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## MORPHOLOGICAL CHARACTERISTICS AND GENETIC DIVERSITY OF ETHIOPIAN SESAME GENOTYPES

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### ABSTRACT

Sesame (*Sesamum indicum* L.) is produced worldwide, although more than 96% of the world sesame seed is produced in Africa and Asia. The objective of this study was to determine morphological properties and identify the genetic diversity of cultivated sesame genotypes grown in different parts of Ethiopia. Three hundred sesame genotypes collected from diverse ecologies of Ethiopia and introduced from different African and Asian countries, were used in this study. Genotypes showed wide variability for most morphological traits, except for plant growth type, leaf glands, anther filament colour, anther connective tip gland, and anthocyanin colouration of the capsule. Genetic divergence using Mahalanobis D2 statistics was computed, and the genotype lines were grouped into six different clusters. Clustering was not associated with the geographical distribution; instead genotypes were grouped mainly based on morphological differences. The lowest divergence was noticed between cluster I and V (10.06). Maximum inter-cluster distance was observed between clusters IV and VI (D2 =342.56, followed by clusters I and VI (D2 =217.9783), and III with IV (D2 =190.8707). Maximum genetic recombination and variation in the subsequent generation, is expected from crosses that involve parents from the clusters characterised by maximum distances. Thus, maximum distances or variation could maximise opportunities for transgressive segregation, since unrelated genotypes would contribute unique desirable alleles at different loci.

**Key Words:** Inter-cluster distance, Mahalanobis D2 statistics, transgressive segregation

### RÉSUMÉ

Le sésame (*Sesamum indicum* L.) est produit dans le monde entier, bien que plus de 96% de la graine de sésame mondiale soit produite en Afrique et en Asie. L'objectif de cette étude était de déterminer les propriétés morphologiques et d'identifier la diversité génétique des génotypes de sésame cultivés dans différentes régions d'Éthiopie. Trois cents génotypes de sésame collectés dans diverses écologies d'Éthiopie et introduits dans différents pays africains et asiatiques ont été utilisés dans cette étude.

Les génotypes ont montré une grande variabilité pour la plupart des caractères morphologiques, à l'exception du type de croissance des plantes, des glandes foliaires, de la couleur du filament d'anthère, de la glande de l'extrémité conjonctive de l'anthère et de la coloration anthocyanique de la capsule. La divergence génétique à l'aide des statistiques de Mahalanobis D2 a été calculée, et les lignées génotypiques ont été regroupées en six groupes différents. Le regroupement n'était pas associé à la répartition géographique; au lieu de cela, les génotypes ont été regroupés principalement en fonction des différences morphologiques. La divergence la plus faible a été observée entre les grappes I et V (10,06). La distance maximale d'inter-grappes a été observée entre les grappes IV et VI (D2 = 342,56, suivie des grappes I et VI (D2 = 217,9783) et III avec IV (D2 = 190,8707). La recombinaison génétique maximale et la variation dans la génération suivante est attendu de croisements qui impliquent des parents des grappes caractérisées par des distances maximales. Ainsi, les distances maximales ou la variation pourraient maximiser les possibilités de ségrégation transgressive, puisque des génotypes non apparentés apporteraient des allèles uniques souhaitables à différents locus.

*Mots Clés:* Distance inter-grappe, statistiques Mahalanobis D2, ségrégation transgressive

## INTRODUCTION

Sesame (*Sesamum indicum* L.) is a major oil seed crop worldwide, with more than 96% of seed production accounted for by Africa and Asia (FAOSTAT, 2017). Sesame seeds are richly endowed with oil (44-57%), protein (18-25%) and carbohydrates (13-14%) (Borchani, 2010). Cultivated sesame has been described as a self-pollinated species; however, varying degrees of natural crossing to the extent of 2 to 48% have been reported (Daniel and Parzies, 2011).

In Ethiopia, sesame accounts for about 44% of the total acreage and 34% of gross production of major oilseeds cultivated in the country. It grows in almost all regions of the country with altitudes less than 2000 m above sea level (Yebiyo, 1985; Adefris *et al.*, 2011). Farm level national average productivity in Ethiopia is lower (0.68 t ha<sup>-1</sup>) (CSA 2019) than the potential yield of 2 t ha<sup>-1</sup> (Mkamilo and Bedigian, 2007). Improved varieties released in Ethiopia reportedly yield 0.3 to 1.3 t ha<sup>-1</sup> under rainfed, and 1 to 2.4 t ha<sup>-1</sup> under irrigated conditions (Gebremichael, 2017).

Sesame is one of the major indigenous oilseeds, displaying considerable diversity in Ethiopia (IBC, 2012). Moreover genetic diversity in crop plants is essential to sustain high productivity (Rabbani *et al.*, 2010). A large number of Ethiopian sesame germplasm,

locally collected and introduced, are held in the gene bank and in breeders' stock. There have been efforts on the use of agro-morphological characterisation, since the agro-morphological marker has been a primary tool for estimating genetic differences among Ethiopian sesame genotypes (Desawi *et al.*, 2014; Abate and Mekbib, 2015; Abate *et al.*, 2015).

Genetic diversity with agro-morphological characterisation and eco-geographic distribution; and microcentres of the diversity have a number of limitations. Despite the huge amount of locally collected and introduced germplasm held in the Ethiopian gene bank, and in breeders' stock, sesame morphological characterisation has only been done on a limited number of genotypes. The objective of this study was to determine the level of morphological variation and to cluster the Ethiopian sesame genotypes of different origins into similarity groups and assess the extent and pattern of diversity of the genotypes.

