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Research Article

Genetic diversity analysis using agro-morphological traits of *Brassica juncea* subspecies rugosa (Pahari rai) from North-Eastern Himalayan region

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Abstract
Twenty genotypes of *Brassica juncea* subspecies rugosa were grown in a randomized block design (RBD) for assessment of diversity using Mahalanobis D^2 and Principal component statistics. Genetic diversity plays an important role in plant breeding because hybrids between parents of diverse origin generally displayed greater heterosis than those between closely related. Geographical diversity may not be the only factor responsible for causing diversification, thus focus should be laid on selecting the parents based on genetic diversity rather than geographical diversity. The Eigenvalue of the first principal component was accounted for 7.46 of variation. The first six principal components axes explained 88.79 per cent of the variation, suggesting considerable diversity among the genotypes for all the characters on the basis of D^2 analysis the genotypes were grouped into four clusters. Based on cluster analysis the results revealed maximum inter-cluster distance (41.68) between cluster II and IV followed by cluster I and IV, thereby paving the chance for them to be used in hybridization breeding programs. As per the biplot, the most stable genotypes are IC-399826, IC-338751, IC-338535, and IC-399839. The outcome of the study indicated that the genotypes having greater genetic diversity will be useful for developing segregants by utilizing in crossing programmes.

Keywords: Pahari rai, D^2 Mahalanobis, Genetic diversity, Principal component analysis

INTRODUCTION

Brassica juncea belongs to the genus Brassica, and it is a cardinal member of the cruciferous family. It is an allotetraploid species containing genomes of *Brassica rapa* (AA, n=10) and *Brassica nigra* (BB, n=8). It exhibits numerous frugal species which produce edible leaves, stems, flowers, buds, roots, and seeds. Various species are utilized as oil crops, while others are exploited as condiments. It is commonly known as rai, raya or laha. It is the foremost oil seed crop in the world with global production of around 72 million tons (Adhikari *et al.,* 2021). *B. juncea* is primarily cultivated for its oil all across the country. *Brassica juncea* subspecies rugosa, a leafy type variant of *Brassica juncea*, is predominantly grown for vegetable purposes. *Brassica juncea* variety rugosa belongs to the family Brassicaceae which is a well-known leafy vegetable and laipatta. Its leaf foliage varies from light green to dark purple in color. It is consumed as a leafy vegetable in American and Asian countries especially in India, China and Nepal. All three Asian countries are the main cultivating regions for this leafy vegetable and are also the leading suppliers. In India, it is mainly cultivated in hilly regions of Uttarakhand, Meghalaya, Mizoram, Assam, Nagaland, Manipur, Arunachal Pradesh, and West Bengal. It is consumable in raw form as salads when its leaves are tender and soft. It is utilized for the preparation of traditional popular saag during the winter season in northern India. In hilly regions, farmers usually grow it in their backyard in small patches for daily consumption. The growth of the leafy vegetable is slow and requires a low temperature for the splendid growth of the leaf. Leaves are broad in size, crumpled, crunchy, and peppery. Like any other leafy vegetable, it contains several phytonutrients for health enhancement and disease preventive properties. It contains numerous nutrients such as protein, iron, calcium, magnesium, manganese, potassium, and also including Vitamins A and E (Pant *et al.,* 2020). Potential consumers who imbibe green mustard as a vegetable on a routine basis are protected from iron deficiency, osteoporosis, and a variety of cardiovascular disorders. They also aid in the treatment of arthritis, asthma, and nervous system diseases (Macready *et al.,* 2014). This leafy vegetable is also employed to make a fermented pickle product known as 'Gundruk', which is well-known and widely enjoyed in Nepali dishes (Bhattarai *et al.,* 2018). Because of its easy availability, it can contribute as a low-cost source of critical nutrients for those in need, particularly women and developing children. With the foregoing in mind, germplasm lines were collected from different hilly regions of the different states of India, and morphological characterization was done to identify suitable lines to boost people's nutritional status, well-being, and breeding purposes. Plant diversity analysis conducted during the study also helps the breeder to identify suitable lines for crop improvement programmes.

MATERIALS AND METHODS

The experiment was conducted at Norma E. Borlaug, Crop Research Centre (C.R.C) of Govind Ballabh Pant University of Agriculture and Technology, Pantnagar during the *rabi* season 2019-2020 and 2020-2021. The experimental material comprises of 20 genotypes of *Brassica juncea* variety rugosa from a diverse location along with two checks. The collected genotypes were from the states of Uttarakhand, Assam, Meghalaya, Mizoram, Nagaland, Arunachal Pradesh, Meghalaya, Himachal Pradesh, Tripura, and Uttar Pradesh. The experimental material was sown in randomized block design with three replications along with two checks, with a spacing of 60 x 15 cm. Observations were recorded for twenty-one quantitative characters *viz*., silique bearing angle, plant height (cm), point to first branch (cm), the number of primary branches, the number of secondary branches, the number of siliquae on the main shoot, siliquae length (cm), the number of seeds per siliquae, 1000 seed weight (g), seed yield per plant, oil content (%), leaf length (cm), leaf blade length (cm), lamina width (cm), leaf index, leaf petiole length (cm), leaf area (cm^2), days of flowering (initiation, 50%, 100%). The data were subjected to principal component analysis, which is a multivariate technique, developed by Hotelling (1933) after its original concept was given by Pearson (1901). D^2 analysis is a measure of the distance between a point P and a distribution D, introduced by Mahalanobis (1936). FactoMineR was used to estimate principal component analysis, and correlations using the corrected mean values of all quantitative characteristics (Lê *et al.,* 2008). The Tocher's method from the cluster package was used to determine the optimum number of clusters.

