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DECIPHERING GENETIC VARIABILITY, CHARACTER ASSOCIATIONS AND DIVERSITY ANALYSIS IN SESAME (*SESAMUM INDICUM* L.) GERMPLASMS

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ABSTRACT

This study evaluated thirty-seven sesame germplasms using genetic variability parameters, correlation, path coefficient analysis, and genetic diversity analysis based on thirteen morphological and biochemical traits. The experiment followed a randomized block design with three replications. The analysis of variance revealed substantial variability among the germplasms under study. Traits such as primary branches per plant, capsules per plant, number of seeds per capsule, 1000 seed weight, seed yield per plant, Fe content, Zn content and crude protein content exhibited moderate to high PCV and GCV, high heritability and moderate to high genetic advance as per cent mean. Seed yield per plant was significantly and positively correlated with primary branches per plant, capsules per plant, days to 50% flowering and Zn content. Capsules per plant showed the strongest positive direct effect on seed yield per plant, followed by primary branches per plant, days to 50% flowering, number of seeds per capsule, crude protein content, and plant height. Therefore, selecting genotypes with these traits could significantly improve sesame yield. Cluster analysis categorized the genotypes into six clusters, highlighting high heterogeneity among the genotypes. Hybridization between genotypes from clusters I and III, having the highest inter-cluster distance, is suggested to maximize heterotic hybrids and generate a broader range of variation in the segregating population. Through the use of genetic variability parameters and the identification of key traits and genotype clusters via diversity analysis, breeders can make strategic selections to develop sesame varieties with enhanced productivity and quality.

***Keywords*:** Sesame, genetic variability, correlation and path analysis, diversity, cluster analysis

Introduction

Sesame (*Sesamum indicum* L.), a vital oilseed crop in India, belongs to the Pedaliaceae family. Known by various names such as Gingelly, Til, and Benniseed, it is one of the oldest oilseed crops, believed to have originated in Africa (Bedigian, 2003). Sesame is a self-pollinated diploid species (2n = 26) but exhibits some cross-pollination depending on environmental factors. It ranks as an important oilseed crop in India after groundnut, rapeseed, and mustard. Often called the "Queen of Oilseeds," sesame seeds contain 40-54% oil, rich in antioxidants and oleic and linoleic acids (Abate and Mekbib, 2015). Despite its nutritional and economic value, sesame has low yields due to factors such as seed shattering, low harvest index, and sensitivity to diseases (Yol and Uzun, 2012). Sesame thrives in temperatures of 25-37°C and well-drained, medium-textured soils with pH 5-8. While drought-tolerant, optimal growth requires adequate soil moisture. High temperatures (≥40°C) during flowering reduce fertilization and capsule formation. It is less suitable for heavy clay, salty, or waterlogged soils (Terefe *et al*., 2012). Nutritionally rich, sesame is a source of calcium, potassium, vitamins, tryptophan, and methionine. Its oil has diverse uses in food, pharmaceuticals, cosmetics, and soaps. The by-product, sesame meal, serves as a protein-rich cattle feed and fertilizer. India produces

sesame seeds in various colours, with white used in confectionery, brown for oil extraction, and black for seasoning (Ashri, 1998). Sesame's versatility and nutritional profile underscore its industrial and agricultural importance, even as its low yield challenges remain a barrier to expanded cultivation. It is a key oilseed crop in tropical and subtropical regions and majorly produced in India, China, and Africa. In India, sesame is grown on 16.226 lakh hectares land along with production of 6.575 lakh tonnes and a productivity of 405 kg/ha. Gujarat contributes 1.657 lakh hectares area in sesame cultivation along with a production of 1.075 lakh tonnes and a productivity of 649 kg/ha, needing improvement to meet demands (Anon., 2020).

To enhance sesame cultivation and profitability, leveraging its vast genetic diversity, adaptability, and varied seed oil characteristics is essential (Hegde, 2012). Despite its potential, sesame remains an orphan crop, receiving limited attention from international agricultural research centres. Genetic variability in economic traits is crucial for effective breeding programs, enabling breeders to develop high-yielding, quality cultivars. Heritability indicates a trait's transmission potential across generations, aiding in selecting traits for yield improvement. Combined with genetic advance, heritability better predicts genetic gain under selection. Sesame yield, a complex polygenic trait, necessitates understanding character associations and their direct and indirect effects on seed yield. Correlation studies help breeders predict how enhancing one trait can simultaneously improve others. Path coefficient analysis further distinguishes direct effects from indirect ones, facilitating a deeper understanding of cause-effect relationships. Genetic diversity, a vital factor, quantifies divergence within experimental samples. This allows breeders to identify and select genetically divergent genotypes for hybridization programs, fostering novel variability. By focusing on these genetic and trait-based approaches, breeders can unlock sesame's full potential, improving its yield and quality while addressing the crop's historical neglect in agricultural research. In context of the crop's current status and future potential, this study aimed to evaluate sesame germplasms based on genetic variability, correlation, path coefficient analysis, and diversity studies for seed yield and its component traits.

Materials and Methods

The present experiment was carried out at College Farm, N. M. College of Agriculture, Navsari Agricultural University, Navsari during summer, 2022. Thirty-seven sesame genotypes including thirty-three genotypes and four check varieties (GT-10, TKG-525, JLS-708, GJT-5) were evaluated in Randomized Block Design (RBD) with three replications. The seeds were obtained from the Niger Research Station, Navsari Agricultural University, Vanarasi, Gujarat. Names of the genotypes evaluated under this experiment are mentioned in the Table 1. All the suggested standard agronomic package of practices along with necessary plant protection measures were followed timely during crop growth period. The observations were recorded on five randomly selected plants from each genotype in each replication except for days to 50% flowering and days to maturity. Data for days to 50% flowering and days to maturity were recorded on a population basis.

Table 1: Genotypes of sesame evaluated under this experiment

Sr. No.	Genotype	Sr. No.	Genotype
1	GT-10 ©	2	RT-383
3	DS-18-46	4	AT-392
5	RT-384	6	TKG-525 ©
7	DS-51	8	OSM-17-22
9	AT-386	10	DS-45
11	DS-53	12	JCS-2420
13	JLS-907	14	JLS-408-3
15	Nana Rajkot-1	16	DS-18-21
17	AT-371	18	JLS-120
19	JLS-2611	20	AT-410
21	AT-382	22	IC-199433
23	JLS-706	24	AT-3661
25	NIC-8476	26	AT-369
27	SVT-333	28	IC-204680
29	AT-411	30	AT-378
31	AT-482	32	AT-468
33	JLS-708 ©	34	LT-17-28
35	LT-17-10	36	GJT-5 ©
37	DS-17-15		

© = Check variety

The analyses for biochemical parameters *i.e.* oil content (%), crude protein content (%), Fe content (ppm) and Zn content (ppm) were carried out at Central Instrumentation Laboratory, Department of Agricultural Chemistry and Soil Science, N.M. College of Agriculture, Navsari Agricultural University, Navsari. Oil content of the sesame seed samples were determined by soxhlet method (Sadasivam and Manickam, 1992) with the soxhlet apparatus (Pelican equipments). Oil from a known quantity of the seed was extracted with petroleum ether. It was then distilled off completely, dried, weighed and the percentage of oil was calculated with the following formula:

$$\text{Total oil content (\% dry weight)} = \frac{(Y - W) \times 100}{W}$$

Where,

W = Weight of the sample (g)

X = Weight of the flask (g)

Y = Weight of the flask with ether extract (g)

Crude protein analysis was done by micro-kjeldahl method as described in Sadasivam and Manickam (1992) by using digestion and distillation apparatus. The nitrogen in protein or any other organic material was converted to ammonium sulphate by sulphuric acid during digestion. This salt, on steam distillation, liberates ammonia which was collected in boric acid solution and titrated against standard acid. Since 1 ml of 0.1 N acid is equivalent to 1.401 mg nitrogen, calculation was made to arrive at the nitrogen content of the sample with the following formula:

$$\text{N (g/kg)} = \frac{(\text{ml } H_2SO_4 \text{ in sample}) - (\text{ml } H_2SO_4 \text{ in blank}) \times \text{Normality of } H_2SO_4 \times 14.01}{\text{Weight of sample (g)}}$$

Crude protein content (%) = N (g/kg) x 6.25

Where,

6.25 = Protein conversion factor

The estimation of Fe and Zn content (ppm) from sesame sample was done by using Atomic Absorption Spectrophotometer (AAS Plus, Motras Scientific) instrument which is a fully automated PC controlled atomic absorption spectrophotometer with flame and hydride system. Wavelengths used for Fe and Zn elements were 248.3 and 213.9 nm, respectively.

