Effect of Mutagenesis on Germination, Survival and Pollen sterility in M_1 Generation of Soybean [*Glycine max* (L.) Merill]

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Research Article

Abstract: M_1 generation of soybean [*Glycine max* (L.) Merill] was raised by treating the dormant seeds of variety of MAUS-71 and JS-335 with varied concentration of chemical mutagen (EMS) and physical mutagen (Gamma rays). A dose dependant decrease was noticed in most of the characters in M_1 generation. The results indicated that the reduction in germination percent over control was noticed in all mutagenic treatments in both the cultivars, while increased pollen sterility was associated with corresponding increases in dose/ concentration of mutagens. Results indicate that higher doses were more effective.

Keywords: soybean, EMS, Gamma rays, germination, survival, pollen sterility.

Introduction:

Soybean [*Glycine max* (L.) Merill, family papilionaceae (fabaceae)], is a crop of great world importance due to widespread applicability of its products and their economical value in the national and international market. Soybean is the world's most important source of edible oil. It accounts for nearly 60 percent of global oil seed production. The protein content of soybean is rich in limiting amino acid lysine (Maloo and Sharma 2007) the productivity of soybean in India is much low in comparison with world average. Low productivity is due to limited genetic diversity, narrow genetic base of Indian soybean varieties, short growing period available in Indian latitude, stagnant genetic potential for yield (Tiwari 2003). Due to small, fragile flowers hybridization is very difficult, tedious and costly. Hence classical breeding methods have got limited application in soybean improvement. Alternatively induced mutagenesis is the best method to enlarge genetic variability within short time. Creation of genetic variability by induced mutagenesis proved best for strengthening crop improvement programme and represents a more efficient source of genetic variability than the gene pool conserve by nature (Brock 1965). Considering the above facts the research programme was therefore, undertaken to induce genetic variability and to screen useful mutants or their use in improvement in soybean. However in early and late generation the germination, survival, pollen sterility are more important as initial indicators.

Material and Method:

The two varieties MAUS-71 and JS-335 of soybean (*Glycine max* (L.) Merill) formed the materials for the present investigation. Germplasm of these cultivars was collected from All India Co-ordinate Research Program of Soybean, Marathwada Agricultural University, Parbhani (M.S.). The investigation envisaged studying the differential sensitivity of the soybean varieties by subjecting them to different mutagens- Gamma rays (Physical mutagen) and ethyl methane sulphonate (Chemical mutagen). Gamma irradiation was done using cobalt 60 sources in the Gamma chamber, installed at Government Institute of Science, Aurangabad (M.S.). The chemical mutagen, ethyl methane sulphonate ($CH_3SO_2OC_2H_5$) with molecular weight 124.16, from the sigma chemical company, USA was used for treating the seeds.

For the assessment of LD_{50} dose three hundred seeds of uniform size were used for (Gamma rays – 10Kr, 20Kr, 30Kr and EMS – 0.05%, 0.10%, and 0.15%) each treatment. In respect of EMS treatment, the seeds were presoaked in distilled water for 6 h. appropriate quantities of EMS were dissolved in distilled water to have the concentrations envisaged in the program. The treatment was performed at room temperature 22 $\pm 2^{O}$C early morning hours with intermediate shaking during the treatment period of 6 h. after the chemical treatment, the seeds were washed thoroughly with running tap water for half an hr to remove the residues of the chemical, if any and the

excess moisture in seed coat was removed by using folds of blotting paper.

About 300 seeds of each treatment were sown in the experiment field along with control following randomized block design in three replicates to rise M_1 generation during Kharif season of 2008. All the treatments including control were raised adopting a spacing of 45cm between two lines and 30cm in between plants.

Germination percentage: the number of seed emergence of the radical was counted and mean was expressed as percentage.

Plant survival: the number of plants reaching maturity in the field was noted and expressed as percentage

Pollen sterility: pollen sterility was determined from 10 randomly selected plants belonging to each treatment. The pollen grains from fresh dehisced anther were stained with 1% acetocarmine. Pollen grains that stained fully were counted as fertile, while the empty, partially stained and shriveled ones were counted as sterile.

