The estimation of genetic variability and genetic divergence of some advance lines of sesame based on morphological traits

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Abstract

In plant breeding research, naturally existing genetic variability has been successfully utilized to some extent for sustainable production of desirable crops and the diversity in plant genetic resources is most useful part to the breeders. The present experiment was conducted to estimate the genetic variation as well as divergence also of 29 sesame genotypes based on morphological traits. The high genotypic co-efficient of variation (GCV) and phenotypic co-efficient variation (PCV) observed for number of branches / plant and also for no. of capsules / plant. High heritability coupled with high genetic advance was estimated against seed yield / plant, number of branches / plant and capsules/ plant and HI indicating predominant role of additive genetic components for their expression and also indicating better scope for utilization of direct selection for those traits. The genotypes are grouped into 5 clusters based on 16 morphological traits i.e. Plant height, root length, vegetative dry weight, leaf area index, days to 50% flowering, no. of branches per plant, number of capsules per plant, capsule length, number seeds per capsule, 100 seed weight, seed yield per plant. The maximum inter-cluster distance noticed between cluster II and IV indicating their most diverse relationship. Genotypes of two clusters with wider genetic diversity and with desirable agronomic traits were selected as promising genotypes for hybridization programmes. Crosses can be made between genotypes of cluster II with genotypes of cluster IV to achieve promising recombinants.

Keywords

sesame, genetic variation, genetic advance, genotypic and phenotypic co-efficient of variation, heritability, genetic diversity

Introduction

The various plant genetic resources are priceless assets of human population that will enhance the food production. With the gradual increase of industrialization, the health awareness of consumers has been increased due to changes of pattern of food consumption of human population. The demand of high-quality oilseeds increases during the last decade due to rising income of growing population and expanding urbanization. From the point of nutritional value most of the people prefers sesame seed oil for their use.

In such situation the demand of sesame seeds increasing globally...
considering food security and we need large scale production of quality seeds to meet the consumers demand.

Sesame being an oldest oilseed crop belonging to Pedaliaceae cultivated in tropical and subtropical regions. It was cultivated and domesticated on the Indian subcontinent since Harappan and Anatolian eras over 4000 years ago (1). Sesame is very nutritious oilseeds containing protein (18-25%), carbohydrates (13.5%), oil (44-58%) (2). Sesame seed is a good source of calcium, phosphorous, zinc, manganese, copper and iron (3). Zinc plays as a co-factor of several enzymes of different biological activities such as cell division, cell development, immunity, gene expression (4). Fe is also vital component of enzymes that acts in energy metabolism and in the maintenance of immunity system (5). Due to the presence of various nutritious compounds the sesame seeds used as healthy foods (6). Most of the Africans prefer the sesame oil as a cooking oil as it contains polyunsaturated fatty acids such as oleic acid (43%), linoleic acid (35%), palmitic acid (11%) and stearic acids (7%) and it also contain sesamol, a unique anti-oxidant (7, 8). The sesame seeds used in the preparation of bread, confection and it may be used in the preparation of an oily paste so called tahini (9). Due to presence of antibacterial activity the sesame oil has been exploited in the pharmaceutical market (10). The oil used in the pharmaceutical product is more refined than cooking oil. Various research works have been done regarding the beneficial aspects of sesame (11). The demand of sesame growing continuously as the consumption of edible oil has been increased throughout the year in India. India was in 3rd position in sesame production in the world (12). Till now the production of sesame is not sufficient as India produces only 12.4% world sesame (13). So it is prime important to improve the productivity of sesame in India. The profound knowledge of genetic variability in crops is very essential for effective selection in breeding programme.

The study of high level of variability noticed in different sesame germplasm based on morphological traits (14, 15). The heritability along with genetic advance supports the careful selection of starting material for breeding programmes. Success through hybridization followed by selection primarily depends on the choice of appropriate parents with high genetic variability for different characters (16). Genetic divergence analysis is used to screen and identify promising genotypes for formulation of crossing programmes. Several countries like India, China, Central Asia, Abyssinia known as sesame diversity centers (17). The genetic diversity have been studied in sesame genotypes based on morphological traits (15, 18, 19). The diversity in plant genetic resources has been exploited to meet subsistence food requirement for growing populations. The reduction in agricultural land for cultivation creates the critical issues that we are facing today and we need continuous development of new plant varieties to fulfill current and future demand of edible oil. In this context, the aims of the present study was to identify suitable traits for effective selection and to analyze the genetic diversity among the sesame genotypes that will enhance the high productivity of sesame.

