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Induced Cholrophyll Mutations in *Lablab niger* Medikus

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A total number of 33,731 M_2 seedlings from 340 M_1 plant progenies obtained through different doses of gamma irradiation and EMS treatments of dry and soaked seeds were studied in M_2 generation. The mutation frequency was higher in EMS treatments than gamma rays. But pre-soaking the seeds prior to gamma irradiation enhanced the mutation rate. The mean segregation ratios were higher and also enhanced the effectiveness and efficiency when the soaked seeds were treated with gamma irradiation. Gamma rays were found to be more effective than EMS and also efficient on the basis of injury. The occurence of *chlorina* and *viridis* mutants was frequent in all the treatments of gamma rays and EMS. Gamma irradiation of dry seeds induced a wide spectrum of all the four types of chlorophyll mutants viz., *albina*, *viridis*, *chlorina* and *xantha*.

A vast literature is available on induced chlorophyll mutations in various pulse crops. However, the Indian Pulses like redgram (*Cajanus cajan L* Millsp) blackgram (*Phaseolus mungo* L.) green gram (*Phaseolus aureus* Roxb.), cowpea (*Vigna sinensis* L. Savi) and chickpea (*Cicer arietinum* L.) lack such studies and have no studies on induced chlorophyll mutagenesis in lablab (*Lablab niger* Medikus) particularly. So an attempt has been made in this direction, and the results are presented.

MATERIALS AND METHODS

The strain CO.6 lab lab, a hybrid derivative developed from the cross between kitchen garden lab lab CO.5 (Var. *typicus*) and field lab lab DL. 3196 (Var. *lignosus*) was chosen for the study. Dry seeds with a moisture content of 10 per cent and 12 hours presoaked seeds, were exposed to Cobalt 60, a gamma source. The treatments were given for various durations depending on the doses desired, the dose rate being 0.3×10^6 rads per hour.

The seeds were pre-soaked for 10 hours before EMS treatment, prepared with deionized water and without buffer. The period of treatment was 4 hours at room temperature 26 $\pm$ 2°C with intermittent shaking. After treatment the seeds were thoroughly washed with running tap water for half an hour and were sown to raise M_1 generation.

