

BIOLOGICAL EFFECTS OF MUTAGENIC TREATMENTS IN RICE

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Experiments were carried out with rice variety Co.37 to study effects of gamma irradiation and EMS on germination, survival, pollen and seed fertility in M_1 generation. A progressive reduction in germination and survival was observed as the doses of gamma rays and EMS were increased. The reduction in germination percentage was more following combination treatments than individual treatments and the maximum reduction (10.86%) was observed (40 Krad + 50mM). The lower concentrations of EMS did not affect the survival of seedlings and the reduction in survival percentage was more due to gamma irradiation and combination treatments than EMS. For the combination treatments, the effect was additive on germination and survival. Pollen and seed fertility was markedly reduced in gamma ray treatments compared to the EMS treatments. The combination treatments produced additive effect on pollen and seed fertility in M_1 generation.

Induced mutagenesis is an important complementary and often unique approach to plant breeding. The combined effects of gamma rays and EMS have been studied only to a limited extent. So, an attempt has been made to study the effects in this direction.

MATERIAL AND METHODS

The seeds of rice (*Oryza sativa* L) variety CO37 were presoaked in distilled water for 24 hours prior to the mutagenic treatments. Treatments with EMS were done by keeping the pre-soaked seeds immersed for 6 hours in the requisite concentration of mutagen with intermittent shaking. Immediately after the completion of treatment the treated seeds were

thoroughly washed in running tap water for half an hour. For combination treatments, the dry seeds were exposed to 40 krad of gamma rays and then presoaked for 24 hours in distilled water. The presoaked seeds were soaked for 6 hours in EMS, Simultaneously with the individual EMS treatment under similar conditions. The irradiated seeds were soaked in beaker for 24 hours before sowing, whereas the seed treated with EMS and combined treatment were sown as such. Ninety seeds from each treatment were sown in petridishes with wet filter paper and was replicated three for making counts of germination. In the nursery, 300 seeds from each treatment were sown in small polts of 60x40 cm size. Twentytwo day old seedlings were transplanted in the main field at

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a spacing of 20x10 cm in randomised block design with three replications. The mutagenic effects in M_1 generation was studied in term of germination survival, pollen and seed fertility. The effect of combining the two mutagens on radio sensitivity was determined by the deviation of the observed value of the combination treatment from the sum of values of the corresponding single treatments. The overall statistical significance of the deviation of the observed values from expected values in each series of combination treatments was tested by chi-square method. The interaction effects of combined treatments on M_1 biological damages were calculated according to the formula (Sharma; 1970)

$$K = \frac{a+b}{(a)+(b)}$$

K = Interaction coefficient

(a) and (b) = mutation frequencies of individual mutagens

(a+b) = mutation frequency of combination treatment

If $K = 1$, additive; $K < 1$, synergistic

$K > 1$ antagonistic.

RESULTS AND DISCUSSION

The effect of single mutagens and combination treatments have been presented in Tables 1 and 2 respectively.

a. Germination:

In the present study, inhibition rate for germination increased with the increased doses of both gamma rays and EMS. Significant differences in germination of seeds were observed due to individual and combined treat-

ments of gamma rays and EMS. The reduction in germination following gamma irradiation was higher than in treatments with EMS. Similar findings were observed by Vereschagin (1974). Acharya & Acharya (1977) reported a greater reduction in percentage of germination due to EMS treatments. This was not observed with present study.

The reduction in germination percentage was more following combination treatments than individual treatments. The maximum reduction (10.86%) was observed at the combination treatment of 40 krad+50 mM. As regards combination treatments, the data on the reduction in germination indicated that the deviation was not significant from the observed values to that of expected values ($X^2 = 0.64$, $0.99 > P > 0.98$). This implies that the effect of two mutagens were additive in their effect. The interaction coefficients (K values) showed the near additive effect on M_1 generation in combination treatments which ranged from 0.83 to 0.98. When the gamma rays and EMS were used in combination, decrease in germination percentage appears to indicate near additive effect of the combination treatments on germination. Similar results were recorded by Doll and Sandfaer (1969) and Mohan Rao (1972) in barley for combination of EMS and gamma rays.

