



SHORT COMMUNICATION

Induced Chlorophyll Mutations in Soybean *Glycine max* (L.) Merrill**A.B. Tambe¹, M.V. Pachore¹, S.P. Giri¹, B.S. Andhale¹ and Dr. B.J. Apparao²**¹Padamshree Vikhe Patil College of Arts, Commerce and Science, Pravaranagar Tal- Rahata Dist- Ahmednagar (MS), India
Pin- 413713² Jijamata College of Arts and Science, Bhende Tal- Newasa Dist- Ahmednagar (MS), India**ABSTRACT**

*In the present work attempt were made to induce chlorophyll mutations in soybean [*Glycine max* (L.) Merrill] by employing EMS and Gamma rays. The germplasm of soybean cultivar MACS 450 was procured from Agharkar Research Institute, Pune Maharashtra. The uniform presoaked seeds of soybean were treated with different concentrations of EMS (10, 20, 30 and 40 mM) for 08 hrs and dry seeds were irradiated with different doses (100, 200, 300 and 400 Gy) of gamma rays at Government Institute of Science, Aurangabad. Treated as well as untreated seeds were sown in the experimental field in RBD during kharif 2006 in the spacing of 45 × 15 cm to raise M₁ population. Seeds threshold from individual M₁ plants were sown in kharif 2007 to raise M₂ generation. M₂ population was screened seems from the first day of emergence of the seedlings up to the harvesting for chlorophyll mutations and viable mutations. Four different types of chlorophyll mutations were observed in the M₂ progeny of soybean. They are albina, viridis, chlorina and xantha. The variety responded differently to the mutagens. The frequency of chlorophyll mutations increased with an increase in the concentration of the mutagens except at 20 mM EMS concentrations. The frequency of chlorophyll mutations 2.12 to 8.12% in EMS, and 4.04 to 7.78% in Gamma rays. The maximum frequency of chlorophyll mutations (8.12%) was observed at 40 mM EMS concentration and it was maximum (7.78%) at 400 Gy dose of gamma rays. The overall spectrum of induced chlorophyll mutations as observed in soybean was in the following order. Chlorina (17.61%) > Viridis (15.67%) > Xantha (11.07%) > Albina (0.56%)*

KEYWORDS: Soybean, chlorophyll mutant, EMS and Gamma rays.

INTRODUCTION

Soybean [*Glycine max* (L.) Merrill, Family, Papilionaceae (Fabaceae)], vernacularly also known as golden bean is an important oil seed crop widely cultivated in Indian subcontinent. In Maharashtra, it is widely cultivated in Ahmednagar district. Soybean forms one of the important constituents in the dietary practices of local communities. Soybean is consumed in the form of vegetable oil. It is mainly used in the preparation of idly, papad, dosa, paneer, soya flour, soya milk and other regional fermented foods. Nutritional composition of soybean indicates that it has protein content as high as 40% and 20% oil. Its beans form a nutritious item of the food, while the whole plant gives rich feed for cattle and is a good manure as well as conservation crop.

Soybean is also prescribed as a medicine to fulfill the need of malnutrition. In spite of its nutritional importance, the yield of soybean did not witness much appreciation during the past decade. It has been argued that one of the chief reasons for failure to achieve breakthrough in productivity of soybean is lack of its genetic variability. Genetic variability is the most essential prerequisite for any successful crop improvement programme as it provides a spectrum of variants for an effective selection process. Mutation breeding techniques are the best methods to enlarge the genetically conditioned variability of a species within a short time and have played a significant role in the development of many crop varieties.

The role of induced mutations in developing new and better cultivars has now been well recognized. Therefore the most popular method employed for creating genetic variability is induced mutagenesis through gamma irradiation [1] and chemical mutagen like EMS. Chlorophyll mutations are one among the few dependable parameters for evaluation of genetic effects of various mutagens and are widely used as genetic markers in basic and applied research. The present study reports the induction of different chlorophyll mutants in M₂ generation in soybean cultivar MACS 450.