## MATERIALS AND METHODS

**Materials used.** A total of 300 sesame genotypes, comprising 225 local Ethiopian collections, including 16 released varieties and 75 exotic collections received from the Biodiversity Conservation Institute (IBC) of Ethiopia, and different federal and regional

research centre of Ethiopia, were used in this study. The collections were mainly from different regions of Ethiopia, and different countries of Africa and Asia. The number of sesame genotypes used in this study and their countries of origin are listed in Table 1.

The genotypes were planted at the Metema trial site (120 39'N, 360 17' E) in the 2017/18 cropping season, and this was repeated in the 2018/19 cropping season at Metema and T/ Armacho trial (13088'N, 370 43'E) sites. Metema is located at 760 meters above sea level and receives 1030.2 mm of rainfall per *annum* and its soil is a Vertisol. T/Armacho is located at 1022 meters above sea level and receives 970.88 mm of rainfall per *annum*, also with a Vertisol.

The study was laid out in an alpha lattice design, with each plot consisting of two rows of 4 m length with a spacing of 40 cm between rows and 10 cm between plants. Up to 65 kg ha<sup>-1</sup> of Urea, with two times split application, was applied manually based unpublished site specific recommendations. Thinning and hand weeding were carried out.

All quantitative (plant height, primary branch, secondary branch, length of basal leaf, width of basal leaf, length of middle leaf, width of middle leaf, length of top leaf, width of top leaf, petiole length of basal leaf, petiole length at middle (mid-level/mid-height) leaf, petiole length of top leaf, days to flower initiation, days to 50% flowering, number of capsules per plant, mean capsule length, mean capsule width, mean capsule thickness, seeds per capsule, 1000-seed weight, days to physiological maturity, pod bearing zone, seed yield, bacterial blight and qualitative), plant growth type (plant growth habit, root system, main stem colour, stem hairiness, shape of hair, stem shape in cross section, stem fasciation, stem branching, branching pattern, leaf colour, leaf hairiness, leaf arrangement, middle leaf shape ,top leaf shape, basal leaf profile, basal leaf margin, lobe incision of basal leaf, leaf glands, leaf angle to main stem, petiole colour, petiole hairiness, shape of petiole hair, number of flowers per leaf axil, extra-floral

TABLE 1. List of origin countries and number of sesame genotypes used for this study

Collection from Asia	China	Japan	Israel	Philippines
Number of genotypes	2	1	3	1
Collection from different Africa countries except Ethiopia	North Africa	South Africa	West Africa	East Africa
	Egypt	Zambia	Burkina Faso	Kenya
Number of genotypes	27	3	17	3
		Zimbabwe		Somalia
				Sudan
				2
Collection from different regions of Ethiopia	Amhara	Benshangul-Gumuz	Improved	Oromia
				SNNP
Number of genotypes	56	38	16	52
				3
				60
				Tigray

nectary development, extra-floral nectar colour, calyx tip colour, calyx hairiness, shape of calyx hair, corolla hairiness, shape of corolla hair, exterior corolla colour, interior corolla colour, corolla interior pigmentation, lower lip colour, absence/presence of foveolar, anther filament colour, anther connective tip gland, style length, number of carpels per capsule, bicarpellate capsule shape, capsule arrangement, capsule hairiness, shape of capsule hair, anthocyanin colouration of capsule, colour of dry capsules, capsule dehiscence at ripening, type of capsule beak, and thickness of capsule mesocarp data) were recorded according to the sesame descriptors list of IPGRI and NBPGR (IPGRI and NBPGR, 2004).

All measurements were done after tagging five randomly selected plant in each plot. Yield, growth period, plant and capsule variation and disease reactions, were recorded using standard procedures (IPGRI and NBPGR, 2004). The early flowering dates were recorded as the number of days from sowing to observation of the first flower on 50% of the individuals. Flower and leaf-related traits (days to flower initiation, days to 50% flowering and days to physiological maturity) were observed and measured in the full-bloom stage. After harvesting manually, yield-related traits (Number of carpels per capsule, Seeds per capsule, 1000-seed weight and seed yield) were measured in the laboratory. Seed yield was collected per plot and later converted into metric tonnes per hectare.

### Data analysis

**Analysis of variance.** Data obtained from the different environments were analysed separately, and thereafter combined for after the error homogeneity test, conducted between testing locations using SAS computer software (SAS, 2002). Statistically significant genotype means were separated using LSmeans package of SAS at  $P < 0.05$  level.

Multivariate analyses, including Cluster Analysis and Principal Component Analysis,

were used to group the 300 sesame genotypes into respective categories. The Principal Components Analyses were meant to identify large contributing traits to the total variation among the populations. Hierarchical clustering of accessions based on the Average Linkage Method were performed using JMP SAS software (JMP®, 2002) to group sesame genotypes. Statistics, pseudo F statistic and pseudo t<sup>2</sup> statistic generated by SAS were examined to decide the number of optimum clusters.

**Divergence analysis.** The patterns of distribution of morphological variation were analysed using Mahalanobis Generalised Distances ( $D^2$ ). The  $D^2$  were applied to estimate the distances between and within clusters, using the SAS computer software package as per the following formula:

$$D^2_{ij} = (X_i - X_j)' S^{-1} (X_i - X_j)$$

Where:

$D^2_{ij}$  is the distance between class  $i$  and  $j$ ;  $X_i$  and  $X_j$  are the vector means of the traits for the  $i^{\text{th}}$  and  $j^{\text{th}}$  groups, and  $S^{-1}$  is the inverse of the pooled covariance matrix.

The  $D^2$  analysis was based on the mean values of all morphological traits across locations. The  $D^2$  values obtained for pairs of clusters were considered as the calculated values of Chi-square ( $\chi^2$ ) and were tested for significance at  $P < 5\%$  against the tabulated value of  $\chi^2$  for 'P' degree of freedom, where P is the number of parameters considered (Singh *et al.*, 1985).