RESULTS AND DISCUSSION

Table 1 depicted the genotypic coefficient of variation and phenotypic coefficient of variation was high for 1000 seed weight and seed yield per plant and moderate for flowering initiation, 50% flowering, 100% flowering and leaf petiole length, while the low for siliqua bearing angle, plant height, the number of primary branches, siliquae on the main shoot, siliqua length, oil content, and, leaf index. The heritability was found steep (>70%) for traits point to first branching, the number of secondary branches, main shoot length, the number of seeds per siliquae, 1000 seed weight, seed yield per plant, leaf length, leaf width, leaf petiole length, Flowering initiation 50% flowering and 100% flowering. A minor difference in genotypic coefficient of variation estimates compared to phenotypic coefficient of variation implies that observed variance is mostly attributable to genetic causes, whereas a significant difference reveals the impact of the environment. The genetic advance as a percentage of mean at 5% was observed more at 123.888 and at 1% it was 158.769 for 1000 seed weight. The characteristics 1000 seed weight, seed yield per plant, flowering initiation, 50% flowering and 100% flowering exhibited substantial heritability and genetic progression, indicating additive gene action for these variables. The characters point to the first branch, leaf length, leaf petiole length, and 100% flowering exhibited significant heritability with intermediate genetic advance, whereas siliqua bearing angle, main shoot length, the number of seeds per siliqua, and leaf width exhibited higher heritability with modest genetic advance, indicating the relevance of dominance and epistatic effects in the inheritance of these variables. The principal component analysis, also known as canonical root analysis, is a multivariate statistical technique that attempts to expedite and analyze the interrelationship between a large set of variables in terms of a relatively small set of variables or components, while retaining all of the authentic data sets vital information (Kumar and Kumar, 2021).

The principal component analysis breaks down a massive collection of data into a lesser number of components by looking for groups with high inter-correlation in a set of variables, with each component explaining a percentage of the overall variability. Among the total variation in the population, principal component 1 is the biggest contributor followed by subsequent components. Clifford and Stephenson (1975) noted that the first three main components are frequently the most essential in representing variation patterns throughout accessions,

Table 1. Basic descriptive statistics for eight quantitative traits of 20 germplasm lines of *Brassica juncea* subspecies rugosa

	SBA	PH	PFB	PB	SB	MSL	SMS	SL	SPS	1000SW	SYP
GCV (%)	8.676	12.025	22.071	13.933	15.846	13.259	10.148	11.381	12.04	60.718	52.802
PCV (%)	10.700	13.006	22.828	14.975	16.933	13.772	13.226	12.439	12.816	61.302	53.250
Variance (P)	22.575	358.500	1.142	0.859	3.888	39.404	8.438	0.186	2.102	0.200	15.242
Variance (G)	14.842	306.467	1.067	0.744	3.405	36.523	4.967	0.156	1.855	0.196	14.986
h^2 (Broad Sense) (%)	0.657	0.855	0.935	0.866	0.876	0.927	0.589	0.837	0.882	0.981	0.983
GA as % of mean (5%)	14.491	22.903	43.957	26.704	30.548	26.296	16.038	21.450	23.299	123.888	107.856
GA as % of mean (1%)	18.571	29.351	56.334	34.222	39.149	33.700	20.554	27.489	29.859	158.769	138.223

Table 1. Continued..

	OC	LL	LBL	LW	LI	LPL	LA	FI	50 %	100%
GCV (%)	2.354	13.761	15.607	14.755	11.494	20.686	20.936	32.036	25.875	20.822
PCV (%)	2.970	14.598	16.859	15.629	12.522	21.916	22.502	32.134	25.975	21.246
Variance (P)	1.466	19.752	12.811	4.730	0.037	4.227	3295.734	526.642	483.091	433.980
Variance (G)	0.921	17.551	10.979	4.216	0.031	3.766	2853.002	523.454	479.38	416.797
h^2 (Broad Sense) (%)	0.628	0.889	0.857	0.891	0.843	0.891	0.866	0.994	0.992	0.960
GA as % of mean (5%)	3.844	26.721	29.764	28.696	21.735	40.221	40.127	65.794	53.098	42.035
GA as % of mean (1%)	4.926	34.244	38.144	36.776	27.855	51.545	51.425	84.319	68.048	53.870

SBA=siliquae bearing angle; PH=plant height; PFB=point to first branch; PB=number of primary branches; SB=number of secondary branches; MSL= Main shoot length; SMS= number of siliquae on main shoot; SL=siliqua length; SPS=number of seed per siliqua; 1000SW=1000 seed weight; SYP= seed yield per plant; OC=oil content; LL=leaf length; LBL=leaf blade length; LW=leaf width; LI=leaf index; LPL=leaf petiole length; LA=leaf area; FI=flower initiation; 50%=50% flowering from initiation; 100%= 100% flowering from initiation

and the features linked with these are more beneficial in distinguishing the accessions, as confirmed by Guei *et al.* (2005). As a result, rather than going after all the characteristics under research, it is more effective for genetic enhancement of essential features with bigger contributions to variability. Out of twenty-one, only eight principal components (PC) exhibited more than 0.5 eigenvalues and showed about 94. 3184 per cent total variability among the characters studied (here only 5 PC is considered to have the most variability in the proposed characters/traits). Eight PCs were further explained due to their importance in variability. The PC1 showed 35.25 per cent followed by PC2 (18.71%), PC3 (17.09%), PC4 (6.96%), PC5 (5.18%), PC6 (4.50%), PC7 (3.54%), PC8 (2.78%) variability among the genotypes for the traits under study **(Table 2)**.

