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Devesh Yadav
Department of Genetics and Plant Breeding, C. S. Azad University of Agriculture and Technology, Kanpur, Uttar Pradesh, India

Lokendra Singh
Department of Genetics and Plant Breeding, C. S. Azad University of Agriculture and Technology, Kanpur, Uttar Pradesh, India

Syed Kulsoom Fatima Jafri
Department of Genetics and Plant Breeding, C. S. Azad University of Agriculture and Technology, Kanpur, Uttar Pradesh, India

Upendra Kumar Mishra
Department of Genetics and Plant Breeding, C. S. Azad University of Agriculture and Technology, Kanpur, Uttar Pradesh, India

Vineet Singh
Department of Genetics and Plant Breeding, C. S. Azad University of Agriculture and Technology, Kanpur, Uttar Pradesh, India

Corresponding Author:
Devesh Yadav
Department of Genetics and Plant Breeding, C. S. Azad University of Agriculture and Technology, Kanpur, Uttar Pradesh, India

Study of genetic divergence in different genotypes of Indian mustard [*Brassica juncea* (L.) Czern and Coss]

Devesh Yadav, Lokendra Singh, Syed Kulsoom Fatima Jafri, Upendra Kumar Mishra and Vineet Singh

Abstract
An experiment involving 18 genotypes of Indian Mustard (*Brassica juncea* L.) was conducted in randomised block design with three replications, during *Rabi* 2020. Data were recorded and analysed for fourteen characters. All the 18 genotypes were grouped into 5 clusters using D^2 statistics. Cluster II, III and IV had equal number of genotypes (5) while, cluster I had 2 genotypes. The minimum number of genotypes (1) was present in cluster V. Maximum intra-cluster distance was found in cluster I while, there was no intra-cluster distance in cluster V. The inter cluster distance was found maximum between cluster II and V followed by cluster IV and V & cluster III and V.

Keywords: Genetic divergence, D^2 statistics and Indian mustard

Introduction
Indian mustard [*Brassica juncea* (L.) Czern and Coss] is the second most important oilseed crop of the world as well as India after groundnut. It is a natural amphidiploid (2n=36) containing complete genome of *Brassica campestris* (2n=20) and *Brassica nigra* (2n=16) having self-compatible and mainly self-pollinated nature (85-90%). Indian mustard is popularly known as rai, raya or Laha and it occupies major acreage about 75-80% of the total area under rapeseed-mustard in the country.

Mustard oil has a strong aroma and is considered a 'strong' oil source due to its dominating flavour. Due to the mustard oil composition, it is very well suited to spicy Indian curries and is used widely across Indian households. Indian mustard oil is rich in MUFA or monounsaturated fatty acids that are considered as good fats and help your body in fighting cardiovascular ailments. Moreover, mustard oil also exhibits anti-microbial properties due to the presence of glucosinolate that further enhances the health benefits of the oil. It also contains allyl isothiocyanate which have anti-inflammatory properties. In general, 100g of mustard oil composition has around 20%-28% oleic acid, 10%-12% linoleic acid, 9%-9.5% linolenic acid and 30%-40% erucic acid. Various studies published by NCBI reflect the importance of oleic acid, linolenic acid and linoleic acid.

Genetic diversity is a necessity for hybridization in crop development programmes. The use of a variety of parents aids in the isolation of superior recombinants. D^2 analysis and cluster analysis are two multivariate analysis approaches that have been reported. A greater understanding of the nature and magnitude of line diversity will assist in the selection of better parents for hybridization. Improved hybrids and excellent recombinants are expected to result from inter-crossing genetically diverse inbreds. Mahalanobis' D^2 statistic of multivariate analysis is recognized as a powerful tool in quantifying the degree of genetic divergence among the inbreds (Hemavathy *et al.*, 2006) [4].

In order to generate cultivars with higher yields, broader adaptation, desired traits, and pest and disease resistance, genetic divergence research is required. Introducing more diversified parents into the hybridization programme increases the probability of achieving maximum heterosis and provides a wide range of variability in segregating generations. Thus, this study was framed with the objective of studying the genetic divergence and clustering of different genotypes.

Material and Methods
The field experiment was carried out at oil seed research farm of Chandra Shekhar Azad University of Agriculture and Technology, Kanpur during *Rabi* season 2020-21.

The experimental site was located at 26.28° N latitude, 80.20° E longitude and about 126 meters above the sea level lying in the lower Ganges-Yamuna Doab at the bank of Ganges river. This place falls in the Central Plain zone of Agro-Ecological sub region (ICAR) and Upper Gangetic Plain Zone of Agro-Climatic zone (Planning Commission). The soil type of this site is deep, loamy with proper irrigation and drainage facility which is favorable for raising good crop.