**Principal Component Analysis.** Principal components based on correlation matrix, and Euclidian distances were calculated using PAST software (Hammer *et al.*, 2001). One of the major reasons that analyses of principal components on correlation matrix was to standardise each variate (by subtracting its mean and dividing by its standard deviation), which is useful as the parameters considered

in this study did not share a common scale of measurement. Principal components having Eigen values >1 were considered as significant and presented in the results.

## RESULTS

**Morphological characteristics.** All genotypes showed wide ranges of variation for most of the morphological traits studied; except plant growth type, leaf glands, anther filament colour, anther connective tip gland and anthocyanin colouration of capsule (Table 2). All tested genotypes consisted of the plants with shattering and indeterminate growth habits. There was only white anther filament colour and absence of leaf glands and anther connective tip gland. Most of the genotypes had one flower per axil (95.64%). Of the 300 sesame genotypes, only 13 that were introduced from one of the African countries, Egypt had three flowers per axil.

There were large variations in stem, leaf, calyx, capsule and corolla hairiness among the accessions (Table 2). Most of the genotypes showed glabrous (hair absent) and weak or sparse hairiness; and short and straight shape of hair of stem, petiole and capsule. Strong or profuse hair was observed in a few genotypes (Table 2); on the other hand, uniqueness of hairiness and shape of hair of stem, petiole and capsule were recorded on certain genotypes that were introduced from Egypt.

There were large variations in sesame branching across all genotypes (Table 2), with 88.09% for Ternate, 1.36% for Alternate, 3.4% for Opposite and 7.48% as mixed branching patterns. All 21 genotypes that recorded mixed branching pattern were introduced from different African countries; and out of 21 germplasm, 13 were from Egypt alone. Some genotypes had Tetracarpellate capsule structure. (Table 2).

**Yield and yield components.** There were significant differences ( $P < 0.05$ ) among the study genotypes for yield and yield components; with the exception of number of

secondary branches, petiole length at middle leaf, mean capsule width and mean capsule thickness (Table 3).

The maximum values for days to flower initiation, days to 50% flowering and days to maturity were 72, 82 and 128 days, respectively. The highest seed yield was found in 9026 genotypes collected from Benshangul-Gumz Region. The average seed yield was 0.52 t ha<sup>-1</sup> with a range of 0.034 and 1.24 t ha<sup>-1</sup>. The mean and range values of all quantitative characters are presented in Table 3.

**Cluster analysis.** The 300 genotypes were grouped into 6 clusters based on 22 sesame parameters (Fig. 1). Five of the six clusters comprised of more than one germplasm; whereas one cluster was singleton (containing single accession). Cluster 1 contained 115 genotypes (38.3%) out of 300; and was followed by clusters II, V, IV and III containing 107, 36, 31 and 10 accessions, respectively. Whereas the cluster VI contained only one genotype (0.33%).

Cluster I constituted the largest number of germplasm, mainly collected from the different regions of Ethiopia 73(64.34 %); namely Amhara (n=36), Benshangul-Gumz (11), Oromia (4), Tigray (22) and 1 improved variety, 35 (30.43%). Those registered as introduced from different African countries included Burkina Faso (8), Egypt (10), Kenya (1), Somalia (1), Sudan (1), Zambia (1) and Zimbabwe (13). The remaining 6 genotypes were introduced from different Asian countries.

Most genotypes of Cluster II were collected from the different regions of Ethiopia 93(86%); - Amhara (n=9), Benshangul-Gumz (21), Oromia (35), SNNP (2), Tigray (26) and 10 improved varieties. Only 4 genotypes were introduced from three African countries; namely Burkina Faso (2), Egypt (1) and Kenya (1) belonged to Cluster II. All genotypes of Cluster III were collected from three different regions of Ethiopia; namely Amhara (n=4), Oromia (3), Tigray (2) and 1 improved variety.

TABLE 2. Predominant morphological characters for the sesame genotypes used in a study in Ethiopia

Plant growth type	100% Indeterminate				
Plant growth habit	0.3% Prostrate	0.3% Semi-erect	99.32% erect		
Root system	Shallow fibrous	94.01% Deep thin taproot	5.63% Tuberos thick taproot		
Main stem colour	0.36% Green	50% Yellow	1.07% Purplish green	48.20 Purple	0.36% Other
Stem hairiness	0.35% Glabrous (hair absent)	96.52% Weak or sparse	2.09% Medium	1.05% Strong or profuse	
Shape of hair	97.19% Short and straight	2.10% Medium and straight	0.7% Long and bent		
Stem shape in cross section	0.7% Round	99.3% Square			
Stem fasciation	99.3% Absent	0.7% Present			
Stem branching	3.4% Opposite	1.36% Alternate	88.09% Ternate	7.48% Mixed	
Branching pattern	4.60% Non-branching	16.31% Basal branching	31.91% Top branching	47.16% Other	
Leaf colour	40.88% Green	18.24% Green with yellowish cast	40.20% Green with blue-gray cast	0.67% Green with purple cast	
Leaf hairiness	82.37% Glabrous (hair absent)	17.62% Weak or sparse			
Leaf arrangement	4.73% Opposite	18.92% Alternate	8.11% Ternate	68.24% Mixed	
Leaf shape (middle)	Linear	31.9% Lanceolate	0.34% Elliptic	67.69% Ovate	
Leaf shape (Top)	30.45% Linear	69.55% Lanceolate	Elliptic		
Basal leaf profile	13.17% Flat	4.73% Cup shaped (concave)	82.09% Reverse cup shaped (convex)		
Basal leaf margin	5.76% Entire	93.22% Serrate	1.02% Dentate		
Lobe incision of basal leaf	6.84% Absent (leaf entire)	2.74% Weak	26.71% Medium	63.7% Strong (three or more lobes)	
Leaf glands	100% Absent				

TABLE 2. Contd.

