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Induced Mutations in Foxtail Millet (*Setaria italica* Beauv.)

I. Chlorophyll Mutations Induced by Gamma rays, EMS and DES

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Summary. Chlorophyll mutations induced by gamma rays, EMS and DES were studied in foxtail millet (*Setaria italica*), using two cultures, MU-1 (bristled) and MU-2 (non-bristled). No major differences in the mutagenic response of the two cultures were observed. The treatments included four doses of gamma rays (10Kr, 20Kr, 30Kr, 40Kr) and four durations (6 hrs, 12 hrs, 18 hrs, 24 hrs) each of EMS (0.1%) and DES (0.1%). The combined treatments of gamma rays + EMS and gamma rays + DES were also given.

Frequencies of chlorophyll mutations were recorded by three different methods, viz. (a) mutations per cent M_1 plants, (b) mutations per cent M_1 spikes and (c) mutants per cent M_2 plants. No significant differences in the results obtained by these three methods were observed. The frequencies and spectrum of mutations are discussed. *Chlorina* type were most frequent and *viridoalbina* least frequent. *Striata* and *virescens* were also quite common. *Albinos*, reported frequently in other crops, were found to be less frequent in foxtail millet during the present study. Number of sectors per spike were also determined from segregation ratios and only one sector per spike was found at all doses. Efficiency and effectiveness of mutagens were also determined and discussed. The results are also discussed with respect to mutagen specificity.

Introduction

In order to induce and utilize useful mutations for plant breeding, the systematic study of induced mutagenesis in a variety of crop plants is essential. Although extensive studies on induced mutations in barley and similar cereal crops have been undertaken in the past, limited reports are available on millets, such as studies in *Sorghum vulgare* by Sharma (1965), Kapoor (1967), Goud, Nayar and Rao (1970) and Sree Ramulu (1972), and in *Eleusine coracana*, by Goud, Nayar and Rao (1969, 1971). However, no reports on induced mutations are available for foxtail millet (*Setaria italica*), which is an important crop in the arid areas of India.

Material and Methods

Commercial seed of foxtail millet (*Setaria italica*) was selfed for several generations and two distinct cultures, namely (a) bristled (designated as MU-1) and (b) non-bristled (designated as MU-2), were isolated. The seeds has a moisture content of 9.6% in MU-1 and 10.8% in MU-2. The seed from the two cultures was subjected to the following treatments of gamma rays, EMS (ethyl methane sulphonate) and DES (diethyl sulphate):

(1) Irradiation of fresh seed with gamma rays at the following doses: 10Kr, 20Kr, 30Kr, 40Kr.

(2) Soaking of fresh seed in 0.1% aqueous solution of EMS for the following durations: 6 hrs, 12 hrs, 18 hrs, 24 hrs.

(3) Soaking of fresh seed in 0.1% aqueous solution of DES for the following durations: 6 hrs, 12 hrs, 18 hrs, 24 hrs.

(4) Soaking of seed irradiated with 10Kr, 20Kr, 30Kr and 40Kr, in 0.1% aqueous solution of EMS for 6 hrs and 12 hrs.

(5) Soaking of seed irradiated with 10Kr, 20Kr, 30Kr, and 40Kr, on 0.1% aqueous solution of DES for 6 hrs and 12 hrs.

Gamma rays were secured from gamma cell-200 having a 2000 curie Co^{60} source located at the Division of Genetics, IARI, New Delhi. EMS of Eastman Kodak Chemicals, USA, and DES of BDH Chemicals Limited were used for preparing aqueous solutions of chemical mutagens. Treatments with chemical mutagens were given with intermittent shaking. In each treatment, 200 seeds were treated and grown in a randomized block design with four replications. Seeds were initially sown in nursery beds and the surviving seedlings were transplanted on the 25th day. In the plots, the rows were placed two feet apart, with the plants in a row spaced 10 inches apart.

The seed from each spike of a plant in the M_1 generation was harvested separately and the first two spikes from each plant were studied for chlorophyll mutations in progeny tests. Chlorophyll mutations were scored in the nursery beds before transplantation.