The analysis of variance (ANOVA) for randomized block design (RBD) was done for each character with the method suggested by Panse and Sukhatme (1978) by taking the average values on individual characters. Phenotypic and genotypic coefficient of variation (PCV and GCV) was calculated from the ANOVA table (Burton and Devane, 1953). The heritability in broad sense (h^2_{bs}) and genetic advance (GA) was estimated according to the method given by Allard (1960). The expected genetic advance expressed as percent of mean (GAM) was calculated by the method suggested by Johnson *et al.* (1955). Genotypic (r_g) and phenotypic (r_p) correlation coefficients were calculated by adopting the method explained by Miller *et al.* (1958). Using seed yield per plant as the dependent character and rest of the variables as the independent characters, path coefficient analysis was performed at genotypic and phenotypic levels in accordance with the method recommended by Dewey and Lu (1959). The Mahalanobis D^2 statistic method was adopted to assess the genetic divergence among the sesame genotypes (Rao, 1952). In addition, the agglomerative hierarchical clustering (AHC) was done based on Ward's method. All the statistical analysis carried out under this experiment were done using the R statistical software.

Results and Discussion

Genetic variability is a vital requirement for the success of every crop improvement initiative. ANOVA for different characters studied is presented in Table 2. Analysis showed significant mean sum of square values for all the traits among all the genotypes. It also indicated that sufficient variability is present among the genotypes for all the traits covered under this study. The relative mean performances are depicted graphically in Figure 1-4. The genetic variability parameters were illustrated in Table S1 and Figure 5. The extent of variability as measured by PCV and GCV, gives information regarding the relative amount of variation in the genotypes. Coefficients of variation studies indicated that the estimates of PCV were slightly higher than the corresponding GCV estimates indicating that the characters were less influenced by the environment. Therefore, selection on the basis of phenotype alone can be effective for the improvement of these traits. In the present study, genotypes showed higher values of GCV and PCV for primary branches per plant, capsules per plant, number of seeds per capsule, Fe content and Zn content indicated the greater scope of improving these characters by applying the selection in an appropriate direction and also showed greater variability among the genotypes for these traits. Similar results were also observed by Begum *et al.* (2017), Pandey *et al.* (2017), Agrawal *et al*. (2018), Alake (2018), Patil and Lokesha (2018), Umamaheswari *et al.* (2019), Kumari *et al.* (2020b), Mohanty *et al.* (2020), Pavani *et al.* (2020), Saravanan *et al.* (2020), Sirisha *et al.* (2020), Umate (2020), Ahmed *et al.* (2022), Patel *et al.* (2022), Sahu *et al.* (2022), Singh *et al*. (2022), Sundari *et al.* (2022), Thouseem *et al.* (2022b) and Patel *et al.* (2023). Moderate GCV and PCV values were observed for the traits *viz.,*1000 seed weight, seed yield per plant and crude protein content. This indicated that the extent of response of these traits for selection would be less than previous characters. Similar results were also recorded for 1000 seed weight by Begum *et al.* (2017), Patil and Lokesha (2018), Umamaheswari *et al.* (2019), Ranjithkumar *et al*. (2022) and Thouseem *et al.* (2022b); for seed yield per plant by Ahmed *et al.* (2022) and Sundari *et al.* (2022); for crude protein

content by Kumari *et al.* (2020b). Low GCV and PCV values were recorded for the characters *viz.,* days to 50% flowering, days to maturity, capsule length, oil content except plant height showing moderate PCV values indicating a narrow range of variability for these traits and restricting the scope of selection for these traits. Similar results were observed by Manjeet *et al.* (2020), Mohanty *et al.* (2020), Patidar *et al*. (2020), Durge *et al.* (2022), Patel *et al.* (2022) for days to 50% flowering; Kadvani *et al.* (2020), Pavani *et al.* (2020), Sirisha *et al.* (2020) for days to maturity; Abate *et al.* (2021), Srikanth and Ghodke (2022) for capsule length; Kadvani *et al.* (2020), Ranjithkumar *et al*. (2022) for oil content and Patel *et al.* (2023) for plant height.

Heritability is an important indicator of the extent to which traits are transferred from parents to their progeny. However, genetic advance can help to predict the extent of improvement that can be achieved for the traits. The high genetic advance along with high heritability would suggest better conditions for making an effective selection. Table S1 and Figure 5 shows heritability in the broad sense and genetic advance as percent of mean for all characters and their performance was adjudged using the categories provided by Johnson *et al*. (1955). All of the traits in this experiment showed moderate to high heritability. High heritability values were observed for days to 50% flowering, primary branches per plant, capsules per plant, capsule length, number of seeds per capsule, 1000 seed weight, seed yield per plant, oil content, crude protein content, Fe content and Zn content indicating that there is very less environmental influence on these traits. Whereas, moderate heritability was observed for days to maturity and plant height. The high heritability with high genetic advance as per cent of mean was observed for primary branches per plant, capsules per plant, number of seeds per capsule, 1000 seed weight, crude protein content, Fe content and Zn content indicating the role of additive gene effects and less effect of environmental factors on the expression of the traits. Thus, the improvement of these traits could be achieved through direct phenotypic selection. Similar results were also observed by Agrawal *et al*. (2018), Umamaheswari *et al.* (2019), Sahu *et al*. (2022) for primary branches per plant; Mohanty *et al.* (2020), Patel *et al.* (2022), Sundari *et al.* (2022) for capsules per plant; Sirisha *et al.* (2020), Singh *et al*. (2022) for number of seeds per capsule; Srikanth and Ghodke (2022), Thouseem *et al.* (2022b) for 1000 seed weight; Kumari *et al.* (2020b) for crude protein content and Pandey *et al.* (2017) for Fe content, Zn content. High heritability coupled with moderate genetic advance as per cent of mean was observed for capsule length, seed yield per plant and oil content, suggesting the predominance of non-additive gene action in governing these traits. Thus, there is limited scope of improvement of these traits by means of simple phenotypic selection. Similar results were also observed by Patil and Lokesha (2018), Kadvani *et al.* (2020), Sasipriya *et al.* (2022) for capsule length; Begum *et al.* (2017) for seed yield per plant and Patidar *et al*. (2020), Kumar *et al*. (2022) for oil content. High heritability with low genetic advance as per cent of mean was observed for days to 50% flowering, indicative of non-additive gene action. Moderate heritability with moderate genetic advance as per cent of mean was recorded for plant height, similarly, moderate heritability with low genetic advance as per cent of mean was observed for days to maturity indicating the non-additive gene effect for these traits. Therefore, possibility of improvement through selections for these traits is negligible. Similar results were also recorded by Manjeet *et al.* (2020), Saravanan *et al.* (2020), Abate *et al.* (2021), Kant *et al.* (2021), Vamshi *et al.* (2021) and Ranjithkumar *et al*. (2022).

