Classification for Core Collection of Rapeseed-Mustard (*Brassica juncea* L.) Germplasm

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Based on passport data of maturity (early, medium and late), stratified random sampling procedure was adopted to select 385 accessions of Indian mustard (*Brassica juncea* L.) maintained at NRCRM, Bharatpur, India, which was taken for this study. Observations of 17 characters were taken into consideration for the classification. All the accessions were classified in 9 clusters. Core collections of different cluster were selected with the help of following method, which is the main objective of the project. Mean ± SD of each character of different clusters were calculated and taken into consideration for selecting the core accessions of each cluster. All the characters in different clusters were studied and the character that has more than Mean ± SD was marked rank first. All the accession that ranked first in a minimum of four characters, were selected as core collection of that group. This way, out of 385 accessions only 116 accessions were selected as core collection, which is 30.13%. Now, it will be easy to maintain and easier for the breeder to select for breeding purposes. Viewing the Euclidean square distance, different accession of *Brassica* can be selected for breeding purposes as per the requirements of the breeder.

Key Words: Rapeseed-Mustard, Agro-morphological characters, Variability, Cluster analysis, Core collection, Euclidean square distance, DAS (days after sowing)

Introduction

Among the oilseeds, *Brassica* group of crop is a highly diverse group of crop with great economic value. After groundnut oilseed *Brassica* are the second most important source of edible oil production in India. Productivity of *Brassica* in our country has not increased despite selections and intensive breeding programme, reason being most of the parents selected in hybridization programme were selected for seed yield, which is almost homogeneous in nature. Hence, pedigree method could not provide the desirable improvements in the yield. The selection of pedigree by the breeder from the huge germplasm for hybridization programme depends mainly on specific character or yield, which is not the proper way of selection by which we are lagging in improving the productivity in comparison to world scenario. Maintenance and selection of pedigree from the huge *Brassica juncea* L. germplasm, which is maintained at National Research Center on Rapeseed-Mustard, Bharatpur, India, is not only a cumbersome task but also quite difficult for the breeder to select the pedigree for breeding programme. Hence, this study was carried out in *Brassica* germplasm, considering principal component analysis as a tool, in which all the 17 characters were considered together to calculate principal component score to classify the entire population into different groups. A methodology was given to select core collection which have distinctly variable, agro-morphological characters useful for the breeder for selecting them for their breeding programme to attain maximum productivity, viewing the crop productivity level in rest of the world (Mishra *et al.*, 1991).

Materials and Methods

Based on passport data of maturity (early, medium and late), stratified random sampling procedure was adopted to select three hundred eighty three germplasm accessions of *Brassica juncea* L. along with two check varieties, Bio-902 and PCR-7, were grown for characterization, under irrigated condition in augmented design at National Research Centre on Rapeseed-Mustard, Sewar Bharatpur (India) during *rabi* 2002-03-04. Each accession was grown in two-row; a plot of three-meter length and plant and row distance was maintained at the spacing of 30 cm×10 cm, respectively. Standard agronomic and plant protection measures were followed to raise a healthy crop.

The observations were recorded on randomly tagged five plants for different traits such as initiation of flowering (DAS); 50% flowering (DAS); maturity duration (days); plants height (cm); primary branches per plant (no); secondary branches per plant (no); main shoot length (cm); siliquae on main shoot (no); siliqua length (cm); siliqua beak length (cm); seed per siliqua (no); total yield (g); seed yield per plant (g); 1000-seed weight (g); harvest index (%); oil content (%) and protein content (%) at appropriate growth stages of the crop. 1000-seed was counted by the electronic seed counter (the old mill

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company, USA) and weighed by electronic balance. Further, oil and protein content were analyzed by NIR (Dickey-Johan, USA) method. All the observed data were tabulated in Microsoft Excel programme and considered for cluster analysis.

