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REGULAR ARTICLE

CHANGES ON QUANTITATIVE TRAITS OF BLACK GRAM (*VIGNA MUNGO* (L.) HEPPER) INDUCED BY EMS IN M_2 GENERATION

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SUMMARY

The systematic collection of black gram is displayed inadequate variability for biotic and abiotic desirable genes. It is possible that genes for high productivity could have been lost due to overriding role of natural selection and genetic base of the present day collection remains poor due to lack of genetic variability owing to their autogamous nature. Mutagenesis has been widely used as a potent method of enhancing variability for crop improvement. In the present investigation, the genetic variability was induced to improve quantitative traits of black gram in M_2 generation induced by EMS. The results showed that a significant enhancement in quantitative mean performance archived at 0.1% EMS concentration.

Keywords: Black gram, Genetic variability, Ethylmethane Sulphonate, M_2 generation

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1. Introduction

Black gram is an important pulse crop occupying unique position in Indian agriculture. Among the pulses, it stands fourth in production and acreage [1]. In the past, there were attempts to increase the productivity of this crop using conventional breeding approaches at different agricultural research centers. However, the national productivity of black gram is alarmingly remaining around 500 kg/ha due to restricted cultivation in the marginal lands [2, 3]. Black gram, under cultivation in India is about 3.25 million hectares with an annual production is 1.45 million tons. About 70% of the total area is in the central and southern part of the country, which contributes about 77% of the total production [1].

The systematic collection of black gram is displayed inadequate variability for biotic and abiotic desirable genes. It is possible that genes for high productivity could have been lost due to overriding role of natural selection [4] and genetic base of the present day collection

remains poor [5] due to lack of genetic variability owing to their autogamous nature. So, the creation of variability is difficult through hybridization due to its high self-pollination and more flower drop [1].

Research on Black gram is lagging behind than that of cereals and other legumes. In order to improve yield and other polygenic characters, mutation breeding can be effectively utilized [1]. The efficiency of selection depend on the nature and magnitude of variability in a population and extend to which desirable characters are heritable [6]. Mutation induction has become an establishment tool in plant breeding to supplement existing germplasm and to improve cultivars in certain specific traits [7]. Induction of mutation forms an important part of breeding programme as it widens the gene pool through creation of genetic variability. Therefore, the genetic variability is the basic requirement for making progress in crop breeding [8]. Hence, induced mutation using physical and chemical mutagen is one method to create genetic variation resulting in new varieties with better characteristics [9].

2. Materials and Methods

Collection of seeds

Black gram (*Vigna mungo* (L.) Hepper) variety Vamban-1 was selected to induce mutation by EMS to analyze quantitative traits in M_2 generation. The certified seeds of black gram were collected for this investigation from Vamban Pulse Researhch Station (Pudukottai), Tamilnadu, India.

Chemical mutatgen

One of the chemical mutagens namely Ethylmethane sulphonate (EMS) was used for induction of mutation on seed propagules. Ethylmethan sulphonate was obtained from Himedia Laboratory Limited, Mumbai, India which having a dosimetry/half-life period is 30 hours with a molecular weight is 124.16 and density is 1.20.

Induction of mutation (EMS)

Six hundred well matured healthy and uniform size of non-dormancy seeds were subjected to the mutagenic treatment. The solution of EMS was prepared with corresponding to the required concentration in distilled water. The volume of solution was about three times than that of volume of seeds. The seeds were pre-soaked in double distilled water for five hours at room temperature (28 ± 2^{o}C) prior to treatment. After the pre-soaking the excess of moisture in the seeds were removed by filter paper. Then seeds were soaked in the freshly prepared aqueous solution of EMS in the following concentrations (%) Viz 0.02, 0.04, 0.06, 0.08, 0.1, 0.12, 0.14, 0.16 and 0.18 % for six hours at room temperature (28 ± 2^{o}C) with an hour intermittent shaking. The pH of aqueous solution was adjusted at 8.5 by using 0.2 M solution of sodium tetra borate (Borax). After the treatment, the seeds were washed thoroughly with distilled water for eight to ten times and sown in the field as randomized block design with three replication to rise M_1 generation.

