Journal of Phytology 2009, 1(6): 444–447
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ISSN: 2075-6240
Available Online: www.journal-phytology.com

REGULAR ARTICLE

EFFECTIVENESS EFFICIENCY AND BIOCHEMICAL CONTENT OF PHYSICAL AND CHEMICAL MUTAGENS IN SOYBEAN (*GLYCINE MAX* (L.) MERR.)

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SUMMARY

In a mutation breeding experiment, in the CO-1 variety of soybean (*Glycine max* (L.) Merr.) the efficiency and effectiveness of physical and chemical mutagens viz., gamma rays, ethyl methane sulphonate (EMS), diethyl sulphate (DES) and Colchicine (COH) were examined. Gamma rays were found to be more effective than other mutagens in producing chlorophyll and viable mutants. The efficiency was observed based on lethality and injury. Gamma rays were found to be more effective than other treatments. The chlorophyll mutants viz., Albino, Viriscence and Xantha and viable mutants viz., plant type, days to maturity, early flowering, seed color, seed shape, pod shape & male sterility etc. were recorded with various frequencies. The protein and oil content was recorded maximum at 50 KR of gamma rays.

Keywords: Soybean, mutation, frequency, effectiveness, efficiency.

P. Pavadai et al. Effectiveness Efficiency and biochemical content of Physical and Chemical Mutagens in Soybean (*Glycine max* (L.) Merr.). J Phytol 1 (2009) 444-448
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1. Introduction

Soybean (*Glycine max* (L.) Merr.) is an important crop on account of its high nutritive value. Broadening the genetic base for crop improvement can be quickly achieved through induced mutagenesis. The first news of the soybean was brought to the Western Hemisphere (1). As per the FAO/IAEA database, 41 mutant varieties of soybean have been released, out of which three have emanated from crosses of mutants (2). Chlorophyll mutants are most convenient for evaluating the genetic effects of mutagens in plants (3). It may serve as a good index for determining the doses of different mutagens. In this present study deals with the observations on seedling growth injury (I) and seedling survival (L) bases in M_1 generation. The chlorophyll mutations in M_2 generations seedling of soybean (*Glycine max* (L.) Merr.) were induced by gamma rays, EMS, DES and Colchicine.

2. Materials and Methods

The dry and dormant seeds of soybean (*Glycine max* (L.) Merr.) var. CO-1 were used for mutagenic treatments. Hundred seeds were

taken for each treatment. Both physical and chemical mutagens were given. The seeds were packed in polythene bags and exposed to 10, 20, 30, 40, 50 and 60 KR doses of gamma rays in 60CO gamma source. The gamma radiation was carried out at sugarcane breeding institute (ICAR), Coimbatore, India.

Three chemical mutagens namely, ethyl methane sulphonate (EMS), diethyl sulphate (DES) and Colchicine (COH) were used in various concentrations. The chemical mutagens were subjected to six different concentrations of ethyl methane sulphonate (0.1, 0.2, 0.3, 0.4, 0.5 and 0.6 per cent), diethyl sulphate and Colchicine (0.01, 0.02, 0.03, 0.04, 0.05 and 0.06 per cent). For chemical treatments, seeds were presoaked in distilled water for 6 hours to ensure complete hydration of the seeds. The seeds were treated with solution of EMS, DES and COH for duration of 6 hours. After the treatment, seeds were washed thoroughly for one hour in running tap water to terminate the residual effect of the mutagenic chemicals. The treated seeds were sown in the field along with the control (untreated seeds) in a randomized block design (RBD) with three replications. All the treatments including control were raised adopting a spacing of 35cm in between rows and 15cm in between plants, respectively. The seedling height reduction (I) in different M_1 treatment was studied (4). The height in all the seedlings in a treatment was recorded 30 days after sowing and their average was computed and expressed as percentage of control. The plant survival percentage (L) was computed as the percentage of plant surviving till maturity out of total number of plants recorded after germination. The M_1 plants were harvested, separately and the seeds were sown in the next season in plant progeny rows to raise M_2 generation in a randomized block design with three replications. Chlorophyll mutations were calculated per 100 M_1 plants and 100 M_2 seedling bases. The chlorophyll mutants were classified (5, 6). Frequency of viable mutations was calculated on M_1 and M_2 plant bases. Data on biological abnormalities such as injury and lethality in M_1 generation and chlorophyll mutation frequency in M_2 generation were used to determine the mutagenic effectiveness and efficiency was calculated on the basis of formulae (7).

Mutagenic effectiveness = Mf×100/krad (or) c × t

Mutagenic efficiency = Mf/I×100

3. Results and Discussion

Chlorophyll and viable mutations were studied in segregating progenies in M_2 generation of soybean. Three types of chlorophyll mutants were recorded and classified (5). They are Albino, Viriscence and Xantha. The viable mutants were recorded at tall, dwarf, stunted, early and late flowering, early and late maturity, green colour seed, wrinkled seed, single seeded pod, long pod, tetra foliate leaf and male sterility. The chlorophyll mutants for M_1 plant and M_2 seedling bases and viable mutants for M_2 plant were bases observed (Table 1). Generally chlorophyll mutants were identified in all treatments i.e., gamma rays and chemical mutagens. The frequency increased with the elevation of mutagenic level which confirms most of earlier reports in barley (8), mungbean (9) and lentil (10).

On applying different doses of gamma rays, the frequency increased from 30 to 50 KR on M_1 plant and M_2 seedling bases and thereafter reduction in frequency was observed. On M_2 seedling basis, maximum mutation frequency was observed at 10 KR (3.51) of gamma rays. Whereas in chemical mutagens maximum mutation frequency was recorded at 0.05 per cent (3.41) followed by 0.14% of EMS (3.23) and 0.04% of DES (2.67) respectively (Table 2). Frequency of chlorophyll mutants was

highest on M_1 plant basis when compared to M_2 seedling basis (11, 12).