Materials and Methods

The field experiment was conducted at District Seed Farm, D-Block, Kalyani. The sesame seeds of 8 parental genotypes (Supplementary Table 1) and their 21 advance lines (Supplementary Table 2) were taken for experiment.

The seeds were sown in randomized block design (RBD) with three replications in 5 rows plot for each genotype with row to row spacing of 25 cm and plant to plant spacing of 12-15 cm. The irrigation and other agronomic practices were followed during crop growth period observations were recorded on the basis of five random competitive plants selected from genotypes in every replication for seed yield and its attributing characters. The observations were recorded on the following characters such as plant height (cm) at 30, 60 and 80 days after germination (DAG) as well as at harvest, Root length (cm) at 30 and 60 DAG, Days to 50% flowering, Total vegetative dry weight (g) at both 30 and 60 DAG, Leaf area index (LAI) on the average at 30 and 60 DAG. No. of branches per plant, No. of capsules per plant Length at the capsules (cm), Root length (cm) at 30 and 60 DAG, Days to 50% flowering, Total vegetative dry weight (g) at both 30 and 60 DAG, Leaf area index (LAI) on the average at 30 and 60 DAG, No. of branches per plant, No. of capsules per plant, Length at the capsules (cm), No. of seeds per capsule, 100 Seed weight (g), Seed yield/plant (g) and Harvest index (Supplementary Table 7).

**Determination of Vegetative dry weight**

Total vegetative dry weight is the sum of root, shoot and leaf dry weight. To determine the total vegetative dry weight, roots, shoots and leaves of five randomly selected plants for each genotype per replication were oven dried at 50°-60 °C for 16-18 hrs till a constant weight and the average was computed for each genotype. The average values for no. of branches per plant and no. of capsules per plant were computed on the basis of 5 plants of each genotype per replication. Length of the capsules (cm) was measured on randomly selected 15 capsules per plant i.e. 5 each from base, middle and top of the plant. Seeds from randomly selected 5 capsules from each plant were threshed and counted separately and weighed for number of seeds per capsule and 100 seed weight (g). Total number of capsules from each randomly selected plant were threshed separately and weighed for seed yield of individual plant.

Leaf area index (LAI) and harvest index (HI) were calculated as –

\[
LAI = \frac{\text{Total leaf area (LA)}}{\text{Ground area (GA)}}
\]

and

\[
\text{Average LAI} = \frac{L_{A1} + L_{A2}}{2} \times \frac{1}{\text{GA}}
\]

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Where, \( G.A = \pi r^2, r = D/2, \) D = Diameter in cm,
And LA\(_1\) and LA\(_2\) = Leaf area at two different stage
(i.e. 30 and 60 Days After Germination respectively).

\[
H.I = \frac{\text{Economic yield (g)}}{\text{Biological yield (g)}}
\]

**Estimates of variability, heritability (broad sense) and genetic advance (GA)**

The co-efficient of variation (C.V), being a unit less measurement, is a good basis of comparing the extent of variation amongst different characters and estimated as –

\[
\text{C.V.} \times 100 = \frac{\text{S.D.}}{\text{Grand mean}} \times 100
\]

Where, S.D. = Standard deviation = \( \sqrt{\text{MSE}} \) And X = Grand mean.

Genetic divergence was estimated by the use of Mahalanobis D\(^2\) statistics (20). The genotypic (GCV) and phenotypic co-efficient of variation (PCV) were calculated by the formula given bellow

\[
\text{GCV} \times 100 = \frac{\text{Genotypic standard deviation}}{\text{Grand mean}} \times 100
\]

\[
\text{PCV} \times 100 = \frac{\text{Phenotypic standard deviation}}{\text{Grand mean}} \times 100
\]

Where,

- Genotypic standard deviation \( (\sigma_g) = \sqrt{\text{Varialce of G - Error MS}} \)
- Phenotypic standard deviation \( (\sigma_p) = \sqrt{\sigma_g^2 + \sigma_e^2} \)
- \( \sigma_e^2 = \text{MSE} \)

The heritability (in broad sense) was estimated (21), as –

\[
\text{Heritability, } h^2(\%) = \frac{\sigma_g^2}{\sigma_p^2} \times 100
\]

Where, \( \sigma_g^2 \) and \( \sigma_p^2 \) were genotypic and phenotypic variance respectively.

The genetic advance (G.A) was calculated (22), as –

\[
\text{GA} = k \times h^2 \times \sigma_p
\]

Where, \( \sigma_p = \text{Phenotypic standard deviation}, k = \text{Selection differential a constant}, 2.06 \text{ and 1.70 selection intensity respectively.} \)

\[
\text{Genetic gain} = \frac{\text{GA}}{\text{Grand Mean}} \times 100
\]

Grouping of genotypes in to different clusters based on morphological traits was done followed by Torcher’s method (23).