Results and Discussion

A total of 385 accessions of Indian mustard germplasm including two check varieties of Bio-902 and PCR-7 were taken for the study. The 17 characters, which are presented in Table 1, along with Characters' mean, C.V. and Percentage contribution to divergence were considered to be the base for the classification. Using Principal component analysis 385 accessions were classified into 9 groups/clusters. These 9 groups were taken, considering F-Statistics (Rao, 1952), in the process of statistical analysis, which is higher in the case of cluster # 9. The analysis revealed that the most variable cluster is cluster # 8 followed by cluster # 4, cluster # 6, cluster # 7, cluster # 5, cluster # 9 etc viewing within mean sum of square (Table 2). It also revealed that most variable character are Seed weight/plant (CV 38.65) followed by Secondary branches (CV 36.04), Total weight (CV 32.46), Siliqua beak length (CV 21.27), Seed/Siliqua (CV 21.16) etc. (Table 1). Arranging percentage contribution of the characters to genetic divergence in descending order (Table 1) it reveals, 10 characters namely, Siliqua beak length (SBL); Siliqua length (SL); Seed/siliqua (SS); Day to maturity (DM); Protein content (PC); 1000-seed weight (1000WS); Initial flowering (IF); Harvest index (HI); Plant height (PH); Secondary branches (SB) explained 67.54%, where as 5 principal component explained 65.06% and 10 principal component explained 88.07% towards genetic divergence (Hepziba *et al.*, 1995; Mishra *et al.*, 1998). Hence, classification done through principal component analysis is justified. Initial flowering is highly correlated with 50% flowering (r=0.859) followed by seed weight/plant, total weight (r=0.811); secondary branches, primary branches (r=0.652); total weight, secondary branches (r=0.548); plant height, 50% flowering (r=0.518); harvest index, seed weight/plant (r=0.484); siliqua on main shoot, plant height (r=0.482); day to maturity, plant height (r=0.445); protein content, 50% flowering (r=0444); day to maturity, 50% flowering (r=0.434); primary branches, plant height (r=0.431); total weight, primary branches (r=0.430) etc. (Table 4). Considering Table 4, description of quantitative characters for each cluster were classified as, >Over all mean + Standard deviation = VL; > Over all mean = L; < Over all mean = M; < Over all mean – Standard deviation = S (Table 5). Descriptions of quantitative characters (Table 5) were tested with Chi-square. Calculated Chi-square = 22.11. Tabulated Chi-square value for 8 d.f. at 1% level = 20.09 (Mishra *et al.*, 1998).

Hence, Chi-square is significant at 1% level, which justifies the classification of accession in 9 clusters with different characteristics.

Table 1. Characters, their means, coefficient of variance (CV) and percentage contribution to divergence

S.No.	Character Name	Mean	CV	Percentage contribution to divergence
1.	Initial flowering (IF)	52.89	17.01	6.55
2.	50% flowering (50%F)	65.12	18.79	4.50
3.	Plant height (PH)	148.00	13.65	6.11
4.	Primary branches (PB)	6.11	17.96	5.20
5.	Secondary branches (SB)	9.67	36.04	5.29
6.	Main shoot length (MSL)	49.49	15.38	5.07
7.	Day to maturity (DM)	141.93	2.67	6.99
8.	Siliqua beak length (SBL)	0.72	21.27	8.15
9.	Siliqua length (SL)	3.60	13.16	7.51
10	Siliqua on main shoot (SMS)	36.48	16.22	5.23
11.	Seed weight/plant (SW/P)	7.50	38.65	4.96
12.	Total weight (TW)	33.61	32.46	4.55
13.	1000 seed weight (1000SW)	3.74	19.54	6.58
14.	Harvest index (HI)	22.42	21.15	6.43
15.	Seed/siliqua (SS)	14.09	21.16	7.16
16.	Protein content (PC)	22.16	3.23	6.77
17.	Oil content (OC)	39.25	5.01	295