Results

The frequencies of chlorophyll mutations were recorded using the following three methods:

(a) mutations per cent M_1 plants,

(b) mutations per cent M_1 spikes

(c) mutants per cent M_2 plants.

In the first two methods, mutations were recorded on the basis of segregation observed in particular

Table 1. *Frequencies of chlorophyll mutations in Setaria italica (MU-1)*

Treatment	Dose/ duration	Population size			Mutation frequency (%)		
		M_1 plants	M_1 spikes	M_2 seedlings	M_1 plants	M_1 spikes	M_2 seedlings
Control	—	80	158	7,900	—	—	—
Gamma rays	10Kr	120	231	30,723	7.5	4.7	1.57
	20Kr	80	149	12,367	10.0	7.3	2.50
	30Kr	61	108	4,752	14.7	9.2	3.11
	40Kr	39	67	2,627	15.3	11.9	4.22
EMS (0.1%)	6 hrs	82	159	20,352	8.5	6.2	2.82
	12 hrs	86	170	17,687	11.6	8.8	3.15
	18 hrs	85	161	13,057	14.1	11.1	3.76
	24 hrs	58	102	6,322	15.4	12.7	4.58
DES (0.1%)	6 hrs	103	191	26,398	6.7	4.1	1.61
	12 hrs	116	219	27,942	8.6	6.8	2.23
	18 hrs	127	241	21.693	11.0	9.1	2.84
	24 hrs	112	203	16,766	13.3	10.3	3.46
Gamma rays (10Kr) + EMS (0.1%)	6 hrs	81	148	10,656	11.1	9.4	3.04
	12 hrs	65	117	17,656	12.3	11.2	3.80
Gamma rays (20Kr) + EMS (0.1%)	6 hrs	51	94	6,175	15.6	11.5	4.25
	12 hrs	53	95	4,940	16.9	12.6	4.87
Gamma rays (30Kr) + EMS (0.1%)	6 hrs	34	66	3,894	17.4	13.6	5.31
	12 hrs	45	81	4,510	17.7	14.8	6.34
Gamma rays (40Kr) + EMS (0.1%)	6 hrs	22	40	1,862	18.1	15.3	7.08
	12 hrs	18	28	1,324	16.6	14.3	8.15
Gamma rays (10Kr) + DES (0.1%)	6 hrs	74	141	11,388	10.8	7.8	2.74
	12 hrs	61	107	8,024	13.1	11.2	3.56
Gamma rays (20Kr) + DES (0.1%)	6 hrs	48	91	5,888	14.5	11.9	3.97
	12 hrs	43	76	5,508	13.9	11.5	4.13
Gamma rays (30Kr) + DES (90.1%)	6 hrs	33	61	3,162	15.1	12.9	5.09
	12 hrs	39	71	4,252	17.9	14.0	5.66
Gamma rays (40Kr) + DES (0.1%)	6 hrs	22	37	1,786	18.1	15.7	6.83
	12 hrs	16	28	947	17.6	14.2	7.81

progeny (Figs. 1, 2), while in the third method, mutants were scored from the total M_2 population. The frequencies of chlorophyll mutations on the basis of the three methods are presented in Tables 1 and 2 for cultures MU-1 and MU-2, respectively.

The spectrum of chlorophyll mutations was classified following Gustafsson (1940) and the following different kinds were identified:

(i) *albino*: seedlings white in colour; survive up to three leaf stage and die within 5—15 days (Figs. 1, 2).

(ii) *virescens*: seedlings yellowish-green in colour, gradually changing to light green and finally to normal.

(iii) *chlorina*: seedlings slow growing and yellowish-green in colour; the colour persists till the seedlings die within 30 days.

(iv) *alboviridis*: seedlings with leaves white at the tip and yellowish-green in the lower region (Figs. 3, 4).

(v) *viridoalbina*: seedlings with leaves white at the base and yellowish-green in the upper region.

(vi) *striata*: seedlings with leaves characterized by the presence of longitudinal zones of normal green, alternating with yellow (Fig. 5).

