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Screening for pod shattering in mutant population of Soybean (*Glycine max* L.)

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ABSTRACT
The extent of yield losses due to pod shattering in soybean may range from 34 to 99 per cent depending upon delayed harvesting after maturity, the environmental conditions during harvesting and genotype. The identification of resistant sources for pod shattering is one of the most important aspects in the management of pod shattering. Induced mutations, have offered a single and short alternative to conventional breeding. In this study, variability was induced by gamma rays in three soybean genotypes viz. Phule Agrani (KDS-344), Phule Sangam (KDS-726) and KS-103. Screening for pod shattering was carried out in M_2 populations. The shattering effect recorded at field level by exposing to sunlight. The shattering percentage ranged from of 6.70 per cent in Phule Agrani (300 Gy) to 64.44 per cent) of Phule Agrani(400 Gy. A total of 93 shattering tolerant mutants were selected from field observations. These mutants were again scored under laboratory condition as per IITA method. In Phule Agrani 18 mutants showed tolerant reaction whereas, in Phule Sangam 12 mutants and in KS-103 with 15 mutants were showing tolerant reaction. Screening of mutants for pod shattering revealed 45 mutants as tolerant types, 35 mutants as moderately shattering and 13 mutants as highly shattering categories in M_2 generation. This study on identification and screening of the mutants tolerant to pod shattering with high yielding potential will help to increase the production of the pods to a greater extent.
Key words: *yield losses, pod shattering, soybean, resistant sources and mutants*

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INTRODUCTION

Soybean (*Glycine max* (L.) Merrill) is known as "golden bean" and the miracle crop of the 21st century with yield potential (40-45 q/ha), protein (40 per cent), cholesterol free oil (20 per cent) omega-6 and omega-3 fatty acids, 6 to 7 per cent total minerals, 5 to 6 per cent crude fiber and 17 to 19 per cent carbohydrates reported by Chauhan *et al*., [4]. In India area, production and productivity of soybean during 2017 was 101.5 lakh ha, 91.4 lakh million tones and 900 kg ha^{-1}, respectively. While in Maharashtra it was 34.4 lakh ha, 31.8 lakh million tones and 925 kg ha^{-1}, respectively Anonymous [2].

Pod shattering refers to the opening of mature pods along the dorsal or ventral sutures and dispersal of seed as the crop reaches maturity, as well as during harvesting. The yield losses due to pod shattering ranged from 34 to 99 per cent depend on environmental conditions during harvesting and genotype reported by Tiwari and Bhatnagar, [11]. Fully mature pods of soybean are extremely sensitive to opening, resulting in seed loss. This can take place in susceptible varieties prior to harvest due to disturbance of the during dry weather conditions, leading to seed losses of 50-100 per cent reported by IITA (6). Pod shattering is aggravated if there is rain followed by dry weather, low humidity, high temperature, rapid temperature changes, wetting and drying reported by Agrawal *et al*., [1]. The loss of seeds by pod dehiscence is one of the major reasons for low yield in soybean. Most of the soybean genotypes are prone to shattering. In this crop hybridization is a tedious process as small and fragile flower leads to injuries to the flower parts and causes heavy flower shedding over 75 per cent reported by Johnson and Bornard, [7].

Mutation breeding is substitute to conventional breeding for varietal improvement reported by Sanjay Gandhi *et al*., [10]. Mutation breeding offers scope for achieving accomplishments through backcross breeding and selection reported by Lavanya *et al*., [9]. Mutation using physical and chemical mutagens

help to create genetic variation is the basis of improvement in quantitatively inherited characters which help in plant improvement without altering the original genetic makeup. It is speedy method to improve the crop varieties, without resorting to hybridization and back crossing. The identification of resistant sources for pod shattering is one of the most important aspects in the management of pod shattering. Hence, the present investigation was carried out with mutant population of soybean for screening of elite mutants tolerant to pod shattering.

MATERIAL AND METHODS

Experimental site:

The research work was carried out at Post-graduate research farm, RCSM College of Agriculture, Kolhapur (M. P. K. V. Rahuri) during Summer and Kharif 2017-2018 for M_1 and M_2 generation respectively. The materials used and methods adopted for various studies under this investigation are presented below.

Biological material:

Three diverse genotypes of soybean *viz.,* Phule Agrani (KDS344), Phule Sangam (KDS-726) and KS-103 were used for mutagenic treatment. The source seed for first two genotypes were Agriculture Research Station Kasabe-Digraj, Dist-Sangli and KS-103 from Agril. Botany Section, RCSM College of Agriculture, Kolhapur.

Physical mutagen:

The mutagen Gamma rays (cobalt-60) was used. The seeds of soybean varieties *viz*., Phule Agrani, Phule Sangam and KS-103 were treated with gamma rays at Mutation Breeding Section, Department of Nuclear and Agriculture, BARC, Trombay, Mumbai 400 085.

