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Genetic divergence studies for seed yield and component traits in brown sarson (*Brassica campestris* var. brown sarson) under northwestern Himalayas

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Abstract
In order to assess the genetic diversity, an experiment was conducted with 26 brown sarson genotypes grown in randomized complete block design with three replications. The genotypes could be grouped into 6 clusters namely cluster I (5 genotypes), cluster II (11 genotypes), cluster III (5 genotypes), cluster IV (3 genotypes), cluster V (1 genotype) and cluster VI (1 genotype). This indicates that the genotypes grouped within a particular cluster are more or less genetically similar to each other and apparent wide diversity is mainly due to the remaining genotype distributed over rest of the other clusters. The maximum intra cluster distance exhibited for cluster III. Maximum inter-cluster distance was observed between clusters IV and V followed by between clusters II and V and clusters IV and VI. Cluster II showed the highest number of siliquae per plant which suggested that the genotype falling in this cluster can be selected directly and used in hybridization programme. The genotypes falling under clusters III and V can be used as a source population for early flowering and better yield based upon cluster means.

Keywords: Genetic divergence, D^2 statistic, cluster distance, genotypes, *brassica*

Introduction
Brassica species, commonly called as rapeseed-mustard, are the third most important oilseed crops of the world after soybean and palm. The species *Brassica campestris* var. brown sarson (2n=2x=20; genome AA) is one progenitor of the both amphidiploids *Brassica juncea* L. (2n=4x=36; genome AABB) and *Brassica napus* L. (2n=4x=38; genome AACC). It is one of the important winter oilseed crop cultivated in a wide range of agro-climatic conditions. It is also referred to as toria, sarson, summer turnip rape, polish rape, and so on. Brown sarson is an often cross-pollinated crop and belongs to the family *Brassicaceae* and believed to have been widely distributed with secondary centers of diversity in Europe and Central Asia.
Brown sarson is primarily used for human consumption as an edible oil and serve as an important raw material for the manufacture of soaps, paints and varnishes, hair oils, lubricants, textiles, auxiliaries and pharmaceuticals. In world, brown sarson occupies an area of 33.58 million hectares with a total annual production of 67.76 million tones. In India, the crop occupies an area of 6.50 million hectares with a total production of 6.80 million tonnes (Anonymous 2016) [1].
Genetic diversity plays an important role in plant breeding because hybrids between lines of diverse origin generally display a greater heterosis than those between closely related strains (Singh, 1986) [10] which permits to select the genetically divergent parents to obtain the desirable recombination in the segregating generations (Uddin and Chowdhury, 1994) [11]. Estimates of genetic divergence provide the extent of diversity existed within the available germplasm and moreover, evaluation of genetic diversity is important to know the sources of genes for a particular trait. Keeping this in view, the present investigation was carried out to assess the nature and magnitude of genetic diversity in brown sarson which would help in selection of efficient genotypes with desirable traits for utilization in hybridization programme.

Materials and methods
1. Plant material and experimental site
A total of 26 genotypes of *Brassica campestris* were used in present study. All the genotypes, including 2 checks viz., KBS-3 and HPBS-1 were raised at the experimental farm area of Department of Crop Improvement, Chaudhary Sarwan Kumar Himachal Pradesh Krishi Vishvavidyalaya, Palampur (H.P.) which is situated at an elevation of 1290.8 m above mean sea level with latitude 32°6´ N and 76°3´ E longitude represents the mid hill zone of Himachal Pradesh

(Zone-II) during winter 2015. The soils are clay loam to silty clay loam in texture. The reaction of soil is acidic with pH ranging from 5.0 to 5.6.

2. Design and layout

Experiment layout was a randomized complete block design with three replications in the plot size of 2.0 × 0.9 m^2. Each genotype was raised in a plot consisting of 2 rows with spacings of 30cm between rows and 15cm between the plants. Irrigation was given whenever required and regular weeding was done to keep the trial free from weeds.