The frequencies of the various kinds of chlorophyll mutations obtained in culture MU-1 and culture MU-2 are presented in Table 3 and Table 4, respectively. The frequencies of different kinds of chlorophyll mutations were recorded only as per cent M_2 mutant seedlings, following Gaul's (1964) suggestion that this is the most reliable estimate of mutation frequencies at different doses of treatment.

To ascertain the number of sectors forming a spike, the segregation data for chlorophyll mutations were analysed, using the data for four different doses of gamma rays pertaining to culture MU-1 only. The number of sectors was worked out as expected ratio/ observed ratio. The expected segregation ratio was taken to be 25% on the basis of a 3:1 ratio. The analysis is presented in Table 5 and suggests that only one sector is found at the different doses of treatment.

Efficiency and effectiveness of different mutagens were worked out using formulae by Konzak, Nilan,

Table 2. *Frequencies of chlorophyll mutations in Setaria italica (MU-2)*

Treatment	Dose/ duration	Population size			Mutation frequency (%)		
		M_1 plants	M_1 spikes	M_2 seedlings	M_1 plants	M_1 spikes	M_2 seedlings
Control	—	80	157	7,850	—	—	—
Gamma rays	10Kr	106	193	29,722	8.4	5.1	1.70
	20Kr	101	187	17,268	10.8	7.7	2.22
	30Kr	92	171	14,053	14.1	10.4	3.46
	40Kr	48	81	4,797	16.6	12.6	4.69
EMS (0.1%)	6 hrs	117	218	26,984	8.5	6.4	1.77
	12 hrs	134	251	28,265	10.3	8.9	2.54
	18 hrs	137	247	20,925	13.1	10.9	3.47
	24 hrs	128	238	17,616	16.4	13.0	4.71
DES (0.1%)	6 hrs	126	237	30,094	7.1	4.9	1.48
	12 hrs	123	231	27,268	9.7	6.9	2.13
	18 hrs	109	207	20,162	12.3	9.6	2.89
	24 hrs	112	204	17,012	13.4	10.4	3.60
Gamma rays (10Kr) + EMS (0.1)%	6 hrs	64	116	8,626	10.9	8.6	2.97
	12 hrs	62	103	6,027	14.5	10.3	3.91
Gamma rays (20Kr) + EMS (0.1%)	6 hrs	51	92	5,704	13.7	11.9	4.24
	12 hrs	57	97	5,159	15.7	14.3	5.03
Gamma rays (30Kr) + EMS (0.1%)	6 hrs	47	84	2,912	17.0	14.2	5.63
	12 hrs	30	52	1,431	16.6	15.0	5.93
Gamma rays (40Kr) + EMS (0.1%)	6 hrs	27	47	1,246	22.2	17.1	6.58
	12 hrs	23	41	847	21.7	17.4	7.31
Gamma rays (10Kr) + DES (0.1%)	6 hrs	94	178	14,643	9.5	7.3	2.64
	12 hrs	78	137	10,412	13.1	11.6	3.98
Gamma rays (20Kr) + DES (0.1%)	6 hrs	66	119	6,987	13.6	10.7	4.12
	12 hrs	57	105	3,787	14.0	12.1	5.06
Gamma rays (30Kr) + DES (0.1%)	6 hrs	51	87	3,152	15.6	13.7	5.61
	12 hrs	34	61	2,493	17.6	14.7	6.77
Gamma rays (40Kr) + DES (0.1%)	6 hrs	34	52	1,979	17.6	15.4	7.33
	12 hrs	31	54	1,504	16.1	14.3	7.91

Fig. 1. A spike progeny showing segregation for albino mutant

Fig. 3. A spike progeny showing segregation for *viridoalbina* mutant;

Fig. 2. Leaves from control and albino seedlings

Fig. 4. A seedling of *viridoalbina* and a control seedling

Fig. 5. A *striata* seedling and a control seedling

Fig. 2

Fig. 4

Fig. 5

Figs. 1—5. Chlorophyll mutations in *Setaria italica*

Table 3. *Frequencies of different kinds of chlorophyll mutations in Setaria italica (MU-1)*