The experimental germplasm material was sown on 18th of November 2020. The experiment was carried out in a randomized block design with 3 replications. The plot for each treatment has six rows of 5-meter length. The spacing between rows and plants was maintained 45 and 20 cm respectively. Also, the recommended dose of fertilizer 80:60:20:20 N:P:K:S kg/ha was applied for good plant growth. Recommended agronomic package and practices were followed to raise healthy and competitive plants population.

The material utilized in this experiment consist of 18 genotypes of *Brassica juncea* (L.) Czern and Coss which are KMRL-20-501, KMRL-20-502, KMRL-20-503, KMRL-20-504, KMRL-20-505, KMRL-20-506, KMRL-20-507, KMRL-20-508, KMRL-20-509, KMRL-20-510, KMRL-20-511, KMRL-20-512, KMRL-20-513, KMRL-20-514, KMRL-20-515, KMRL-20-516, VARDAAN and ASHIRVAAD. This material was obtained from the breeder of section of oilseed of the department of Genetics and Plant breeding, Chandra Shekhar Azad University of agriculture and technology, Kanpur.

Five competitive plants from each plot were randomly taken for recording observations for all the quantitative characters except days to flowering and days to maturity which were recorded on the plot basis. Oil content, methionine content, tryptophan content and protein content were estimated by using pre-caliberated near infrared reflectance spectroscopy (NIR, Dickey John Instalab 600) [1]. For studying genetic divergence among these genotypes Mahalanobis D^2 statistics was used and the clustering of D^2 values was done as elaborated by Murty and Arunachalam (1966) [9]. The genotypes were grouped into different clusters by following Tocher's method as described by Rao (1952) [10]. The intra and Inter cluster distances were calculated using the formula given by Singh and Chaudhary (1985).

Characters under observation

The data was collected from 5 competitive plants from each plot and data was recorded for the following characters

- Days to 50% flowering
- Days to maturity
- Plant height (cm)
- Number of primary branches/plant
- Number of secondary branches/plant
- Number of siliquae/plant
- Number of grains/siliqua
- Economic yield/plant (gm)
- Biological yield/plant (gm)
- Oil content (%)
- 1000 seed weight (gm)
- Protein content (%)
- Methionine content (%)
- Tryptophan content (%)

Results and Discussion

The analysis of variance revealed highly significant differences among all the genotypes for all 14 characters Wilk's criterion revealed significant difference among the genotypes for combined effect of 14 characters studied (Wilk's criterion, X^2 = 12190.894E-23 for 234 d. f.). Hence, the study was further extended to D^2 analysis.

All the 18 genotypes were grouped in to 5 clusters (Table 1). Out of them cluster II, III and IV had the maximum number of genotypes (5 each) while cluster I had 2 genotypes. There was only 1 genotype in cluster V. Lodhi *et al.* (2016) [6] also grouped ninety genotypes of Indian mustard in nine clusters while, Singh *et al.* (2010) divided 33 genotypes into nine clusters by Tocher's method.

The average intra and inter cluster distances are depicted in table 2. The maximum intra-cluster distance has been seen in cluster I (1562.48) followed by cluster II (1324.19), cluster IV (1182.09) and cluster III (1054.54). Cluster V did not show any intra-cluster distance because of single genotype in it (fig.-1). The inter-cluster D^2 values showed that the maximum inter-cluster distance was seen between cluster II and V (17143.86) followed by cluster IV and V (14716.91), cluster III and V (14505.84), cluster I and V (10286.65) & cluster I and IV (3561.21). The minimum inter-cluster distance value was found between cluster III and IV (1680.57) indicating that these two clusters were the least diverse. The genotypes grouped into identical cluster showed the lowest degree of divergence from each other, and in case crosses are made among such genotypes, no transgressive segregant can be obtained from such combinations. Therefore, hybridization programmes should constantly be planned in such manner that the parents belonging to distinct clusters with maximum divergence should be crossed to get suitable transgressive segregants and heterotic F_1s. The genotypes for hybridization should always be chosen from widely separated clusters (fig. 2), as it is always seen that there are multiple genotypes included in the crossing programme from widely separated clusters. Although, for final selection of the parents for breeding programme, the genotypes to be used may be selected almost without exception or its proven performance in the areas of intended use including quantitative characters and include in crossing with the existing varieties for their further improvement (Allard, 1960) [1].

Among all the characters, Number of secondary branches (39.22%) contributed the maximum towards genetic diversity followed by methionine content (20.26%) and 1000 seed weight (19.61%). Economic yield /plant, biological yield/plant, plant height, oil content, number of grains/siliqua and number of primary branches contributed 7.19%, 5.88%, 3.27%, 2.61%, 1.31% and 0.65% respectively. These results were in close confirmation with the findings of Vaishnav *et al.* (2006) [14] and Sutariya *et al.* (2011) [13].

