

Correlation, Genetic Advance and Heritability studies in EMS induced mutants of Blackgram (*Vigna mungo* L Hepper) for seed yield in M2 generation

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Abstract: *In the current study, correlations, genetic advance and heritability were estimated in order to determine the level of EMS-induced genetic variability in the black gram accession IC-436524. For the purpose of estimating genetic parameters data were gathered based on the Phenotypical characteristics that differed between the control plant (untreated) and the T-9 blackgram variety. Mutants were categorised as high mutants if their investigated quantitative characters had a high mean value. Character variability was shown to be significantly different in M2 generations according to correlation, heritability studies, and genetic advance estimations. This increased genetic variability for seed yield and its contributing traits shows there is a lot of scope for development in black gram varieties, which could be helpful for further crop improvement and selection. Lower doses of EMS-induced mutants (0.2 and 0.3%) were exposed to be effective for generating the appropriate variability in the yield and yield contributing traits.*

Keywords: Blackgram, Phenotypic and Genotypic correlation, Genetic advance and Heritability

Introduction:

Blackgram, sometimes known as 'urdbean,' is a type of pulse that belongs to the Leguminaceae family. Especially for vegetarians, legumes and pulses have proven to be a significant source of protein (Sonu Goyal et al., 2020). Legumes possess a special trait that facilitates the fixation of atmospheric nitrogen and increases soil fertility (Deewanjee and Sarkar, 2017). Pulses are frequently referred to as "poor man's meat" because they are a widely available source of protein for the general public. They are mostly grown in dry soil in places with little annual rainfall. Because pulse crops are more profitable, easier to grow, require fewer inputs, require less water for irrigation, are shorter-lived crops, and are generally healthier due to their high protein content and ease of digestion, farmers find it easier to cultivate pulse crops than other food crops. They are also very nutrient-dense with plenty of minerals and the amino acids cysteine and methionin, which contain less sulphur. Due to issues like low yielding and limited genetic variety, the relevance of pulse crops has significantly diminished from previous decades. Urbanization, deforestation, conversion of agricultural lands to non-agricultural lands for transportation, industrialization, use of low-quality yielding seeds that are more susceptible to diseases, irregular

poor management practises, change in climatic conditions, etc. are additional factors that have contributed to a decrease in productivity and production of pulses. There is an urgent need for the production and development of pulse crops to satisfy the demand by making use of the available agricultural area because such output is not even adequate to keep up with the demand. Genetic variation among genotypes and relationship between major yield contributing characters is of vital importance to crop breeding programmes for developing new varieties (Tah, 2006).

To overcome the above problem for increasing the crop yield and productivity induced mutations has played an important role in increasing the yield. Several researches have been carried out on different crops to increase the productivity after Green Revolution has reported success results in wheat and cereals. According to FAO/IAEA 2021 report there was only 9 blackgram varieties released out of 466 released legume varieties. This shows that this blackgram crop is less exploited for creating genetic variabilities to develop new varieties. Therefore strong efforts are needed to develop new varieties by controlling economically important characters, especially useful for agriculture purposes. (Laskar and Khan, 2017). The chemical agent EMS to induce mutations for creating genetic variability in black gram as earlier reports has proved that EMS is a highly potent chemical mutagen to induce variability in plants. Some research works have been carried out in black gram using gamma rays, combinations with EMS other chemical alkalizing agents to increase the yield and yield contributing traits and good results have been reported. Induced mutations are highly used to create genetic variability with desirable agronomical characters in the plants like black gram as these are self pollinated crops having less chance of genetic variability.(Wani, 2017, 2021; Raina et al., 2020; Sonu Goyal et al., 2019; Kumar et al., 2019; Amin et al., 2019; Laskar et al., 2019)). Similarly in other crops like chickpea (Raina et al., 2019), lentil (Laskar et al., 2018), mungbean (Singh, 2009), pigeon pea (Etther et al., 2019),blackgram (Sonu Goyal et al., 2020). The present investigation was carried with EMS to induce mutations for creating genetic variability in seed yield and yield contributing traits. Through mutations new allelic combination of genes without altering the basic genomic structure of the plants or organism can be created. The effective mutations depends on the concentration of mutagen used (Sonu Goyal et al 2020). Optimum doses of EMS induces desired and successful mutations which could be utilized for the isolating promising mutant lines with desirable agro-economical traits and development of blackgram crop (Sonu Goyal et al., 2020). Many researchers has also reported that EMS a chemical mutagen, and an alkalizing agent at lower doses acts as a super mutagen for inducing mutations in mungbean (Singh, 2007), chickpea (Shah et al 2008), urdbean (Goyal et al., 2019), lentil (Solanki and Sharma 1994). Knowledge on genetic variability for the characters of economic importance that effects the relationship of yield and yield components for the available genotypes are of utmost importance which helps in planning the future breeding programme for genetic improvement for yield potential of any crop species. Inheritance of quantitative characters is regulated by different genes interactions with additional effects on phenotypic variability (Laskar et al., 2018; Sonu Goyal et al., 2019).