**Results and Discussion**

The average performance of genotypes based on Plant height at 30 days after germination, 60 days after germination, 80 days after germination and at harvest shown in the Supplementary Table 3.

It is to be noted that for plant height at all stages of growth, the advanced lines showed superior performance for tallness over their parents indicating persistence for acquired inherited heterotic effect for this character over their parents after a long gap of generations. The coefficient of variation (CV\%) was however greater at early growth stage.

Maximum root length at 30 days after germination, as revealed from Supplementary Table 4, was recorded by line 20 in 1\(^{st}\) year, line 12 in 2\(^{nd}\) year and line 19 in pooled condition followed by either line 18, 19, 21 though they seemed to be at par statistically. At 60 DAG, line 20, 21 and 15 exhibited the same trend in respective years and pooled condition. The root length was consistently longer for lines 21 and 19 irrespective of growth stages and changed environment with regard to the total biomass production, lines 13, 14, 18 and 2 at 30 days after germination and line 12, 13, 2 and 6 at 60 days after germination consistently occupied the higher position.

With regard to leaf area index (LAI), Line 21, 18, 16, 17, 10 and 7 showed consistency in their higher performance over the years and pooled condition. It is to be noted that few parents viz. 4, 3 and 6 (B-67, B-9 and IDP-51 were also superior for LAI over the years and pooled condition as well.

Supplementary Table 5 shows average performance of the genotypes for days to 50% flowering, number branches per plant, number of capsules per plant and capsule length. It was noted that the parent B-14 and IDP-51 as the earliest genotypes as evidenced by days to 50% flowering. Line 18 and 21 were the most late flowering genotypes, both selected from the cross : Parent-7 (IET-2) x 1 (R-9), the late flowering genotypes.

Maximum branching habit was noted against line 9 followed by 20 in both the years and pooled conditions, the later being the product of the both less branching genotypes: R-9 x T-12 (1x5).

Significant differences for both number of capsules per plant and capsule length were reduced not only amongst all the genotypes, but also amongst the parents in both the years as well in pooled condition. The parents with dark seed coat colour produced higher number of capsules per plant in comparison to those with light coloured seeds. The highest number of capsules was recorded by line 5 followed by line 3 consistently over the years both of which were derived from the crosses involving poor performing genotypes as one of the parents.

Maximum elongated capsule (2.95 cm) was recorded against line 10 in both years indicating its high heterosis over both the parents for this character as its parents (5 and 1) produced smallest capsules (2.3 and 2.38 cm respectively). Persistence of high heterosis was also noted for all the
advanced lines over their superior parents for the capsule characters.

From Supplementary Table 6, it is revealed that maximum co-efficient of variation was recorded for seed yield per plant amongst the four parameters. Consistent higher number of seeds per capsules was recorded against 10 advance lines 5 and 6 (71.33) in 1st year against lines 12 and 18 (70.33) in 2nd year and against lines 6 (70.67) in pooled condition. The superior performing lines derived from the crosses involving at least one poor performing parent, showed heterosis over its superior parents.

When 100 seed weight is concerned, consistently maximum value was recorded against parent 2 (B-14) and line 15 followed by 2, 7, 12 and 17 lines out of which lines 15, 2 and 17 were derived from the crosses involving poor performing genotypes, ether 5 or 8, as one of the parents.

With regard to seed yield per plant, maximum value was noted against line 5 followed by line 3, 9, 12 and 6 over the years as well as in pooled condition. The highest yield (line-5) was selected from the segregants of the cross between T-12 and B-9. All these superior performing lines showed higher heterosis over its superior parents.

In case of harvest index, almost similar observations as seed yield per plant were recorded irrespective of the year of experiment i.e. change of environmental situation.

Characters are developed by the interaction between genotype and environment and partitioning the influence of each component is essential to elucidate the penetrance and expressivity of a character. Estimates of different genetic parameters, viz., genotypic and phenotypic co-efficient of variation (GCV and PCV), heritability (h²) and genetic advance (GA) (%) over mean were made to study the genetic control on individual parameter as well as the environmental influence on their expression. The heritability coupled with genetic advance is more effective to the plant breeders to take the decision of selection in breeding procedure.