Table 2: Analysis of variance for seed yield per plant and yield attributing traits in sesame

Sl. No.	Characters	Mean sum of square			C.V. (%)
		Replication	Genotype	Error	
1	Days to 50% flowering	1.414	9.319**	1.377	3.05
2	Days to maturity	3.144	8.863**	2.005	1.60
3	Plant height (cm)	90.070	363.870**	77.000	8.54
4	Primary branches per plant	0.021	0.623**	0.027	11.43
5	Capsules per plant	22.503	108.949**	7.629	10.20
6	Capsule length (cm)	0.026	0.145**	0.018	5.18
7	Number of seeds per capsule	12.220	402.750**	8.870	6.51
8	1000 seed weight (g)	0.024	0.670**	0.007	3.20
9	Oil content (%)	0.372	46.108**	0.384	1.42
10	Crude protein content (%)	0.030	22.191**	0.087	1.45

11	Fe content (ppm)	26.700*	4591.200**	3.300	1.22
12	Zn content (ppm)	0.610	1713.510**	1.110	1.59
13	Seed yield per plant (g)	0.003	0.542**	0.067	6.74

** Significant at P = 0.01

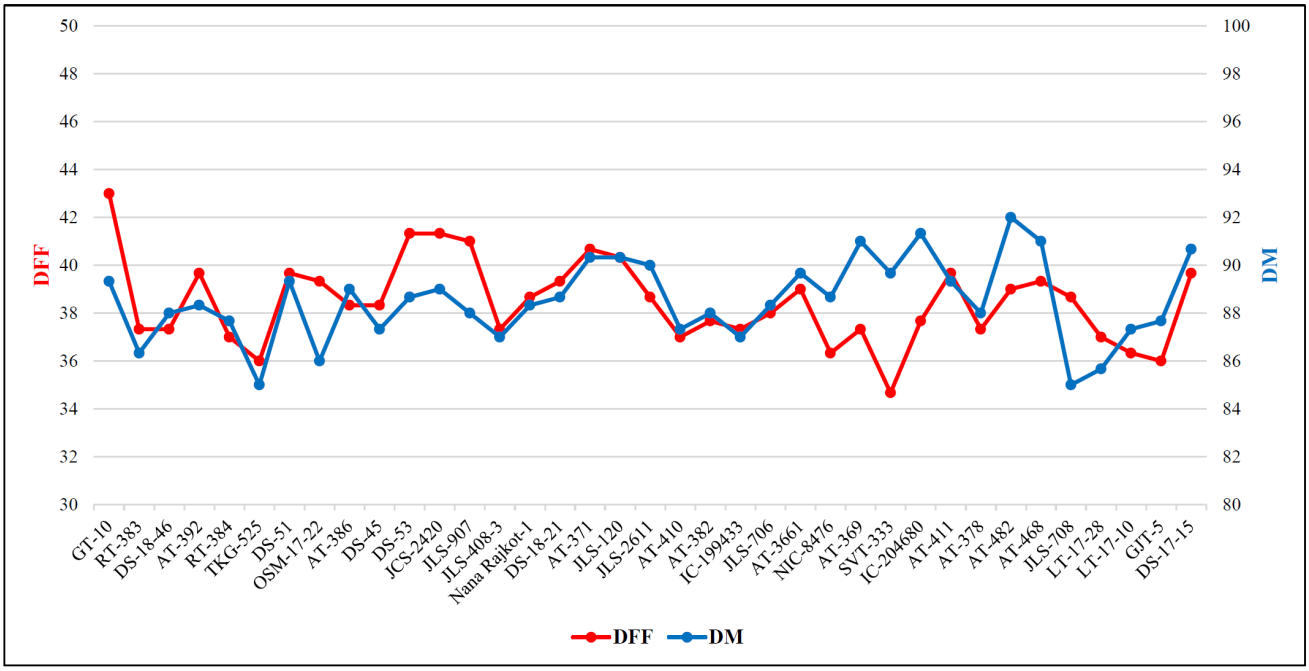

Fig. 1 : Comparative mean performance of thirty-seven genotypes for days to 50% flowering and days to maturity in sesame

DFF=Days to 50% flowering DM=Days to maturity

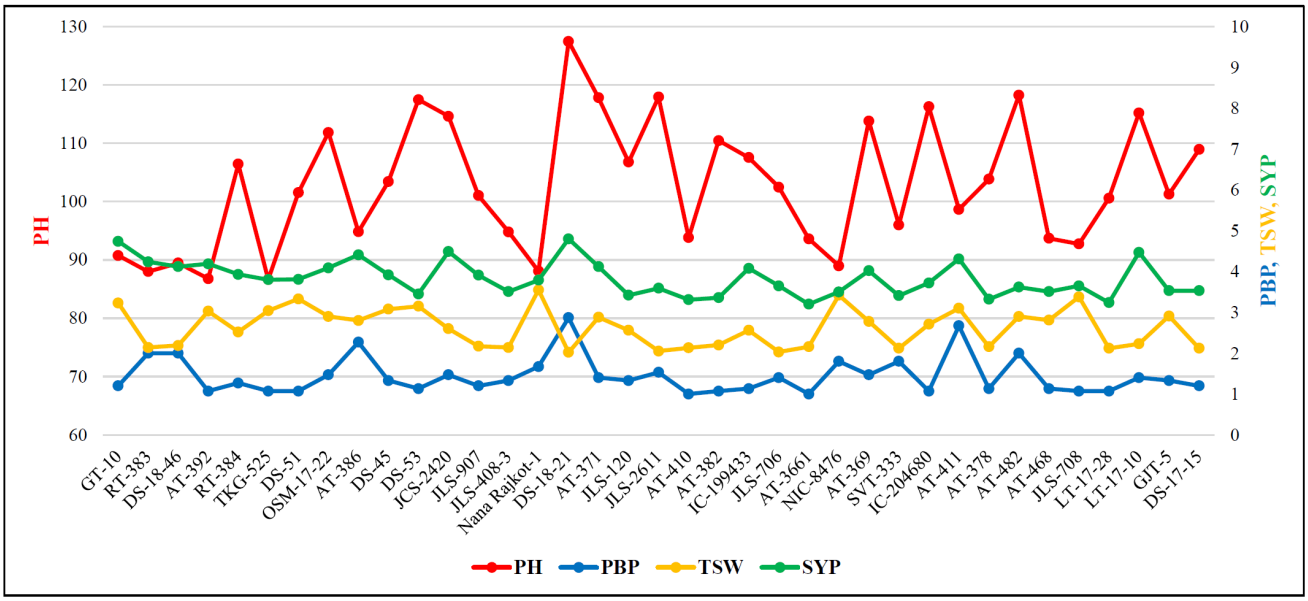

Fig. 2: Comparative mean performance of thirty-seven genotypes for plant height, primary branches per plant, 1000 seed weight and seed yield per plant in sesame

PH=Plant height (cm) PBP=Primary branches per plant

TSW=1000 seed weight (g) SYP=Seed yield per plant (g)