Scree plot explained the percentage of variance associated with each principal component obtained by drawing a graph between eigen values and principal component number. PC1 showed 35.52 per cent variability with eigen value of 7.45 which then declined gradually **(Table 2)**. Semi curve line is obtained after nine PC tended to become straight with little variance observed in each PC. From the graph, it is clear that the maximum variation was observed in PC1 in comparison to the other nineteen PCs. So, a selection of lines from this PC will be desirable **(Fig. 1 and Table 2)**.

Cluster analysis divides a vast number of accessions into a small number of homogenous clusters, making it easier to choose among a variety of accessions. Cluster analysis by the Tocher method **(Fig. 2)** based on principal component analysis was found to be a more useful measure for estimating divergence. Genetic divergence was studied in twenty genotypes in respect of various economic traits using cluster analysis by theTocher method. The distribution of genotypes in each cluster is presented in **Table 3**. Cluster number I had the highest number of genotypes (13) followed by cluster II (5), cluster III (1), and cluster IV (1). The genotypes were grouped into four clusters and the averages of inter and intra-cluster distances have been presented in **Tables 3 and 4**. The maximum intra-cluster distance was noted in cluster II (14.31) followed by cluster I (13.92), while the minimum intra-cluster distance was recorded in cluster III, and cluster IV (0.00). The maximum inter-cluster distance (41.68) was noted between clusters II and IV followed by clusters II and IV (40.6), clusters I and IV (34.95), clusters I and II (26.29), and clusters I and III (22.96).

Table 2. Principal component analysis (PCA) based on standard data for 21 quantitative traits of *Brassica juncea* subspecies rugosa

Components	PC1	PC2	PC3	PC4	PC5	PC6	PC7	PC8	PC9	PC10
Eigen value	7.46	3.93	3.59	1.46	1.09	0.95	0.75	0.58	0.34	0.33
Proportional variance (%)	35.52	18.71	17.09	6.97	5.19	4.50	3.55	2.78	1.63	1.55
Cumulative variance (%)	35.52	54.24	71.33	78.29	83.48	87.99	91.54	94.32	95.95	97.51
Traits										
SBA	0.563	0.062	0.206	-0.667	0.222	0.263	-0.058	-0.118	0.033	-0.020
PH	0.793	0.277	-0.180	-0.003	-0.103	0.328	0.154	-0.138	-0.032	0.265
PFB	0.459	-0.569	0.267	-0.423	-0.087	0.288	0.175	0.174	-0.019	-0.099
PB	0.785	-0.004	0.409	-0.017	0.184	-0.177	0.016	-0.085	-0.366	-0.012
SB	0.446	0.122	0.772	-0.065	0.249	0.075	0.060	0.043	-0.018	-0.228
MSL	0.439	-0.327	-0.549	0.062	-0.083	-0.127	0.445	0.345	-0.104	0.044
SMS	0.741	-0.109	-0.291	0.091	-0.339	0.219	0.182	-0.125	0.254	-0.240
SL	0.794	0.067	0.213	0.299	0.042	-0.089	0.355	-0.068	-0.079	-0.027
SPS	0.727	0.041	-0.018	0.431	0.332	-0.299	0.012	-0.032	0.189	-0.105
1000SW	0.245	-0.122	0.897	0.237	-0.037	-0.041	0.021	-0.141	0.121	0.102
SYP	-0.015	0.051	0.919	0.139	-0.142	0.099	0.129	0.049	0.114	0.241
OC %	0.099	-0.157	-0.003	0.631	0.203	0.611	-0.268	0.253	-0.101	-0.051
LL	0.936	-0.092	-0.046	-0.031	-0.123	-0.129	-0.249	0.114	0.027	0.041
LBL	0.840	-0.323	-0.275	0.071	-0.157	-0.028	-0.228	-0.150	-0.019	0.028
LW	0.543	-0.654	-0.375	-0.041	0.313	0.010	-0.038	-0.086	0.063	0.142
LBL/LW	0.595	0.375	0.106	0.084	-0.621	-0.051	-0.208	-0.076	-0.161	-0.090
LPL	0.622	0.337	0.311	-0.219	-0.065	-0.228	-0.199	0.475	0.132	0.046
LA	0.789	-0.476	-0.268	-0.064	0.145	-0.074	-0.182	-0.030	0.033	0.077
FI	0.268	0.883	-0.215	-0.079	0.265	-0.010	0.002	-0.075	-0.003	-0.062
50% flowering	0.306	0.879	-0.310	-0.005	0.098	0.077	0.078	0.054	0.019	0.033
100% flowering	0.336	0.877	-0.244	0.003	0.040	0.138	0.018	0.049	0.041	0.083

SBA=siliquae bearing angle; PH=plant height; PFB=point to first branch; PB=number of primary branches; SB=number of secondary branches; MSL= Main shoot length; SMS= number of siliquae on main shoot; SL=siliqua length; SPS=number of seed per siliqua; 1000SW=1000 seed weight; SYP= seed yield per plant; OC=oil content; LL=leaf length; LBL=leaf blade length; LW=leaf width; LI=leaf index; LPL=leaf petiole length; LA=leaf area; FI=flower initiation; 50%=50% flowering from initiation; 100%= 100% flowering from initiation