Materials and Methods:

Collection of Seeds

Blackgram (accession IC-436524) seeds were obtained from NBPGR regional centre Hyderabad and T9 (check) seeds were obtained from ICAR-CRIDA, Hyderabad.

Treatment of Seeds using EMS

M1 generation was raised by treating the seeds with EMS prepared at different known concentration *Viz.*,0.2%,0.3%,0.4%,0.5% and Control(untreated EMS) along with T9(check). All agronomic practices were strictly followed during preparation of field, sowing and subsequent management of M1, M2 and M3 generations. Quantitative characters of M1 generation are useful in determination of the mutations created in subsequent generations (Neelavathi and Govindarasu 2010) to determine the desired traits.

M1 generation

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Data was prepared accordingly and mutants with high range of morphological characters for high yielding in each row of each concentration like plant height per plant (cm), number of branches per plant, number of leaves per plant, number of clusters per plant, number of seeds per plant, Seed yield(g) per plant, 100 seed weight(g), root length(cm) and root nodules per plant were screened and isolated in M1 generation(raised by inducing EMS).Seeds of M1 plants were harvested separately and preserved in paper bags for next generation studies(M2 generation).

M2 generation

Data was prepared accordingly and mutants with high range of morphological characters for high yielding like plant height per plant (cm), number of branches per plant, number of leaves per plant, number of clusters per plant, number of seeds per plant, Seed yield(g) per plant, 100 seed weight(g), root length(cm) and root nodules per plant in each row of each concentration were screened and isolated in M2 generation (raised by M1 progeny). Seeds of M2 plants were harvested separately and preserved in paper bags for next generation studies (M3 generation).Fifty seeds from each treatment viz.,0.2% mutants, 0.3% mutants, 0.4% mutants, 0.5% mutants, Control and T-9(totally 300 seeds) were sown in field located at Kodad, Suryapet, Telangana to evaluate the seed yield and yield contributing traits.

For raising M2 generation total 180 seeds of 30 healthy seeds from each treatment were collected from high yielding mutant and control along with T9 and were sown with 10cm x 30 cm distance between plants per row in a field in RBD (Randomized Block Design) with three replications each along with control. Similar agronomical parameters as M1 generation were practiced to raise M2 generation. M2 population was evaluated for agronomic and morphological characters by phenotypical observations which are yield and yield contributing traits Plants which have high quantitative characters with high yielding in each row of each concentration were separated and seeds from those plants were collected and data was prepared based on the yield produced and yield contributing characters. For raising M3 generation total 300 seeds of 50 healthy seeds from each treatment were collected from high yielding mutant and control along with T9 and were sown with 10cm x 30 cm distance between plants per row in a field in RBD (Randomized Block Design) with three replications each along with control. Similar agronomical parameters as M1 generation were practiced to raise M3 generation.