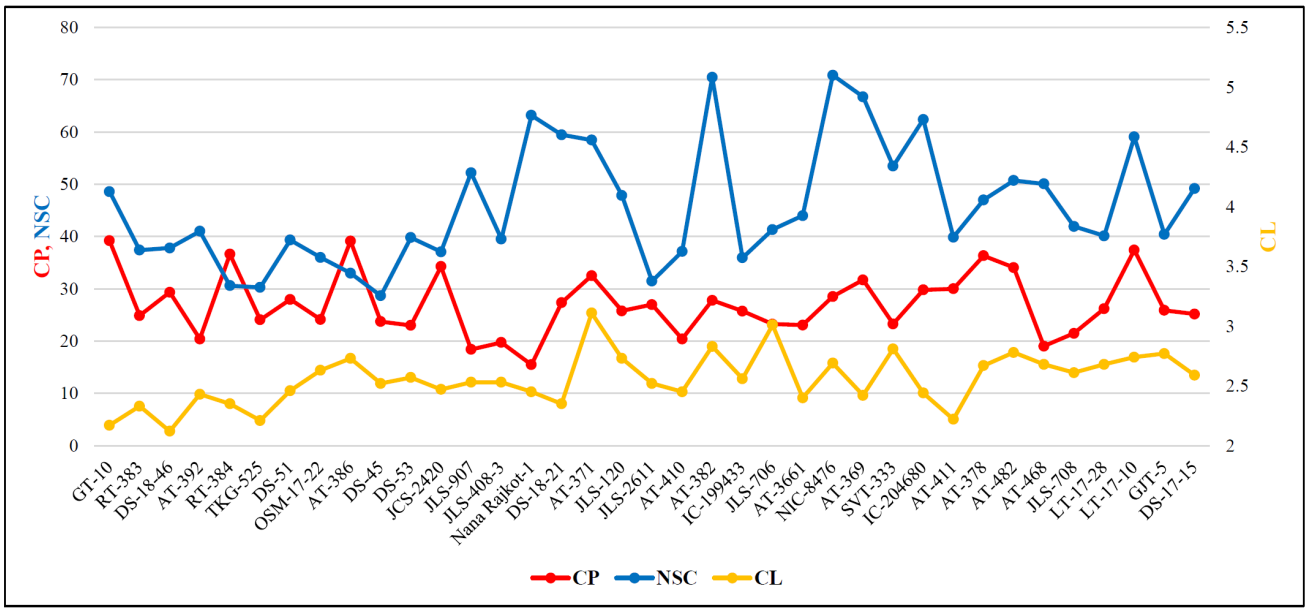

Fig. 3: Comparative mean performance of thirty-seven genotypes for capsules per plant, number of seeds per capsule and capsule length in sesame

CP=Capsules per plant NSC=Number of seeds per capsule
CL=Capsule length (cm)

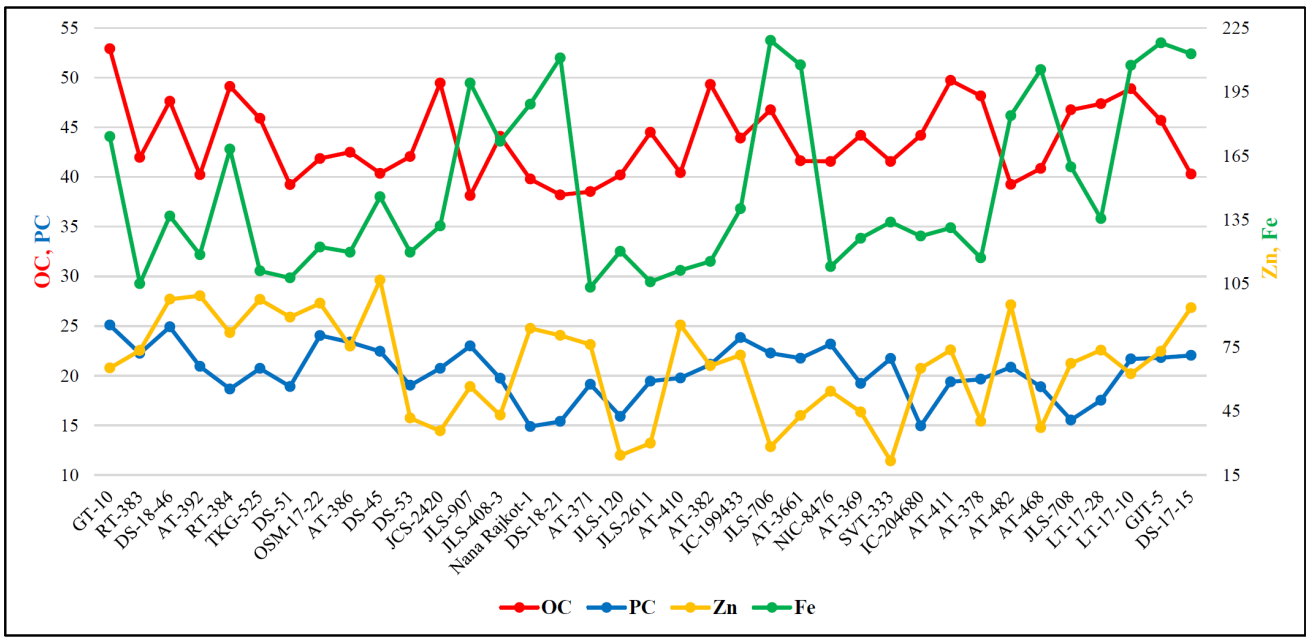

Fig. 4: Comparative mean performance of thirty-seven genotypes for oil content, crude protein content, Zn content and Fe content in sesame

OC=Oil content (%) PC=Crude protein content (%)
Zn=Zn content (ppm) Fe=Fe content (ppm)

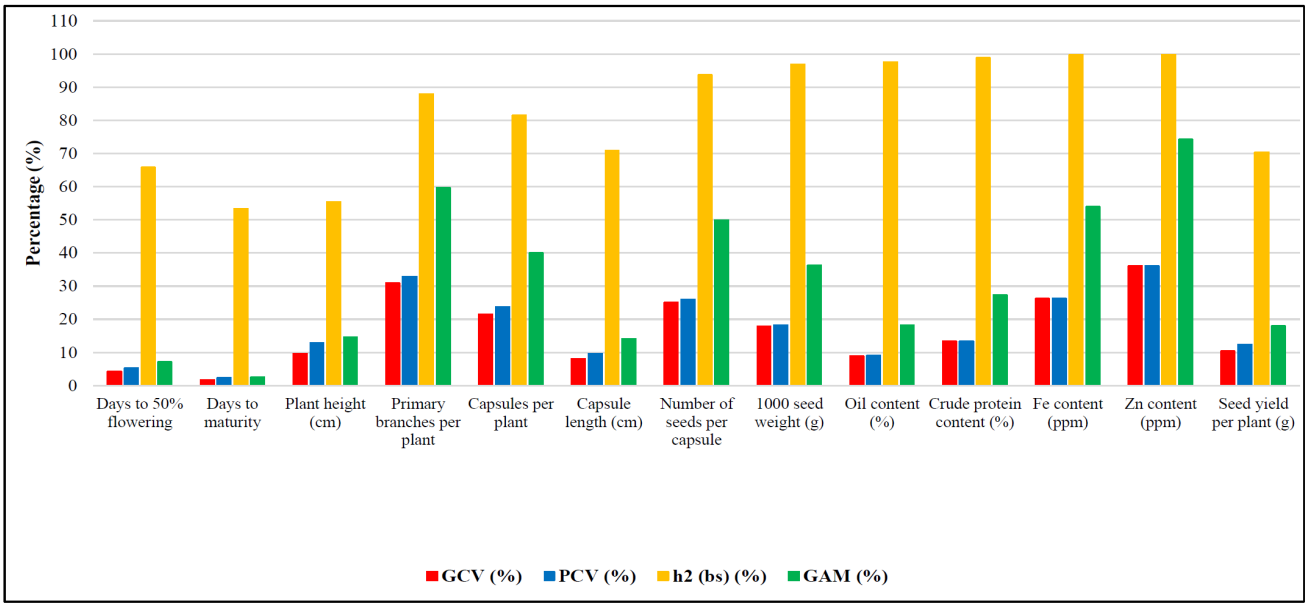

Fig. 5: GCV, PCV, $h^2_{(bs)}$ and GAM for thirteen characters in thirty-seven genotypes of sesame

