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A Comparison of Mutation Induction in Diploid and Tetraploid Rice

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Summary. A comparative assessment of the frequency and spectrum of chlorophyll mutations in the M_2 and M_3 of three diploids and one tetraploid of rice after X-irradiation was made. As well as a linear relationship of the frequency of mutations with the dose of mutagen, a saturation effect was also evident. Among the three diploids, the maximum frequency of mutations was observed in T. N. 1, followed by G. E. B. 24 and A. S. D. 8. The diploid of G. E. B. 24 showed a higher frequency of mutations than the tetraploid when measured on the M_1 plant basis only. The diploids showed a higher rate of mutations in the M_2 than in the M_3.

There was no relationship between the frequency of different types of chlorophyll mutations and the dose of X-rays. *Albina* occurred in greater proportion than the other types of mutations in the diploids as well as in the tetraploid. The diploid showed a wider spectrum in the M_2, whereas the tetraploid recorded the maximum types of mutations in the M_3. Striking differences in the spectrum and the relative frequency of each type were observed among the three diploids and also between the diploid G. E. B. 24 and its tetraploid.

The frequency and spectrum of induced mutations in the diploid and autotetraploid provided an insight into the genetic behaviour of the diploid and autotetraploid of G. E. B. 24, indicating that the genetics of diploidisation of the existing diploid rice may give evidence on the nature of the evolutionary pathway.

Introduction

Advances in knowledge of a wide range of physical and chemical mutagens have accelerated the study of induced mutagenesis. In a recent review, Gustafsson and Gadd (1966) listed over 350 papers on induced mutations in rice, *Oryza sativa*, alone. Beginning with Stadler (1929), many workers have studied the influence of polyploidy on the physiological and genetical effects of radiation (Bhaskaran and Swaminathan, 1962). MacKey (1954) found that polyploidy is an advantage rather than a handicap in mutation breeding in view of the superior ability of polyploids to tolerate chromosome aberrations and the buffering action of genes. Since then, the relationship between polyploidy and response to mutagenic treatments has been considered in detail by many authors (Swaminathan, 1965). The present study was taken up with three diploids and one tetraploid of the *indica* type of *Oryza sativa* L. to make a comparative assessment of the frequency and spectrum of chlorophyll mutations induced by X-rays.

Material and Methods

The material comprised three diploid improved strains (A. S. D. 8, T. N. 1 and G. E. B. 24) and the autotetraploid of G. E. B. 24 of *indica* rice. The autotetraploid of G. E. B. 24 was induced by colchicine treatment and is being maintained for fifteen years. Dry dormant seeds with a moisture content of 11.5 per cent were treated with X-rays at doses of 30 to 65 KR. A Phillips' X-ray machine operated at 50 KV delivering the dose rate of 500 R/Sec. was used. Three of the first formed spikes of each M_1 plant were bagged and seeds were pooled to raise M_2. Seed samples from randomly selected M_2 plants were grown in M_3 in family rows. The chlorophyll mutations were scored in M_2 and M_3 when the seedlings were 7 to 12 days old.

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Results

Frequency of chlorophyll mutations: A linear relationship of mutation frequency with dose and also a saturation effect were evident in M_2 in the three diploids. The maximum frequency of mutations in M_2

Table 1. *Frequency of chlorophyll mutations in the M_3 of X-irradiated diploids, T. N. 1, A. S. D. 8 and tetraploid of G. E. B. 24*

Treatment* (KR)	Mutation frequency (%)				
	T(N) 1		A.S.D. 8		G.E.B. 24 (tetraploid)
	M_2 plant progeny basis	M_2 spike basis	M_2 plant progeny basis	M_2 spike basis	M_2 plant progeny basis
Control	—	—	—	—	—
30	35.0	12.9	40.0	30.0	48.0
35	15.0	6.7	45.0	21.3	—
40	25.0	9.6	75.0	36.3	40.0
45	45.0	16.3	45.0	19.4	—
50	30.0	10.3	60.0	21.7	25.0
55	20.0	7.5	50.0	25.8	—
60	50.0	28.8	30.0	12.9	21.0
65	35.0	15.1	25.0	11.3	—

* In each treatment, 40 M_2 plant progenies and 140 M_2 spikes in T. N. 1, 40 M_2 plant progenies and 120 M_2 spikes in A. S. D. 8 and 100 M_2 plant progenies in G. E. B. 24 were scored.

