidence of chlorophyll mutations data from linkage analysis in barley (Robetson, 1963 and Nilan, 1964) and maize (Neuffer, 1966) and the chromosomes aberrations studies (Natrajan and Upadhyaya, 1964) have provided evidence in support of this view. Ramanathan and Rathinam (1983) have reported high frequency of chlorophyll mutations in M2 generation by combined dose of gamma rays and EMS. The more or less similar results were also reported by Chopde (2009), Venkatachalam*et al.* (1999), Levy and Ashri (1975) and Shivasubramaniam (1978).

Viable Mutations: The various types of viable mutations with altered plant habit were isolated in M2 generation and important ones were confirmed in M3 generation. The frequency of macro mutations expressed on M2 plant basis as well as M1 family basis was found increased as the dose of mutagens increased (Table 3). The observations recorded are in agreement with those of Vannirajan *et al* (1993) in blackgram, Vandana *et al* (1994) in Lentil and Nadarajan *et al* (1982) in pigeonpea.

Study of spectrum of viable mutations showed that numbers of viable mutations were induced for plant type, leaf modifications, growth habit, flowering, pod characters, seed and economic mutants. The mutations confirmed in M_3 generation. The maximum mutation rate on M_1 family basis was observed in combined dose of 300 Gy gamma rays + 0.05 percent EMS while, it was found maximum in 300 Gy gamma rays on M_2 plant basis. It was predicted that increased mutation rate responsible for increased doses or concentrations of the mutagens. The frequency of viable mutations was recorded higher under gamma ray treatment in comparison with ethyl methane sulphonate. Gregory (1968) found 11,502 visible mutations in M2 out of 84, 213 plants following the treatment with 18.5 kR of X-rays. Ashri and Levy (1972) found that gamma rays gave a higher mutation rate than EMS in groundnut. In present study higher mutation on M2 plant basis was obtained in combined treatment might be due to higher dose of 300 Gy gamma rays.

Table 2
Frequency and spectrum of the chlorophyll mutations in M_2 generation

Treatments	*Total No. of M_1 families sown*	*No. of M_1 families segregating*	*Total No. of M_2 plants*	*Spectrum of Chlorophyll mutations*						*Total No. Mutations*	*Mutation frequency*	
				Albina	*Albaviridis*	*Xantha*	*Viridis*	*Xantha-Viridis*	*Cholorina*		*M_1 Family basis*	*M_2 plant basis*
Gamma rays												
T_1– 150gy	50	4	670	0.45	0.45			0.30		1.19	8	0.18
T_2– 250gy	50	6	456	0.44		0.22	0.22	0.88		1.75	12	0.38
T_3– 300gy	50	9	286		0.35		0.70		0.35	1.40	18	0.49
EMS												
T_4– 0.05% EMS	50	5	1125				0.18	0.09		0.27	10	0.02
T_5– 0.1% EMS	50	5	826		0.24	0.12	0.48			0.85	10	0.10
T_6– 0.2% EMS	50	7	620		0.48	0.32	0.48	0.48		1.77	14	0.29
Combination												
T_7– 150 Gy x 0.2% EMS	49	5	312	0.32			0.32		0.64	1.28	10.20	0.41
T_8– 250 Gy x 0.1% EMS	32	5	195			1.54		0.51		2.05	15.63	1.05
T_9– 300 Gy x 0.05% EMS	23	6	142	1.41		0.70			0.70	2.82	26.09	1.98
Control												
T_{10}– Control Dry	50	-	1432									
T_{11}– Control Presoaked	50	-	1463									

Table 3
Frequency and spectrum of macro mutation in M_2 generation

.	*Total No. of M families sown*	*No. of M1 families segregating*	*Total No. of M_2 plants*	*Leaf mutants*				*Growth habit mutants*				
				Multifoliate	*Broad*	*small*	*Total*	*Tall erect*	*Unbranched*	*miniature*	*Dwarf bushy*	*Total*
Gamma rays												
T_1 – 150Gy	50	4	670	-	0.60	0.30	0.90	-	-	0.45	0.45	0.90
T_2 – 250Gy	50	6	456	0.22	0.22	0.88	1.32	0.22	0.66	-	-	0.88
T_3 – 300Gy	50	9	286		0.70	1.05	1.75	-	-	1.05	1.05	2.10
EMS												
T_4 – 0.05% EMS	50	5	1125	-	0.36	0.36	0.71	-	0.09	0.44	-	0.53
T_5 – 0.1% EMS	50	5	826	0.12		0.48	0.61	-	-	0.61	-	0.61
T_6 – 0.2% EMS	50	7	620	-	-	0.61	0.61	-	0.32	0.48	0.48	1.29
Combination												
T_7 – 150 Gy + 0.2% EMS	49	5	312	0.96	-	0.24	1.20	0.96	-	0.32	-	1.28
T_8 – 250 Gy + 0.1% EMS	32	5	195	0.51	-	0.51	1.03	-	-	1.03	-	1.03
T_9 – 300 Gy + 0.05% EMS	23	6	142	0.70	-	-	0.70	-	0.70	1.41	-	2.11
Control												
T_{10} – Dry	50	-	1432	-	-	-		-	-	-	-	-
T_{11} – Presoaked	50	-	1463	-	-	-		-	-	-	-	-

Table 3: Continued....

Mutants of economic intrest							*Other mutants*								*Total number of mutations*	*Mutation rate*	
Big podded and bold seeded	*Non shattering*	*Early maturing*	*Late maturing*	*High yielding*	*Profuse flowering*	*Total*	*Small podded and seeded*	*Bunchy flower*	*High pubescence (Hairy)*	*changed flower colour*	*Early flower*	*Deformed*	*Variegated seed colour & shape*	*Total*		*M1 family basis*	*M2 plant basis*
0.35	1.49	-	0.75	-	0.60	3.18	0.45	0.45	-	-		0.45	0.30	1.64	45.00	8.00	12.34
0.66	-	-	0.66	0.44	0.44	2.19	-	-	0.44	0.44	-	-	-	0.88	24.00	12.00	9.21
-	0.35	2.10		0.70	-	3.15	1.75	-	-	-	2.10	0.70	-	4.55	33.00	17.00	21.33
0.36	0.42	-	-	-	-	0.77	0.53	-	-	-	-	0.44	0.25	1.23	37.00	10.00	5.78
0.36	0.33	0.61	-	0.12	-	1.42	0.36	-	-	-	0.61	-	0.36	1.33	33.00	10.00	7.32
0.32	-	0.81	0.97	0.59	-	2.68	-	-		0.65	-	0.32	-	0.97	37.00	14.00	10.49
1.28	0.64		-	1.92	-	3.85	0.96	0.96	-	-	-	-	0.32	2.24	28.00	10.20	15.95
-	-	1.03		2.05	0.94	4.02	1.03	0.51	-	-	1.03	1.54	-	4.10	20.00	15.63	19.32
-	-	2.11	-	0.70	-	2.82	0.70		-	-	2.11	1.41	-	4.23	14.00	26.09	19.01
-	-	-	-	-	-	0.00	-	-	-	-	-	-	-	0.00	-	0.00	0.00
-	-	-	-	-	-	0.00	-	-	-	-	-	-	-	0.00	-	0.00	0.00

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