GCV (%) = Genotypic coefficient of variation PCV (%) = Phenotypic coefficient of variation

GAM = GA as per cent of mean (%) h^2_{bs} (%) = Heritability (broad sense)

The correlation coefficient measures the relationship between variables, aiding breeders in selecting strategies for trait improvement. Positive correlations allow simultaneous enhancement of desirable traits, while negative correlations hinder it. Seed yield per plant often correlates with its components, making multi-trait selection crucial. Genotypic correlation, unaffected by environmental factors, provides a true and stable association. It is valuable for identifying correlated traits and isolating superior genotypes, enabling breeders to improve key traits effectively. In the present study, both the phenotypic and genotypic correlation coefficients were estimated among thirteen characters of total thirty-seven genotypes to find out the association of the characters with seed yield per plant at genotypic and phenotypic levels (Table 3). At phenotypic level, seed yield per plant recorded a positive and highly significant correlation with days to 50% flowering, primary branches per plant, capsules per plant and Zn content, while it had a positive and significant correlation with crude protein content. It had a positive and non-significant correlation with days to maturity, plant height, 1000 seed weight, oil content and Fe content. Seed yield per plant recorded a negative and highly significant correlation with capsule length, while it has showed a negative and non-significant correlation with number of seeds per capsule. At genotypic level, seed yield per plant recorded a positive and highly significant correlation with primary branches per plant, capsules per plant and a positive and significant correlation with days to 50% flowering, Zn content, while it had a positive and non-significant correlation with plant height, 1000 seed weight, oil content, crude protein content and Fe content. Negative and highly significant correlation was observed with capsule length, while a negative and non-significant correlation was showed with days to maturity and number of seeds per capsule. A similar trend for correlation of seed yield per plant were observed by Patil and Lokesha (2018) for primary branches per plant; Ahmed *et al.* (2022), Thouseem *et al.* (2022a) for capsules per plant; Patel *et al.* (2022) for days to 50% flowering; Iqbal *et al.* (2017) for Zn content; Singh *et al.* (2022) for plant height and days to maturity; Kalaiyarasi *et al.* (2019), Umamaheswari *et al.* (2019) for 1000 seed weight; Kumari *et al.* (2020a) for number of seeds per capsule; Agrawal *et al.* (2017) for oil content; Patel *et al.* (2023) for crude protein content; Pandey *et al.* (2017) for Zn content and Ramprasad *et al.* (2019) for capsule length.

The correlation coefficient may not give a truly comprehensive picture in a complex situation which creates difficulty in deciding the breeding procedure to be adapted for simultaneous selection for crop improvement. Correlation gives only the relation between the two variables whereas path coefficient analysis facilitates the partitioning of correlation coefficients into direct and indirect effects of various yield component characters on seed yield. In the present experiment, the genotypic correlation coefficient of various traits with seed yield per plant under study was subjected to genotypic path coefficient

analysis to estimate the direct effect of component traits on dependent variable seed yield per plant (Table S2 and Figure 6). The overall genotypic path analysis revealed that the highest positive direct effects on seed yield per plant were exhibited by capsules per plant followed by primary branches per plant, days to 50% flowering, number of seeds per capsule, crude protein content and plant height. Therefore, selection for such traits in the genotypes would be useful to bring about the improvement in sesame. These results are in agreement with Ramprasad *et al.* (2019), Disowja *et al.* (2020), Sasipriya *et al.* (2022), Thouseem *et al.* (2022a) for capsules per plant; Kumari *et al*. (2020a) for primary branches per plant; Patel *et al.* (2022) for days to 50% flowering; Kalaiyarasi *et al*. (2019), Kumar *et al*. (2022) for number of seeds per capsule; Agrawal *et al.* (2017) for crude protein content and Kant *et al.* (2021) for plant height. The highest negative direct effects on seed yield per plant were recorded by days to maturity followed by capsule length, oil content and Zn content indicating less significance of these traits in selection for higher seed yield, except days to maturity. As the trait days to maturity exhibited high negative direct effects on seed yield, direct selection for higher yield could be resulted into indirect selection for early maturing genotype and which is desirable in improvement of sesame. These results are in agreement with Kumari *et al.* (2020a) for days to maturity and oil content; Patel *et al.* (2022) for capsule length. Path coefficient analysis for seed yield revealed low residual effect of 0.174, which indicated that most of the traits contributing to seed yield per plant were included in this study to cover most of the available variation in case of sesame.

Table 3: Estimates of genotypic (r_g) and phenotypic (r_p) correlation coefficients among the characters in sesame

Characters	DFF	DM	PH	PBP	CP	CL	NSC	TSW	OC	PC	Fe	Zn	SYP
DFF	**1.000**	0.332*	0.329*	-0.025	0.108	-0.111	-0.037	0.290	-0.119	-0.042	0.070	-0.026	0.401*
DM	0.337**	**1.000**	0.439**	0.193	0.268	0.230	0.484**	0.025	-0.300	-0.149	0.122	-0.334*	-0.064
PH	0.082	0.304**	**1.000**	0.122	0.319	0.376*	0.246	-0.254	-0.085	-0.290	0.060	-0.148	0.111
PBP	-0.034	0.141	0.126	**1.000**	0.240	-0.164	0.086	-0.031	-0.093	0.022	0.043	0.106	0.525**
CP	-0.008	0.227*	0.378**	0.246**	**1.000**	-0.006	0.055	0.004	0.521**	0.213	-0.162	0.020	0.450**
CL	-0.144	0.078	0.238*	-0.120	0.023	**1.000**	0.318	-0.124	-0.240	-0.022	0.094	-0.339*	-0.437**
NSC	-0.015	0.346**	0.177	0.098	0.053	0.242*	**1.000**	0.056	-0.137	-0.246	0.150	-0.213	-0.066
TSW	0.245**	0.018	-0.199*	-0.044	-0.005	-0.098	0.051	**1.000**	-0.069	-0.125	-0.184	0.313	0.179
OC	-0.112	-0.246**	-0.071	-0.092	0.460**	-0.186	-0.137	-0.068	**1.000**	0.173	-0.017	-0.160	0.181
PC	-0.035	-0.112	-0.214*	0.024	0.191*	-0.035	-0.238*	-0.124	0.171	**1.000**	0.020	0.139	0.227
Fe	0.057	0.092	0.048	0.039	-0.145	0.080	0.145	-0.180	-0.017	0.020	**1.000**	-0.055	0.037
Zn	-0.015	-0.242*	-0.116	0.098	0.014	-0.284**	-0.205*	0.307**	-0.159	0.138	-0.054	**1.000**	0.326*
SYP	0.261**	0.005	0.172	0.478**	0.431**	-0.266**	-0.032	0.143	0.148	0.188*	0.028	0.274**	**1.000**

** Significant at 1% level of probability, * Significant at 5.0 % level of probability

Genotypic correlation coefficients (r_g) in above diagonals (blue shaded) and Phenotypic correlation coefficients (r_p) in below diagonals (yellow shaded)

DFF=Days to 50% flowering | DM=Days to maturity | PH=Plant height (cm) | PBP=Primary branches per plant
CP=Capsules per plant | CL=Capsule length (cm) | NSC=Number of seeds per capsule | TSW=1000 seed weight (g)
OC=Oil content (%) | PC=Crude protein content (%) | Fe=Fe content (ppm) | Zn=Zn content (ppm)
SYP=Seed yield per plant (g)