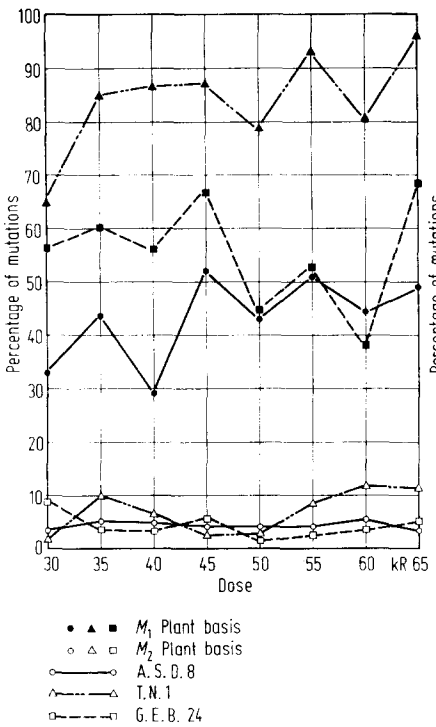

Fig. 1. Frequency of M_2 chlorophyll mutations in diploid rice

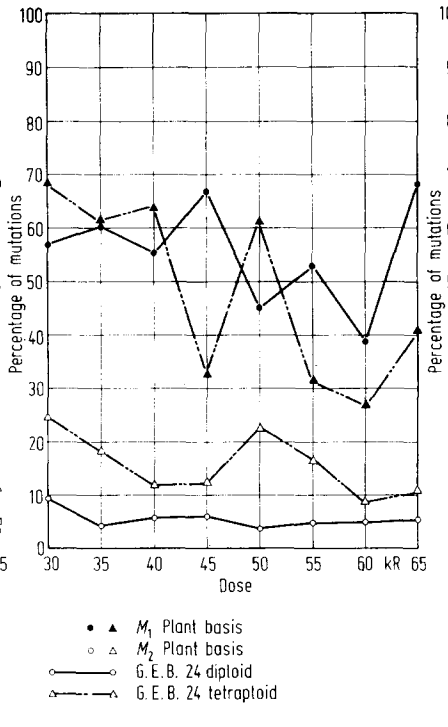

Fig. 2. Frequency of M_2 chlorophyll mutations in diploid and tetraploid rice (G.E.B. 24)

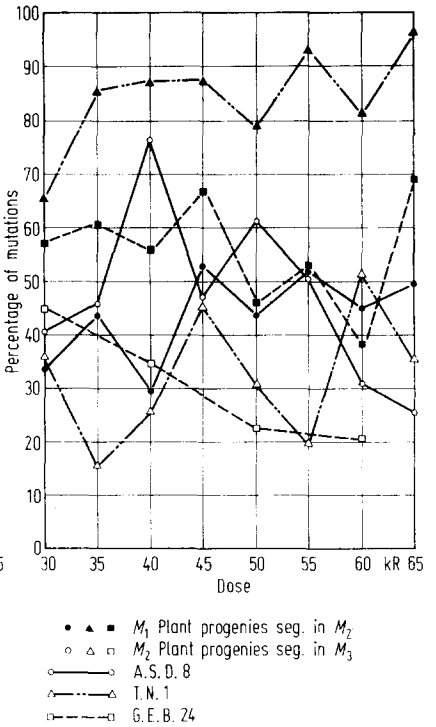

Fig. 3. Frequency of chlorophyll mutations in the M_2 and M_3 generations of diploid rice

was observed in T. N. 1 followed by G. E. B. 24 and A. S. D. 8 (Fig. 1). Among the diploids, T. N. 1 and G. E. B. 24 showed a higher frequency of mutations in M_2 than in M_3, whereas in A. S. D. 8 an opposite trend was noticed (Fig. 3). The mutation rate was higher in M_3 when adjudged on an M_2 plant progeny basis rather than on an M_2 spike basis in A. S. D. 8 and T. N. 1 (Table 1). The maximum frequency of mutations was observed in A. S. D. 8 in M_3.

The diploid of G. E. B. 24 gave a higher rate of chlorophyll mutations than the tetraploid on the M_1 plant basis, whereas a reverse trend was noticed when the mutation rate was computed on an M_2 plant basis (Fig. 2).