Data collected on the quantitative phenotypic characters like plant height per plant (cm), number of branches per plant, number of leaves per plant, number of clusters per plant, number of seeds per plant, Seed yield(g) per plant, 100 seed weight(g), root length(cm) and root nodules per plant were studied to assess the extent of induced phenotypic and genotypic variations and by genetic advance, heritability. The significance of differences between the mean values of treated mutants, Control and T-9 in M2 generations. Genotypic coefficient of variation (GCV) and phenotypic coefficient of variation (PCV) (Burton, 1952), heritability (Allard, 1960), genetic advance (Johnson *et al.*, 1955) were estimated.

Results and Discussion:

Data collected and recorded and analysed for correlation studies, genetic advance, and heritability estimates revealed that there was highly significant differences among the mutants and showed genetic variability among the treated mutants (Radhikaben et al., 2020) when compared with Control and T-9. This reflects that EMS induced mutations in M1 generation had a positive effect on the mutants in M2 generation for increasing the seed yield along with their contributing traits.

Table1. Blackgram Mutants In M2 Generations Show Phenotypic Association For Seed Yield And Yield Contributing Characteristics.

“Phenotypic	Pl.Ht(cm)	No.Br/Pl	No.L/Pl	No.Cl/Pl	No.P/Pl	No S/Pl	S.Y(g)/Pl	100 S.Wt(g)	R.L(cm)/Pl
Pl.Ht(cm)	1.000								
No.Br/Pl	0.792**	1.000							

No.L/Pl	0.831**	0.751**	1.000						
No.Cl/Pl	0.371**	0.234NS	0.203NS	1.000					
No.P/Pl	0.617**	0.372**	0.472**	0.875**	1.000				
No S/Pl	0.575**	0.403**	0.537**	0.445**	0.686**	1.000			
S.Y(g)/Pl	0.576**	0.399**	0.481**	0.442**	0.668**	0.960**	1.000		
100 S.Wt(g)	0.747**	0.548**	0.550**	0.385**	0.593**	0.796**	0.883**	1.000	
R.L(cm)/Pl	0.882**	0.611**	0.787**	0.280*	0.597**	0.557**	0.551**	0.682**	1.000
No.R.N/Pl	0.777**	0.462**	0.723**	0.370**	0.623**	0.539**	0.545**	0.652**	0.915**

*Statistically significant at the 0.05 percent and ** level (0.01 percent and 0.05%, respectively)

Table 2. M2 Generation Blackgram Mutant Seed Yield and Contributing Characteristics Analyzed Genetically

“Genotypic	Pl.Ht(cm)	No.Br/Pl	No.L/Pl	No.Cl/Pl	No.P/Pl	No S/Pl	S.Y(g)/Pl	100 S.Wt(g)	R.L(cm)/Pl
Pl.Ht(cm)	1.000								
No.Br/Pl	1.034**	1.000							
No.L/Pl	0.785**	0.693**	1.000						
No.Cl/Pl	-0.629**	-0.642**	-0.656**	1.000					
No.P/Pl	-0.144NS	-0.189NS	-0.037NS	0.830**	1.000				
No S/Pl	0.477**	0.340**	0.598**	0.418**	0.877**	1.000			
S.Y(g)/Pl	0.481**	0.375**	0.594**	0.452**	0.919**	1.004**	1.000		
100 S.Wt(g)	0.923**	0.994**	0.853**	0.002NS	0.630**	0.989**	0.952**	1.000	
R.L(cm)/Pl	0.867**	0.877**	1.089**	-0.825**	-0.218NS	0.574**	0.539**	0.771**	1.000
No.R.N/Pl	0.373**	0.350**	1.180**	-0.923**	-0.232NS	0.748**	0.614**	0.230NS	0.873**