Twenty M_1 plants in each treatment and in control were advanced to M_2

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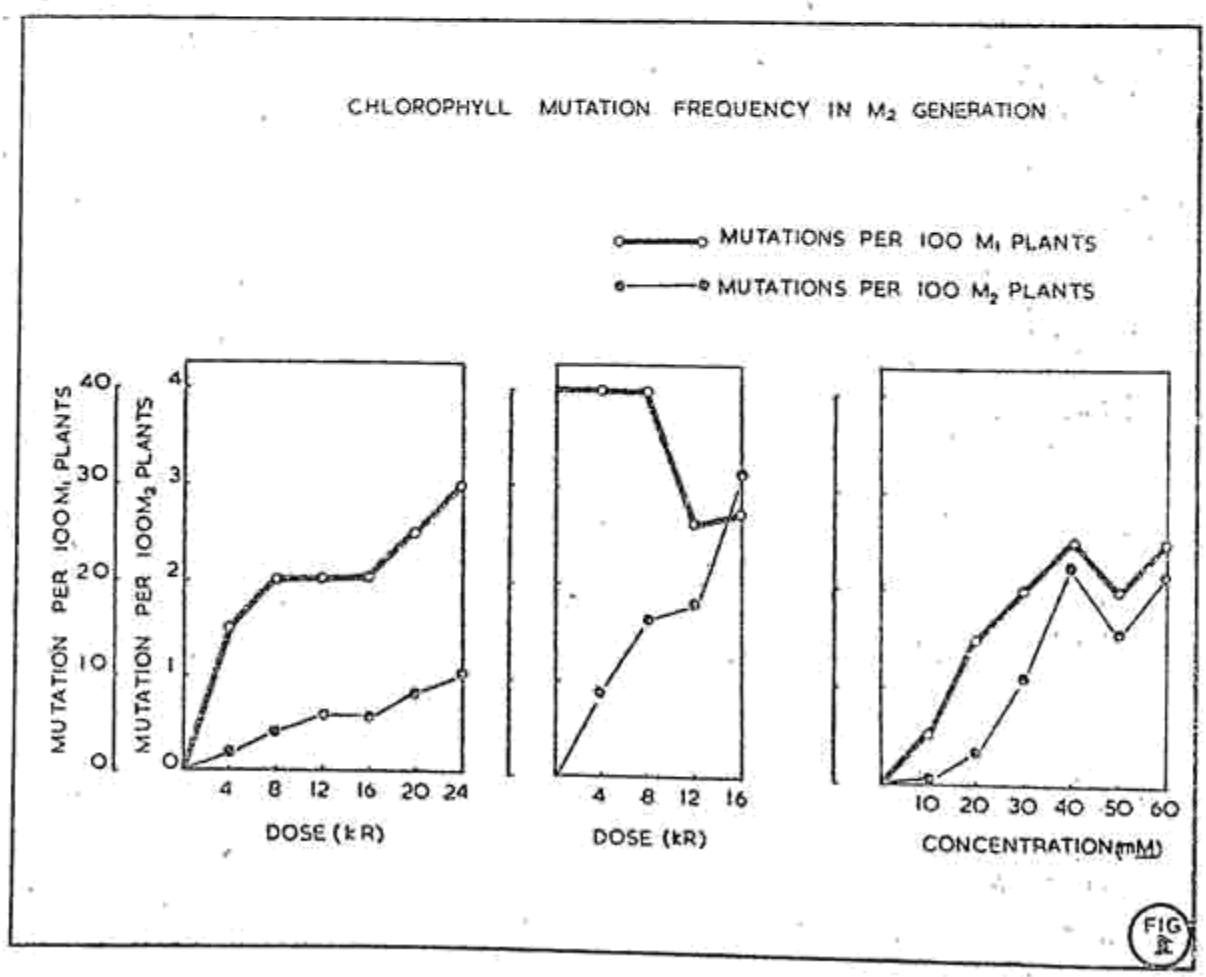
generation and raised as individual progeny rows. The M_2 seedlings for individual M_1 plant progenies were examined from the day of germination upto 12th day. The chlorophyll mutants were classified according to the system proposed by Gustaffson (1940) and Blixt (1961). The effectiveness and efficiency of mutagens in inducing chlorophyll mutants were estimated after Konzak *et al*.(1965).

RESULTS AND DISCUSSION

a) **Chlorophyll mutation frequency :** A total number of 33,531 M_2 seedlings, from 340 M_1 plants was studied for estimating chlorophyll mutation frequencies (Table I). The rate of mutation was, in general, higher when expressed as percentage on M_1 plant basis. The chlorophyll mutation frequencies showed a proportionate increase and reached a maximum at the highest dose of gamma rays on dry seeds (Fig. 1). But in the case of EMS, the mutation rate increased upto middle dose and slightly dropped at higher doses thereby indicating the elimniation of mutations at higher concentrations. The mutation frequency was higher in EMS than in gamma rays. Similar results were reported in *Vicia* by Zannone (1965). The presoaking enhanced the mutation rate as compared to dry seeds treated with gamma rays. Rao (1968) obtained similar results in cowpea irradiated with X-rays.



TBALE I. Chlorophyll Mutations, Mutation rate, Segregation ratio, Effectiveness and Efficiency in M_2 Generation

Mutagen	Dose/conc.	Mutation rate M_1 plant basis	Mutation rate M_2 plant basis	Mean segregation ratio	Effectiveness M x 100 / tc or Krad	Efficiency X x 100 / L	Efficiency M x 100 / I	Efficiency M x 100 / S
Gamma rays								
Dry seeds								
	4 krad	15.0	0.20	0.92	5.0	3.3	20.0	3.7
	8 krad	20.0	0.45	1.84	5.5	3.7	15.5	2.8
	12 krad	20.0	0.63	2.62	5.3	3.5	4.1	3.5
	16 krad	25.0	0.61	2.93	3.8	1.9	2.9	2.8
	20 kred	25.0	0.85	3.04	4.3	2.4	3.4	3.0
	24 krad	30.0	1.01	3.98	4.2	1.9	3.4	3.9
	Control	—	—	—	—	—	—	—
Soaked seeds								
	4 krad	40.0	0.93	1.33	23.3	6.8	22.7	11.6
	8 krad	40.0	1.74	3.18	21.8	6.5	11.2	11.8
	12 krad	26.7	1.85	6.14	15.4	3.0	7.8	9.9
	16 krad	27.3	3.16	9.06	19.8	4.2	9.8	13.1
	Control	—	—	—	—	—	—	—
EMS								
	10 mM	5.0	0.06	1.60	0.2	0.5	0.6	5.0
	20 mM	15.0	0.37	1.39	0.5	1.5	2.6	3.9
	30 mM	20.0	1.17	3.02	1.0	4.9	5.9	7.6
	40 mM	25.0	2.26	4.01	1.4	7.0	8.8	15.9
	50 mM	20.0	1.66	6.27	0.8	4.0	5.6	8.4
	60 mM	25.0	2.19	4.26	0.9	3.6	6.7	9.6
	Control	—	—	—	—	—	—	—