Table 1. Mutagenic effectiveness and efficiency of soybean var. CO 1 using mutagenic treatments

Mutagens Doses/Conc.	Total no. of plant mutants	Total no. of plant studied	Mutation frequency % (M)	Lethality %(L)	Injury % (I)	Effectiveness $\frac{Mf}{KR\ (or)\ c \times t} \times 100$	Efficiency $\frac{Mf}{L} \times 100$	Efficiency $\frac{Mf}{I} \times 100$
Gamma rays								
10 KR	19	540	3.51	16.56	15.10	35.1	21.19	23.24
20 KR	9	615	1.46	21.38	17.06	7.30	6.82	8.55
30 KR	12	466	2.57	37.99	20.56	8.56	6.76	12.50
40 KR	16	732	2.18	48.17	31.60	5.45	4.52	6.89
50 KR	17	624	2.72	55.33	36.55	5.44	4.91	7.44
60 KR	14	750	1.86	65.33	41.99	3.10	2.84	4.42
EMS								
0.1 %	10	616	1.62	12.98	10.19	27.0	12.48	15.89
0.2 %	8	407	1.96	21.89	15.29	16.33	8.95	12.81
0.3 %	15	528	2.84	34.88	21.50	15.77	8.14	13.20
0.4 %	12	371	3.23	43.98	26.58	13.45	7.34	12.15
0.5 %	12	427	2.81	53.67	35.88	9.36	5.23	7.83
0.6 %	10	575	1.94	65.09	40.13	5.38	2.98	4.83
DES								
0.01 %	11	670	1.64	14.69	14.83	27.33	11.16	11.95
0.02 %	6	585	1.02	24.03	18.30	8.50	4.24	5.57
0.03 %	16	627	2.55	32.40	25.90	5.55	7.87	9.84
0.04 %	11	411	2.67	47.63	36.62	11.25	5.60	7.29
0.05 %	9	384	2.34	63.19	40.10	7.80	3.70	5.83
0.06 %	10	440	2.27	71.25	49.31	6.30	3.18	4.60
COH								
0.01 %	6	510	1.17	19.75	14.08	19.51	5.92	8.30
0.02 %	11	575	1.91	33.17	21.22	15.91	5.75	9.00
0.03 %	14	660	2.12	34.74	27.34	11.77	6.10	7.75
0.04 %	15	494	3.03	57.54	30.93	12.65	5.26	9.79
0.05 %	18	527	3.41	70.13	36.29	11.36	4.86	9.39
0.06 %	9	660	1.36	76.25	40.37	3.79	1.78	3.36

Effectiveness means the rate of mutation induction as dependent upon the mutagenic does and efficiency refers to the mutation rate in mutation to the various biological effects usually a measure of damage (13). In general the effectiveness decreased with increasing dose or concentration. Mutagenic effectiveness of chlorophyll and viable mutations on M_1 and M_2 plant basis is given in Tables 1. In M_2 plant basis maximum effectiveness was observed at 10 kR gamma rays (35.1) followed by 0.1% of EMS (27.0), 0.001% of DES (27.3) and 0.01% of COH (19.5).

Table 2. Protein and oil content of soybean variety CO 1 using mutagenic treatments

Treatments	Protein content (%)	Oil content (%)
Control	38.72	18.71
Gamma rays		
10 KR	38.19	18.32
20 KR	38.75	18.65
30 KR	39.06	19.27
40 KR	39.41	18.94
50 KR	40.56	20.32
60 KR	39.52	18.91
EMS		
0.1%	38.52	17.56
0.2%	39.77	18.17
0.3%	39.17	18.32
0.4%	38.75	19.27
0.5%	40.67	20.02
0.6%	40.02	18.56
DES		
0.01%	38.14	17.11
0.02%	38.46	17.87
0.03%	39.08	18.32
0.04%	39.39	18.71
0.05%	40.00	19.05
0.06%	38.52	18.37
COH		
0.01%	38.62	17.77
0.02%	39.17	17.92
0.03%	39.34	18.00
0.04%	39.71	18.06
0.05%	38.35	17.57
0.06%	38.18	17.32

However, the effectiveness of chlorophyll and viable mutations were higher in chemical mutagens in both M_1 and M_2 generation on the basis of seedling stage (7-15 days) and throughout the growth period, respectively. In the present study, the effectiveness decreased with increase in doses and concentration of gamma rays and chemical mutagens. This was in confirmation with the findings of black gram (11), cowpea (14, 15, 16), soybean (17).

Efficiency

The mutagenic efficiency was worked out based on injury and lethality. The mutagenic efficiency gives an idea of the proportion of mutations in relation to other associated undesirable biological effects such as injury, lethality and sterility induced by the mutagen (7). The maximum efficiency of chlorophyll and viable mutant on M_2 generation was

recorded at 10 KR of gamma rays (21.19 and 23.24) followed by 0.1 per cent of EMS (12.48 and 15.89), 0.01 per cent of DES (11.16 and 11.95) and 0.03 and 0.04 per cent of COH (6.10 and 9.79) respectively.

According to the present study 10 KR gamma rays dose was more efficient on lethality and injury basis on inducing chlorophyll and viable mutations. Gamma rays were more efficient than EMS in inducing chlorophyll and viable mutations in black gram (11,18). In general, both the positive and negative shift was observed in most of the mutagenic treatments than the control. The highest protein content was measured at 0.5% of EMS and 50 kR of gamma rays than the other mutagenic treatments. In most of the treatments efficiency increased with increase in dose / concentration.

In the present study more amount of protein content was measured in 0.5% of EMS. Reports of previous work done in soybean also shows similar results (19, 20).

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