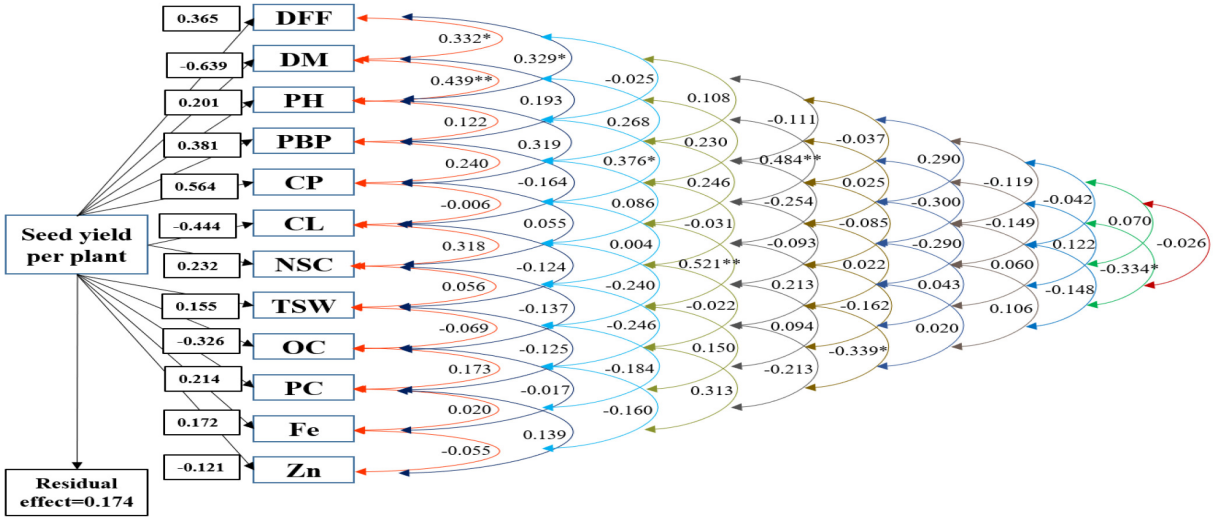

Fig. 6 : Genotypic path diagram for seed yield per plant in sesame

DFF=Days to 50% flowering | DM=Days to maturity | PH=Plant height (cm) | PBP=Primary branches per plant
CP=Capsules per plant | CL=Capsule length (cm) | NSC=Number of seeds per capsule | TSW=1000 seed weight (g)
OC=Oil content (%) | PC=Crude protein content (%) | Fe=Fe content (ppm) | Zn=Zn content (ppm)

Cluster analysis is a widely used method for genetic diversity studies, grouping genotypes based on their similarities. Understanding genetic diversity within and between closely related crop germplasm is essential for effectively utilizing genetic resources. Thirty-seven genotypes of sesame were grouped into 6 different non-overlapping clusters as presented in Table 4 and Figure 7. Cluster II is the biggest cluster with 11 genotypes, followed by cluster IV, which includes 7 genotypes. Three clusters *i.e.* I, III and V have 6 genotypes each and lastly, cluster VI is the smallest group containing only one genotype. The significant genetic diversity observed in the genotypes studied highlights their value for selecting diverse parents in hybridization programs, aiming to isolate desirable segregants and develop improved sesame varieties.

Table 4: Grouping of thirty-seven genotypes of sesame into different clusters using D^2 statistics

Cluster No.	No. of Genotypes	Genotypes
I	6	GT-10, RT-383, DS-18-46, AT-386, JCS-2420, AT-411
II	11	AT-392, TKG-525, DS-51, OSM-17-22, DS-45, JLS-408-3, Nana Rajkot-1, AT-410, IC-199433, JLS-708, LT-17-28
III	6	RT-384, AT-382, NIC-8476, SVT-333, AT-378, LT-17-10
IV	7	DS-53, AT-371, JLS-120, JLS-2611, AT-369, IC-204680, AT-482
V	6	JLS-907, JLS-706, AT-3661, AT-468, GJT-5, DS-17-15
VI	1	DS-18-21

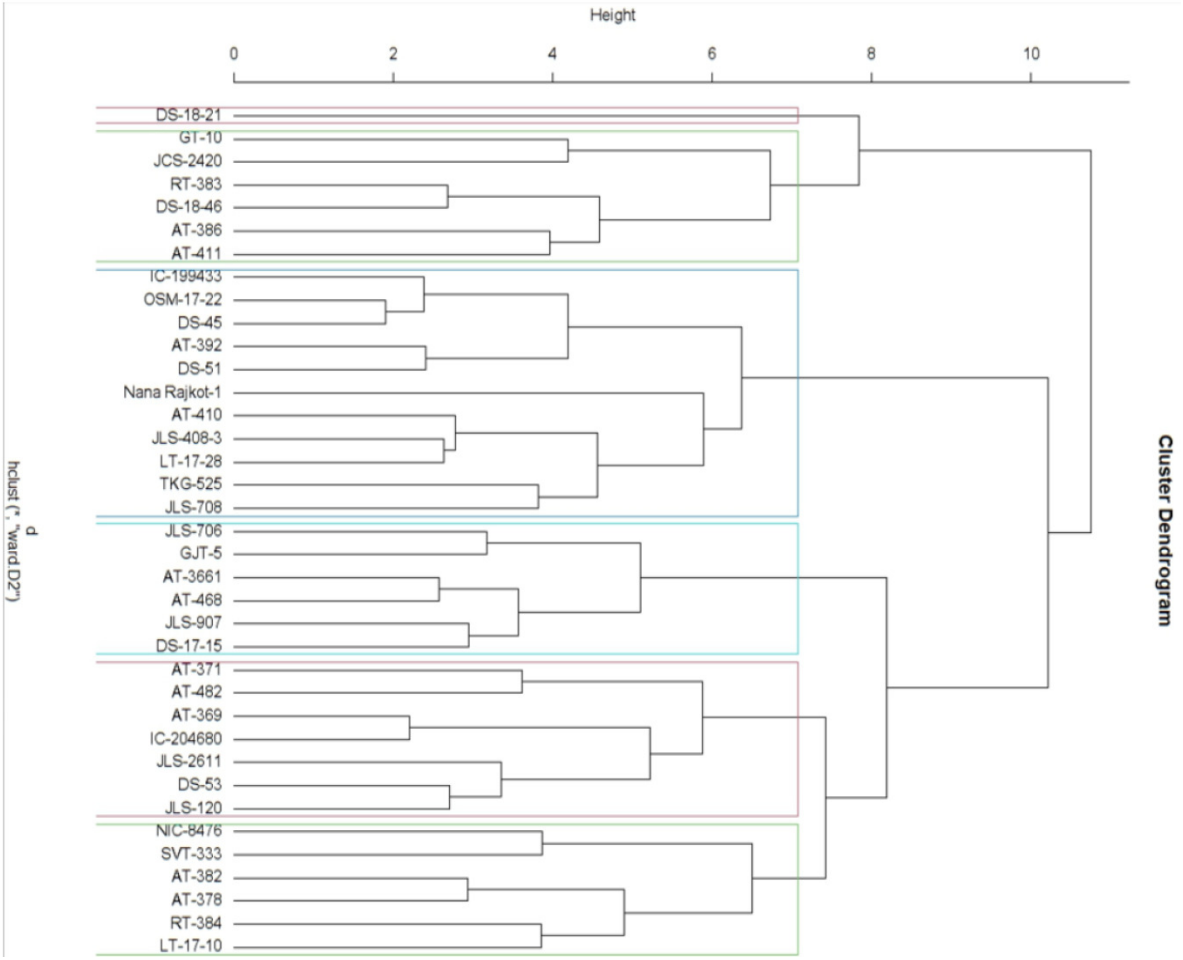

Fig. 7 : Dendrogram of thirty-seven sesame genotypes based on thirteen traits

Table S3 and Figure 8 indicates that cluster I exhibit maximum intra-cluster distance (6.516), which refers wide genetic diversity within the genotypes of this cluster. Whereas, cluster VI possess intra-cluster distance 0.000, as it consists only one genotype. Clusters I and III exhibit the maximum inter-cluster distance (8.346), followed by clusters II and VI (8.333), clusters I and VI (8.311), clusters I and II (7.721) and clusters III and VI (7.646). Genotypes from these diverse clusters can produce heterotic

hybrids by combining diverse genes in their hybrid progeny. Clusters III and V exhibited the smallest inter-cluster distance (5.879), indicating a close phylogenetic relationship among the genotypes within this cluster pair. Rahna *et al*. (2023), Hemanth *et al*. (2024) and Khuntia *et al*. (2024) also suggested selecting genotypes from clusters with the greatest inter-cluster distance for hybridization.