Table 3. Distributing pattern of twenty genotypes of *Brassica juncea* subspecies rugosa

Cluster number	Number of genotypes	Genotypes
I	13	IC-399826, IC-399839, Pusa Sag, IC-338751, IC-417128, IC-597917, IC-597873, IC-350800, IC-410471, IC-413486, IC-338535, IC-276011, IC-524259
II	5	EEC-25, IC-298019, IC-597933, IC-399880, IC-363758
III	1	IC-597821
IV	1	PR-15

The cluster means for all the twenty-one characters are given in **Table 5**. Siliqua bearing angle had shown the highest cluster mean value in cluster II (47.47) and lowest in cluster III (34). The highest cluster mean value for plant height was observed for cluster II (161.35) and minimum for cluster III (86.23). Cluster I (6.07) showed maximum cluster mean value for point to first branch while cluster III (2.7) the least. The mnmber of primary branches showed the highest cluster mean value in cluster IV (7.07) and minimum for cluster III (4.5). The character number of

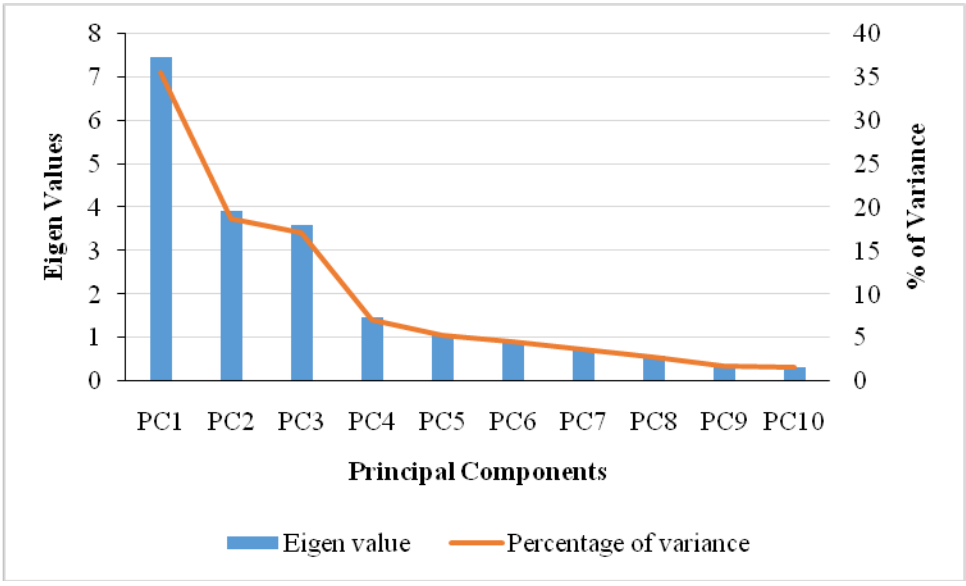

Fig. 1. Scree plot diagram using principal components of *Brassica juncea* subspecies rugosa

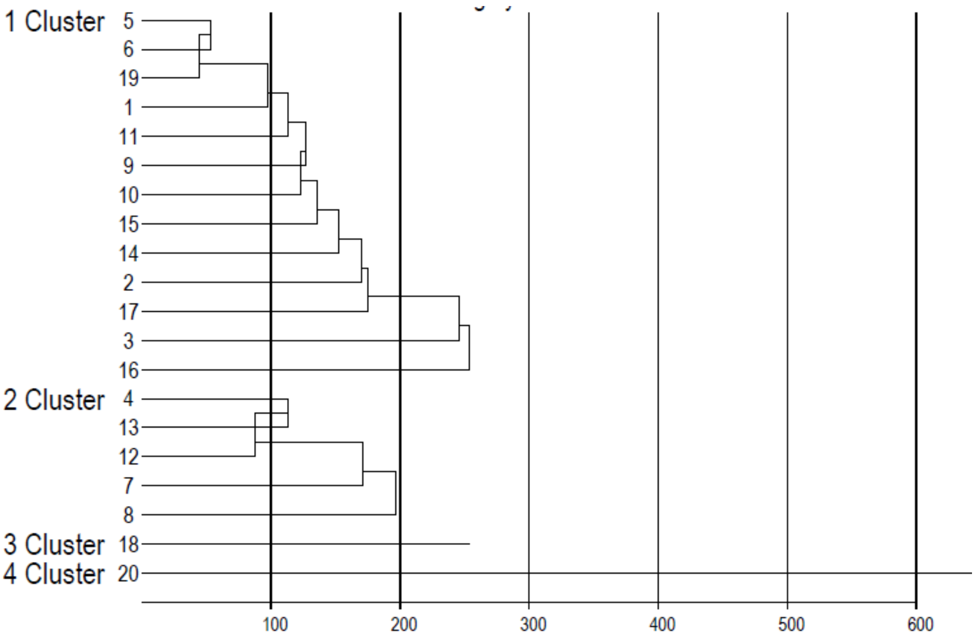

Fig. 2. Cluster of *Brassica juncea* subspecies *rugosa* prepared by Tocher Analysis

Table 4. Inter and intra-cluster distances

Cluster	I	II	III	IV
I	13.92	26.29	22.96	34.95
II		14.31	33.76	41.68
III			0	40.6
IV				0