MATERIAL AND METHODS

The experimental plant material used in the present investigation is local variety of soybean [*Glycine max* (L) Merrill], MACS-450. Germplasm of this cultivar of soybean was procured from the 'Agharkar Research Institute (ARI), Pune, Maharashtra state, India). Seeds presoaked in water for 6 hours, were treated with different concentrations of EMS (10mM, 20mM, 30mM and 40mM) for 8 hours at room temperature. Dry seeds were irradiated with 100Gy, 200Gy, 300Gy, and 400Gy gamma radiation doses at Government Institute of Science, Aurangabad (M.S., India). Seeds not treated with the mutagens served as control. About 300 seeds of each treatment were sown in the experimental field, along with controls, following randomized block design in 3 replications to rise M₁ generation during kharif season of 2006. The individual seeds of M₁ plant progenies were sown in the field to raise M₂ progeny. Uniform cultivation methods and agricultural practices were followed for all M₁ and M₂ generations. Various chlorophyll mutants were identified according to Gustafsson [2]. Frequencies were calculated according to Kozak *et al.*, (1965). Using the data on chlorophyll mutation frequency was calculated.

RESULTS

The M₂ progeny raised from the M₁ seed showed the presence of four types of chlorophyll mutations. They are: viridis, chlorina, xantha and albina.

Viridis

The seedlings were golden yellow-green in colour and survived for a reasonably long period.

Chlorina

The seedlings were light yellowish green (pale green) in colour. They survived for a reasonably long period.

Xantha

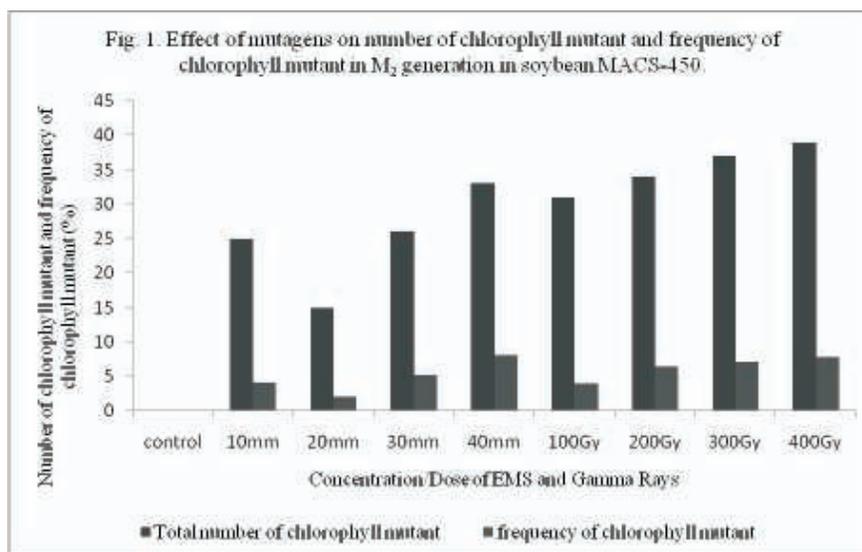
The seedlings were completely yellowish and leaves are larger than viridis. The seedlings survived for only 7-8 days.