Plant growth type	100% Indeterminate				
Leaf angle to main stem	16.55% Acute (<90°)	83.11% Horizontal (=90°)			
Petiole colour	21.33% Green	33.92% Greenish purple	40.56% Purple	4.19% Pink	
Petiole hairiness	2.09% Glabrous (hair absent)	96.15% Weak or sparse	1.39% Medium	0.35% Strong or profuse	
Shape of petiole hair	98.21% Short and straight	1.43% Medium and straight	0.36% Long and bent		
Number of flowers per leaf axil	95.64% One	4.36% More than one			
Extra-floral nectary development	0.34% Rudimentary	59.12% Small	39.18% Medium	1.35% Large	
Extra-floral nectar colour	0.34% Light yellow	99.66% Yellow			
Calyx tip colour	3.31% Green	96.69% Purple			
Calyx hairiness	0.83% Glabrous (hair absent)	97.93% Weak or sparse	1.24% Medium		
Shape of calyx hair	99.58% Short and straight	0.42% Medium and straight			
Corolla hairiness	35.37% Weak or sparse	37.75% Medium	26.87% Strong or profuse		
Shape of corolla hair	38.09% Short and straight	40.47% Medium and straight	21.43% Long and bent		
Exterior corolla colour	6.78% White	70.85% White with pink shading	22.37% White with deep pink shading		
Interior corolla colour	13.65% White	60.75% White with pink shading	25.59% White with deep pink shading		
Corolla interior pigmentation	4.41% Absent	31.86% Pigmented throughout	49.83% Pigmentation along the lip region of corolla tube	4.40% Pigmentation in the supra foveolate region	9.49% Pigmentation in the infra foveolate region

TABLE 2. Contd.

Plant growth type	100% Indeterminate		
Lower lip colour	16.27% Colourless	83.73% Coloured	
Absence/presence of foveolar	99.66% Absent	0.34% Present	
Anther filament colour	100% White		
Anther connective tip gland	100% Absent		
Style length	1% Short (stigma terminating below the position of anthers)	96.99% Medium (stigma position at anther's level)	2% Long (stigma protruding outside the position of anthers)
Number of carpels per capsule	98.99% Bicarpellate	0.66% tricarpellate	0.33% Tetracarpellate
Bicarpellate capsule shape	24.49% Narrow oblong	75.51% Broad oblong	
Capsule arrangement	95.65% Monocapsular	4.35% Multicapsular	
Capsule hairiness	97.32% Weak or sparse	2.67% Medium	
Shape of capsule hair	96.98% Short and straight	3.02% Medium and straight	
Anthocyanin colouration of capsule	100% Present		
Colour of dry capsules	29.89% Straw/yellow	69.56% Brown/tan	0.54% Purple
Capsule dehiscence at ripening	15.05% Partially shattering	84.95% Completely shattering	
Type of capsule beak	99.33% Curved	0.67% Cleft	
Thickness of capsule mesocarp	98.91% Thin	1.09% Thick	

TABLE 3. Range of variation and F- value of analysis of variance for quantitative characters in sesame genotypes

Character	Min	Max	Mean	Range	F Value
PTH	47.36	181.32	136.81	133.96	2.56***
PBR	0.12	6.5	3.5	6.38	2.16***
SBR	0	2.2	0.37	2.2	1
LBL	5.51	13.72	9.91	8.21	1.34**
WBL	2.51	9.52	6.76	7.01	1.54***
LML	5.49	12.68	9.3	7.19	1.84***
WML	2	6.44	3.53	4.44	1.42***
LTL	3.67	7.25	5.17	3.58	1.36***
WTL	0.46	7.25	0.72	6.79	1.31**
PLBL	2.84	8.44	5.89	5.6	1.46***
PLML	0.95	4.67	2.67	3.72	1.04
PLTL	0.24	1.44	0.47	1.2	1.89***
DFI	30	72	49.28	42	1.37***
DF	35	82	53.76	47	1.35**
NCPP	7.14	57.06	34.33	49.92	2.29***
CAPL	2.01	3.81	2.83	1.8	2.47***
CW	0.55	1.01	0.71	0.46	1.15
CT	0.33	0.74	0.5	0.41	1.06
SPC	48.44	82.4	67.36	33.96	1.4***
TSW	1.66	3.33	2.25	1.67	2.32***
DM	79	128	103.57	49	1.73***
PBZ	20.56	83.46	45.24	62.9	2.67***
YLD	0.034	1.24	0.52	1.204	3.95***
BBL	8.64	69.1	21.3	60.46	1.54***

PTH = plant height in centimeter; PBR = primary branch; LBL = length of basal leaf; WBL = width of basal leaf; LML = length of middle leaf; WML = width of middle leaf; LTL = length of top leaf; WTL = width of top leaf; PLBL = petiole length of basal leaf; PLTL = petiole length of top leaf; DFI = days to flower initiation; DF = days to 50% flowering; COL = corolla length; LLL = length of longest lip; NCPP = Number of capsules per plant; CAPL = capsule length; SPC = seeds per capsule; TSW = 1000 seed weight in gram; DM = days to maturity; PBZ = Pod bearing zone; YLD = yield in tonnes per hectare; BBL = bacterial blight reaction. \*\* Significant at  $P < 0.01$ ; \*\*\* Significant at  $P < 0.001$

Most genotypes of Cluster IV were collected from different countries of Africa 22 (70.96%) (Burkina Faso (6), Egypt (12), Sudan (1), Zambia (2) and Zimbabwe (2)); and 9 genotypes were collected from three regions of Ethiopia (Oromia (4), SNNP (1) and Tigray (4)).

