J Indian bot Soc Vol 72 (1993) 25-28

A COMPARATIVE STUDY OF MICROSPERMA AND MACROSPERMA LENTILS. I. CHLOROPHYLL MUTATIONS

V.R.K. REDDY, *P.J. THRESIAMMA AND RENU EDWIN
Cytogenetics Laboratory, Department of Botany, Bharathiar University, Coimbatore - 641 046.
(Accepted December 1992)

Using gamma rays and EMS, chlorophyll mutation were induced in microsperma lentil var. LL 1204 and macrosperma lentil var. PRECO-2. Five types of chlorophyll mutants viz. albina, chlorina, xantha, virescens and redina were isolated. Redina was found only in macrosperma lentil. The frequency and spectrum of mutants were increased with the dose and duration of the mutagens. Combined treatments produced wider and high frequency of mutants. Frequency of albina mutants were mainly dependent on dose of gamma rays while other mutants were found more in EMS and combined treatments. Presence of wider spectrum and high frequency of all the mutants and presence of `redina' mutants exclusive in PRECO-2 it was concluded that, macrosperma lentil was more sensitive and more mutable than microsperma lentil LL 1204.

Key Words : Chlorophyll mutants, microsperma, macrosperma lentils.

Chlorophyll mutations have been used as an index in evaluation of mutagens in order to select suitable mutagen at appropriate dose/concentration so as to use them in applied plant breeding. Chlorophyll mutations have been reported in different microsperma lentil cultivars (Sharma & Sharma, 1981; Reddy & Annadurai, 1991 a,b). However, no comparative study with reference to chlorophyll mutations were made in microsperma and macrosperma lentil cultivars. The present communication reports the response of both the small and large seeded lentils to different mutagens in terms of chlorophyll mutations.

MATERIALS AND METHODS

Seeds of microsperma lentil cultivar `LL 1204' and macrosperma lentil cultivar `PRECO-2', procured from IARI, New Delhi, were treated with three doses of gamma rays (15, 25, 35 kR) and three durations of 0.5% aqueous solution of EMS (8,10,12h) (at room temperature) and with combination of gamma rays with EMS for three treatments viz. 15kR + 12 h, 25kR + 10h, 35 kR + 8h. After EMS treatment, seeds were washed throughly in running tap water for one hour and planted immediately in the field along with respective control. For each treatments, one hundred seeds were taken. All M_1 plants were harvested separately and sown as plant progenies in M_2 generation. Chlorophyll mutants were identified according to standard classification. Mutation frequencies were calculated on M_1 plant basis as well as M_2 plant progeny level. Chi-square test was applied to test the goodness of fit for normal to mutant plants.

RESULTS AND DISCUSSION

Chlorophyll mutations were induced in one variety each of microsperma lentil var. LL 1204. and macrosperma var. PRECO-2. Both the varieties differ greatly in seed size (Fig. 1). Mutation frequencies are calculated on M_1 plant on M_2 plant progeny (Table 1). Irrespective of the method used, in both the varieties, the mutation frequency increased with increase in dose of gamma rays (in individual treatment only) and duration of EMS. Similar observations were made earlier in lentil for several type of mutations including chlorophyll mutants (Reddy & Annadurai, 1991 a,b). A total of four chlorophyll mutants in LL 1204 and five mutants in PRECO-2 were isolated (Table - 2). Chlorophyll mutant `Redina' (Figs. 2 & 3) were exclusively found only in PRECO-2 and was absent in LL 1204.

Spectrum and frequency of individual chlorophyll mutants (Table - 2) suggest that, in combined treatments, the frequency and spectrum of chlorophyll mutants were high in both the varieties. Similar observations were made in crops like *Arbidopsis* (Bhatia, 1967), mungbean (Bahl & Gupta, 1982) and lentil (Dixit & Dubey, 1983). Among individual treatments, EMS produced more number of chlorophyll mutants than gamma rays, which further confirms EMS is highly potent mutagen in inducing chlorophyll mutants (Ehrenberg, 1960). Reddy & Gupta (1989) suggested that, EMS preferentially acts on genes responsible for chlorophyll development. However, if a close look to the spectrum of

Received May 1992
* Prof. & Head, Dept. of Botany, Mercy College, Palghat, Kerala.

Table 1: Frequency of chlorophyll mutations on the basis of M_1 plant progeny and M_2 plant population in various mutagenic treatments in two lentil cultivars.