b. Survival of plants on 30th day:

The survival of M_1 plants, in the present study showed a gradual reduction for increasing dose of gamma rays and EMS treatments when used indivi-

dually. This is in agreement with results of Yamagato *et. al.*, (1965). Reduction in survival even at the advanced stages of growth was recorded and this indicated prolonged lethal action of gamma rays. The lower concentrations of EMS however did not affect the survival of seedlings in the present investigation and is in accordance with that reported by Soriano (1972). The reduction in survival percentage was more due to gamma irradiation and combination treatments than EMS alone. The individual treatment of 50 krad of gamma rays recorded the highest reduction of 74.74 per cent in survival whereas, the maximum reduction of 68.43 per cent on control was observed in the combination treatment of 40 krad + 50 mM.

The overall chi-square analysis of deviations showed that the combination treatments produced additive effect on M_1 plant lethality ($X^2 = 1.04, 0.98 > P > 0.95$) as the deviation was not significant. The interaction coefficients showed a near additive effect on M_1 plant lethality which ranged from 0.98 to 0.99 in combination treatments. The percentage of survival in either of the combination treatments did not differ considerably from values expected on additive basis. The combined effect of gamma rays and EMS on survival in the M_1 was additive. This is in conformity with the results of Gopinathan Nair (1977). A comparatively greater reduction in survival was observed in combination treatments than individual treatments. This is in agreement with Chakrabarti (1975).

The LD50 for survival on 30th day was 40 krad due to gamma irradiation and 40 krad+20 mM of combination treatment. But, EMS treatment failed to cause 50 per cent reduction in survival of plants. Fujii and Matsumura (1958) noticed that LD50 with gamma rays ranged from 20 to 50 krad in *japonica* varieties, and in *indica* it varied from 20 to 65 krad. Co37 being an *indica* the present results concurred with that of the above authors.

c. Pollen and seed fertility:

A linear reduction in pollen fertility with increasing dose was evident. Among all the treatments, the highest reduction in pollen fertility (i.e., 80.00 per cent) was observed in 50 krad gamma rays treatment. In the case of EMS and combination treatments, the reduction in pollen fertility ranged from 1.54 to 3.51 per cent and 56.61 to 60.64 per cent respectively. Significant variation on pollen fertility was observed between control and treated populations due to gamma rays, EMS alone or in their combinations.

The percentage of seed fertility ranged from 18.53 to 81.15 in gamma rays, 94.11 to 98.56 in EMS and 30.55 to 35.77 in the case of combination treatments. The seed sterility was more due to combination treatments than individual treatments of 40 krad and EMS.

A linear reduction in pollen and seed fertility was noticed with increase in dosage of gamma rays in the present investigation. Rahman and Soriano (1972) reported similar trends in

pollen and seed fertility of rice. Reduction in pollen and seed fertility was observed in EMS treatments also but it was to a lesser degree than gamma rays. The concentrations of EMS adopted in the present study were insufficient to cause 50 per cent reduction in pollen and seed fertility whereas, 50 per cent reduction in pollen and seed fertility was observed in 30 to 40 krad of gamma rays and 40 krad+10mM to 40 krad + 20 mM of combination treatments,

Most of the combination treatments produced additive effect on pollen fertility ($X^2 = 0.06$, $P > 0.99$). The additive effect was confirmed by interaction coefficients which ranged from 0.08 to 1.02. The deviation was not significant ($X^2 = 0.48$, $P > 0.99$) in combination treatments. This shows that the two mutagens were combined to produce additive effect on pollen and seed fertility in the present investigation. This additive effect may be due to the independent action of the two mutagens probably by different mechanisms. Earlier workers like Khalatkar and Bhatia (1975) however proposed antagonistic effect on pollen and seed fertility in the M_1 generation,

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Table 1 Effect of single Treatments on Biological characters