Spectrum of chlorophyll mutations: The different types of chlorophyll mutations were *albina*, *xantha*, *chlorina* and *xanthalba*. No relationship was observed between the frequency of different types of chlorophyll mutations and the dose of *X*-rays (Table 2 and Fig. 4 and 5). *Albina* mutations occurred in greater proportion than the other types in diploids (A. S. D. 8 and G. E. B. 24) as well as tetraploid rice. The occurrence of different types of mutations in the M_2 of the three diploids was in the following order:

A. S. D. 8 = *albina* > *chlorina* > *xantha* > *xanthalba*
T. N. 1 = *xantha* > *albina* > *xanthalba* > *chlorina*
G. E. B. 24 = *albina* > *xantha* > *chlorina* > *xanthalba*.

In the diploid of G. E. B. 24, a maximum of four types were observed in M_2 as compared with *albina* and *xantha* only in the tetraploid. *Albina* mutations occurred more often than the rest in the diploid of G. E. B. 24. In the tetraploid, the incidence of *xantha* was higher than of *albina*.

The spectrum was wider in the M_2 than in the M_3 in the diploids, whereas a reverse trend was observed in the tetraploid (Fig. 4). Only two types, *albina* and *xantha* in T. N. 1, and three types, *albina*, *xantha* and *chlorina* in A. S. D. 8 and G. E. B. 24 (Fig. 3 and 4), occurred in the M_3 as against the four types in the M_2. Conversely, in the tetraploid three types of mutations, *albina*, *xantha* and *chlorina*, were observed in the M_3, whereas only the two types, *albina* and *xantha*, occurred in the M_2.

Discussion

Frequency of chlorophyll mutations: The frequency of mutations in the M_2 increased in the diploids with increase in dose, showing a linear relationship between the *X*-ray dose and the mutation frequency. This was strictly the case with T. N. 1. A linear or exponential dose relationship was obtained in a series of experiments after irradiation in barley (Gaul, 1960) and rice (Fujii, 1962; Bekendem, 1961). A critical comparison of the chlorophyll mutations also revealed that the mutation rate increased with an increase in dose up to a certain dose level only, beyond which it decreased (Fig. 1). The frequency of mutations in the M_2 increased at middle doses in A. S. D. 8 in the diploid as well as in the tetraploid. The mutation

Table 2. *Frequency and relative percentage of different types of chlorophyll mutations in the M_2 and M_3 of X-irradiated A. S. D. 8 and G. E. B. 24 (diploid)*

Treatment (KR)	M_2(%)				M_3(%)		
	Albina	*Xantha*	*Chlorina*	*Xanthalba*	*Albina*	*Xantha*	*Chlorina*
A. S. D. 8							
30	62.5	21.5	7.1	8.9	94.3	5.7	—
35	71.2	9.9	10.8	8.1	47.6	33.3	19.1
40	73.3	20.0	—	6.7	30.0	57.5	12.5
45	67.1	—	25.3	7.6	64.7	35.3	—
50	69.8	15.9	7.9	6.4	91.5	8.5	—
55	61.2	13.4	14.9	10.5	80.9	19.1	—
60	71.2	6.4	21.3	1.1	71.4	28.6	—
65	52.2	20.0	21.1	6.7	78.9	21.1	—
G. E. B. 24							
30	82.8	13.3	3.9	—	69.3	13.3	17.4
35	76.5	20.2	3.3	—	—	—	—
40	76.8	21.7	1.5	—	85.4	11.0	3.6
45	87.2	11.3	1.5	—	—	—	—
50	78.6	12.9	7.1	1.4	62.5	37.5	—
55	88.7	7.5	3.8	—	—	—	—
60	87.1	12.9	—	—	37.9	44.8	17.2
65	84.6	14.5	0.9	—	—	—	—