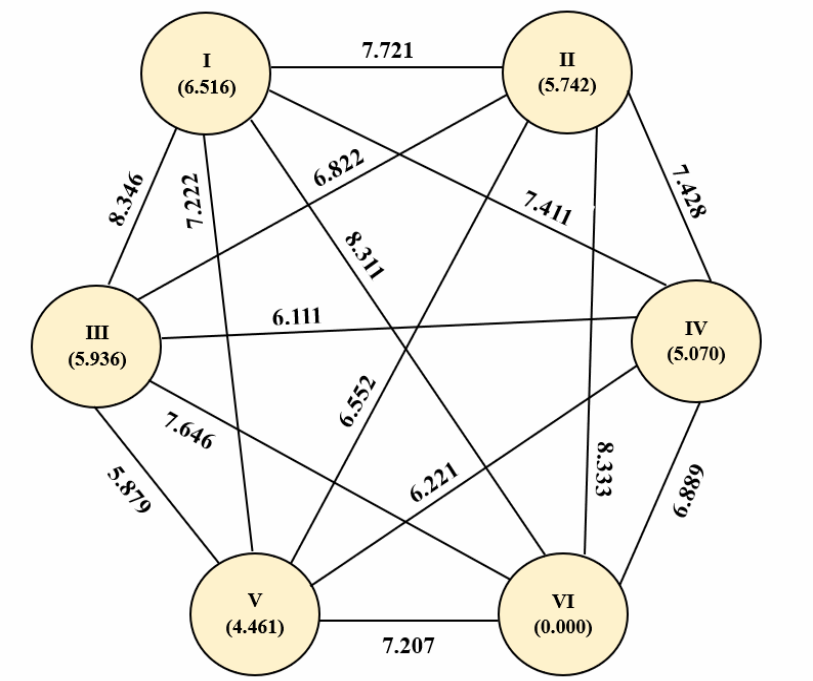

Fig. 8: Cluster diagram depicting intra and inter-cluster distances among various clusters of sesame genotypes

Genotypes from clusters with high mean values for the target trait should be prioritized for hybridization programs aimed at trait improvement. A review of Table 5 showed notable differences in cluster means for various characters. A closer analysis reveals that cluster II achieved the highest cluster means for most of the desirable traits *viz.* days to 50% flowering, primary branches per plant, capsules per plant, oil content, crude protein content and seed yield per plant. Thus, genotypes from these clusters can be directly chosen for immediate utilization. Similar findings were reported earlier in sesame by Jadhav and Mohrir (2012), Tripathi *et al*. (2013), Swathy *et al*. (2018), Tanwar and Bisen (2018), Bhattacharjee *et al*. (2019), Ramya *et al*. (2020) and Khuntia *et al*. (2024).

Table 5: Cluster wise mean values of thirteen characters in sesame genotypes

Characters	Clusters					
	I	II	III	IV	V	VI
Days to 50% flowering	37.067	40.583	38.125	39.292	38.000	37.889
Days to maturity	87.333	89.167	89.000	90.292	87.185	87.333
Plant height (cm)	107.311	99.713	98.979	116.975	96.556	89.924
Primary branches per plant	1.187	1.900	1.300	1.600	1.348	1.511
Capsules per plant	32.807	35.567	22.217	28.896	24.515	21.822
Capsule length (cm)	2.655	2.397	2.666	2.616	2.413	2.586
Number of seeds per capsule	49.453	39.633	46.275	52.117	35.948	58.667
1000 seed weight (g)	2.246	2.932	2.308	2.631	2.711	3.449
Oil content (%)	48.580	48.658	42.383	41.396	42.396	42.711
Crude protein content (%)	19.729	22.149	21.401	17.988	21.979	17.862
Fe content (ppm)	148.687	139.225	196.021	137.146	121.974	153.989
Zn content (ppm)	64.920	62.567	49.554	57.196	90.681	68.567
Seed yield per plant (g)	3.665	4.488	3.533	3.843	3.951	3.647

Figure 9 indicates how individual characters contribute to overall genetic diversity. Among the examined characters, Zn content contributed maximum (45.8%) towards genetic divergence followed by Fe content (36.6%) and crude protein content (6.9%). Contribution from days to maturity, plant height and seed yield per plant towards genetic divergence were reported very low *i.e.* 0.1% each. The present findings are supported by earlier reports of Parameshwarappa *et al*. (2012), Narayanan and Murugan (2013), Kiranmayi *et al*. (2016), Soundharya *et al*. (2017) and Hemanth *et al*. (2024).

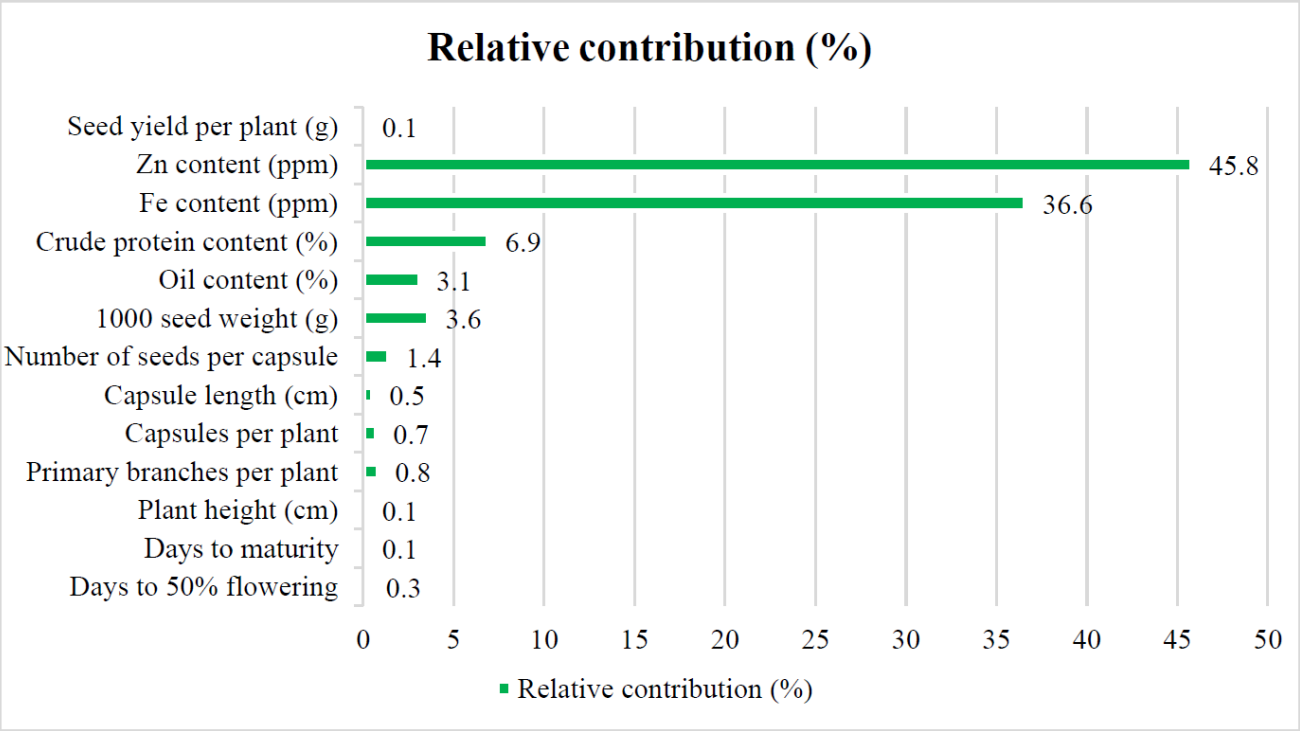

Fig. 9 : Relative contribution (%) of different characters to total genetic divergence in sesame