The genetic parameters viz., GCV, PCV, h² and GA as well as GA (%) over means for different plant characters are presented in the Supplementary Table 7. Amongst plant height at different stages of growth, both GCV and PCV were high at 60 DAG in both the years as well as in pooled condition except PCV value in 1st year. A slightly higher value of PCV than that of GCV over the years for almost all the characters indicated negligible environmental influence on plant height at later stages of growth for their expression, while the reverse situation was noted at the earliest growth stage, i.e. plant height at 30 DAG. Heritability in broad sense (h²) was also high for all those characters irrespective of environmental situation except at 30 DAG. But genetic advance (%) was high only after 60 DAG in all environmental situations and it was moderate for all other characters. For root length, GCV as greater at 60 DAG than the same of 30 DAG in both the years as well in pooled condition, where as it was reverse in case of PCV indicating greater environmental influence at early stage created in root rhizosphere which is initially important for crop establishment. The negligible differences between magnitudes of PCV and GCV for root length at 60 DAG as well as greater differences between the same at 30 DAG also indicated favorable environmental influence at early growth stages supporting the earlier supposition.

High heritability along with high GA% value at 60 DAG or low heritability along with moderate GA (%) lead to draw the conclusion about the greater environmental influence on root length at early stage. Considering both h² and GA (%) values, it also indicated the additive genetic control on the expression of both plant height and root length at later stages.

So far as vegetative dry weight is concerned higher GCV and PCV values were recorded at 30 DAG than the same at 60 DAG. The same hold true also for genetic advance (%) and heritability, though the GA (%) was low and moderate respectively in spite of high h² values irrespective of all environmental situations, excepting medium h² value in 1st year at later growth stage. The values of all the genetic parameters for days to 50% flowering, as estimated over different years and pooled condition, were low in comparison to other plant characters, the values estimated for the same in pooled condition were GCV 0.82, PCV 2.06, GA (%) 0.34 and heritability 15.6%. Inspite of all, greater environmental influence was indicated for its expression as PCV values were higher than that of GCV consistently over the years as well as low values of both h² and GA (%) as well.

The low value of h² coupled with low GA (%) value, also indicated non-additive genetic control for its expression. A slight difference in magnitude of PCV and GCV were noted for LAI, whereas the value were same for HI irrespective of year of experiment indicating little or no environmental interaction with the genetic make up for expression of those two characters respectively. Heritability values were high in both the cases for all situations moderate h² for LAI in 2nd year, but high GA (%) value indicated the nature of genetic control by additive components for HI and either dominance/epistatic component for LAI as GA (%) value was low, the high h² value for LAI may be due to its greater sensitivity to environmental influence.

Though the values of both GCV and PCV were high irrespective of year of experiment for both numbers of branches per plant and number of capsules per plant and little higher magnitude of PCV than that of GCV indicates less environmental influence for their expression. At the same time, consistently higher values of both h² and GA (%) indicated proper expression of those characters through additive genetic background. High co-efficient of variation for number of branches per plant was also reported (25, 26).

In case of capsule length and number of seeds/capsule, the positive difference in magnitude of GCV and PCV were consistently little as in no of branches and capsules/plant with similar indication of environmental influence for their phenotypic expression. But for both the characters, heritability was high and GA (%) was moderate. It indicated the non-additive control for both the characters and high h² value may be due to environmental interaction.

Consistently high GCV and PCV values were estimated
for seed yield/plant over the years and pooled condition than the same for 100 seed weights. For both the characters, the PCV value was close to that of GCV indicating minimum environmental influence for their phenotypic expression. It was observed that high genotypic coefficient of variation and high phenotypic coefficient of variation by 1000 seed weight (27). Reports are on both GCV and PCV are high for capsules per plant followed by seed yield per plant (28). Moderate values of PCV were reported for plant height, number of seeds per capsule and 1000 seed weight (29, 30). In the present investigations heritability was very high for both the characters but high and moderate GA (%) were estimated against seed yield/plant and 100 seed wt. respectively indicating the reverse genetic control as well as their interaction with environment for their expression (Supplementary Table 7).

High heritability coupled with high genetic advance as percent over mean noticed for seed yield per plant that was reported (31-34).

High \( h^2 \) estimates coupled with moderate to high genetic advance (%) over mean for plant height and root length at 60 DAG number of branches and capsules per plant, 100 seed weight, seed yield/plant and harvest index indicated greater scope for direct selection for those traits which was also supported by earlier observations (35, 36). High \( h^2 \) estimates with low GA (%) for LAI, plant height at harvest, capsule length indicated that those characters were governed by non-additive genes and exhibition of high \( h^2 \) was due to favorable environmental influence rather than genotype. Direct selection may not be effective for improvement of such traits and heterosis breeding may be advocated in such a situation.