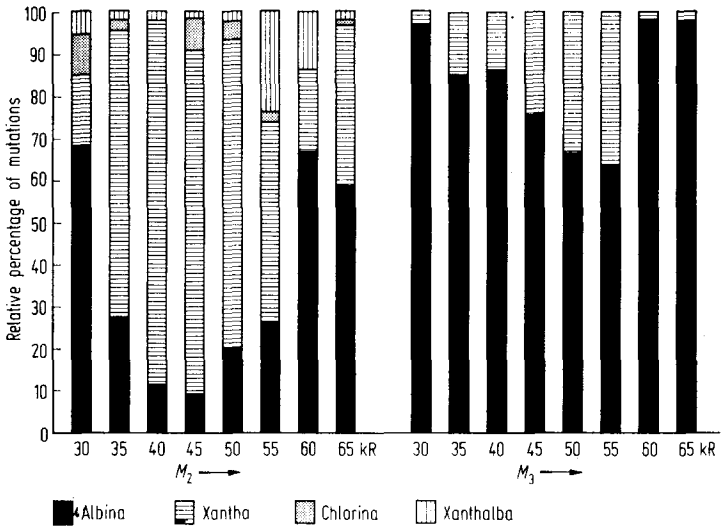

Fig. 4. Spectrum of chlorophyll mutants in the M_2 and M_3 of T. N. 1

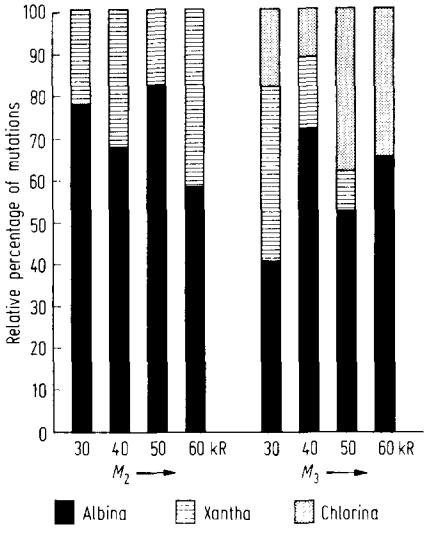

Fig. 5. Spectrum of chlorophyll mutants in the M_2 and M_3 of tetraploid of G.E.B. 24

rate in the M_3 of the three diploids also showed an increase at middle doses and a considerable decrease at high dose. Matsuo (1962), Kawai (1963) and Yamaguchi (1964) observed that mutation frequency reached a maximum at middle doses and decreased at high doses. Ehrenberg and Nybom (1954) using barley, Wellensiek (1965) in peas and Hildering (1963) in tomato also reported the occurrence of a saturation effect. A saturation effect in the induced mutation frequency beyond a particular dose level is attributed to the rigour of both diplontic and haplontic selection in the biological material (Swaminathan, 1961).

Of the three diploids, the maximum mutation rate in the M_2 was observed in T. N. 1 followed by G. E. B. 24 and A. S. D. 8, while in the M_3, the maximum frequency of mutations was observed in A. S. D. 8 followed by T. N. 1 and G. E. B. 24. Such differences in the mutation rate in the varieties of the same species may be due to genetic differences governing them. The variety T. N. 1 is distinctly different from the other two types. Previous studies revealed that the frequency of mutations is governed by the genetic architecture of the organism (Davies, 1962; Nilan, 1964; Chao and Chai, 1961).

The diploid of G. E. B. 24 showed a higher rate of mutations in the M_2 than its tetraploid as measured on the M_1 plant basis, while a reverse trend was observed when the mutation rate was computed on M_2

plant basis. However, for the diploids, the mutation rate was lower in the M_3 than in the M_2.

A lower frequency of chlorophyll mutations in the M_3 than in the M_2 in diploids would indicate that with regard to segregation for chlorophyll mutations, rice behaves like a genetic diploid, since in polyploids mutations find phenotypic expression more readily in the M_3 and later generations (Swaminathan, 1965). Siddiq and Swaminathan (1968) reported that rice behaves more like a diploid than a polyploid based on its radio-biological response as revealed by the lower frequency of chlorophyll mutations in the M_3 than in the M_2.

Spectrum of chlorophyll mutations: The frequency of different types of chlorophyll mutations exhibited no relationship with the dose of *X*-rays. Gustafsson (1963) in barley and Bhatia and Swaminathan (1963) in wheat had observed that the spectrum of mutations remained similar irrespective of dosage, type of mutagen and M_1 fertility. *Albina* mutations occurred in greater frequency than the other types of mutations in the diploids as well as in the tetraploid. The differences in the proportions of different types of mutations occurring among the three diploids indicated the influence of genotypic constitution over the spectrum of mutations. *Albina* mutations occurred in greater proportion in the M_2 of A. S. D. 8 and G. E. B. 24, whereas *xantha* was observed more often than the other types of mutations in T. N. I. The spectrum of mutations was found to be wider in diploids in the M_2 than in the M_3, whereas a reverse trend was observed in the tetraploid. *Albina* mutations occurred with greater frequency than the other types of mutations in the tetraploid. The lower frequency and narrower spectrum of chlorophyll mutations in the M_3 than in the M_2 in diploid, and the reverse trend in the tetraploid, suggest that the cultivated diploid rice *O. sativa* behaves like a genetic diploid or highly diploidised polyploid.

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