Treatment	Dose/ duration	Mutation frequencies (mutants, % M_2 seedlings)					
		albino	virescens	chlorina	albo-viridis	virido-albina	striata
Control		—	—	—	—	—	—
Gamma rays	10Kr	0.22	0.35	0.66	0.09	—	0.25
	20Kr	0.44	0.11	0.50	—	—	0.48
	30Kr	0.46	0.99	0.57	—	—	1.09
	40Kr	0.27	0.72	1.52	0.42	—	1.29
EMS (0.1%)	6 hrs	0.15	1.00	0.45	0.17	0.26	0.79
	12 hrs	0.16	1.03	0.66	0.46	—	0.83
	18 hrs	0.15	1.13	1.31	—	—	1.18
	24 hrs	0.20	1.61	0.92	0.27	0.14	1.44
DES (0.1%)	6 hrs	—	0.48	0.62	0.11	—	0.40
	12 hrs	0.15	0.84	0.65	—	—	0.59
	18 hrs	0.21	0.80	1.10	0.07	—	0.66
	24 hrs	0.13	0.89	1.21	0.08	0.10	1.06
Gamma rays (10Kr) + EMS (0.1%)	6 hrs	0.44	0.30	1.54	—	—	0.76
	12 hrs	0.25	0.76	1.91	—	—	0.89
Gamma rays (20Kr) + EMS (0.1%)	6 hrs	0.28	1.36	1.17	—	0.33	1.23
	12 hrs	0.22	1.31	1.76	0.30	—	1.28
Gamma rays (30Kr) + EMS (0.1%)	6 hrs	0.36	1.16	2.11	0.31	—	1.39
	12 hrs	0.20	1.69	2.31	—	—	2.15
Gamma rays (40Kr) + EMS (0.1%)	6 hrs	—	1.21	3.60	—	—	2.20
	12 hrs	1.06	1.21	2.57	—	—	3.32
Gamma rays (10Kr) + DES (0.1%)	6 hrs	0.20	0.50	1.06	—	0.15	0.83
	12 hrs	0.67	0.55	1.69	—	—	0.65
Gamma rays (20Kr) + DES (0.1%)	6 hrs	0.36	0.41	2.01	0.24	—	0.97
	12 hrs	0.22	1.24	1.59	—	—	1.11
Gamma rays (30Kr) + DES (0.1%)	6 hrs	0.51	0.98	1.80	0.35	—	1.45
	12 hrs	0.33	0.99	2.45	—	0.24	1.67
Gamma rays (40Kr) + DES (0.1%)	6 hrs	—	0.61	3.19	—	—	3.02
	12 hrs	—	0.88	4.01	—	—	3.30

Wagner and Foster (1965). The results are presented in Table 6. It is obvious that efficiency as well as effectiveness goes down with increasing doses of gamma rays or with increases in the duration of treatment with chemical mutagens. The efficiency goes down at higher doses because of an increase in lethality without any appreciable increase in mutation frequency. Similar, effectiveness declines because the mutation frequency does not increase in the same proportion with increases in dose or duration.

The data on the frequencies of different types of chlorophyll mutation were pooled for each mutagen in order to study the specificity of mutagens. The pooled mutation frequencies are presented in Table 7.

Discussion

Among millets, radiation-induced chlorophyll mutations have been studied in *Eleusine coracana* by Goud, Nayar and Rao (1969) and in *Sorghum vulgare* by Goud, Nayar and Rao (1970) and Sree Ramulu (1972). Similarly EMS-induced chlorophyll mutations in *Sorghum vulgare* were reported by Kapoor (1967). These reports mainly dealt with mutation frequencies and several other aspects were not studied. The present study is the first attempt to study induced chlorophyll mutations in *Setaria italica*, using radiation as well as chemical mutagens.