Cluster V consisted of genotypes mainly collected from different regions of Ethiopia 29 (80.55 (Amhara (n=7), Benshangul-Gumz (5), Oromia (6) and Tigray (6); and 4 were

improved varieties. Only 7 genotypes were introduced from three African countries; namely Burkina Faso (2), Egypt (4) and Kenya (1). The remaining one genotypes was from Israel. Cluster VI consist only one genotype collected from Benshangul-Gumz from Ethiopia.

**Cluster mean analysis.** The clusters for different traits indicated wide variations for all the characters considered (Table 4). The

## Hierarchical Clustering

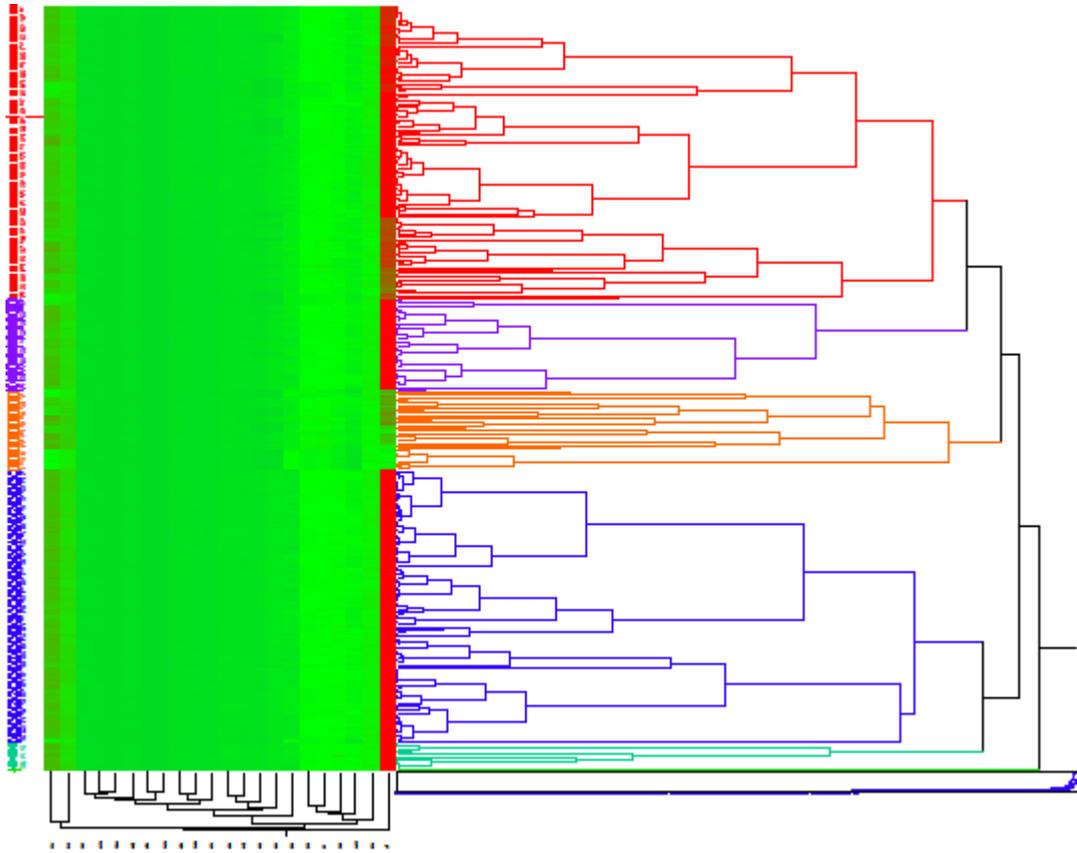


Figure 1. Dendrogram of 300 sesame genotypes based on average linkage hierarchical cluster analysis between groups of sesame in Ethiopia.

highest values for grain yield were recorded from Cluster VI (1.24 t ha<sup>-1</sup>) and the lowest from Cluster IV (0.132 t ha<sup>-1</sup>). The next highest yield to Cluster VI were recorded by Cluster III (0.973 t ha<sup>-1</sup>). Clusters means for all characters in sesame genotypes presented in Table 4.

**Divergence analysis.** Pair wise generalised squared distances (D<sub>2</sub>) among six clusters are presented in Table 5. There were 15 possible pair wise genetic distances between any two clusters. Among these, only 4 genetic distances (between clusters I and IV, I and V, II and III, II and V) were not significant ( $P > 0.05$ ); but the remaining genetic distances were highly significant ( $P < 0.01$ ). The maximum distance

was found between clusters IV and VI ( $D_2 = 342.56$ ), and the distances between I, IV and V clusters with cluster VI and cluster II and III with IV were maximum and very highly significant ( $P < 0.01$ ).

**Principal Component Analysis (PCA).** The first five principal components were found to be significant (Eigen value greater than 1) and accounted for about 75.4% of the total variation (Table 6). The first PCA component explained 34.8% of the total variance; while the first and second PCA components accounted for 54.6% of the variation (Fig. 2). Parameters that contributed relatively more with an Eigen vector value for the first PCA were plant height, length and width of basal leaf, petiole

TABLE 4. Cluster means for parameters in sesame genotypes used in a study of sesame germplasm conducted in Ethiopia