Raising of M_2 Generation

From seeds of M_1 generation, M_2 generation was raised to study quantitative traits. The optimum concentration such as, 0.08, 1.0 and 1.2 % was selected and these sets of seed sown in the field on randomized block design. All the control measures were maintained through out the growth period.

Control

Healthy, well-matured, non-dormant, untreated seeds were used as control.

Experimental design

The chemically treated (EMS) and control seeds were grown at the Breeding field of Department of Botany, Annamalai University, Annamalainagar, TN, India.

Harvest of M_2 generation

At 60th day (maturity) triplicates (30 plants/plot for each dose) of all 0.08, 1.0 and 1.2 % EMS treated population with control were separately harvested and the following quantitative traits were duly analyzed such as, plant height, number of branches/plant, number of leaves/plant, number of fruit clusters/plant, number of pods/plant, number of seeds/pod, yield/plant (g) and 100 seed weight (g) The bulked seeds collected form each dose/control were saved and raised to M_2 generation were grown in suitable season for RBD with three replications.

Statistical analysis

Analysis of variance (ANOVA for RBD) was to use to analyze yield and its component traits calculated using the software NPRCSTAT, developed in National Pulse Research Center, Vamban, Pudukottai, TN, India. The variance observed among the replication was exclusively non-heritable and hence treated as environmental variance. The variance of (EMS treated) M_2 populations was partitioned into heritable and non-heritable components [10]. Phenotypic and genotypic coefficient of variation (PCV & GCV) was computed using the formula adopted by Burton [11] and categorized of the range of variation was done as proposed by Sivasubramanian and Madhavamenon [12]. Heritability (h2) was computed using the formula according to Lush [13] and it was classified according to Robinson [14]. Genetic advance was estimated adopted the method suggested by Johnson et al [15]. The significance was assessed at the 5% and 1% probability level, unless otherwise stated.

3. Results and Discussion

Quantitative parameters

An estimation of the extent of variability induced in M_2 generation will be of great value in providing useful information for carrying out further selection. In view of this, the present study was investigated to estimate the effect of mutagens and their impaction in mean performance of quantitative and qualitative characters on M_2 generation.

Table 1.Quantitative traits variation induced by EMS in M2 generation

EMS	0.08%	0.1%	0.12%
Plant height (cm)	52.86 ± 2.87	55.72 ± 1.58	48.96 ± 3.12
Number of branches	4.86 ± 1.66	4.92 ± 1.95	3.97 ± 2.22
Number of leaves	26.58 ± 1.63	29.57 ± 1.25	23.37 ± 1.47
Number of days taken for 50% flowering	32.76 ± 1.52	30.34 ± 2.68	33.24 ± 2.36
Number of fruit cluster	16.46 ± 1.25	18.12 ± 0.87	13.74 ± 0.67
Number of pods	30.95 ± 1.25	33.97 ± 1.79	31.21 ± 2.87
Number of seeds/ pod	8.57 ± 0.28	9.25 ± 0.63	8.12 ± 0.34
100 seed weight (g)	4.966 ± 0.68	5.214 ± 0.39	4.827 ± 0.42
Yield/Plant (g)	7.11 ± 0.06	9.56 ± 0.02	6.11 ± 0.06

± Standard error

A significant enhancement in mean performance was observed in plant height, number of branches per plant, number of leaves per plant, number of days taken for 50% flowering, number of fruit cluster per plant, number of pods per plant, number of seeds per pod, 100 seed weight (g), yield per plant, with effect of EMS than control (Table 1). Quantitative

characters in the M_2 generation such as number of pods per plant, number of seeds per plant, 100 seed weight and yield per plant were higher in EMS treatment in green gram than the control [16]. Plant height, number of branches per plant, number of pods per plant, 100 seed weight (g) and seed yield were increased with effect of EMS in soybean on M_2 generation [17]. Among different concentrations (0.08, 0.1 and 1.2%) 0.1 % was showed high quantitative mean performance in M_2 generation. This was confirmed with earlier reports on legumes with effect of different mutagen [18 -20]. Improved quantitative traits namely, plant height, number of branches per plant, 100 seed weight and plant yield with effect of EMS in M_2 generation of chick pea [21].