Diagonal values indicates intracluster D^2 value

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Table 5. Cluster mean for different economic traits in *Brassica juncea* variety rugosa

S. No.	Characters	Clusters			
		I	II	III	IV
1	Siliqua bearing angle	43.83	47.47	34.00	47.00
2	Plant height (cm)	144.05	161.35	86.23	146.13
3	Point to first branch (cm)	4.89	4.27	2.70	6.07
4	Number of primary branches	6.12	6.55	4.50	7.07
5	Number of secondary branches	10.96	12.73	9.52	17.2
6	Main shoot length (cm)	48.11	44.48	30.00	33.73
7	Number of siliquae on main shoot	22.61	21.88	14.38	21.6
8	Siliqua length (cm)	3.42	3.7	2.39	4.00
9	Number of seed per siliqua	11.27	11.93	8.12	12.00
10	1000 seed weight (g)	0.63	0.67	0.45	2.55
11	Seed yield per plant (g)	6.07	7.68	7.38	21.92
12	Oil content (%)	40.73	40.75	41.03	40.95
13	Leaf length (cm)	30.95	31.86	15.45	31.85
14	Leaf blade length (cm)	22.39	20.62	9.75	20.71
15	Lamina width (cm)	14.79	12.94	8.78	12.55
16	Leaf index	1.52	1.61	1.11	1.65
17	Leaf petiole length (cm)	8.80	11.28	5.70	11.14
18	Leaf area (cm^2)	273.65	245.35	80.38	237.99
19	Days to flower initiation	61.28	104.00	66.00	45.67
20	Days 50% flowering	76.03	115.13	73.00	55.33
21	Days 100% flowering	89.95	126.07	83.33	78.00

secondary branches had the highest cluster mean value in cluster IV (17.2) and the lowest cluster mean value in cluster III (9.52). For main shoot length cluster I (48.11) had the highest cluster mean value and cluster III (30) the least. The number of siliquae on the main shoot was noted to have a maximum cluster mean value in cluster I (22.61), while a minimum cluster mean value in cluster III (14.38). Siliqua length had the highest cluster mean value in cluster IV (4), while the lowest cluster mean value in cluster III (2.39). The maximum cluster mean value for the number of seed per siliqua was observed for cluster IV (12) and minimum in cluster III (8.12). Cluster IV (2.55) showed the highest cluster mean value for 1000 seed weight, while cluster III (0.45) possessed the least cluster mean value. Seed yield per plant cluster IV (21.92) acquired the maximum cluster mean value and cluster I (6.07) the minimum. The cluster mean value for oil content was observed to be highest for cluster III (41.03) and the rest of the clusters had almost the same values. The character leaf length had the highest cluster mean value in cluster II (31.86) followed by cluster IV (31.85) and minimum for cluster III (15.45). Cluster I (22.39) showed the highest cluster mean value for leaf blade length, whereas cluster III (9.75) possessed the minimum cluster mean value. The cluster mean value for lamina width was found to be highest for cluster I (14.79) and the minimum cluster mean value for cluster III (8.78). The highest cluster mean value for leaf index was observed in cluster IV (1.65), while the minimum cluster mean value in cluster III (1.11). The cluster mean for leaf petiole length was maximum for cluster II (11.28) and least for the cluster (5.7). Leaf area was observed highest in cluster I (273.65) and minimum in cluster III (80.38). Flowering time at zero per cent was highest in cluster II (104) but lowest in cluster IV (45.67). At 50% flowering time, the cluster mean was maximum for cluster II (115.13) and minimum for cluster III (73). Cluster II (126.07) showed a maximum cluster mean value for 100% flowering time and the least cluster mean for cluster IV (78).

In **Table 6**, it was observed that flowering time at 0 % (51.05 %) attributed maximum per cent towards divergence followed by flowering time at 50 % (14.21 %), 1000 seed weight (8.42 %), lamina width (7.37 %), seed yield per plant (4.21 %), the number of seed per siliqua (3.16 %), main shoot length (2.63 %), point to first branch leaf length (2.63 %), leaf index (1.58 %), the number of secondary branches (0.53 %) and leaf area (0.53 %). The current study concluded that the very high contribution of flowering in time at 0 % (at initiation) to diversity was seen, which was most likely owing to the genotypes being of various origins and having more or less distinctive

Table 6. Per cent contribution of characters towards divergence in Twenty *Brassica juncea* subspecies rugosa germplasm accessions

S. No.	Characters	Per cent contribution
1	Siliqua bearing angle	0.00
2	Plant height	0.00
3	Point to first branch	2.63
4	Number of primary branches	0.00
5	Number of secondary branches	0.53
6	Main shoot length	2.63
7	Number of siliquae on main shoot	0.00
8	Siliqua length	0.53
9	Number of seed per siliqua	3.16
10	1000 seed weight	8.42
11	Seed yield per plant	4.21
12	Oil content	0.00
13	Leaf length	2.63
14	Leaf blade length	0.53
15	Lamina width	7.37
16	Leaf index	1.58
17	Leaf petiole length	0.00
18	Leaf area	0.53
19	Days to flower initiation	51.05
20	Days 50% flowering	14.21
21	Days 100% flowering	0.00

features for most of the parameters. Shalini *et al.* (2000) and Somu (2001), on the other hand, found that the characters number of siliquae per plant, plant height and days to 50% flowering were the most crucial contributors to genetic divergence in the same order.