Treatment	LL 1204				PRECO -2			
	Population size		Mutation frequency (%)		Population size		Mutation frequency (%)	
	M_1 plants	M_2 seed-lings	M_1 plants	M_2 seed-lings	M_1 plants	M_2 seed-lings	M_1 plants	M_2 seed-lings
Gamma rays								
15 kR	72	5310	1.38	0.395	64	4814	3.12	0.789
25 kR	66	4911	3.03	0.813	56	4489	5.35	1.4870
35 kR	59	4938	3.38	1.068	52	3914	8.92	2.120
EMS								
8h	57	4289	5.26	1.072	51	3796	9.80	2.265
10h	53	3869	7.54	1.808	46	3481	13.04	3.417
12h	52	3831	7.69	2.530	43	3299	18.60	4.848
G.R.+EMS								
15 kR + 12h	45	3369	11.11	3.857	36	2701	22.22	8.217
25 kR + 10h	49	3619	10.20	3.591	43	2988	18.60	7.494
35 kR + 8h	54	3987	9.25	2.857	46	3116	17.39	5.906
Total								
Gamma rays	197	14619	3.04	0.738	172	13217	4.06	1.414
EMS	162	11989	6.17	1.776	140	10576	8.57	3.451
G.R. + EMS	148	10975	9.45	3.407	125	8905	13.60	7.175

Table 2: Frequency spectrum of chlorophyll mutants (per cent M_2 plants) in different mutagen treatments in two lentil cultivars.

Treatments	LL 1204					PRECO-2					
	Albina	Chlo-rina	Xantha	Vires-cens	Total	Albina	Chlo-rina	Xantha	Vires-cens	Redina	Total
Gamma rays											
15 kR	0.395	-	-	-	0.395	0.789	-	-	-	-	0.789
25 kR	0.488	0.325	-	-	0.813	1.047	0.423	-	-	-	1.470
35 kR	0.636	0.432	-	-	1.068	1.303	0.817	-	-	-	2.120
EMS											
8h	-	0.746	0.326	-	1.072	-	1.001	0.992	0.342	-	2.265
19h	-	0.801	0.646	0.361	1.808	-	1.034	1.005	0.976	0.402	3.417
12h	-	0.835	0.730	0.965	2.530	0.333	1.151	1.273	1.091	1.000	4.848
G.R. + EMS											
15 kR + 12h	0.474	0.979	0.920	1.484	3.857	0.962	1.332	1.777	2.184	1.962	8.217
25 kR + 10h	0.525	0.884	0.801	1.381	3.591	1.271	1.271	1.639	1.874	1.439	7.494
35 kR + 8h	1.128	0.777	0.501	0.451	2.857	1.679	1.492	0.746	0.808	1.181	5.096
TOTAL											
Gamma Rays	0.499	0.239	-	-	-	1.028	0.3985	-	-	-	-
EMS	-	0.792	0.558	0.425	-	0.104	1.059	1.059	0.784	0.444	-
G.R. + EMS	0.728	0.874	0.728	1.075	-	1.325	1.370	1.358	1.583	1.078	-
All Treatment combined	0.407	0.601	0.391	0.449	1.848	0.810	0.871	0.712	0.685	0.553	3.631

individual chlorophyll mutants is given it is clear that, certain mutants are exclusively found in some mutagenic treatments and absent in others. Albina and chlorina mutants were found only in individual treatments of gamma rays and in combined treatments, while xantha, virescens and redina mutants only in individual treatments of EMS and in combined treatments. The presence of certain chlorophyll mutants in some mutagenic treatments and absence in others indicates differences in the availability of mutagenic loci to the mutagen. Presence of albina mutants exclusively in gamma rays treatments, while

Table 3: Segregation pattern of various chlorophyll mutants (pooled data) in M_2 generation in two lentil cultivars (First line is number of segregating rows and ratio of normal to mutant seedlings).

Chlorophyll mutant	P = 3 : 1	
	LL 1204	PRECO-2
Albina	7,457 : 153 (0.90-00.95)	12,777 : 265 (0.70-0.80)
Chlorina	9,686 : 226 (0.80-0.90)	12,883 : 285 (0.50-0.70)
Xantha	7,456 : 147 (0.70-0.80)	11,722 : 233 (0.50-0.70)
Virescens	8,514 : 169 (0.80 - 0.90)	9,676 : 224 (0.90-0.95)
Redina		9.559 : 181 (0.70-0.80)