1. *Methods for Scoring Mutation Frequencies*

During the present study mutation frequencies were recorded using three methods, namely, (a) mutations per cent M_1 plants, (b) mutations per cent M_1 spikes and (c) mutants per cent M_2 plants. From studies in barley and similar crops, Gaul (1964) suggested that mutation frequencies scored by methods (a) and (b) would be relatively underestimated at higher doses. This was considered to be due to three factors: (i) a reduction in the number of spikes per plant at higher doses; (ii) a reduction in the number of seeds obtained from a single spike at higher doses; and (iii) a reduction in the number of sectors in each spike at higher doses. While all three factors will contribute to an underestimate of mutation frequencies as per cent M_1 plants, the first factor, i.e. number of spikes per plant, will not contribute to the underestimate if frequencies are worked out as per cent M_1 spikes. Gaul (1964), however, argues that none of these fac-

Table 4. *Frequencies of different kinds of chlorophyll mutations in Setaria italica (MU-2)*

Treatment	Dose/ duration	Mutation frequencies (mutants, % M_2 seedlings)					
		albino	virescens	chlorina	albo-viridis	virido-albina	striata
Control	—	—	—	—	—	—	—
Gamma rays	10Kr	0.31	0.29	0.57	—	0.12	0.43
	20Kr	0.33	0.42	0.87	—	—	0.57
	30Kr	0.33	0.67	1.37	0.15	—	0.95
	40Kr	0.58	0.54	2.11	0.33	—	1.13
EMS (0.1%)	6 hrs	0.23	0.59	0.53	—	—	0.42
	12 hrs	0.31	0.57	0.71	0.21	—	0.73
	18 hrs	0.32	0.71	1.31	0.25	—	0.88
	24 hrs	0.14	1.22	1.79	0.22	0.10	1.24
DES (0.1%)	6 hrs	0.12	0.50	0.46	0.11	—	0.31
	12 hrs	0.23	0.59	0.73	—	0.08	0.50
	18 hrs	0.20	1.07	0.93	—	—	0.60
	24 hrs	0.27	1.05	1.27	—	—	1.01
Gamma rays (10Kr) + EMS (0.1%)	6 hrs	0.28	0.78	1.08	—	—	0.85
	12 hrs	0.22	0.68	1.73	0.28	—	1.01
Gamma rays (20Kr) + EMS (0.1%)	6 hrs	0.30	0.63	1.61	—	0.26	1.44
	12 hrs	0.60	0.79	2.17	—	—	1.57
Gamma rays (30Kr) + EMS (0.1%)	6 hrs	0.38	1.37	2.16	0.27	—	1.44
	12 hrs	0.63	1.40	2.38	—	—	1.54
Gamma rays (40Kr) + EMS (0.1%)	6 hrs	—	1.04	2.65	0.72	—	2.17
	12 hrs	—	2.13	3.42	—	—	1.77
Gamma rays (10Kr) + DES (0.1%)	6 hrs	0.49	0.31	0.80	0.23	—	0.81
	12 hrs	0.53	0.61	1.60	—	—	1.24
Gamma rays (20Kr) + DES (0.1%)	6 hrs	0.39	0.73	1.70	0.37	—	1.03
	12 hrs	0.37	0.92	2.06	0.29	—	1.43
Gamma rays (30Kr) + DES (0.1%)	6 hrs	—	1.54	2.70	—	0.29	1.30
	12 hrs	0.48	1.85	2.73	—	—	1.72
Gamma rays (40Kr) + DES (0.1%)	6 hrs	—	2.02	2.73	—	0.56	2.07
	12 hrs	0.60	1.26	4.05	—	—	2.00

tors will be effective, if mutation frequencies are worked out as mutants per cent M_2 plants.

Applying these arguments in the present study of foxtail millet, it should be realized that only the first two spikes of a plant were used to raise the M_2 population and, therefore, a reduction in the number of spikes at higher doses would not influence the results. Moreover, since a large number of seeds are obtained from a single spike, reduced fertility at higher doses would still give enough seed to show segregation, if a mutation were present. Lastly, there is evidence from the present study that perhaps a spike consists of a single sector in foxtail millet, so that the question of a reduction in the number of sectors in a spike at higher doses does not arise. It can, therefore, be suggested that the three factors discussed by Gaul (1964), in connection with the relative underestimation of mutation frequency at high doses when recorded as per cent M_1 plants or as per cent M_1 spikes, do not hold good in the case of foxtail millet, and that each of the three methods should give an equally reliable estimate of mutation frequencies in foxtail millet at all doses.