Parameters	Cluster						
	I	II	III	IV	V	VI	GM
PTH	125.6	142.48	149.8	105.1	133.47	144.32	133.46
PBR	3.35	3.17	3.26	3.13	2.99	6.08	3.66
LBL	10.31	10.63	11.24	8.37	10.31	10.52	10.23
WBL	6.76	6.96	7.35	5.4	6.76	6.61	6.64
LML	9.43	9.96	10.85	7.78	9.71	9.42	9.52
WML	3.94	3.8	3.87	3.4	3.9	4.16	3.85
LTL	5.32	5.3	5.5	4.96	5.39	5.24	5.28
WTL	0.8	0.82	0.83	0.99	0.84	0.96	0.87
PLBL	5.81	6.2	6.61	4.3	5.98	6.52	5.9
PLTL	0.51	0.52	0.57	0.69	0.57	0.43	0.55
DFI	48.21	46.54	45.07	49.38	45.77	48	47.16
DF	52.81	50.74	49.03	53.1	50.42	53.33	51.57
COL	13.17	16.45	16.27	8.21	14.58	16.37	14.18
LLL	7.89	9.85	10.12	4.85	8.9	10.07	8.62
NCPP	31.47	38.85	45.2	21.71	34.41	51.36	37.17
CAPL	2.94	2.85	2.95	2.63	2.86	2.38	2.77
SPC	68.92	69.36	68.14	63.3	68.74	63.8	67.04
TSW	2.33	2.3	2.45	2.17	2.43	2.62	2.38
DM	100.46	99.54	98.46	100.75	97.85	110	101.18
PBZ	42.79	52.95	58.27	33.62	48.32	45.48	46.91
YLD	0.393	0.771	0.973	0.132	0.569	1.24	0.679
BBL	24.34	17.08	18.1	34.46	21.73	9.14	20.81

PTH = plant height in centimeter; PBR = primary branch; LBL = length of basal leaf; WBL = width of basal leaf ; LML = length of middle leaf; WML = width of middle leaf ; LTL = length of top leaf ; WTL = width of top leaf; PLBL = petiole length of basal leaf; PLTL = petiole length of top leaf ; DFI = days to flower initiation; DF = days to 50% flowering ; COL = corolla length; LLL = length of longest lip; NCPP = Number of capsules per plant; CAPL = capsule length; SPC = seeds per capsule; TSW = 1000 seed weight; DM = days to maturity; PBZ = pod bearing zone; YLD = yield in tonnes per hectare; BBL = bacterial blight reaction

length of basal leaf, number of capsule per plant and grain yield. Most of the variations attributed to the second PCA were contributed by length of middle and top leaf, petiole length of top leaf and pod bearing zone.

The third PCA explained about 9.5% of the variation traits, such as width of middle leaf, capsule length and bacterial blight reaction contributed much of its variation (Fig. 2). The fourth PCA were explained by corolla length, length of the longest lip and seed per capsule;

while the fifth PCA explained by 1000 seed weight.

## DISCUSSION

**Morphological characteristics.** There was a high genetic variability among the tested genotypes for most of the evaluated traits that showed the potential of these genotypes for the sesame breeding programme to improve grain yield through direct selection and

TABLE 5. Intra- (bolded diagonals) and inter-cluster distance between sesame genotypes categorised into clusters

From class	Generalised Squared Distance to class					
	I	II	III	IV	V	VI
I						
II	41.14					
III	97.6344	14.6981				
IV	19.7745	108.429	190.871			
V	10.0594	12.566	48.6915	52.3529		
VI	217.978	88.4217	50.936	342.561	149.602	

$X^2 = 38.932$  at 1% probability level and  $X^2 = 32.671$  at 5% probability level

TABLE 6. Eigen vectors, explained variance and Eigen values of the first significant Principal components for parameters of sesame genotypes

Parameter	Eigen vectors				
	PCA1	PCA2	PCA3	PCA4	PCA5
Plant height	0.304	-0.09	-0.124	0.002	0.068
Primary branch	0.147	-0.292	-0.048	-0.251	0.123
Length of basal leaf	0.296	0.117	0.209	-0.001	0.108
Width of basal leaf	0.294	0.028	0.229	-0.121	0.074
Length of middle leaf	0.222	0.241	0.09	-0.281	-0.06
Width of middle leaf	0.121	0.069	0.296	-0.407	-0.424
Length of top leaf	0.035	0.304	0.217	-0.179	-0.009
Width of top leaf	-0.19	0.201	0.035	-0.316	-0.055
Petiole length of basal leaf	0.276	0.082	0.107	-0.206	0.156
Petiole length of top leaf	-0.236	0.21	0.011	-0.153	-0.136
Days to flower initiation	0.156	-0.368	0.15	-0.027	-0.171
Days to 50% flowering	0.162	-0.384	0.122	-0.068	-0.121
Corolla length	0.277	0.07	0.006	0.354	0.012
Length of longest lip	0.278	0.069	-0.013	0.351	0.001
Number of capsules per plant	0.222	0.07	-0.399	-0.18	-0.096
Capsule length	0.092	0.261	0.331	0.265	-0.107
Seeds per capsule	0.143	0.152	0.099	0.318	-0.521
1000 seed weight	0.118	0.073	0.323	-0.005	0.589
Days to maturity	0.173	-0.371	0.018	-0.101	-0.083
Pod bearing zone	0.178	0.269	-0.347	-0.054	0.055
Grain yield	0.209	0.205	-0.351	-0.092	0.093
Bacterial blight	-0.278	-0.017	0.258	0.045	0.18
Eigenvalue	7.672	4.3453	2.1004	1.3935	1.0867
Explained variance %	34.9	19.8	9.5	6.3	4.9
Cumulative variance	34.9	54.6	64.2	70.5	75.4