EMS	0.08%		0.1%		1.2%	
	PCV	GCV	PCV	GCV	PCV	GCV
Plant height	18.11	19.74	23.18	21.67	16.66	18.34
Number of branches	19.59	17.54	29.31	27.33	7.58	6.81
Number of leaves	21.26	19.80	32.31	30.91	19.11	17.52
Number of days taken for 50% flowering	28.16	20.56	33.33	30.58	17.75	14.23
Number of fruit cluster	30.16	28.55	13.45	12.79	11.72	10.13
Number of pods	16.73	15.66	46.74	45.79	19.28	19.12
Number of seeds/ pod	16.62	14.26	22.47	20.32	15.97	13.11
100 seed weight	19.16	18.52	25.44	23.83	17.24	15.44
Yield/Plant	10.43	9.56	26.55	25.17	10.29	10.16

Table 2. Phenotypic and genotypic variation induced by EMS in M_2 generation

Variability analyses

Phenotypic and genotypic co-efficient variation (PCV and GCV), heritability and genetic advance (GA %) as percent of mean

Association among phenotypic and genotypic co-efficient of variation (PCV and GCV), heritability and genetic advance appear to be good criteria for selection in crop improvement programme. The observed variability is a combined measure of genetic and environmental to generation. Heritability may give useful indication for relative value of selection among the materials in hand [22].

PCV and GCV

Among the dose/concentration of mutagenic treatments, quantitative and qualitative traits showed high and moderate PCV and GCV in M_2 generations. The quantitative traits such as, plant height, number of branches per plant, number of leaves per plant, number of fruit cluster per plant, number of pods per plant, number of seed per pod, number of seeds per pod and seed yield seed protein showed PCV and GCV were significantly higher in EMS (Table-2). Singh et al (1998) reported high PCV and GCV values in plant height, primary branches per plant, number of seeds per plant and yield per plant in okra (Abelmoschus esculentus) with effect of gamma rays and EMS. However, low PCV and GCV were recorded in days for 50 % flowering. Deepalakshmi and Anandakumar (2004) recorded high PCV and GCV value in plant height, number of primary branches per plant, number of cluster per plant, number of pods per plant, pod length per plant and seed yield per plant in black gram with effect of gamma rays and EMS. In M2 generation, a significant variation with high PCV and GCV values were recorded almost in EMS at 0.1%.

EMS	0.08%		0.1%		1.2%	
	h2	GA (%)	h2	GA (%)	h2	GA (%)
Plant height	29.29	42.64	92.35	60.62	71.48	39.31
Number of branches	36.73	65.43	79.85	92.68	71.46	54.40
Number of leaves	10.18	4.93	26.35	20.89	18.69	37.06
Number of days taken for 50% flowering	29.75	47.13	89.36	63.90	30.84	55.80
Number of fruit cluster	25.33	20.59	81.84	30.13	20.26	31.44
Number of pods	66.39	23.13	96.28	58.02	76.94	18.72
Number of seeds/ pod	40.16	38.85	48.33	48.54	32.66	20.32
100 seed weight	30.88	63.01	68.52	79.78	61.33	76.85
Yield/Plant	29.98	12.35	86.55	17.91	31.14	14.26

Table 3. Heritability (h2) and genetic mean (%) induced by EMS in M_2 generation

Heritability (h2) and genetic advance as % of mean (GA %)

In the present study, heritability and genetic advance (%) showed high to moderate level among the dose/concentrations in M_2 generation. High h2 and GA as % of mean observed in plant height, number of branches per plant, number of leaves/plant, number of days for 50% flowering, number of fruit cluster per plant, number of pods per plant, number of seeds per pod, 100 seed weight and yield per plant in gamma rays and EMS treatments (Table 3). Deepalakshmi and Ananadakumar (2004) observed high heritability and genetic advance as % of mean in plant height, number of primary branches per plant, number of clusters per plant, number of pods per plant, pod length, and seed yield per plant in black gram with different dose/concentrations of EMS.

Conclusion

In the present investigation, chemical mutagen EMS was employed through mutagenesis, which clearly showed genetic variation in black gram genotype with respective control. Among the dose/concentrations, 0.1% EMS provided most significant enhancement in mean performance of quantitative traits with genetic variation.

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