

## RADIATION INDUCED CHLOROPHYLL MUTATIONS IN RICE\*

G. BARI, G. MUSTAFA, A.M. SOOMRO AND A.W. BALOCH

*Atomic Energy Agricultural Research Centre,  
Tandojam, Sind, Pakistan.*

### Abstract

Air dried grains of four local varieties of rice were treated with gamma-rays and fast neutrons for determining their mutagenic effectiveness through the occurrence of chlorophyll mutations. Fast neutrons were more effective in inducing chlorophyll mutations and the rice variety Basmati 370 produced maximum number of mutations followed by varieties Sonahri Sugdasi, Jajai 77 and Sada Gulab. The highest frequency of chlorophyll mutations was that of albina types followed by striata types. The xantha, viridis and tigrina types of mutations were less frequent.

### Introduction

Several types of chlorophyll deficiencies occur in seedlings of mutagenically treated plant material. Some of these deficiencies like albina are lethal, while some of the seedlings with partial deficiencies like virescent and chlorina continue to survive, though with reduced vigour. The frequency of occurrence of these chlorophyll deficient seedlings in the treated population is generally considered an appropriate measure for determining the effectiveness of a mutagen. The present studies were undertaken to determine the mutagenic effectiveness of gamma-rays and fast neutrons for inducing mutations in different varieties of rice.

### Material and Methods

Air dried, unhusked grains of four aromatic, fine grain, tall growing, local varieties of rice (*Oryza sativa* L.) viz., Basmati 370, Jajai, Sada Gulab and Sonahri Sugdasi were treated with gamma-rays and fast neutrons. The moisture content of the grains at the time of irradiation was 11%, and the doses given were 150, 200, 250 and 300 Gy of gamma-rays from the  $^{60}\text{Co}$  radiation source and 10, 15, 20 and 25 Gy of fast neutrons from the nuclear reactor. The radiation treatments were given at the Seibersdorf

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Table 1. Spectrum and frequency of chlorophyll mutations induced by gamma rays in rice.

Variety	Radiation dose (Gy)	Number of $M_2$ seedlings analysed.	Types and frequency of chlorophyll mutations (%)					Total frequency of mutations (%)
			Albina	Xantha	Viridis	Striata	Tigrina	
Basmati 370	0 (Control)	1030	0.38	--	--	--	--	0.38
	150	2083	1.58	0.04	0.09	0.62	--	2.33
	200	2773	1.84	0.18	--	0.68	--	2.70
	250	2182	0.36	--	--	0.18	--	0.54
	300	1586	0.56	--	--	0.13	--	0.69
Jajai 77	0 (Control)	1163	--	--	--	--	--	--
	150	6585	0.89	0.13	0.01	0.04	--	1.07
	200	6016	0.11	--	--	0.07	0.09	0.27
	250	3654	0.27	--	--	0.38	--	0.65
	300	2947	1.69	--	0.10	0.11	--	1.90
Sada Gulab	0 (Control)	1919	--	--	--	--	--	--
	150	4355	0.52	--	0.04	0.03	--	0.59
	200	11360	0.37	--	--	--	--	0.37
	250	6182	0.29	--	--	0.03	--	0.32
	300	9859	0.23	--	--	0.04	--	0.27
Sonahri Sugdasi	0 (Control)	1744	0.05	--	--	0.12	--	0.17
	150	11254	0.77	0.01	0.02	0.03	0.04	0.87
	200	15414	1.29	0.01	0.04	0.03	--	1.37
	250	7498	0.73	0.05	0.19	--	--	0.97
	300	4134	2.23	0.07	--	0.02	--	2.32

Laboratory of the International Atomic Energy Agency, Vienna, Austria. Five hundred grains per treatment were taken from each variety for these studies. The treated grains were sown along with untreated grains in nursery beds, and one month old individual seedlings were transplanted in the field keeping a uniform distance of 20 cm between plants and between rows. At maturity, the five first formed panicles of each  $M_1$  plant from each treatment as well as from the untreated control plants were harvested individually. The  $M_1$  panicles thus harvested were subsequently sown separately, without threshing, in the seed beds and the  $M_2$  seedlings raised therefrom were scored for chlorophyll deficiencies in the second week after their germination. The non-irradiated grains were also sown along with the irradiated material in the seed beds for comparative studies.

The chlorophyll deficiencies in the seedlings were classified into five groups viz., albina, xantha, viridis, striata and tigrina. The albina were white seedlings, and the xantha had pale yellow leaves. Both of these types survived only for one or two weeks after germination. In the viridis type of seedlings, there was some delay in the development of chloroplastid pigments in their leaves. They were pale yellow in colour, but gradually turned green. The striata seedlings had white longitudinal streaks on normally green leaves, while the tigrina seedlings had variegated green and yellow patches on the leaves. The mutation frequency was expressed as the number of chlorophyll mutants per 100  $M_2$  seedlings.