Cluster I possessed high mean value for number of primary branches/plant (8.93), protein content (26.55); cluster II for plant height (179.61), number of secondary branches/plant (389.13). The cluster I genotypes were having maximum number of primary branches and high protein content which means the genotypes of this cluster can be selected for abovesaid characters. Cluster III showed high mean values for number of grains/siliqua (14.98) and tryptophan content (1.97%) while, cluster IV was found best for methionine

content (2.50%). Also, cluster V exhibited highest mean values for biological yield/plant (92.60), 1000 seed weight (0.533), oil content (39.43%) and economic yield/plant (27.48). Cluster V was also the best for days to 50% flowering (56.33) and days to maturity (121.00). Genotypes of cluster V had lesser life cycle and reproduction phase thus very suitable for early maturity selection. Gadi *et al.* (2020) [3], Kumar and Pandey (2013) [5] and Malviya *et al.* (2021) [8] also reported similar findings.

Table 1: Number of genotypes in each cluster

Clusters	Number of genotypes	Genotypes
I	2	KMRL-20-501, KMRL-20-508
II	5	KMRL-20-506, KMRL-20-515, KMRL-20-509, KMRL-20-511, KMRL-20-516
III	5	KMRL-20-502, ASHIRVAAD, KMRL-20-507, KMRL-20-514, KMRL-20-505
IV	5	KMRL-20-503, KMRL-20-504, KMRL-20-510, KMRL-20-513, VARDAAN
V	1	KMRL-20-512

Table 2: Cluster mean values of different characters

Cluster	Plant Height	Number of primary branches	Number of secondary branches	Number of siliquae/plant	Number of grains/ siliqua	Biological yield/ plant	1000 seed weight	Days to 50% flowering	Days to maturity	Protein content	Oil content	Methionine content	Tryptophan content	Economic yield/plant
I	169.16	8.93	16.26	332.13	14.63	88.50	0.402	61.66	122.16	26.55	36.33	2.16	1.95	25.44
II	179.61	8.04	19.37	389.13	14.93	81.76	0.279	64.26	122.60	26.29	36.80	2.36	1.87	19.56
III	174.78	7.54	15.02	297.02	14.98	63.34	0.266	60.86	125.40	26.01	36.24	2.06	1.97	16.97
IV	157.52	7.21	10.78	249.33	13.65	56.94	0.319	59.60	123.86	26.08	36.85	2.50	1.97	14.55
V	156.06	7.20	10.33	318.86	10.66	92.60	0.533	56.33	121.00	24.07	39.43	0.10	1.64	27.48

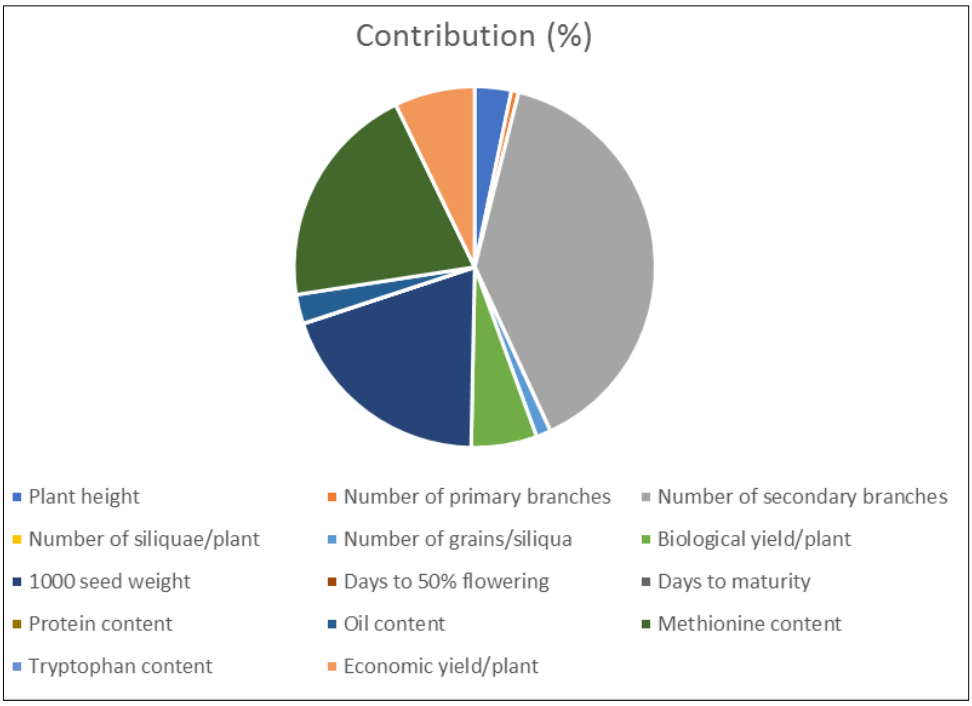
Table 3: Intra and Inter Cluster Distances