However, an important factor, which might have contributed to a biased estimate of mutation frequency at high doses in the present study, is that only the first two spikes of a plant were used to raise the M_2 population. Gaul (1961), using barley, and Osone (1963), in rice, recorded higher mutation frequencies in the later tillers. The obvious explanation for such a difference would be diplontic selection during growth. Contradictory reports in this connection are also available. For instance, lower mutation frequen-

Table 5. *Number of sectors per spike at different doses of gamma rays in Setaria italica (MU-1)*

Treatment	Number of seedlings examined	Number of mutant seedlings	Segregation ratio (%)	Number of sectors per spike*
10Kr	2,178	483	22.7	1.10
20Kr	1,593	309	19.3	1.29
30Kr	678	148	21.8	1.14
40Kr	514	111	21.1	1.16

* Number of sectors $= \frac{\text{expected segregation ratio (25\%)}}{\text{observed segregation ratio}}$

Table 6. *Efficiency and effectiveness of different mutagens in Setaria italica (MU-1, MU-2)*

Treatment	Dose/ duration	Culture MU-1		Culture MU-2	
		efficiency	effectiveness	efficiency	effectiveness
Gamma rays	10Kr	—*	0.16	0.07	0.17
	20Kr	0.10	0.13	0.09	0.11
	30Kr	0.08	0.10	0.11	0.12
	40Kr	0.07	0.11	0.08	0.12
EMS (0.1%)	6 hrs	0.12	4.70	0.10	2.95
	12 hrs	0.24	2.63	1.21	2.12
	18 hrs	0.25	2.09	4.96	1.93
	24 hrs	0.10	1.91	0.47	1.96
DES (0.1%)	6 hrs	0.30	2.67	0.31	2.47
	12 hrs	—*	1.86	0.26	1.78
	18 hrs	—*	1.58	0.13	1.18
	24 hrs	—*	1.33	0.21	1.50
Gamma rays (10Kr) + EMS (0.1%)	6 hrs	0.12	—	0.04	—
	12 hrs	0.10	—	0.07	—
Gamma rays (20Kr) + EMS (0.1%)	6 hrs	0.09	—	0.07	—
	12 hrs	0.10	—	0.09	—
Gamma rays (30Kr) + EMS (0.1%)	6 hrs	0.09	—	0.09	—
	12 hrs	0.12	—	0.08	—
Gamma rays (40Kr) + EMS (0.1%)	6 hrs	0.10	—	0.08	—
	12 hrs	0.10	—	0.09	—
Gamma rays (10Kr) + DES (0.1%)	6 hrs	0.08	—	0.09	—
	12 hrs	0.08	—	0.10	—
Gamma rays (20Kr) + DES (0.1%)	6 hrs	0.07	—	0.08	—
	12 hrs	0.07	—	0.09	—
Gamma rays (30Kr) + DES (0.1%)	6 hrs	0.07	—	0.08	—
	12 hrs	0.10	—	0.09	—
Gamma rays (40Kr) + DES (0.1%)	6 hrs	0.09	—	0.10	—
	12 hrs	0.09	—	0.10	—

* In these treatments, survival was either equal to or improved upon control.

cies in the ontogenetically later tillers were recorded by Sarvella, Nilan and Konzak (1962) and by Frydenberg and Jacobsen (1966) in barley and by Reddy and Reddy (1971) in rice. No differences in mutation frequencies between early and late tillers were observed by Frydenberg, Doss and Sandraer (1964) in barley or by Khan and Doll (1968) in wheat.