## Results

In these experiments, mutations were induced effectively by gamma-rays as well as by fast neutrons (Tables 1,2). The frequency of the occurrence of mutations, however, indicated that fast neutrons were more effective in inducing chlorophyll mutations. The highest frequency of mutations was that of albina types, followed by the striata types. The remaining three types i.e. xantha, viridis and tigrina were less frequent. The combined average occurrence of albina mutants in all the four varieties of rice was 2.75% for gamma-rays, and 3.78% for fast neutrons, followed by striata mutants in which case the frequency was 0.48% for gamma-rays and 0.51% for fast neutrons. The averages for the remaining three types i.e. xantha, viridis and tigrina were less than 0.51% in treatments of both gamma-rays and fast neutrons.

Comparing the mutability of different varieties, it was observed that Basmati 370 produced maximum number of chlorophyll mutations in case of gamma-rays as well as fast neutrons, followed by varieties Sonahri Sugdasi, Jajai 77 and Sada Gulab. The most effective doses of gamma-rays for inducing chlorophyll mutations were 200 Gy for variety Basmati 370, 300 Gy for Jajai 77 and Sonahri Sugdasi, and 150 Gy for Sada Gulab.

Table 2. Spectrum and frequency of chlorophyll mutations induced by fast neutrons in rice.

Variety	Radiation dose (Gy)	Number of $M_2$ seedlings analysed	Types and frequency of chlorophyll mutations (%)					Total frequency of mutations (%)
			Albina	Xantha	Viridis	Striata	Tigrina	
Basmati 370	0 (Control)	1030	0.38	---	---	---	---	0.38
	10	9160	5.01	---	---	1.50	---	6.51
	15	4566	1.05	0.04	---	0.13	---	1.22
	20	1311	2.44	0.30	---	0.30	---	3.04
	25	1801	1.12	---	---	0.12	---	1.24
Jajai 77	0 (Control)	1163	---	---	---	---	---	---
	10	10193	0.47	0.01	---	0.08	---	0.56
	15	6271	0.05	---	---	0.09	---	0.14
	20	1722	0.58	---	---	---	---	0.58
	25	1519	0.47	0.07	0.32	0.13	---	0.99
Sada Gulab	0 (Control)	1919	---	---	---	---	---	---
	10	4169	1.87	0.08	0.02	0.04	---	2.01
	15	9654	0.33	---	0.03	---	---	0.36
	20	9215	0.47	---	0.02	0.08	---	0.57
	25	6754	0.20	---	0.01	0.02	---	0.23
Sonahri Sugdasi	0 (Control)	1744	0.05	---	---	0.12	---	0.17
	10	12331	0.75	---	0.01	0.01	---	0.77
	15	11408	0.62	0.08	---	0.01	---	0.71
	20	8285	1.66	0.08	0.03	0.02	0.51	2.30
	25	2953	0.81	---	---	---	0.20	1.01

In case of fast neutrons, the most effective doses were 10 Gy for varieties Basmati 370 and Sada Gulab, 25 Gy for Jajai 77, and 20 Gy for Sonahri Sugdasi.

### Discussion

The occurrence of chlorophyll mutations in plants is a complex phenomenon. The chlorophyll mutations could result from the interference in chlorophyll synthesis (von Wettstein, 1960), due to obstruction in the formation of accessory pigments (Koski & Smith, 1951; Anderson & Robertson, 1960), due to anomalies in the structural development of chloroplasts and consequently the photosynthetic pigments (von Wettstein, 1961), or due to the interference in the formation of essential metabolites that are necessary for the development of normal chloroplast structure and chloroplast pigmentation (Wallis, 1963; Redei, 1965).

The mechanism of mutability of different varieties of crop plants is considered closely related to their radiosensitivity, and several factors like physiological properties inside the cell and the nucleus (Gustafsson, 1944), nuclear volume and DNA content (Sparrow & Evans, 1961; Sparrow *et al.*, 1961), chromosome size (Bari & Godward, 1969), and ploidy level (Conger *et al.* 1982) have been shown to influence the radiosensitivity of plants.

In the present studies, vast differences were observed in the response of different varieties of rice to radiation treatments. Although the fast neutrons were generally more effective in inducing chlorophyll mutations than gamma-rays, but nonetheless, in some treatments, the gamma-rays proved more effective than fast neutrons. Some varieties produced more chlorophyll mutations than others, and different treatments of radiation produced diversely different spectra and frequencies of chlorophyll mutations in different varieties. Such differences in radiation response indicate that several physical and biological factors are involved in the mutational process, and that makes it extremely difficult to predict the occurrence of mutations in different varieties of crop plants. Nevertheless, it can be concluded that radiation doses used in the present studies were, by and large, most appropriate for inducing mutations in rice.

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