Clusters	I	II	III	IV	V
I	1562.48 (39.52)	2390.70 (48.89)	3076.66 (55.46)	3561.21 (59.67)	10286.65 (101.42)
II		1324.19 (36.38)	2059.25 (45.37)	3264.01 (57.13)	17143.86 (130.93)
III			1054.54 (32.47)	1680.57 (40.99)	14505.84 (120.44)
IV				1182.09 (34.38)	14716.91 (121.31)
V					0.00 (0.00)

Bold values show Intra cluster distances.
Normal values show Inter cluster distances.
Normal values are D^2 values while, values in bracket are D values.

Table 4: Percentage contribution of each character towards genetic divergence

S.no.	Source	Time Ranked 1st	Contribution (%)
1.	Plant height	5	3.27
2.	Number of primary branches	1	0.65
3.	Number of secondary branches	60	39.22
4.	Number of siliquae/plant	0	0.01
5.	Number of grains/siliqua	2	1.31
6.	Biological yield/plant	9	5.88
7.	1000 seed weight	30	19.61
8.	Days to 50% flowering	0	0.01
9.	Days to maturity	0	0.01
10.	Protein content	0	0.01
11.	Oil content	4	2.61
12.	Methionine content	31	20.26
13.	Tryptophan content	0	0.01
14.	Economic yield/plant	11	7.19



Graph 1: Graphical representation of percentage contribution of each character towards genetic divergence.

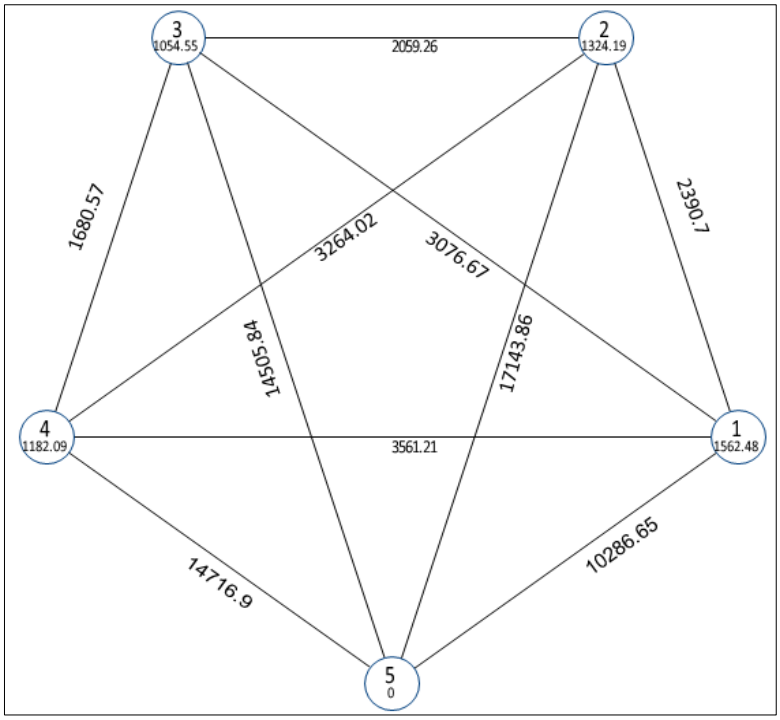


Fig 1: Euclidean cluster Distance diagram

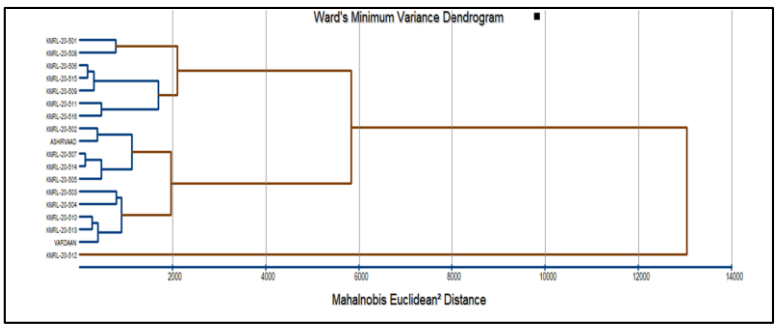


Fig 2: Euclidean average linkage dendrogram

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