Conclusion

The present study aimed to assess the genetic variability parameters along with correlation, path coefficient analysis and genetic diversity using cluster analysis among thirty-seven germplasm of sesame. The ANOVA revealed the highly significant differences among the mean sum of squares with respect to sesame genotypes for all the traits under study. This exhibited that a sufficient amount of variability is present within the material under study. Primary branches per plant, capsules per plant, number of seeds per capsule, 1000 seed weight, seed yield per plant, Fe content, Zn content and crude protein content showed moderate to high GCV and PCV, high heritability and moderate to high genetic advance as per cent mean. So, improvement of these traits could be achieved through direct phenotypic selection. At the genotypic level, seed yield per plant was significantly and positively correlated with primary branches per plant, capsules per plant, days to 50% flowering and Zn content, which concluded that these characters can be improved simultaneously with seed yield per plant by direct selection. The overall genotypic path analysis revealed that the highest positive direct effects on seed yield per plant were exhibited by capsules per plant followed by primary branches per plant, days to 50% flowering, number of seeds per capsule, crude protein content and plant height. Therefore, selection for such characters in the genotypes would be helpful to bring about the improvement in sesame. The genetic diversity study through cluster analysis grouped the genotypes into six clusters, indicating high heterogeneity among the genotypes. Therefore, using the germplasms from such heterotic group could be effective for increasing the variation in hybridization programme. Divergence study revealed the major contribution of Zn content, Fe content and crude protein content towards total diversity in the population. Considering the inter-cluster distance, hybridisation programme may be carried out between the genotypes from cluster I and cluster III to achieve maximum heterotic hybrids and to produce a broader range of variation in the segregating population. The information on genetic variability and the grouping of genotypes into different clusters can be crucial for breeding programs focused on enhancing yield potential in sesame.

Table S1: Genetic variability parameters of various characters in sesame

Characters	Range	Mean	V_g	V_p	GCV (%)	PCV (%)	h^2_{bs}(%)	GA	GAM
Days to 50% flowering	34.67-43.00	38.45	2.65	4.02	4.23	5.22	65.77	2.72	7.07
Days to maturity	85.00-92.00	88.50	2.29	4.29	1.71	2.34	53.27	2.27	2.57
Plant height (cm)	86.64-127.47	102.75	95.62	172.63	9.52	12.79	55.39	14.99	14.59
Primary branches per plant	1.00-2.87	1.44	0.20	0.23	30.88	32.92	87.95	0.86	59.66
Capsules per plant	15.47-39.20	27.08	33.77	41.40	21.46	23.76	81.57	10.81	39.93
Capsule length (cm)	2.12-3.11	2.56	0.04	0.06	8.07	9.59	70.88	0.36	14.00
Number of seeds per capsule	28.67-70.87	45.74	131.29	140.17	25.05	25.88	93.67	22.85	49.94
1000 seed weight (g)	2.02-3.55	2.63	0.22	0.23	17.90	18.18	96.89	0.95	36.29
Oil content (%)	38.13-52.93	43.72	15.24	15.63	8.93	9.04	97.54	7.94	18.17
Crude protein content (%)	14.88-25.09	20.37	7.37	7.45	13.32	13.40	98.83	5.56	27.29
Fe content (ppm)	103.17-219.20	149.34	1529.29	1532.63	26.19	26.22	99.78	80.47	53.89
Zn content (ppm)	21.60-106.60	66.24	570.80	571.91	36.07	36.11	99.81	49.17	74.23
Seed yield per plant (g)	3.20-4.80	3.83	0.16	0.23	10.38	12.38	70.36	0.69	17.94

Table S2: Genotypic path coefficient analysis of component characters towards seed yield per plant in thirty-seven sesame genotypes

Characters	DFF	DM	PH	PBP	CP	CL	NSC	TSW	OC	PC	Fe	Zn	Correlation with seed yield per plant
DFF	**0.365**	-0.212	0.066	-0.010	0.061	0.049	-0.008	0.045	0.039	-0.009	0.012	0.003	**0.401***
DM	0.121	**-0.639**	0.088	0.074	0.151	-0.102	0.112	0.004	0.098	-0.032	0.021	0.040	**-0.064**
PH	0.120	-0.281	**0.201**	0.047	0.180	-0.167	0.057	-0.040	0.028	-0.062	0.010	0.018	**0.111**
PBP	-0.009	-0.123	0.025	**0.381**	0.135	0.073	0.020	-0.005	0.030	0.005	0.007	-0.013	**0.525****
CP	0.040	-0.171	0.064	0.091	**0.564**	0.003	0.013	0.001	-0.169	0.046	-0.028	-0.002	**0.450****
CL	-0.040	-0.147	0.075	-0.062	-0.004	**-0.444**	0.074	-0.019	0.078	-0.005	0.016	0.041	**-0.437****
NSC	-0.013	-0.309	0.049	0.033	0.031	-0.141	**0.232**	0.009	0.045	-0.053	0.026	0.026	**-0.066**
TSW	0.106	-0.016	-0.051	-0.012	0.002	0.055	0.013	**0.155**	0.022	-0.027	-0.032	-0.038	**0.179**
OC	-0.044	0.192	-0.017	-0.035	0.293	0.107	-0.032	-0.012	**-0.326**	0.037	-0.003	0.019	**0.181**
PC	-0.015	0.095	-0.058	0.008	0.120	0.010	-0.057	-0.019	-0.056	**0.214**	0.003	-0.017	**0.227**
Fe	0.026	-0.078	0.012	0.016	-0.092	-0.042	0.035	-0.029	0.006	0.004	**0.172**	0.007	**0.037**
Zn	-0.009	0.213	-0.030	0.040	0.011	0.150	-0.049	0.049	0.052	0.030	-0.009	**-0.121**	**0.326***

** Significant at 1.0 per cent level of probability, * significant at 5.0 per cent level of probability, Residual = 0.174, Bold diagonal figures are the direct effects

DFF=Days to 50% flowering DM=Days to maturity PH=Plant height (cm) PBP=Primary branches per plant
CP=Capsules per plant CL=Capsule length (cm) NSC=Number of seeds per capsule TSW=1000 seed weight (g)
OC=Oil content (%) PC=Crude protein content (%) Fe=Fe content (ppm) Zn=Zn content (ppm)

Table S3: Intra (bold) and inter cluster D values for various clusters of sesame genotypes

Cluster No.	Cluster I	Cluster II	Cluster III	Cluster IV	Cluster V	Cluster VI
Cluster I	**6.516**	7.721	8.346	7.411	7.222	8.311
Cluster II		**5.742**	6.822	7.428	6.552	8.333
Cluster III			**5.936**	6.111	5.879	7.646
Cluster IV				**5.070**	6.221	6.889
Cluster V					**4.461**	7.207
Cluster VI						**0.000**

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