Mutagenic treatment with gamma rays:

Sr.No	Genotype	Dose of Irradiation with Gamma rays (Gray)
1	Phule Agrani (KDS-344)	0 (Control), 200, 300, 400
2	Phule Sangam (KDS-726)	0 (Control), 200, 300, 400
3	KS-103	0 (Control), 200, 300, 400

Details of experiment:

Design : R.B.D. (Randomized Block Design)
Treatments : 12 (9 Treatments + 3 control)
Replications : Three
Plot size : 3 x 2.5 cm
Fertilizer : 50:75:25 kg ha^{-1}
Spacing : 45 x 5 cm
Date of sowing : 14 February 2018

Screening for pod shattering resistance:

At field level: Visual screening is the most effective and efficient method for identifying mutant phenotypes. In the present study, 15 mutants were selected in each treatment and observations of shattering were taken at 5 days interval. Similar method of screening was reported by Yamada, *et al*. [12] and Khan *et al*. [8].

Under Laboratory Condition: The pod shattering resistance was recorded with mutant plants which were showing no shattering at field level after delaying harvesting and exposing to sunlight. The screening was done under laboratory condition by following the methodology adopted by IITA [6]. A sample of pods from each mutant plant were collected and kept in oven at 40°C for 7 days.

On the 7th day the number of shattered pods was counted and expressed in percentage as below,

Pod shattering percentage (%) = Number of pods shattered / Total number of pods x 100

The genotypes were classified into different categories based on their reaction to pod shattering. The scoring rate was followed according to method adopted by IITA [6].

Sr.No	Category	Resistant reaction
1	No pod shattering	Shattering resistant
2	<25% pod shattering	Shattering tolerant
3	25-50% pod shattering	Moderately shattering
4	51-75% pod shattering	Highly shattering
5	>75% pod shattering	Very highly shattering

RESULTS AND DISCUSSION

Pod shattering resistance was evaluated both in laboratory and field conditions

Table 1: Screening for shattering tolerance at field level

Genotype	Shattering 5th DAM	Shattering % 10th DAM	Shattering % 15th DAM
Phule Agrani (Control)	4.26	33.06	64.10
Phule Agrani 200 Gy	2.19	26.33	49.35
Phule Agrani 300 Gy	2.65	23.80	51.87
Phule Agrani 400 Gy	1.85	26.38	51.14
Phule Sangam (Control)	3.84	22.81	54.70
Phule Sangam 200 Gy	2.48	27.97	60.73
Phule Sangam 300 Gy	2.81	25.07	51.27
Phule Sangam 400 Gy	2.84	26.44	52.99
KS-103 Control	4.27	27.01	68.37
KS-103 200 Gy	5.83	22.35	51.84
KS-103 300 Gy	4.19	21.14	50.98
KS-103 400 Gy	4.06	22.76	48.24
Mean	**3.43**	**25.42**	**54.63**
Range	**1.85-5.83**	**21.14-33.06**	**48.24-68.37**

Screening on 5th day after maturity

The mean shattering was 3.43 per cent with a range from 1.85 to 5.83 per cent. Genotypes were classified in three groups based on shattering percentage on the 5th day after maturity viz., no shattering (0); 0.1 -5 per cent shattering (11 treatments) and > 5 per cent shattering (1 treatment). Maximum shattering was observed in genotype KS-103 200 Gy (5.83%).

Screening on 10th day after maturity: The mean shattering was 25.42 per cent (Range 21.14 to 33.06%). Genotypes were classified in three groups i.e. no shattering (none);

15 to 25 per cent shattering (5 treatments) and > 25% shattering (7 treatments). Maximum shattering was observed in genotype Phule Agrani Control (33.06%) followed by Phule Sangam 200 Gy, (27.97%), KS 103 Control. (27.01%) and Phule Sangam 400 Gy (26.44%).

Screening on 15th day after maturity: Mean shattering on 15th day after maturity was 54.63 per cent (Range 48.24 to 68.37 %). Genotypes were classified in three groups based on shattering percentage 15 days after maturity *viz*., less than 25 per cent shattering as tolerant (none treatments ;25 50 per cent shattering as moderately tolerant (2 treatments) and more than 50 per cent shattering as sensitive (10 treatments). Minimum shattering was observed in genotype KS-103 with 400 Gy. (48.24%) followed by Phule Agrani 200 Gy (49.35%), whereas highest shattering in KS 103 Control (68.37%) followed by Phule Agrani Control (64.10%).

Under Laboratory Condition: The plants not sowing shattering at field level after delaying harvesting and exposing to sunlight. In the present study, 93 mutants were selected based on field observations to shattering. In laboratory lowest shattering was recorded by M_{18} (300 Gy) of Phule Agrani (6.70%) followed by M_8 (200 Gy) of Phule Agrani (10.58%) and M_{15} (300 Gy) of Phule Agrani (11.76%). Highest shattering recorded by the mutants M_{22} (400 Gy) of Phule Agrani (64.44%) followed by M_{39} (200 Gy) of Phule Sangam (60.87%), M_{32} (200 Gy) of Phule Sangam (60.81%) and M_{65} (200 Gy) of KS-103 (60.27%). Phule Agrani was under very highly shattering type (64.44%), Phule Sangam was under very highly shattering type (60.87%) and KS 103 showed highly shattering percentage of 60.27 % in M_2 generation.