High heritability coupled with high genetic progress for point to the first branch, main shoot length, 1000 seed weight, seed yield per plant, flowering initiation, 50% flowering and 100% flowering revealed additive type gene action and growth can be accomplished through simple selection. According to principal component analysis, seven principal components generated 87 per cent of the overall variance across the genotypes tested for twenty-one agronomic traits. PC1 contributed the most to the variability (35.52 %), followed by PC2 (18.71 %) and PC3 (17.09 %). Cluster analysis clustered the accessions into four major clusters, indicating a decent relationship between genetic diversity and accessions. Based on phenotypic and genotypic variation, principal component analysis and cluster analysis, sufficient diversity was identified in the genotypes, which researchers can exploit in future *Brassica juncea* variety rugosa breeding programmes. The degree of variability determines the potential for crop improvement. The presence of genetic diversity is the foremost decisive element in a plant breeding programme. Therefore, knowledge of available explainable variability is the basis to formulate a strategic improvement programme for the crop.

Fig. 3 represents the score plot of the two principal components of 18 *Brassica juncea* variety rugosa with two checks (PR-15 and Pusa saag). In the PC1 versus PC2 score plot, quarter 1 strains had a lot of variance for traits that contributed positively to PC1 and PC2, but quarter 2 strains Pusa Saag, IC-338751 and IC-597821 had a lot of variability for attributes that contributed positively to PC2 and traits that had negative weight for PC1. Strains in third quarter IC-597873, IC-524259 and IC-597917 had significantly much variance for traits bestow negatively for PC1 and for PC2. Similarly, strains in fourth quarter PR-15, IC-410471, IC-417128, IC-413486, IC-276011 and IC-350800 had enough variance for traits with negative weights for PC2. Those genotypes which are in the close vicinity of the origin are highly stable as compared to those which are far from the origin. IC-399826 genotype within an ace of origin focused as the enduring genotype out of all the genotypes examined. Whereas, IC-597821, IC-298019 and IC-276011 are away from the origin, which are less stable and more diverse strains comparable to others in their own quarter.

The correlation matrix among the traits along with the correlation coefficient showed 66 associations and

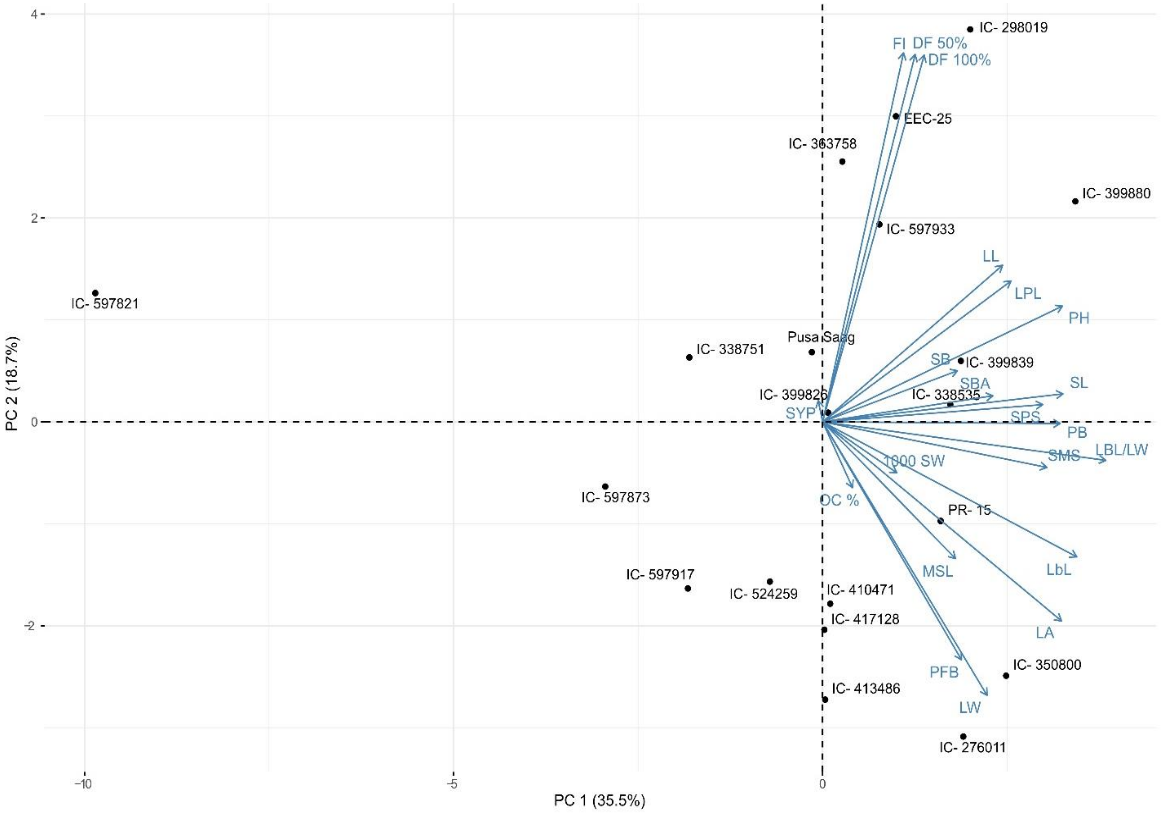

Fig. 3. Biplot of first two principal components in *Brassica juncea* subspecies rugosa genotypes

had 21 positive associations **(Fig. 4)**. Point to the first branch showed significant association with leaf area and positively correlated with each character. The number of primary branches showed a highly significant positive association with the number of secondary branches, and siliquae length but the number of seeds per siliqua, 1000 seed weight, leaf index and leaf area are less significantly associated with primary branches and positively associated with the remaining characters. A positive association of secondary branches with siliqua length, seed weight, seed yield per plant and main shoot length has a significant association with the number of siliquae on main shoot and leaf area. Siliquae on main shoot had highly significantly associated with siliqua length, leaf area, the number of seeds per siliqua and leaf index. Whereas, siliqua length had a significant association with the number of seeds per siliqua, leaf index and leaf area. The number of seeds per siliqua and 1000 seed weight were associated with leaf area and seed yield per plant, respectively.