In the present study, mutation frequencies increased with increases in dose or duration and the same trend was observed irrespective of the method used (Tables 1, 2). A critical analysis, however, indicates that at higher doses mutations recorded as per cent M_1 plants did not show an increase in the same proportion as when scored as mutants per cent M_2 plants. This suggests a relative underestimation of mutation frequency (mutations per cent M_1 plants). No suitable explanation for this can be given in view of the above discussion.

2. *Mutation Frequencies*

The pooled data presented in Table 7 indicate clearly that the response of the two cultures (MU-1 and MU-2) in chlorophyll mutations was similar except for minor differences. It is obvious that the total frequencies of chlorophyll mutations in the two cultures were not very different (3.09% for MU-1 and 2.97% for MU-2), though slightly higher in culture MU-1. Whether such a difference could be attributed to a difference in the moisture content of the seed of the two cultures would be difficult to say. It is also obvious that the mutation frequencies in general were much higher in the combined treatments than in the treatments with individual mutagens. However, no synergistic effect was observed.

The mutation frequencies obtained during the present study in foxtail millet (*Setaria italica*) can be compared with those reported earlier in other millet crops. Goud, Nayar and Rao (1969) reported a mutation frequency as high as 15.6% with 20Kr dose in a particular variety of ragi (*Eleusine coracana*). Since this frequency was recorded as per cent M_1 plants, this will amount to only 4%—5% mutants per cent M_2 plants. In jowar (*Sorghum vulgare*), Goud, Nayar and Rao (1970) reported a mutation frequency of 6.23% mutants per cent M_2 plants. In the present study, at 20Kr, the mutation frequencies were 2.50% in MU-1 and 2.22% in MU-2. At 40Kr, these frequencies were 4.22% in MU-1 and 4.69% in MU-2.

Table 7. *Frequencies of different kinds of chlorophyll mutations from pooled data in Setaria italica (MU-1, MU-2)*

Treatment	Culture	Mutation frequencies (mutants, % M_2 seedlings)*						
		albino	virescens	chlorina	albo-viridis	virido-albina	striata	Total
Gamma rays	MU-1	0.30 (14.4)	0.61 (29.3)	0.66 (31.7)	0.08 (3.8)	—	0.44 (21.1)	2.08 (100.0)
	MU-2	0.34 (13.9)	0.42 (17.2)	0.93 (38.2)	0.06 (2.5)	0.05 (2.1)	0.62 (25.9)	2.43 (100.0)
EMS	MU-1	0.16 (4.8)	1.10 (33.1)	0.76 (22.8)	0.23 (9.9)	0.11 (3.3)	0.96 (28.9)	3.33 (100.0)
	MU-2	0.26 (8.9)	0.73 (24.9)	1.00 (34.1)	0.15 (5.1)	0.02 (0.7)	0.77 (26.3)	2.93 (100.0)
DES	MU-1	0.12 (4.9)	0.74 (30.5)	0.85 (35.1)	0.06 (2.4)	0.02 (0.8)	0.64 (26.4)	2.42 (100.0)
	MU-2	0.20 (8.5)	0.75 (31.5)	0.78 (33.1)	0.03 (1.3)	0.02 (0.9)	0.57 (24.3)	2.35 (100.0)
Gamma rays + EMS	MU-1	0.32 (7.0)	0.98 (24.4)	1.89 (41.3)	0.07 (1.5)	0.03 (0.7)	1.28 (28.0)	4.57 (100.0)
	MU-2	0.33 (7.6)	0.86 (19.8)	1.73 (39.9)	0.11 (2.5)	0.05 (1.2)	1.26 (29.0)	4.34 (100.0)
Gamma rays + DES	MU-1	0.34 (8.4)	0.70 (17.3)	1.75 (43.2)	0.06 (1.5)	0.07 (1.8)	1.13 (27.8)	4.05 (100.0)
	MU-2	0.42 (10.0)	0.76 (18.1)	1.66 (39.4)	0.14 (3.3)	0.04 (1.0)	1.17 (27.8)	4.21 (100.0)
Total	MU-1	0.22 (7.1)	0.82 (26.5)	1.07 (34.7)	0.10 (3.2)	0.94 (1.4)	0.83 (27.0)	0.39 (100.0)
	MU-2	0.29 (9.7)	0.69 (23.5)	1.09 (36.7)	0.09 (3.0)	0.03 (1.0)	0.76 (26.2)	2.97 (100.0)

* Figures in parentheses are relative per cent values.