Table 2: Pod shattering at laboratory

Mutant	Genotype	Shattering %	Category
1	Phule Agrani 200 Gy	18.60, 24.28, 23.40	Tolerant
2		25.36	Moderately Shattering
3		21.69, 20.25	Tolerant
4		25.59	Moderately Shattering
5		10.58	Tolerant
6		40.48	Moderately Shattering
7	Phule Agrani 300 Gy	38.51	Moderately Shattering
8		24.24	Tolerant
9		31.34, 39.32	Moderately Shattering
10		15.38, 11.76	Tolerant
11		26.51	Moderately Shattering
12		15.13, 6.70	Tolerant
13	Phule Agrani 400 Gy	13.43, 23.91, 20.29	Tolerant

14		64.44	Highly Shattering
15		34.40	Moderately Shattering
16		24.68, 21.69	Tolerant
17		52.53	Highly Shattering
18		31.48	Moderately Shattering
19		55.13	Highly Shattering
20		39.08	Moderately Shattering
21		23.53, 16.67	Tolerant
22	Phule Sangam 200 Gy	33.75	Moderately Shattering
23		60.81	Highly Shattering
24		22.67	Tolerant
25		44.62	Moderately Shattering
26		53.42	Highly Shattering
27		23.29	Tolerant
28		30.77	Moderately Shattering
29		60.87	Highly Shattering
30		28.13	Moderately Shattering
31		20.34, 14.29	Tolerant
32	Phule Sangam 300 Gy	34.07	Moderately Shattering
33		13.79	Tolerant
34		52.78	Highly Shattering
35		19.28	Tolerant
36		30.43	Moderately Shattering
37		24.00, 23.08, 24.39	Tolerant
38		40.51	Moderately Shattering
39	Phule Sangam 400 Gy	58.33	Highly Shattering
40		56.16	Highly Shattering
41		43.37	Moderately Shattering
42		21.88, 22.97	Tolerant
43		27.54	Moderately Shattering
44		23.94	Tolerant
45		32.47	Moderately Shattering
46		55.13	Highly Shattering
47	KS-103 200 Gy	53.40	Highly Shattering
48		33.07	Moderately Shattering
49		23.53	Tolerant
50		55.41, 60.27	Highly Shattering
51		40.78, 28.09, 31.67	Moderately Shattering
52		19.44	Tolerant
53	KS-103 300 Gy	25.00	Moderately Shattering
54		22.22	Tolerant
55		34.04, 26.09	Moderately Shattering
56		24.14, 22.37, 22.97	Tolerant
57		31.07, 35.61	Moderately Shattering
58	KS-103 400 Gy	47.95, 35.40, 29.08	Moderately Shattering
59		23.66, 23.17, 18.46, 22.97	Tolerant
60		40.51	Moderately Shattering
61		16.00, 22.78, 23.08, 18.46	Tolerant
62		30.95, 30.56	Moderately Shattering
63		22.92	Tolerant
64	**Mean**	**30.47**	
65	**Range**	**6.70 to 64.44**	

Among the 93 mutants, 45 mutants were identified as tolerant types, 35 mutants observed to be medium shattering, 13 mutants showed highly shattering. These findings are similar to genotypic studies in Soybean given by Gadde [5] and Khan *et al*., [8]. Highest tolerant type was observed in genotype Phule Agrani followed by KS-103 and Phule Sangam. In the genotype Phule Agrani 18 Mutant was observed which were showing tolerant reaction whereas, In Phule Sangam 12 Mutant and in KS-103 15 mutant were showing tolerant reaction. Screening of mutants for pod shattering revealed 45 mutants as tolerant types, 35 mutants as moderately shattering and 13 mutants as highly shattering categories in M_2 generation. Similar results were reported by Bhara *et al*. [3] in Soybean. These shattering tolerant

mutants can be further evaluated for yield contributing characters in succeeding generations for the selection of elite mutants for resistance to novel trait. None of the mutants showed resistance to pod shattering. Mutant plants displayed a range of reduction in depending upon the genotype. This variation is being utilized for variety development.

CONCLUSION

Sufficient variability obtained through present mutagenic treatment to soybean genotypes, reveals that through mutation breeding extent of variability is possible to increase involving diverse parents in soybean. Pod shattering is one of the major constraints in soybean, which reduces the yield potential considerably. Hence, the identification of resistant sources for pod shattering is one of the most important aspects in the management of pod shattering. Mutagenesis is a well-recognized potential tool to induce high genetic variability for effective selection towards improvement in yield and quality. 45 mutants of three genotypes of soybean were found to be tolerant to pod shattering. The identified mutants can be screened further and used in hybridization for development of resistant variety.

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