The inter-character association between seed yield and its contributing characters helped breeder to improve seed yield indirectly through the improvement of associated traits. Simultaneously many component traits can be considered at a time provided a positive significant relationship was established between the traits. The linkage/ association between traits have also been studied earlier (Roy *et al.,* 2018 and Singh *et al*., 2018) in crops where correlation among the traits was taken a criteria for improvement of complex traits like seed yield. Re-examining the results, it was shown that heritability is substantial for the majority of the variables, while genetic improvement can high, intermediate, or low. High heritability and high genetic advance suggested that simple selection could be used to its fullest potential. While, maintaining heterozygosity through the creation of hybrids would be a preferable method to maximize gain for situations when the genetic gain is moderate or low. These findings are in consonance with those of Singh *et al.,* 2018; Pal *et al.,* 2019; Rout *et al.,* 2019; Gadi *et al.,* 2020; Shah *et al.,* 2021 and Yadav *et al.,* 2021.

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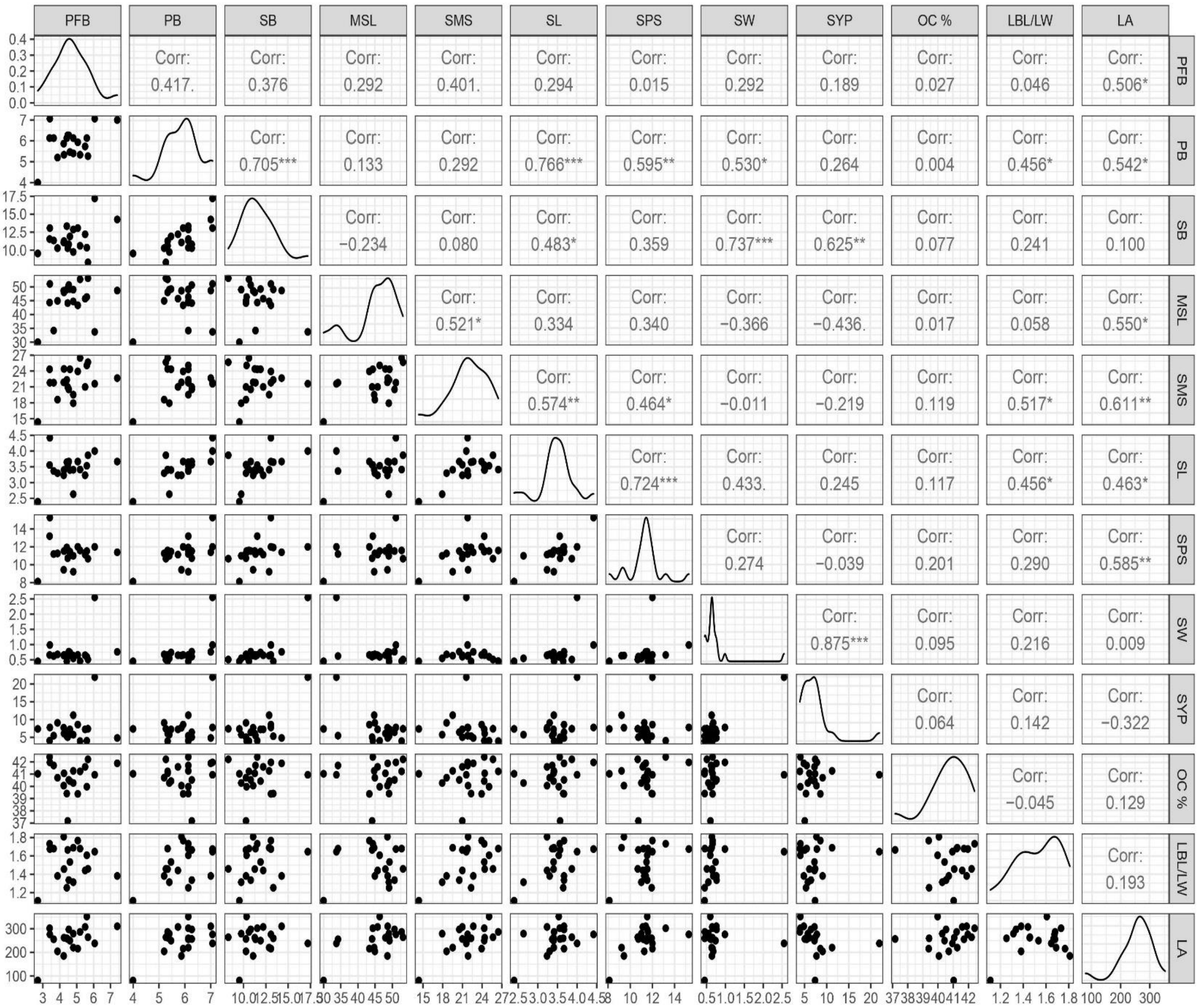