These frequencies are, therefore, lower than the highest mutation frequencies recorded in some other varieties. The greatest mutation frequencies during the present study were recorded in combined treatments of gamma rays — DES. These frequencies were 7.81% in MU-1 and 7.91% in MU-2.

3. *Mutation Spectrum and Mutagen Specificity*

In both the cultures of foxtail millet, *chlorina* mutations were most frequent and *viridoalbina* were least frequent, so much so that no *viridoalbina* mutations were obtained with gamma rays in culture MU-1. The order of relative frequencies of different kinds of chlorophyll mutation can be represented as *chlorina* > *striata* > *virescens* ⋙ *albino* > *alboviridis* > *viridoalbina*. Such a relationship was observed in both the cultures, confirming the kind of mutation response given by foxtail millet. There were, however, minor differences in the response of the two cultures.

In culture MU-1, in the combined treatments, *striata* was more frequent than *virescens*, while in single treatments, *virescens* was more frequent than *striata*. With EMS, in culture MU-1, *virescens* and *striata* exceeded the frequency of *chlorina*, which otherwise was highest in most of the treatments. This situation was not noted in culture MU-2. In culture MU-2, in DES treatments, *striata* was more frequent than *virescens* (Table 7).

It is generally believed that ionizing radiations produce a high frequency of the *albino* type of chlorophyll mutation and the chemical mutagens produce a high frequency of other types, such as *viridis*, *chlorina* and *xantha* (Gustafsson, 1963). Gaul (1964) showed in barley that, with X-rays as well as EMS treatments, *viridis* was more frequent than *albino*. Goud, Nayar and Rao (1969) studied frequencies of radiation-induced chlorophyll mutations in three varieties of ragi (*Eleusine coracana*) and observed that *viridis* was more frequent than *albino* or *xantha* in all three varieties. They also recorded major differences in the mutagenic response of the three varieties.

Basu and Basu (1969) studied frequencies of chlorophyll mutations in rice, induced by X-rays, P^{32} and S^{35}, and observed that the order of the frequencies of different kinds of chlorophyll mutations was *albino* > *virescens* > *xantha* > rare types (*striata*, *chlorina* etc.). Goud, Nayar and Rao (1970) observed in jowar that the relative proportions of different kinds of chlorophyll mutation could differ in different varieties: in one variety *albino* was most frequent, while in another variety *chlorina* was most frequent. They also noticed differences in the total frequencies of

chlorophyll mutations in two varieties of jowar. In the present study, only minor differences were observed in the frequencies and spectrum of chlorophyll mutations in the two cultures of foxtail millet.

4. *Efficiency and Effectiveness of Mutagens*

The efficiency of gamma rays decreased with increasing doses in culture MU-1, but increased in culture MU-2 until the dose reached 30Kr. There was a decline in efficiency at the 40Kr dose in culture MU-2 also. The efficiency of EMS increased with increases in duration in both the cultures up to 18 hrs, after which there was again a decrease in efficiency at 24 hrs duration. The increase in efficiency of EMS in culture MU-2, particularly at 18 hrs duration, was rather conspicuous. Under DES treatments, the survival improved in culture MU-1, while efficiency decreased with increasing duration in culture MU-2. The efficiency of combined treatments was generally low compared with that of EMS and DES, but was close to that of gamma rays. There were no major differences between the efficiencies of different combined treatments. It is obvious that there are differences in the efficiency of different mutagens in the two cultures. These differences are mainly attributable to the differences in the levels of lethality observed between the two cultures in some treatments. Mutation frequencies did not show any major differences.

The effectiveness generally decreased with increases in dose and duration of mutagens, because no corresponding proportionate increase in mutation frequencies was recorded.

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