M=mutation rate, t=time, c=concentration, krad=Kilo rad, L=Percentage of lethality
I=Percentage of injury, S=Peicentage of survival

b) **Segregation ratio** : The mean segregation ratios of chlorophyll mutations increased with increase in dosage of gamma rays and EMS, except at the highest dose of EMS. But, in none of the treatments, the mean segregation ratios approached twenty five per cent indicating the deficites of recessives. The mean segregation ratios were higher after treatment with gamma irradiation with soaked seeds than gamma irradiation with dry seeds. The mean segregation ratio was dose dependant in gamma rays and EMS. The dependence of M_2 segregation ratio on dose was reported by Wellensiek (1965) in peas and Ramaswamy (1973) in blackgram. The increase in the segregation ratios with increase in dose of mutagen could be due to an increase in size of mutated

TABLE II. Spectrum of Chlorophyll Mutation in the M_2 Generation

Mutagens	Dose/Conc.	Total number of chlorophyll mutants	Relative % of chlorophyll mutants			
			Albina	Virdis	Chlorina	Xantha
Gamma rays						
Dry seeds						
	4 krad	6	—	33.3	50.0	16.7
	8 krad	18	—	55.6	44.4	—
	12 krad	20	5.0	45.0	45.0	5.0
	16 krad	15	—	20.0	66.7	13.3
	20 krad	25	—	48.0	52.0	—
	24 krad	32	—	28.1	56.2	15.7
Soaked seeds						
	4 krad	23	8.7	43.5	47.8	—
	8 krad	33	—	24.2	75.8	—
	12 krad	26	—	38.5	34.6	26.9
	16 krad	27	—	85.2	14.8	—
EMS						
	10 mM	1	—	—	—	00.0
	20 mM	5	—	40.0	60.0	—
	30 mM	16	—	25.0	68.8	6.2
	40 mM	35	—	45.7	34.3	20.0
	50 mM	18	—	44.4	55.6	—
	60 mM	22	—	40.9	50.0	9.1

sector resulting from lethality of some of the initial cells of the primordium as explained by Sarvella *et al.* (1962) and Aastveit (1968). Higher segregation ratios obtainable with a change in evironment will be of immense value in mutation breeding. It is evident from the present study that for obtaining higher segregation ratios in lablab, presoaking the seeds prior to gamma irradiation may be more advantageous than treating the dry seeds.

c) Effectiveness and efficiency : Gamma rays were found to be more effective than EMS with respect to induction of chlorophyll mutations. The effectiveness showed an increase in gamma irradiated soaked seeds over that of dry seeds. The high effectiveness of irradiation as compared to chemical mutagens has also been brought out by Siddiq and Swaminathan (1968) and Ramaswamy (1973). Enhancement of effectiveness due to pre-soaking prior to irradiation was also observed by Rao (1968) in cowpea as observed in the present study. Gamma rays were more efficient on the basis of injury but EMS on the basis of lethality and seed sterility. The effectiveness and efficiency were more en-

nanced and manyfold when seeds were presoaked prior to irradiation.

c) Mutation spectrum : The results obtained in the present investigation revealed that the spectrum of induced mutants differ between gamma irradiation and EMS treatments. A low rate of *albina* occurred only in gamma irradiation. The occurrence of *chlorina* and *viridis* mutants was more frequent in all the treatments of gamma rays and EMS. Sjodin (1962) reported that *viridis* was the most common type whereas *albina* was a very rare mutant in *Vicia faba*.

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