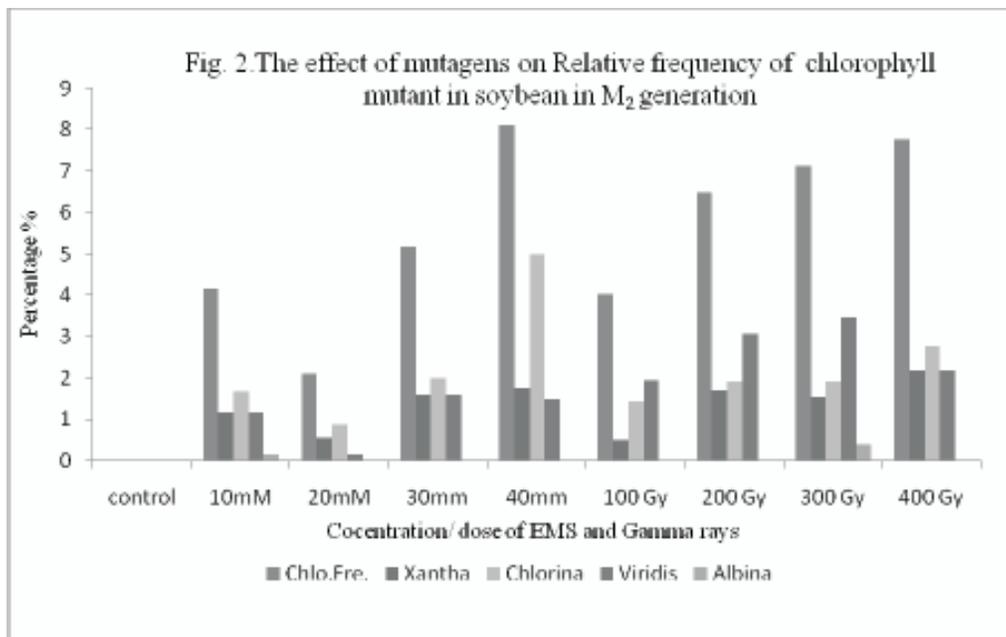
Albina

Leaves are white in colour and the seedlings died after few days.

Frequency of these chlorophyll mutations increased with an increase in concentration/dose of the mutagens. The maximum frequency of chlorophyll mutations was observed at 40mM concentrations of EMS (8.12%) and 400 Gy of gamma rays (7.78%). Chlorina chlorophyll mutation 5% was recorded at 40mM concentration of EMS and 2.79% at 400 Gy doses of gamma rays. Xantha chlorophyll mutations 1.60% at 30mM of EMS and 2.20% at 400 Gy dose of gamma rays. Viridis chlorophyll mutation was recorded 1.60% at 30mM concentration of EMS and 3.47% at 300 Gy doses of gamma rays. Albina type chlorophyll mutation was least and only observed in 10mM of EMS (0.17%) and 300 Gy doses of gamma rays (0.39%). Both the mutagens were found to be equally effective in producing high frequency of chlorophyll mutations. The overall spectrum of induced chlorophyll mutations as observed in soybean was in the following order. Chlorina (17.61%) > Viridis (15.67%) > Xantha (11.07%) > Albina (0.56%).

Increase in the frequencies of chlorophyll mutations with increase in the concentration / dose reported [3-8] see table and fig. 1 & 2.





The high frequency of chlorophyll mutations obtained with mutagens, is due to preferential action of these mutagens on genes for chlorophyll development /or the preferential effect on guanine in the G-C rich chloroplast genome. Biosynthesis of photosynthetic pigments occurs in a series of biochemical reactions. EMS and gamma rays are potent mutagens well known for their action in inducing point mutations, and chromosomal aberrations. Any alteration in the nucleotide composition of the genes, that control the synthesis of enzymes involved in the biosynthesis of pigments, as result of action of the mutagens, would eventually lead to the observed chlorophyll mutations.

Table the effect of mutagens on the spectrum of chlorophyll mutants in M₂ generation

Mutagens	Conc/dose	Frequency of chlorophyll mutants (%)	Relative percentage (%)			
			Xantha	Chlorina	Viridis	Albina
Control	Control	-	-	-	-	-
EMS(mM)	10mm	4.16	1.17	1.67	1.17	0.17
	20mm	2.12	0.57	0.87	0.71	-
Gamma radiation(GY)	30mm	5.18	1.60	2.00	1.60	-
	40mm	8.12	1.75	5.00	1.50	-
	100Gy	4.04	0.52	1.43	1.96	-
	200Gy	6.50	1.72	1.92	3.06	-
	300Gy	7.14	1.54	1.93	3.47	0.39
	400Gy	7.78	2.20	2.79	2.20	-

ACKNOWLEDGEMENTS

We are thankful to Agharkar Research Institution, Pune, for providing the germplasm, Government Institute of Science, Aurangabad for extending irradiation facility and Dr. S.R. Walunj, Principal, Dr. P.G.Reddy H.O.D. Botany Department, Padmashri Vikhe Patil college, Pravaranagar for providing laboratory facilities.

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