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Genetic Divergence in Leafy Mustard (*Brassica juncea* var. rugosa) Germplasm Grown under *Tarai* Condition of Uttarakhand

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ABSTRACT

Background: Mustard represents a rich diversity and widely cultivated in 23 states and union territories of India. However, much of this diversity is concentrated in the Indo-Gangetic plains and the sub-mountain Himalayas. Genetic diversity plays a significant role in plant improvement because a hybrid between the lines of diverse origin usually display a greater heterosis than those between closely related ones which permit the selection of genetically divergent plants to obtain the desirable recombination of segregating generation. Therefore, the present study was undertaken to assess "Genetic Divergence in Leafy Mustard (*Brassica juncea*. var. rugosa) germplasm grown under *Tarai* condition of Uttarakhand" and to identify divergent parents for hybridization program, which would provide superior transgressive segregants from collected germplasm.

Methods: The present investigation consisted of thirty-two genotypes of leafy mustard and the research was carried out at Vegetable Research Centre (VRC), G.B. Pant University of Agriculture and Technology, Pantnagar, U.S. Nagar (Uttarakhand) in *rabi* season of 2015-2016. Mustard genotypes were sown in randomized block design with three replications in field and data were observed for seventeen quantitative and qualitative characters. The estimation of genetic divergence was done with the help of Mahalonobis D^2 statistic as suggested by Rao (1952). Cluster analysis by Tocher method for all the traits was done.

Result: Thirty two germplasm of leafy mustard for different characters and grouped them into six clusters using Mahalanobis D^2 statistic. The analysis revealed the maximum inter cluster distance was (20534.12) between cluster V and cluster VI so, we can create variation by inter mating genotypes from these two clusters to each other and the maximum intra cluster distance in cluster III (441.91) with six germplasm. It means we can intermate genotypes of this cluster with each other (2014/MGVAR-2, FS-13-1, FS-13-4, 2014/MGVAR-4, PRHC-12-9-1, PRHC-12-7-2, FS-13-3 and Pusa Sag 1) to create variation in next generations. The clustering pattern could be utilized in selection of parents for crossing and deciding the best cross combinations which may generate the highest possible variability for various traits.

Key words: Cluster, D^2 statistic, Genetic divergence, Mustard.

INTRODUCTION

Mustard [*Brassica juncea* (L.) Czern.] is an important crop of the *Brassicaceae* family. It is widely cultivated in Asia and Europe (Chen and Chen, 1992). In India, a great diversity exists in *B. juncea* for plant type, seed size, siliqua length, oil content and maturity period (Rana and Singh, 1992; Misra, 2012; Semwal *et al*., 2013). It is mainly self-pollinated, although an average of 7.5-30% out-crossing does occur under natural conditions (Rabbani *et al.,* 1998).

Improvement in crop plants depends upon the magnitude of genetic variability in different quantitative characters (Gupta *et al*., 2015). A breeding program may rightly be formulated on the basis of available information on the extent of genetic diversity (Chauhan *et al*., 2008; Singh *et al*., 2013). *Brassicas* represent a rich diversity, which are cultivated in 23 states and union territories of India (Misra and Kumar, 2008). However, much of this diversity is concentrated in the Indo-Gangetic plains and the sub-mountain Himalayas. Genetic diversity plays a significant role in plant improvement because a hybrid between the lines of diverse origin usually display a greater heterosis than those between closely related ones (Naznin *et al.,* 2015) which permit the selection of genetically divergent plants to obtain the desirable recombination of segregating generation.

The extent of diversity present in the germplasm determines the limit of selection for improvement. The characters

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of economic importance are generally quantitative in nature and exhibit considerable degree of interaction with the environment. Mahalanobis D^2 statistic is a morphometric method and a powerful tool in quantifying the degree of divergence at genotypic level.

Gene banks harbour scanty germplasm collection of *Brassica juncea var. Rugosa* and a very little background information about its genetic diversity has hampered its improvement. Improvement of the crop depends upon the magnitude of genetic diversity of economic characters. Therefore, the measurement and evaluation of genetic diversity in desired direction become extremely important in any crop improvement programme. The extent of genetic diversity in a specific breeding population depends upon the genotype included in it. Therefore, the present study was undertaken on genetic divergence in leafy mustard (*Brassica juncea* var. *rugosa*) germplasm grown under *Tarai* condition of Uttarakhand" to assess the extent of variability in collected germplasm accessions and to identify divergent parents for hybridization program.