3. Observations

Five plants per genotype per replication were randomly selected for recording the observations at appropriate stages of crop growth on characters such as plant height (cm), reproductive phase (days), number of primary branches per plant, number of secondary branches per plant, siliquae per plant, seeds per siliqua, 1000-seed weight (g) seed yield per plant (g), biological yield per plant (g) and harvest index (%). The observations on days to flower initiation, days to 50 per cent flowering and days to 75 per cent maturity were recorded on plot basis.

4. Statistical analysis

The mean data over randomly selected plants from all the replications were subjected to the statistical analysis. Genetic diversity was studied using D^2 statistic as per Mahalanobis, 1936 [5]. The genotypes were grouped into different clusters per Tocher's method as suggested by Rao, 1952 [8]. Intra- and inter-cluster distances and cluster means for different characters were also computed.

Results and discussion

1. Analysis of variance

The analysis of variance showed that there were significant differences among genotypes for all traits. This indicates the existence of considerable genetic variability for selection and breeding.

2. Cluster analysis

Assessment of genetic divergence helps in reducing the number of breeding lines to be maintained and the progenies derived from diverse parents are expected to show a broad spectrum of genetic variability and provide a greater scope for isolating superior segregants. D^2-statistic is a powerful tool for measuring genetic diversity which helps in categorizing genotypes into different groups based on the difference in the character expression. The D^2 statistic resulted all 26 genotypes (including 2 checks) were grouped into six clusters which indicated the presence of adequate genetic diversity among the tested genotypes. Cluster II was largest consisting of eleven genotypes *viz.*, HPBS-1 x 03-472, 03-472, 03-473 x 02-KLM-6-1, HPBS-1 x 03-473, KBS-3, 03-472 x 02-KLM-6, HPKM-04-1 x 02-KLM-6, 03-473, HPKM-04-1, KDH-BS-6 x 03-472 and HPBS-1 x Geeta-1 followed by Cluster I and Cluster III with five genotypes. Cluster IV contained three genotypes *viz.*, 03-473 x 03-472, HPBS-1 x Heera-1 and 03-473 x 02-KLM-6. Clusters V and VI contained one genotype each *viz.*, 02-KLM-6 and KBS-3 x HPKM-04-1, respectively. (Fig1; Table 1). It was concluded that the genotypes within the same clusters were originated from different geographical regions of the country which indicated that the geographical distribution and genetic divergence did not follow the parallelism which might be due to the continuous exchange of brown sarson genetic material among different geographical regions. The results are in confirmatory with the earlier findings of Jahan *et al*. (2013) [3], Shekhawat *et al*. (2014) [9], Neeru *et al*. (2015) [7] and Mohan *et al*. (2017) [6].

Table 1: Distribution of brown sarson genotypes among different clusters on the basis of Mahalanobis D^2 statistic

Clusters	Number of Genotypes	Genotypes
I	5	KBS-3 x HPBS-1, KBS-3 x 03-472, HPKM-04-1 x KDH-BS-6, HPBS-1, HPBS-1 x 02-KLM-6
II	11	HPBS-1 x 03-472, 03-472, 03-473 x 02-KLM-6-1, HPBS-1 x 03-473, KBS-3, 03-472 x 02-KLM-6, HPKM-04-1 x 02-KLM-6, 03-473, HPKM-04-1, KDH-BS-6 x 03-472, HPBS-1 x Geeta-1
III	5	KDH-BS-6 x 03-473, HPKM-04-1 x KDH-BS-6-1, HPBS-1 x HPKM-04-1, KDH-BS-6, KDH-BS-6 x 02-KLM-6
IV	3	03-473 x 03-472, HPBS-1 x Heera-1, 03-473 x 02-KLM-6
V	1	02-KLM-6
VI	1	KBS-3 x HPKM-04-1