Fig. 4. The pairwise correlations of different quantitative traits of *Brassica juncea* subspecies rugosa germplasm. The upper panel showed numerical values of the correlation. The *, **, and * stars indicate correlation significant at 0.05, 0.01, and 0.001 levels of probability, respectively. The lower panel showed the scatter plots.**

PFB= Point to first branch, PB=Primary branches, SB= Number of secondary branches, MSL=Main shoot length, SMS= number of siliquae on main shoot, SL=siliqua length, SPS=Number of seed per siliqua, SW=1000 seed weight, SYP=Seed yield per plant, OC= Oil content, LBL= Leaf blade length, LW= Lamina width, LA=Leaf area

REFERENCES

Adhikari, A., Punetha, H. and Prakash, O. 2021. Status, Cultivation, and Usages of Indian mustard. Nova science publishers Inc, 33.

Bhattarai, B. P., Shing, K. P., Shakya, S. M., Khatri-Chhetri, G. B. and Khadka, Y. G. 2018. Effect of organic and conventional nutrient management on post-harvest status of broad leaf mustard (*Brassica juncea* var rugosa). *International Journal of Horticulture & Agriculture*, **3**(2): 1-4. [Cross Ref]

Clifford, H. T. and Stephenson, W. 1975. An Introduction to Numerical Classification. Academic Press, London. P. 229. [Cross Ref]

Gadi, J., Chakraborty, N. R. and Imam, Z. 2020. To Study the genetic variability, heritability and genetic advance for different quantitative characters in Indian mustard (*Brassica juncea* L. Czern & Coss). *International Journal of Current Microbiology and Applied Sciences*, **9**(10): 1557-1563. [Cross Ref]

Guei, R. G., Sanni, K. A., Abamu, F. J. and Fawole, I. 2005. Genetic diversity of rice (*Oryza sativa* L.). *Agronomie Africaine*, **5**:17-28.

Hotelling H.1933. Analysis of complex of statistical variables into principal components. *Journal of Educational Psychology,* **24**:417-441. [Cross Ref]

Kumar, N. and Kumar, V. 2021. Assessment of genetic diversity in linseed germplasm using morphological traits. *Electronic Journal of Plant Breeding*, **12**(1): 66-73. [Cross Ref]

Lê, S., Josse, J. and Husson, F. 2008. FactoMineR: an R package for multivariate analysis. *Journal of statistical software*, **25**(1): 1-18. [Cross Ref]

Macready, A. L., George, T. W., Chong, M. F., Alimbetov, D. S., Jin, Y., Vidal, A. and FLAVURS Study Group. 2014. Flavonoid-rich fruit and vegetables improve microvascular reactivity and inflammatory status in men at risk of cardiovascular disease-FLAVURS: a randomized controlled trial. *The American Journal of clinical Nutrition*, **99**(3): 479-489. [Cross Ref]

Mahalanobis, P.C. 1936. On the generalized distance in statistics. Proc. National Institute of Science of India, **2**:49-55.

Pal, S., Dubey, N., Avinashe, H., Khan, S. and Reddy, P. J. 2019. Estimation of genetic variability, correlation and path analysis for yield and yield contributing characters in Indian mustard (*Brassica juncea* L.). *Journal of Pharmacognosy and Phytochem*istry, **4**: 102-105.

Pant, U., Bhajan, R., Singh, A., Kulshesthra, K., Singh, A. K. and Punetha, H. 2020. Green leafy mustard: A healthy alternative. *Electronic Journal of Plant Breeding*, **11**(01): 267-270. [Cross Ref]

Rout, S., Kerkhi, S. A. and Gupta, A. 2019. Estimation of genetic variability, heritability and genetic advance in relation to seed yield and its attributing traits in Indian mustard. *Journal of Pharmacognosy and Phytochem*istry, **8**: 4119-4123.

Roy, R. K., Kumar, A., Kumar, S., Kumar, A. and Kumar R. R. 2018. Correlation and Path Analysis in Indian Mustard (*Brassica juncea* L. Czern and Coss) under Late Sown Condition. *Environment and Ecology*, **36** (1A): 247-254.

Shah, M. A., Rehman, F. U., Mehmood, A., Ullah, F., Shah, S. I. and Rasheed, S. M. 2021. Combining ability, heritability and gene action assessment in rapeseed (*Brassica napus* L.) for yield and yield attributes. *Sarhad Journal of Agriculture*, **37**(1): 104-109. [Cross Ref]

Shalini, T. S., Sheriff, R. A., Kulkarni, R. S. and Venkataravana, P. 2000. Genetic divergence in Indian mustard (*Brassica juncea* L. Czern and Coss). *Mysore Journal of Agricultural Sciences*, **34**(3): 251-256.

Singh, S., Dwivedi, A. K., Ashutosh, O. M. and Kumar, K. 2018. Genotypic variability, heritability and genetic advance in Indian mustard [*Brassica juncea* (L.) Czern & Coss.] Genotypes. *Journal of Pharmacognosy and Phytochemistry*, **7**(3): 350-352.

Somu, G. 2001. Genetic divergence in Indian mustard (*Brassica juncea* L. Czern and Coss.). Doctoral dissertation, University of Agricultural Sciences, GKVK, Bangalore.

Yadav, H., Meena, P. K. P., Koli, N. R., Meena, B. S. and Yadav, D. L. 2021. Assessment of selection parameters for yield and oil content in Indian mustard [*Brassica juncea* (L.) Czern & Coss.].