MATERIALS AND METHODS

The present investigation was carried out at Vegetable Research Centre (VRC), G.B. Pant University of Agriculture and Technology, Pantnagar, U.S. Nagar (Uttarakhand) in *rabi* season of 2015-2016. The altitude of the experimental site is geographically situated at an altitude of 243.83 meters above mean sea level and at 29°N latitude and 79.3°E longitudes. This falls in the humid subtropical zone and situated in the *Tarai* belt of North India in the foothills of *Shivalik* hills of the great *Himalayan* range.

The experimental material consisted of 32 leafy mustard genotypes out of which 27 were obtained from the Pantnagar Centre for Plant Genetic Resources of GBPUAT and 4 genotypes from IET, AVT and 1 prominent check, PUSA SAG-1 was received from IARI. The crop was raised in the field in randomized block design with three replications and one check. The recommended package of practices of cultivation was followed. Observations were recorded on five randomly selected plants from each of the plot for twelve characters. Genetic diversity between clusters based on correlation matrix was computed based on multivariate analysis using Mahalanobis D^2 statistic (Mahalanobis, 1936). The important traits in each principal component that significantly contributed to the variation observed were identified as suggested by Johonson and Wichern (1988). Based on the squared distances (D^2), clustering of genotypes was done using Tocher's method as described by Singh and Chaudhary (1999). Squared distance (D^2) for each pair of clusters combinations was computed as per (Singh and Chaudhary, 1999).

RESULTS AND DISCUSSION

The genetic divergence was estimated by Mahalanobis D^2 statistic. Rao (1952) suggested the application of D^2 statistics for the assessment of genetic diversity in plant breeding. Thirty two germplasm of leafy mustard for different characters and grouped them into six clusters using Mahalanobis D^2 statistics (Table 1), whereby different members within a cluster is being assumed to be more closely related in terms of the trait under consideration with each other than those members in different clusters. Among six clusters, cluster I consisted of 9 genotypes, followed by cluster II (8), cluster IV (7) and III (6). Clusters V and cluster VI were mono genotypic. Similarly 60 diverse genotypes of Indian mustard were grouped into eight clusters by Meena *et al*. (2020). The pattern of clustering of these germplasm is shown in Table 1. Cluster I comprised of 9 germplasm *viz*, 2014/MGVAR-1, FS-13-12, FS-13-11, FS-13-5, FS-13-21, PRHC-12-14, EEC-10, FS-13-20 and FS-13-7. Cluster II comprised of eight germplasm, namely PRHC-12-9-2, FS-13-9, PRHC-12-12, FS-13-10, FS-13-8, FS-13-17, FS-13-2 and 2014/MGVAR-3. Cluster III includes six germplasm, 2014/MGVAR-2, FS-13-1, FS-13-4, 2014/MGVAR-4, PRHC-12-9-1 and PRHC-12-7-2. Cluster IV comprised of seven germplasm, FS-13-14, PRHC-12-13, PRHC-12-11, PRHC-12-6, FS-13-15, FS-13-13 and FS-13-18. Cluster V consists of one germplasm, FS-13-3. Cluster VI comprised of only one germplasm, Pusa Sag 1. Rabbani *et al.* (1998) also placed 52 *Brassica juncea* accessions into six clusters while Gupta *et al*. (1991) grouped 48 lines of *Brassica juncea* into five clusters. Rout *et al.* (2019) grouped 71 genotypes of Indian mustard in seven clusters.

Most of the germplasm (28.12%) remained in the first cluster and remaining cluster had 1 to 8 germplasm. The presence of higher number of germplasm in the first cluster indicates presence of lower divergence among the germplasm of that cluster. The experimental material as a whole was found to be more diverse, as the 32 germplasm formed six clusters.

Intra-cluster and inter-cluster average D^2 values and their respective genetic distance (D) have been given in

Table 1: Clustering pattern of 32 germplasm on the basis of D^2 values.