*Statistically significant at the 0.05 percent and ** level (0.01 percent and 0.05%, respectively)

In M2 generation, the correlation stastical analysis data “revealed that highly significant phenotypical and genotypical correlation among all the morphological characters under study. Positive and significant phenotypical and genotypical correlation was observed for the seed yield vs all the characters under study viz., plant height ($r_p = 0.576^{**}$, $r_g = 0.481^{**}$), number of branches per plant ($r_p = 0.792^{**}$, $r_g = 0.375^{**}$), number of leaves per plant ($r_p = 0.481^{**}$, $r_g = 0.594^{**}$), number of clusters per plant ($r_p = 0.442^{**}$, $r_g = 0.452^{**}$), number of pods per plant ($r_p = 0.668^{**}$, $r_g = 0.919^{**}$), number of seeds per plant ($r_p = 0.960^{**}$, $r_g = 1.004^{**}$), 100 seed weight per plant ($r_p = 0.883^{**}$, $r_g = 0.952^{**}$), root length per plant ($r_p = 0.551^{**}$, $r_g = 0.539^{**}$), number of root nodules per plant ($r_p = 0.545^{**}$, $r_g = 0.614^{**}$) respectively (Table 1&2). In M2 generation, the correlation analysis results revealed that among ten quantitative characters, Number of seeds/pl was phenotypically and genotypically “significantly correlated with seed yield ($r_p = 0.960^{**}$, $r_g = 1.004^{**}$) followed by 100 Seed weight(g) ($r_p = 0.883^{**}$, $r_g = 0.952^{**}$), number of Pods per Plant($r_p = 0.668^{**}$, $r_g = 0.919^{**}$), Plant height(cm)($r_p = 0.576^{**}$, $r_g = 0.481^{**}$), number of leaves per plant ($r_p = 0.481^{**}$, $r_g = 0.594^{**}$), number of clusters per plant ($r_p = 0.442^{**}$, $r_g = 0.452^{**}$), number of branches per plant ($r_p = 0.399^{**}$, $r_g = 0.375^{**}$). Significant direction, degree of association between and among the most important traits viz., pods, clusters per plant, seeds and seed yield per plant were reported earlier in blackgram by Goyal et al. 2021; Laskar et al., 2018.

Table 3. Black gramme mutants' quantitative features, including M2 generation heritability, genetic gain, phenotypic coefficient variation, and genotypic coefficient variation.

“Characters	Heritability(%)	Genotypic Coefficient of Variations (GCV)	Phenotypic Coefficient of Variations (PCV)	Genetic Advance	Genetic Advance value % means
Pl.Ht(cm)	22.750	9.032	18.936	2.969	8.874
No.Br/Pl	28.771	14.918	27.812	0.440	16.483
No.L/Pl	37.681	15.815	25.764	2.543	19.998
No.Cl/Pl	41.655	18.486	28.642	1.559	24.577

No.P/PI	32.084	13.552	23.926	4.394	15.813
No S/PI	50.041	18.220	25.756	34.849	26.551
S.Y(g)/PI	35.510	21.878	36.713	1.113	26.856
100 S.Wt(g)	8.643	5.128	17.443	0.094	3.106
R.L(cm)/PI	18.258	7.833	18.332	1.369	6.895
No.R.N/PI	6.513	5.006	19.615	3.225	2.632”