PCA= Principal component analysis

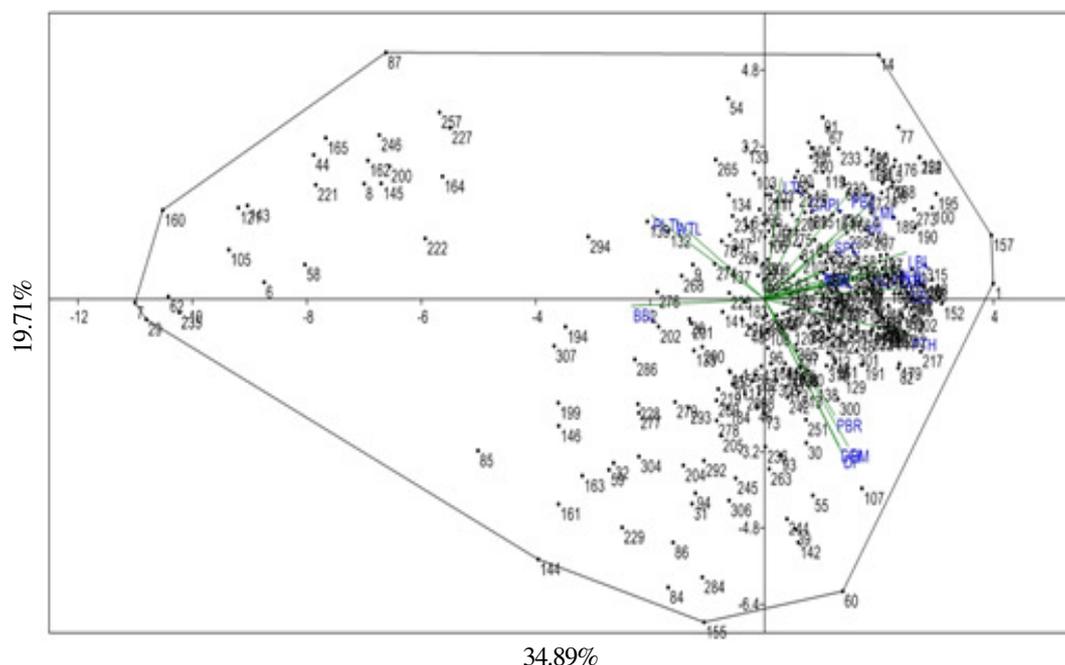


Figure 2. Biplot of PC1 and PC2 showing the overlay of 300 genotypes and the 22 studied parameters. Note: The black color represents the germplasms (n=300) and the blue color represents the parameters under the study.

crossing among variable potential genotypes (Table 2). Among the evaluated traits, grain yield showed high variabilities that can be used for direct selection for release and producing potentially high yielder segregation populations through crossing form different origins. Similar results were reported on 10 morphological and 11 agronomic descriptors of sesame (Uzun and Çađýrgan, 2006; Furat and Uzun, 2010).

Number of flowers per leaf axil (number of capsule per plant) is one of the important characters for plant breeding programmes since it provides sufficient seed per cross for subsequent evaluation activities. In our study, the 13 sesame genotypes introduced from Egypt gave up to three flowers per leaf axil that showed the potential of these genotypes in the future sesame breeding programmes in Ethiopia. The importance of this trait as a yield contributing character was emphasized by (Osman, 1989; Ibrahim *et al.*, 1983).

Large variations were observed in stem, leaf, calyx capsule and Corolla hairiness among

the genotypes; ranging from glabrous to strong or profuse (Table 2). Uniqueness on the hairiness and shape of hair of stem, petiole and capsule were recorded on a certain number of genotypes that were introduced from Egypt. Since hairiness is a typical character of sesame and can be seen in many parts of the plant such as stem, leaf, corolla, and capsule (Weiss, 1983), and the trait has advantages for preventing insect pests and diseases of sesame, it also has positive effect in terms of preventing water loss, and is a desirable trait in breeding. Having this variation is important for further breeding programme.

Some genotypes had Tetracarpellate capsule structure (Table 2). This type of capsule potentially enlarges the space for more seeds to fit and a structural modification converting the two extra-floral nectaries to capsules (Bedigian, 2003). However, plants with Tetracarpellate capsules exhibit flatness on the stem, which is rarely seen in Bicarpellate sesame genotypes.

**Yield and yield components.** The analysis of variance revealed statistically significant differences at 0.1 and 0.5% probability level among three hundred sesame genotypes for yield and yield component characters (Table 2), except for a certain characters (secondary branch, petiole length at middle (mid-level/mid-height) leaf, mean capsule width and mean capsule thickness). This indicates the existence of substantial genetic variation among genotypes in all characters. This result agrees with the observation of Saha *et al.* (2012).

Genotype 203637 had the maximum values for flower initiation (72 days), days to 50% flowering (82 days), and days to maturity (128 days); while several early maturing germplasms existed in the collection (Table 1). These germplasms demonstrated the valuable resource available in the germplasm pool to be used in the breeding programmes to develop sesame genotypes adapted to different environments, as well as for studies on thermo- and photo-period sensitivity (Suddhiyam *et al.*, 1992; Aziz ur Rehman *et al.*, 2009).

The mean seed yield was 0.52 t ha<sup>-1</sup>, with a range of 0.034 and 1.24 t ha<sup>-1</sup>. The mean and the ranges of the genotype values revealed a large genetic diversity useful for the development of varieties. The genotypes with a wide range of variation for agronomic characters had potential to determine the best genotypes for different environments.

**Cluster analysis.** The 300 sesame genotypes were grouped into 6 clusters containing significantly different numbers of sesame genotype, which ranged from 1 to 115. Cluster I and V contained genotypes originating from two different continent countries. Even from the African continent (East, West, South and North Africa), from this we observed a close relationship between genotype from East Africa (majority from different regions of Ethiopia), South Africa, North Africa and West Africa to the genotype from Asia. Clusters II and IV contained genotypes from different countries of Africa that included different regions of Ethiopia. Genotypes from the same

origin were not all grouped into the same cluster. This close genetic relationship observed might be due to the introduction of sesame into many countries and material exchange from widely separated locations (Kim *et al.*, 2002). Moreover, the exchange of plant materials between Asia and East Africa dated back to a long time ago and still occurs (Zohary *et al.*, 2012), with a steady increase in annual exportation of raw sesame seeds mainly for industrial applications.