Cluster no.	Germplasm included	No. of germplasm
I	2014/MGVAR-1, FS-13-12, FS-13-11, FS-13-5, FS-13-21, PRHC-12-14, EEC-10, FS-13-20, FS-13-7	9
II	PRHC-12-9-2, FS-13-9, PRHC-12-12, FS-13-10, FS-13-8, FS-13-17, FS-13-2, 2014/MGVAR-3	8
III	2014/MGVAR-2, FS-13-1, FS-13-4, 2014/MGVAR-4, PRHC-12-9-1, PRHC-12-7-2.	6
IV	FS-13-14, PRHC-12-13, PRHC-12-11, PRHC-12-6, FS-13-15, FS-13-13, FS-13-18	7
V	FS-13-3	1
VI	Pusa Sag 1	1

Table 2 and 3 respectively. Intra-cluster average D^2 values ranged from 0.00 to 441.91. It was maximum in cluster III (441.91) followed by cluster II (339.99), Cluster I (295.97) and cluster IV (0.00). There were also two single germplasm containing clusters having 'zero' intra-cluster distance which is in accordance with finding of Abdisa (2014).

The inter-cluster average D^2 value was maximum (20534.12) between cluster V with one germplasm and cluster VI with one germplasm, followed by average D^2 value of 19421.68 between clusters II and cluster VI (Fig 1).

The minimum inter-cluster average D^2 value of 497.11 was found between cluster I with nine germplasm and Cluster IV with seven germplasm. The magnitude of heterosis largely depends on the extent of genetic diversity in the parental population. Hence, the germplasm belonging to cluster V and VI could be used in hybridization for crop improvement programme. The crosses between genotypes of these diverse clusters might be having more chances of getting desirable segregants (Arunachalam *et al.*, 1984).Whereas Gupta *et al.* (1991) reported average intra-cluster values ranged from a minimum of 13.8 in cluster V to a maximum of 21.9 in cluster III. Inter-cluster values also ranged from as low as 29.7 between clusters IV and V to as high as 81.3 between clusters I and V. Singh *et al.* (2013) from his experimental results reported that the maximum inter-cluster distance was observed between cluster II and IX followed by IV and IX. The genotypes grouped into same cluster displayed the lowest degree of divergence from one another. Therefore, hybridization programmers should always be formulated in such a way that the parents belonging to different clusters with maximum divergence could be utilized to get desirable transgressive segregants. The minimum genetic distance D=22.29 was found between cluster I and IV.

A perusal of these cluster means (Table 4) for different characters indicated considerable distance among clusters for all the characters.

Cluster III with six germplasm had the highest cluster mean value for leaves per plant at 30DAS (5.05). For leaves per plant at 45 DAS, cluster I and VI had the highest mean value (7.00) while cluster V had lowest mean value (6.07). For days to first leaves harvest, highest cluster mean value was shown by cluster VI (50.00) while the lowest was shown by cluster IV (44.00). For days to last leaves harvest, Cluster IV had shown the highest cluster mean value (73.33) while lowest cluster mean value (69.39) was shown by cluster II. Cluster III had shown highest cluster mean value for leaf area (492.76 cm^2) while the lowest cluster mean value was shown by cluster V (361.67 cm^2). For leaf length, cluster III had the highest mean value (51.74 cm) followed by cluster V(49.51 cm) while the cluster VI had lowest mean value (32.48 cm). For leaf width highest cluster mean value (21.66) had shown by cluster III while cluster IV had the lowest (18.21) mean value.

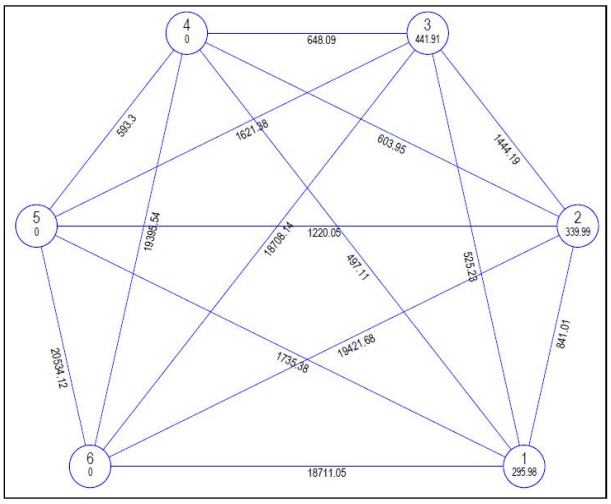

Fig 1: Average intra (in circle) and inter (between circle) cluster D^2 values in 32 genotypes of mustard.

Table 2: Average inter and intra cluster D^2 values.

Clusters	I	II	III	IV	V	VI
I	295.976	841.006	525.229	497.111	1735.381	18711.051
II		339.993	1444.188	603.950	1220.053	19421.680
III			441.910	648.095	1621.381	18708.145
IV				0	593.300	19395.535
V					0	20534.121
VI						0

Table 3: Average intra and inter cluster D^2 values.