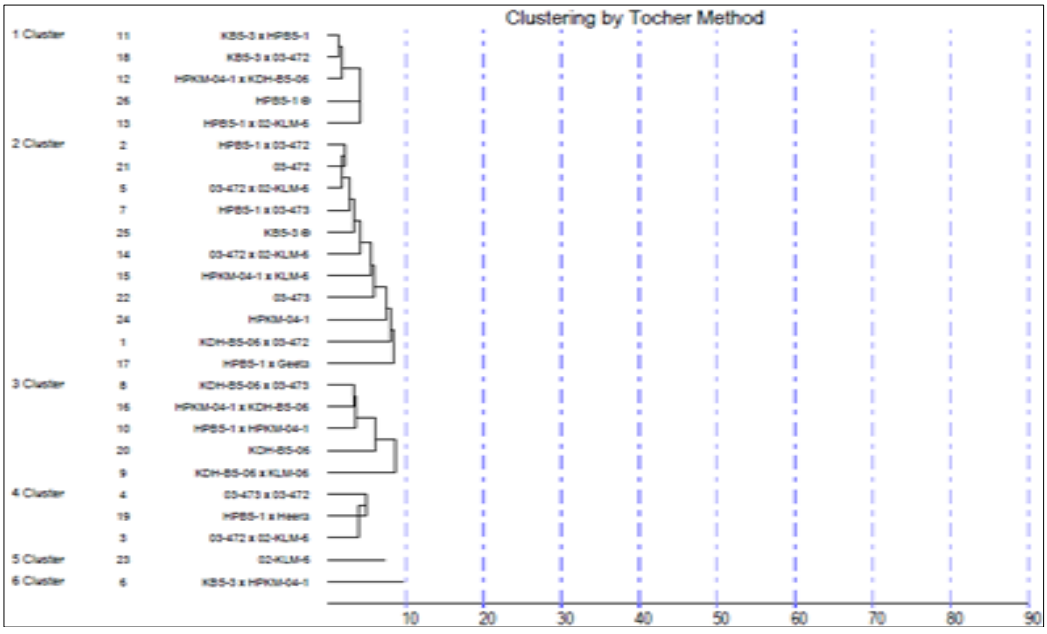

Fig 1: Dendrogram showing diversity of brown sarson genotypes generated using Mahalanobis D^2-cluster analysis (Tocher's method)

3. Estimation of intra and inter cluster square distances (D^2)

The highest average intra-cluster distance √D2 was observed in cluster III (4.66) followed by cluster II (4.34) and cluster IV (3.83). Highest inter-cluster distance was observed between clusters IV and V (11.10) followed by between clusters II and IV (8.87) and clusters IV and VI (8.42). However, the distance between clusters I and III was minimum (5.51) indicating that the genotypes belonging to these clusters were comparatively less diverse (Table 2). Kumari and Kumari (2018) [4] observed the highest inter-cluster distance was observed between clusters III and V (3.41) followed by distance between clusters V and VI (3.36) and clusters II and V (3.14). Dilip *et al*. (2016) [2] reported cluster IV (1553.53) exhibited the maximum intra cluster distance and the maximum inter cluster distance was revealed between cluster VI and III (15909.55) whereas, minimum between clusters IV and III (1917.82). The maximum contribution towards the genetic divergence was exhibited by seeds per siliqua (32.92 %) followed by number of secondary branches per plant (24.62 %) and siliquae/plant (19.68) while the remaining characters contributes <10 per cent to the overall genetic divergence present in the brown sarson genotypes studied (Table 3). Earlier, Zaman *et al*. (2010) [12] observed maximum contribution by days to 50% flowering (81.94%), days to maturity (8.24%), plant height (5.82%), branches per plant (1.91%) and siliqua per plant (1.17%) towards genetic divergence in *brassica* sp.

Table 2: Average intra- and inter-cluster values of D^2 and $\sqrt{D^2}$ among different clusters

Clusters	I	II	III	IV	V	VI
I	11.61 (3.41)	34.38 (5.86)	30.38 (5.51)	50.71 (7.12)	43.14 (6.57)	34.47 (5.87)
II		18.83 (4.34)	35.34 (5.94)	78.74 (8.87)	38.66 (6.22)	54.94 (7.41)
III			21.67 (4.66)	50.32 (7.09)	53.15 (7.29)	58.55 (7.65)
IV				14.69 (3.83)	123.32 (11.10)	70.87 (8.42)
V					0.00 (0.00)	34.01 (5.83)
VI						0.00 (0.00)

Values in bold letters are intra-cluster distances, Values in parentheses are $\sqrt{D^2}$ = D values

Table 3: % Contribution of individual characters in creating diversity in brown sarson