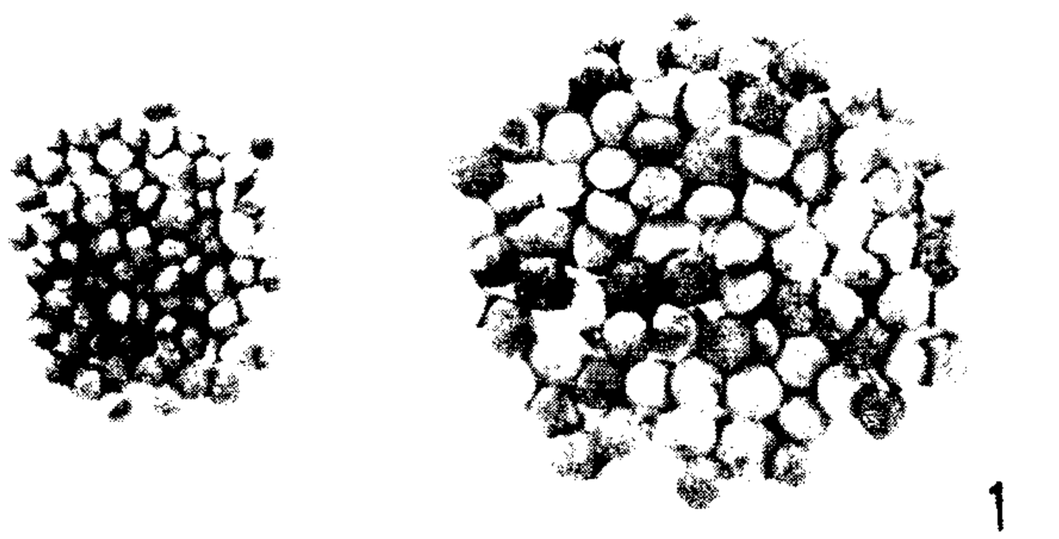

Fig. 1: Seeds of microsperma lentil var. LL 1204 (right) and macrosperma lentil var. PRECO-2 (left).
Figs. 2 & 3 `Redina' chlorophyll mutants (arrow marked).

others in chemical mutagens supports the widely accepted view that physical mutagens produce more albina mutants while chemical mutagens produce other types (Ehrenberg *et al.*, 1961; Reddy & Gupta, 1989).

When compared the spectrum and frequency of chlorophyll mutants of the two varieties, PRECO-2 had wider spectrum and high frequency of mutants. The total frequency of PRECO-2 is almost double than that of LL 1204. Presence of albina mutants in a small frequency at 12h duration treatment of EMS in Preco-2 also suggests the genotypic difference and early mutable nature of PRECO-2. Redina chlorophyll mutants are reported for the first time in lentil. Such type of mutants were, however earlier recorded in hexaploid triticales (Reddy, 1991). Presence of the mutants in PRECO-2 and absence in LL 1204 also supports the view that macrosperma lentil, PRECO-2 is more sensitive to mutagens than microsperma lentil. In LL 1204, the order of highest mutation frequency are chlorina, virescens, albina and xantha, while the order in PRECO-2 is chlorina, albina, xantha, virescens and redina. Such a varietal variation is generally attributed to differences in the mutagenic sensitivity or to a number of genes controlling the chlorophyll development (Bhatia & Swaminathan, 1963; Reddy 1989; Reddy & Gupta, 1989; Reddy & Annadurai, 1991b).

The segregation pattern of all the mutants in both the varieties (Table 3) clearly indicates that, each type of mutants is monogenically controlled with recessive inheritance. High probability values for 3:1 for all the mutants under study indicates gametic or zygotic lethality was considerably low. However, when compare to LL 1204, the probability values in PRECO-2 is slightly low which suggesting lethality was slight more in the later one and which is due to more sensitivity to the mutagens.

P J T is thankful to the Principal, Mercy College, Palghat, Kerala for granting study leave under FIP programme.

REFERENCES

Bahl J R & P K Gupta 1982 Chlorophyll mutations in mungbean (*Vigna radiata* (L.) Wilczek. *Theor Appl Genet* **63** 23-26.

Bhatia C R 1967 Increased mutagenic effect of ethylmethane sulphonate when dissolved in dimethyl sulphoxide. *Mut Res* **4** 375-376.

Bhatia C R & M S Swaminathan 1962 Induced polygenic variability in bread wheat and its bearing on selection procedure. *Z Pflanenzucht* **48** 317-326.

Dixit P & D K Dubey 1983 Seedling mutations induced by separate and combined application of gamma rays and NMU in *Lens culinaris* (Med.) variety T 36. *Pulse Crop Newsletter* **3** 14-17.

Ehrenberg L 1960 Induced mutations on plants - Mechanisms and principles. *Genet Agr* **12** 364-389.

Ehrenberg L, A Gustafsson & U Lundqvist 1961 Viable mutants induced in barley by ionizing radiations and chemical mutagens. *Hereditas* **47** 243-282.

Reddy V R K 1989 Chlorophyll mutants in triticale (x *Triticosecale* Wittmack). *Adv Pl Sci* **2** 161-168.

Reddy V R K 1991 A comparative study of chlorophyll mutations in spring and winter hexaploid triticales. *J Indian Bot Soc* **70** 365-368.

Reddy V R K & P K Gupta 1989 Induced mutations in triticales : Frequency and spectrum of chlorophyll mutations. *Indian J Genet* **49** 183-190.

Reddy V R K & M Annadurai 1991a Mutagenic effects of gamma rays, EMS, sodium azide and their combined treatments in lentil. *J Cytol Genet* **26** 25-29.

Reddy V R K & M Annadurai 1991b Chlorophyll mutants in lentil frequency and segregation. *JCytol Genet* **26** 30-36.

Sharma S K & B Sharma 1981 Induced chlorophyll mutations in lentil. *Indian J Genet* **41** 328-333.