Clusters	I	II	III	IV	V	VI
I	17.203	29.000	22.917	22.295	41.657	136.788
II		18.438	38.002	24.575	34.929	139.361
III			21.021	25.457	40.266	136.777
IV				0	24.357	139.267
V					0	143.297
VI						0

Table 4: Cluster mean for seventeen characters.

Cluster	Leaves per plant at 30 DAS	Leaves per plant at 45 DAS	Days to first leaf harvest	Days to last leaf harvest	Leaf area (cm^2)	Leaf length (cm)	Leaf width (cm)	Petiole length (cm)	Green leaf yield/ plant (g)	Green leaf yield/plot (Q/ha)	Protein content (%)	Phos-phorous content (mg/100 g)	Sulphur content (mg/100 g)	Pota - ssium content (mg/100 g)	Iron content (mg/ 100 g)	Manga-nese content (mg/100 g)	Zinc content (mg/ 100 g)
I	4.97	7.00	44.47	71.29	485.11	48.12	19.99	8.50	216.40	287.81	24.83	1183.93	861.67	2360.67	13.77	5.35	4.20
II	4.97	6.98	46.00	69.39	405.01	45.21	18.66	8.33	189.02	251.40	25.55	1187.83	1986.67	2418.33	13.62	4.62	3.80
III	5.05	6.90	44.92	70.96	492.76	51.74	21.66	9.30	222.73	296.24	26.17	1224.75	535.00	2313.75	21.36	5.24	4.99
IV	4.35	6.47	43.00	73.33	417.63	42.78	18.21	5.90	174.07	231.51	22.28	1173.00	1400.00	2080.00	24.83	6.04	3.58
V	4.73	6.07	44.00	70.67	361.67	49.51	19.85	10.57	180.47	240.02	21.43	1328.00	2030.00	2320.00	37.52	4.91	4.64
VI	5.00	7.00	50.00	70.00	429.37	32.48	18.55	6.97	278.87	370.90	26.62	1140.00	1550.00	2800.00	21.58	4.63	4.63

Cluster V had shown highest cluster mean (10.57 cm) for petiole length while cluster IV had shown the lowest mean value (5.90 cm). For green leaf yield per plant, cluster VI had shown the highest cluster mean (278.87 g) while cluster V had the lowest mean value (180.47 g).

For green leaf yield q/ha, cluster VI had the highest mean value (370.90 q/ha) while cluster IV had lowest mean value (231.51 q/ha). The results are in agreement with findings of Rout *et al.* (2019) in mustard.

Rabbani *et al.* (1998) found 6 clusters in mustard as in present study. I cluster was characterized by small sized leaves and lower number of leaves per plant. The second cluster accessions displayed large sized leaves at seedling. Cluster 3 consisted of three oilseed cultivars. The fourth cluster comprised of only two accessions. Cluster 5 exhibited tall stature, large sized leaves and longer duration. Finally, cluster 6 had large sized leaves and the second cluster accessions showed a lower number of leaves/plant and early maturity. On the other hand Zada *et al.* (2013) reported 7 clusters having first cluster medium number of leaves per plant, cluster two had lower protein contents, cluster three had medium contents of protein, cluster four had minimum ratio between leaf length and width and protein contents. Cluster five had minimum leaf length and protein contents and cluster six had maximum leaves/plant and medium protein contents and cluster seven had maximum protein contents but in this study highest cluster mean value for protein content (26.62) was found in Cluster VI.

CONCLUSION

The analysis revealed the maximum inter cluster distance was between cluster V (FS-13-3) and cluster VI (Pant Sag -1) and the maximum intra cluster distance in cluster III. Degree of genetic divergence would help the plant breeder in selecting the particular parents for hybridization. Apart from the high degree of divergence, the mean performance of genotypes and the characters with maximum contribution towards divergences should also be given due consideration. The best combination of parents for improvement in various economic characters can be recommended on the basis of per se performance of the genotypes and inter-cluster divergence. Based on the maximum intra cluster distance value, the crosses could be made among the genotypes having the highest divergence like 2014/MGVAR-2, FS-13-1, FS-13-4, 2014/MGVAR-4, PRHC-12-9-1, PRHC-12-7-2, FS-13-3 and Pusa Sag 1 from various clusters like III, V and VI to get desirable transgressive segregants.

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