The genetic divergence analysis is generally used to measure the genetic distance amongst the genotypes. The genetic divergence pattern among the genotypes estimated using Mahalanobis \( D^2 \) statistics based on 16 plants characters without harvest index (20). Supplementary Table 8 represents clustering pattern of all the genotypes. All the genotypes were grouped into 5 clusters by applying the clustering technique of which cluster I included maximum number of genotypes (19) comprising almost all the advance lines excepting 5 and 14, cluster IV and V were constituted with single genotypes each advance line no. 5 and 14 respectively and remaining two clusters viz. II and III, were constituted with four parental genotypes each. The specificity of the clustering nature was noted as the adherence of parental genotypes with separate clusters and away from the advance lines. Of these two clusters, clusters II included parents 5, 8, 1, 7 (T-12, HT-1, R-9 and IET-2), all with white/ light coloured seeds and cluster III included the remaining four parents (B-67, B-14, B-9, IDP-51), all genotypes were having dark coloured seeds (Supplementary Table 8). The intra- and inter- cluster distances are presented in Supplementary Table 9. The table value shows the inter cluster distances are larger than intra cluster distance that represents the higher genetic variability among the genotypes of different groups.

The maximum intra-cluster distance 4.6009 was found against cluster – II which may be due to the inclusion of genotypes of mostly divergent geographic origin.

The same was found to be nil against cluster IV and V due to their constitution with single genotype each. The maximum inter-cluster distance (8.9749) was noted between cluster-II and IV indicating their most diverse relationship and the minimum distance was noted between cluster II and III (5.4323) indicating the close relationship between the two clusters with parental genotypes as their sole constitutes. It was preceded by the same between II and V which may be due to the single constitute of cluster V (line 14) which had been selected as a segregant of the cross between parents 1 and 8 (R-9 and HT-1), both were constituents of cluster II. The greater the distance between two clusters, the greater is the divergence and vice-versa. Considering all those inter-cluster distances, cluster III was found to be closely related to all the other 4 clusters followed by cluster V.

The average performance of the different clusters for all the 16 plant characters are presented in Supplementary Table 10. Cluster IV recorded maximum values for most of the characters (9) including seed yield, 100 – seed weight, number of seeds/ capsule, and number of branches/plant and number of capsules per plant. While cluster V exhibited maximum values for plant height at 80 DAG, root length at 30 DAG and vegetative dry weight both at 30 and 60 days after germination (DAG), plant height at 30 days after germination (DAG) and at harvest as well as root length at 60 DAG were found to be maximum against cluster I.

Therefore, the greater the genetic distance between two clusters, the wider the genetic diversity among the parents to be included in hybridization programme. Parents combining high yield potential with wide genetic diversity are likely to yield superior segregants within a reasonable period. Successful breeding can also be made between the members of different clusters rather than between the members within the same cluster. In present case, inter-se mating of advance line 5 with either T-12, HT-1, R-9 and IET-2 or advance line 14 may produce promising segregants, through genetic reconstitution with higher yield potential and its attributes.

**Conclusion**

The present study revealed that the sesame genotypes show sufficient level of genetic variability for seed yield and yield attributing traits. Here high \( h^2 \) coupled with high GA (%) was estimated against seed yield/plant, number of branches per plant and number of capsules per plant, and HI indicating predominant role of additive genetic component for their expression and also indicating better scope for utilization of direct selection on those traits as there was no significant environmental influence on its expression. Among the 5 clusters of present experiment cluster 1 consists of maximum number of genotypes. The maximum inter-cluster distance was noted between cluster II and cluster IV indicating high degree of genetic diversity and the genotypes of these two clusters may be utilized for successful breeding programme to obtain high yielding recombinants
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Authors contributions
BR conducted the whole experiment, designed the experiment and collected the data. AKP supported in the experimental works, writing the manuscript and performed statistical analysis. AKB hypothesized the paper concept.

Compliance with ethical standards
Conflict of interest: The authors have no conflict of interest.

Ethical issues: None.

Supplementary data
Table 1. Plant type and seed colour of parental genotypes of sesame
Table 2. The parentage of advance lines
Table 3. Average performance of the genotypes for plant height at different growth stages
Table 4. Average performance of the genotypes for root length and vegetative dry weight at 30 & 60 DAG and Leaf area index
Table 5. Average performance of the genotypes for days to 50% flowering, no. of branches/plant, no. of capsules/plant, capsule length
Table 6. Average performance of the genotypes for number of seeds/capsule, 100 seed weight, seed yield/plant and harvest index
Table 7. Genetic parameters for different plant characters
Table 8. Clustering pattern, size and constituents involving 29 genotypes for plant characters
Table 9. Average intra- (diagonal) and inter-cluster distances (D values) on the basis of 16 plant characters
Table 10. Cluster means for sixteen plant characters

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