The likelihood of crossover events between materials from different locations grown within the same area is high, knowing that cross-pollination in sesame has been reported to occur at a frequency between 5 and 60% (Wei *et al.*, 2014). This crossing could explain the similarity of genotypes from the eastern part of Africa and Asia. Similar patterns have been observed by other researchers (Kim *et al.*, 2002; Laurentin and Karlovsky, 2006; Park *et al.*, 2011).

Cluster III contained genotypes originating from the same country, but from different three regions (Fig. 1). This result indicates the possibility of exchange of seeds, and seed trade between farmers, and gene flow across boundaries of those areas (Forsberg *et al.*, 2015). The last cluster contained one genotype from Benshangul-Gumz. This might indicate that the genotype from Benshangul-Gumz was more diverse than others. The distribution and pattern of genotype, over significantly different clusters from Africa and Asia major geographic origins, would suggest future collections of local genotypes in those geographic regions is importance, for future national and international collection mission in sesame.

**Cluster mean analysis.** The cluster mean for different traits (Table 4), suggested a wide range of variation for all the characters under the study. Based on Clusters VI and III, the highest mean values for grain yield were recorded from genotypes originating from Ethiopia; but based on clusters IV and I, the lowest mean value for grain yield were recorded from most genotypes introduced from

different African and Asian countries. Several factors may contribute to this observation, including climatic reasons such as temperature (day/night), day length, light intensity, precipitation, altitude and latitude. Photosynthesis is influenced by various biotic and abiotic stresses during grain filling; therefore, decrease or increase photosynthesis capacity is a major limiting factor for yield and all yield components (Beheshti and fard, 2010). There are reasons to believe that the less performing germplasm from different African and Asia countries were not adjusted to the field conditions, and therefore, gave low yields in this study. Basu *et al.* (2009) also reported seed yield to be a complex trait governed by polygene, and therefore is influenced more by environmental factors.

Our observation is in agreement with previous studies, in which sesame was shown to be highly sensitive to day length since it is a short day plant (Narayanan and Reddy, 1982). Suddhiyam *et al.* (1992) also reported about the significant interactions temperature and day length had on the flowering rate. According to them, yield depends on the interactions of different climatic parameters such as solar radiation, temperature, humidity relative to photosynthetically active radiation (PAR) (Beech and Ashri, 1985; Nath *et al.*, 2001). (Yadav *et al.* (1988) also reported close correlations between PAR absorption and yield in sesame.

The germplasm listed in the clusters VI and VII were found valuable to be selected for use in breeding strategy to improve high yielding sesame genotypes.

**Divergence analysis.** The maximum distance among tested sesame genotypes lies between clusters IV and VI ( $D_2 = 342.56$ ). Maximum genetic recombination and variation in the subsequent generation, is expected from crosses that involved parents from the clusters characterised by maximum distances. Thus, it could maximise opportunities for transgressive segregation, since a higher probability that unrelated genotype would

contribute unique desirable alleles at different loci. Genetic distance, as a good indicator of transgression and heterosis, has been reported by several authors on many crops (Mulugeta and Hussein, 2013; Pickup *et al.*, 2013). Hence, the attempt to cluster Ethiopian sesame genotypes using multivariate analyses, in the present study, is a significant precursor to initiating sesame breeding programme. However, the selection of parents for a particular cross should also consider the special advantages of each cluster and accession within a cluster, depending on specific objectives of hybridisation programmes.

Members within a cluster are assumed to be more closely related, in terms of trait under consideration than with members in different clusters (Million, 2012; Habtamu and Million, 2013). This indicates that superior hybrids or recombinants can be realised by mating between the lines of these clusters in a definite fashion. Crossing between genotypes belonging to the same cluster might not be expected to yield desirable segregates. This approach is, however, based on the assumption that suitable parents for crossing may be showing greater amount of genetic divergence. Further research on these selected germplasm will save a lot of time for the breeder in future.

#### **Principal Component Analysis (PCA).**

Principal components analyses in this study showed that the first five PCAs explained about 75.4% of the variation (Table 6). The amount of explained variance by the first PCA and parameters that contributed relatively more, clearly indicated that grain yield and architectural traits of sesame are important traits that could be considered for sesame breeding and selection. Principal components analyses results indicated that the genotype lines studied had a considerable level of variability that could be exploited in future breeding programs. Hybridisation between genetically diverse genotypes in sesame to generate promising breeding material has been suggested by Alarmelu and Ramanathan, 1998.

### CONCLUSION

There is a considerable level of variability among sesame genotypes within the Ethiopian germplasm, as well as genotypes collected from elsewhere in Africa and Asia; a phenomenon that could be exploited in future breeding programmes. All genotypes are grouped into six clusters, with cluster I and II accounting for the largest number of genotypes. Greater genetic divergence is present between clusters IV and VI; followed by distances between clusters I and VI and cluster III with IV; indicating that superior hybrids or recombinants can be realised by mating between the lines of these clusters in a definite fashion. Their high yield potential can subsequently be combined with improvements of other traits such as plant height, number of capsule per plant, oil content, resistance to pests and disease.

The genotype originally collected from four regions of Ethiopia (Amhara, Benshangul-Gumuz, Oromia and Tigray), specially genotype categorised under clusters VI and III, were found interesting and could be candidates for potential immediate breeding sources due to their high seed yield. Further research on these selected genotype will save a lot of time for the breeder in future. Morpho-agronomic traits have some shortcomings in evaluating genetic diversity as these are phenotypic markers and genetically distant genotypes may be morphologically similar. Further research should be done with molecular markers which can be used to determine genetic distance easily and successfully. DNA markers should provide more accurate measures of genetic similarity.

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