In M2 generation, For the character “plant height, the phenotypic coefficients of variation (PCV) recorded was 18.936, Genotypic coefficient of variation (GCV) observed was 9.032. A heritability estimate of 22.75 was found, and the average observed rate of genetic progress was 8.874%. The observed genotypic and phenotypic coefficients of variation for the character "number of branches per plant were 27.812 and 14.917, respectively.” The observed genetic advancement as a percent of the mean was 16.483, and the observed heritability was 28.771 (Table 3). Phenotypic coefficients of variation (PCV) for the character number of leaves per plant were reported at 25.763, whereas genotypic coefficients of variation (GCV) were discovered to be 15.815. Heritability estimations were found to be 37.680, while the observed rate of genetic progress as a percentage of the mean was 19.998. The measured genotypic coefficient of variation (GCV) was 18.486 while the phenotypic PCV (PCV) was 28.641 for the feature of cluster density per plant. Heritability estimations were found to be 41.655%, with genetic improvement as a % of the mean reported at 24.577. The number of pods per plant is a trait with a high PCV of 23.925 and a low GCV of 13.552, both of which were found in the study. Estimates of heritability were 32.084, and genetic improvement was 15.813 percent of the mean. The detected genotypic coefficient of variation (GCV) for the character of seeds per plant was 18.219, whereas the measured phenotypic PCV was 25.756. Estimates for heritability were found to be 50.040%, with genetic progress as a percentage of the mean being 26.50%. Seed yield (g/pl) GCV was measured at 21.878, indicating a high degree of genetic diversity across samples. Whereas the phenotypic PCV was 36.713. Heritability was estimated to be 35.509, and genetic progress was found to be 26.856 percent above the norm. The measured genotypic coefficient of variation (GCV) for the character 100 seed weight (g) was 5.1280, whereas the reported phenotypic PCV was 17.443. Heritability estimations were 8.6430, and genetic progress was 3.1059 percent over the norm. The detected genotypic coefficient of variation (GCV) was 7.8330, whereas the reported phenotypic PCV for the character root length (cm) was 18.332. A heritability estimate of 18.257 and a genetic advance estimate of 6.8946 as a percent of the mean were reported. The PCV for the character root nodule count per plant was reported at 19.614, but the GCV was noted to be just 5.006. Heritability was estimated to be 6.512, and genetic progress was found to be 2.63 percent above the norm. In M2 generation, The heritability and Genetic advance estimations results revealed that the maximum heritability, Genetic advance mean value (%) 50.041% and 26.55% were reported This means that these traits may be further enhanced by means of phenotypic selection. The findings of Kumar et al. (2014), Panigrahi and Baisakh (2014) in blackgram were corroborated. Similar Previously, Singh et al. (2003) in *Abelmoschus esculantus* found an increase in heritability and genetic progress as a percent of the mean for the quantitative features. High genetic advancement for the character seed production, pod per plant, clusters, and quantity of seed was found (Bandi et al., 2018; Mehandi et al., 2013; Pathak et al., 2017; Gupta et al., 2006; Kumar et al., 2010). Comparable work may be found in the works of Kumar et al. (2012), Gupta et al. (2006), Bandi et al. (2018), Kachave et al. (2015), Arulbalachandran and Mullainathan (2016). Similarly, the GCV and PCV data showed that the character seed yield had the highest GCV and PCV at 21.878 and 36.713, followed by the number of clusters per plant at 18.486 and 28.642, the number of seeds per cluster at 18.22 and 25.756, and the number of pods per plant at 18.486 and 28.642. (13.552 and 23.926). The results also revealed that most of the characters under study were recorded higher PCV than GCV which indicates little environmental influence on the characters in the Blackgram mutants in M2 generation. Higher PCV and GCV for the traits clusters, pods and seed yield in blackgram under induced mutagenic treatment was also reported by Devi and Mullianathan, 2012.

Conclusion

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In M₂ generation, the correlation analysis results revealed that among ten quantitative characters, Number of seeds/pl was phenotypically and genotypically “significant correlated with seed yield. The heritability and Genetic advance estimations results revealed that the maximum heritability, Genetic advance mean value (%) recorded for the character number of pods per plant Similarly the GCV and PCV results revealed that maximum GCV and PCV recorded for the character seed yield The results concluded that most of the characters under study were recorded higher PCV than GCV which indicates little environmental influence on the characters in the Blackgram mutants in M₂ generation.

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