Mutagen (Dose/Conc)	Germination		Survival of plants		Pollen Sterility		Seed sterility	
	Percentage on control	Reduction (%)	Percentage on control	Reduction (%)	Percentage on control	Reduction (%)	Percentage on control	Reduction (%)
Gamma rays (Krad)								
Control	100.0	—	100.0	—	100.0	—	100.0	—
10	97.9	2.01	95.6	4.38	87.6	12.31	81.1	18.8
20	95.6	4.36	78.4	21.55	69.9	30.04	64.3	35.6
30	94.6	5.37	74.4	25.59	56.5	43.49	52.6	47.3
40	93.6	6.37	52.5	47.47	43.9	56.06	36.4	63.6
50	93.3	6.70	25.2	74.74	20.0	80.00	18.5	81.4
Value of control	99.3		99.0		93.0		88.5	
C. D.	1.87		2.22		4.56		2.22	
EMR (mM)								
Control	100.0	—	100.0	—	100.0	—	100.0	—
10	98.6	1.34	94.6	5.38	98.4	1.54	98.5	1.44
20	97.6	2.35	92.6	7.40	98.2	1.73	94.1	5.81
30	96.6	3.35	91.2	8.75	97.4	2.51	93.5	6.41
40	95.6	4.36	90.2	9.76	97.5	2.43	98.8	1.12
50	93.6	6.37	73.7	26.26	96.4	3.51	94.1	5.88
Value of control	97.3		99.0		95.2		91.5	
C. D.	1.39		5.96		1.26		1.90	

Table 2 Effect of combination treatment on biological characters

Mutagen (Dose/Conc.)	Percent of Control	Reduction over control					Interaction ⁿ Co-efficient (K)
		Observed (O)	Expected (E)	Deviation (O-E)	Deviation expected (O ² /E)		
a. Germination							
Gamma rays+EMS							
Control	100.0	—	—	—	—	—	
40+10	92.8	7.12	7.71	-0.59	0.05	0.92	
40+20	91.8	8.14	8.72	-0.58	0.04	0.93	
40+30	91.8	8.14	9.72	-1.58	0.26	0.83	
40+40	89.4	10.51	10.73	-0.22	0.01	0.98	
40+50	89.1	10.86	12.74	-1.88	0.28	0.85	
Value of control	98.3						
C, D	1.99						
Chi-Square-value for deviation 0.64, 0.99 p 0.28							
b. Survival							
Control	100.0	—	—	—	—	—	
40+10	51.3	48.6	52.8	-4.19	0.33	0.92	
40+20	49.2	50.7	54.8	-4.10	0.31	0.93	
40+30	44.4	55.5	56.2	-0.71	0.01	0.99	
40+40	43.2	56.7	57.2	-0.51	0.01	0.99	
40+50	31.5	68.4	73.7	-5.30	0.38	0.93	
Value of control	98.67						
C, D	1.72						
Chi-Square Value for deviation 1.04, 0.98 P 0.95							
c. Pollen Sterility							
Control	100.0	—	—	—	—	—	
40+10	43.3	56.6	57.6	-0.99	0.02	0.98	
40+20	41.9	58.1	57.7	0.33	—	1.01	
40+30	41.6	58.3	58.5	-0.23	—	1.00	
40+40	40.3	59.6	58.4	1.12	0.02	1.02	
40+50	39.3	60.6	59.5	1.07	0.02	1.02	
Value of Control	94.2						
C, D,	1.52						
Chi-Square value for deviation 0.06. P 0.99							
d. Seed Fertility							
Control	100.0	—	—	—	—	—	
40+10	35.7	64.2	65.0	-0.81	0.01	0.99	
40+20	34.5	65.4	69.0	-3.98	0.23	0.94	
40+30	32.9	67.0	70.0	-2.96	0.13	0.96	
40+40	32.5	67.4	64.7	2.69	0.11	1.04	
40+50	30.5	69.4	69.4	0.03	—	0.90	
Value of Control	91.5						
C, D,	0.85						
Chi-Square value for deviation 0.48, P 0.99							