Results and Discussion:

Table: 01. Effect of different mutagens in M_1 generation of Soybean Variety MAUS-71 and JS-335

Treatment / Variety MAUS-71	Concentration % Dose	Germination % Mean	Survival % Mean	Pollen sterility Mean
CONTROL	-----	97.33	96.66	--
	0.05%	89.33	85.66	4.46
EMS	0.10%	84.66	80.33	11.3
	0.15%	77.33	72.33	15.16
	10 Kr	86.00	81.66	12.43
Gamma ray	20 Kr	80.00	76.33	14.81
	30 Kr	78.33	71.33	20.93
Variety JS-335				
CONTROL	-----	97.66	97.00	--
	0.05%	91.66	88.00	3.5
EMS	0.10%	86.00	82.66	8.93
	0.15%	79.33	74.00	14.7
	10 Kr	90.00	86.33	13.66
Gamma ray	20 Kr	82.00	76.00	18.23
	30 Kr	81.00	73.00	23.4

Germination percentage

The data on germination percentage in M1 generation for various mutagenic treatments in MAUS-71 and JS-335 are given in table no. 1. In both the varieties, in comparison to the control, the percent germination was low in all treatments, similar results were also reported by Patil *et al.* (1985), Mehtre *et al.* (1994) Padavai and Dhanavel (2004) , Singh and Kole (2005).

The lowest germination of 77.33% and 79.33% was recorded in 0.15 % EMS concentration in both MAUS-71 and JS-335 variety of soybean, which may be due to physiological and acute chromosomal damage (Singh *et al.*1997). Delay in the one set of mitosis (Yadav 1987) and chromosomal aberration induced enzyme activity such as catalase, lipase and hormonal activity results in reduced germination (Ananthaswamy *et al.*1971). Reduction in germination over control in MAUS-71 ranged from 77. 33 to 89.33 for EMS and from 78.33 to 86.00 for gamma radiation. While in JS-335 germination over control ranged from 79.33 to 91. 66 for EMS concentration and from 81.00 to 90.00 for gamma radiation . the findings are close agreement with the earlier reports of Rajib and Jagatpati (2011a, 2011b).

Survival percentage:

Survival (at flowering) due to different mutagenic treatment in MAUS-71 ranged from 71.33 (30Kr) to 85.66 (0.05% EMS), while in JS-335 it ranged from 73.00 (30Kr) to 88.00 (0.05% EMS). (Table No. 1) The decrease in survival percentage was associated with increases in the dose / concentration of the mutagens in both the cultivars. These findings are close agreement with the earlier reports of Wang and Yu (1988), Solanki and Sharma (1999, 2002), Kumar and Selvaraj (2003), Solanki and Phogat (2005), Geeta and Wakode (2011)

Pollen sterility percentage:

However the effect of mutagen was more prominent in terms of pollen sterility, which is an increase as dose

increases in both the mutagens in both varieties of soybean. The maximum sterility was observed in 30Kr gamma ray dose (MAUS-71 20.93 and JS-335 23.4) and 0.15% EMS concentration (MAUS-71- 15.16 and JS-335- 14.7). the increasing pollen sterility has been mainly attributed to chromosomal interchange, chromosomal aberration, gene mutation (Gautam *et al.* 1992), cytoplasmic factors (Malinoveskii *et al.* 1973). In most cases meiotic abnormalities are responsible for pollen sterility (Muthusamy and jayabalan 2002) in cotton and (Khan and Wani 2005) in chickpea. In the present findings, the increase in pollen sterility as a consequence of mutagenesis is in accordance with the findings in (Ignacimuthu and Babu 1989) wild and cultivated urd and mungbean. The gradually increase percentage of pollen sterility with increase dose / concentration was in conformity with the earlier reports in (Dixit and Dube 1988) in lentil, (Kulkarni 2011) horsegram and (Sangle *et al.* 2011) in pigeonpea.

Conclusion:

From present study it can be concluded that both mutagens showed an inhibitory effect on germination, survival and pollen sterility percentage. The concentration/dose used in present study will be effective in induction of wide range of mutants.

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