S. No.	Character	Contribution %
1	Days to flower initiation	1.23
2	Days to 50% flowering	0.62
3	Days to 75% maturity	0.62
4	Reproductive phase	0.01*
5	Plant height	9.20
6	Number of primary branches / plant	4.62
7	Number of secondary branches/ plant	24.62
8	Siliquae / plant	19.68
9	Seeds / siliqua	32.92**
10	1000-seed weight	0.02
11	Biological yield / plant	2.77
12	Seed yield / plant	3.66
13	Harvest index	0.03

** Maximum contribution; * Minimum contribution

4. Cluster mean analysis

The cluster means of brown sarson genotypes falling under different clusters are presented in Table 4. Among six clusters, cluster VI characterterized by maximum 1000 seed weight, seed yield / plant and harvest index with genotype KBS-3 x HPKM-04-1 and genotypes constituting cluster II which exhibited maximum for siliqua per plant and biological yield / plant which suggested that the genotypes falling in these clusters can be selected directly on the basis of these traits and used in hybridization programme. However, clusters III and IV showed the maximum cluster means for plant height and number of primary branches/ plant and number of secondary branches/ plant and number of seed/ siliqua, respectively. Besides, these clusters also showed moderate mean values for other important characters.

Genotype falling in cluster V (02-KLM-6) showed the desirable mean value for days to flower initiation and days to 50 per cent flowering (early). The genotypes falling under cluster V and VI can be used as a source material for increasing yield coupled with early flowering.

Table 4: Cluster means for different traits in brown sarson

Clusters Characters	I	II	III	IV	V	VI	Mean	Minimum	Maximum
Days to flower initiation	53.40	54.88**	52.80	53.56	52.67*	53.33	53.44	52.67	54.88
Days to 50% flowering	66.93**	66.03	64.93	65.56	60.67*	65.67	64.96	60.67	66.93
Days to 75% maturity	142.93*	143.18	144.07	144.22	147.00**	144.67	144.35	142.93	147.00
Reproductive phase	76.00*	77.15	79.13	78.67	86.33**	79.00	79.38	76.00	86.33
Plant height (cm)	115.58*	121.66	129.85**	129.62	117.40	120.53	122.44	115.58	129.85
No. of primary branches/ plant	5.45	5.81	6.28**	4.94	4.87	4.53*	5.31	4.53	6.28
No. of secondary branches/ plant	6.20	4.50	4.53	7.27**	4.33*	6.50	5.56	4.33	7.27
Siliquae / plant	111.07*	145.53**	120.87	128.69	115.20	116.53	122.98	111.07	145.53
Seeds/ siliqua	12.89	13.33	16.08	17.49**	10.47	9.83*	13.35	9.83	17.49
1000- seed weight (g)	3.03	3.10	2.81*	3.09	3.00	3.63**	3.11	2.81	3.63
Biological yield / plant (g)	30.27	34.21**	30.64	33.67	22.00*	24.00	29.13	22.00	34.21
Seed yield / plant	4.67*	4.91	5.27	6.49	4.87	6.53**	5.45	4.67	6.53
Harvest index (%)	15.65	14.56*	17.45	19.51	22.70	27.47**	19.55	14.56	27.47

**Maximum; *Minimum

Conclusion

It can be concluded from present study that considerable genetic variability was found in the present material. All genotypes under study were grouped into 10 clusters which indicated the presence of sufficient diversity among the germplasm lines. The clustering revealed that there was no

correlation between geographical diversity and genetic divergence. The genetic divergence had little to do with the geographic factor as noticed by the random distribution of genotypes into various clusters. The clusters II, III, V and IX exhibited large inter-cluster distances thus, indicating more chances of developing good segregants by crossing the genotypes of the these clusters. Himso-14-132 falling under Cluster IV showed the higher cluster means for plant height and number of pods per plant which suggested that it can be selected directly on the basis of these traits and used in hybridization programme. Three genotypes such as Himso-14-126A, PS-1347 and Himso-14-145 of cluster V and Himso-14-19 of cluster VII exhibited the genes for higher seed yield potential and early flowering. Therefore these genotypes can be used as source population for higher seed yield coupled with earliness.

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