Table: 2. Classified #9 clusters

Cluster	N	Within MSS	Cluster members
1	52	9.31	UPII-81, UPII-132, HP-1, UPI-82, UPII-79, UPII-100, P-32, UPII-87, UPII-33, NCRQR-6-91, UPII-56, UPII-58, KLM 227, UPII-86, RK-2-4, RN 573, P-98, JGM 9005, P-15, KLM-12A, P-72, UPII-80, P-44, P-11, UPII-14,P-50,UPII-9,UPII-123,P-69,UPII-35,NDR200202,UPII-60,UPII-84, UPII-22, UPII-110, UPII-10, P-80, UPI-74, P-48,UPII-78, UPII-76, UPII-44, P-68, P-71, P-58, P-65, P-20, P-45, P-24, JGM-01-04, P-74, P-23.
2	59	8.56	NPJ90, UPII-25, UPII-124, UPII-37, UPII-51, UPI-55, UPII-47, UPI-29, UPII-92, UPI-39, UPII-105, UPI-22, UPI-38, RH-114, TKG-18-64, UPII-29, UPII-48, UPI-78, P-61, UPI-69, UPII-125, RM11, UPI-53, P-25, UPI-72, PR 2001-87, UPII-46, RK02-5, UPII-22, UPII-122, UPI-77, UPI-31, UPII-61, P-33, RH115, P-37, RH118, P-92, PRQ2005-1, NDRE200216, UPII-118, TKG-180-9, UPI-24, 330RKP-2, UPI-51, UPII-26, UPI-76, UPI-63, UP-I-68(BJ), UPI-73UPI-2, UPI-36, NPJ-82, RK-01-3, RGN-74, UPII-50, UPI-14, SKM-158, UPI-26.
3	70	7.41	UPII-40, P-28, UPII-54, UPI-90, UPII-39, PR-2001-84, P-63, PR-2001,UPI-70, P-93, UPII-127, UPII-93, UPII-53, HUJM-01-02, UPI-44, UPI-71, UPII-128, UPI-23, UPII-64, UPI-52, UPI-50, UPI-43, UPI-56, UPII-112, SKM-149, UPII-94, UPII02, JGM2-1, UPII-69, P-42, UPII-75, RK-02-6, UPI-42, RS-2, RH-27, UPII-8,UPI-67,YET22,5084,SKM-9927, UPII-90,UPI-48, UPI-42, UPII-96, P-30, RM-51, UPII-1, UPII-49, UPI-6, UPII-52,UPI-20, UPI-75,NUDHYS-2,UPI-28,UPI-41, UPI-45, UPII-97, P-6, UPII-3, UPI-85, UPII-82, UPII-12, P-60, UPI-81, NDR200203, RK02-2, PR404, SKM2026, UPII-91, UPI-19,
4	2	13.70	RL9927, UPI-61.
5	46	10.30	UPII-38, UPII-41,P-62, UPII-101, UPII-99, UPII-77,UPII-95, UPII-88, UPII-34,RGN-79, UPII-62, JMWR-2-40,UPII-28, Bio-Q-108, UPII-65, RGN-89,UPII-130,UPII-129, UPII-13,UPII-68,UPII-45,UPII-15,UPII-36, P-77, UPII-103, UPII-120, UPII-20, UPI-62, UPII-57, UPII-19, P-53, UPII-43, P-56, P-10, P-103, P-95, NUDH-YJ-1, UPII-73, P-8, UPII-18, RH(OE)-0103, P-106, P-16, P-19, P-82, UPII-71.
6	51	10.63	UPII-85,NDRS-2002,UPII-6,UPII-131,P-31,HUJM-101,P-51,RK02-1, **PCR7**,HUJM105,UPI-1,PR2005,UPI-9UPII-89,P-29,RN510,P-52,DHR9701,JMM927,UPI-57,RM-49,UPI-12,RK-02-3,UPI-49,P-35,UPI-13,UPI-60,UPII-121,RH9615,NDRS2001,UPI-5,YET-17,NDRS-2003,UPI-3,YRN-6,ORM(M)-25-1,UPI-46,UPI-85,UPI-11,PR2001-42,P-38,SKM9816,UPI-89,UPII-4,UPII-115,SAL-9,RH-116,UPI-4,HP-2,NPJ90,UPI-21.
7	33	10.58	RK-2-4, UPI-66, RH(OE)1-1, RH-28, RK2002, UPI-54, UPI-30, SKM9819, UPI-37, UPII-98, NDR-8209, UPII-111, 614-41-4, UPI-34, HUJM-106, 611-1-3-6, P-40, RS-7, UPI-125, RGN-55, UPI-7,UPI-8, UPI-40, UPI-10, RP-2006, PR-2001-78, P-36, UPI-16, RH-0117, **Bio902**, PR2001-65, JMWR-13.
8	31	14.60	UPII-83, UPII-24, P-9, UPII-30, UPII-17, P-26, P-22, P-105, P-99, P-104, P-47, P-7, P-84, P-1, UPII-16, P-102, P-18, P-78, P-83, P-107, P-41, P-13, P-96, P-57, P-5, P-4, P-49, P-14, P-66, P-54, P-79.
9	41	9.74	UPII-67, UPII-63, UPII-66, UPII-42, UPII-32, RN554, P-12, P-27, UPII-126, UPI-58, UPI-33, UPI-33S, SKM9827, UPII-27,P-44, UP11-5, UPI-59 UPII-119UPII-31, NPJ-92, UPI-86, BAUSMM-11, UPII-110, Krishna-2-1, UPI-88, Bio-772R, PRO-2103, UPI-35, UPI-17, Bio-36-99, UPII-23, UPII-11, SALE3, UPI-79, RW-02-01UPI-18, UPI-27, UPI-15, Krishna Shorty, UPI-32, NDRE-200201,RNT-1.

Table 3. Correlations of different characters

Character	IF	50%F	PH	PB	SB	MSL	DM	SBL	SL	SMS	SW/P	TW	1000SW	HI	SS	PC	OC
IF		0.859	0.427	0.279	0.239	-0.398	0.384	-0.144	-0.105	0.138	-0.101	0.023	-0.310	-0.224	0.097	0.386	0.206
50%F			0.518	0.399	0.315	-0.442	0.434	-0.174	-0.178	0.168	-0.085	0.076	-0.350	-0.284	0.101	0.444	0.280
PH				0.431	0.220	-0.040	0.445	-0.081	-0.024	0.482	0.249	0.426	-0.179	-0.232	0.074	0.206	0.332
PB					0.652	-0.312	0.114	-0.182	-0.233	0.153	0.222	0.430	-0.302	-0.281	-0.017	0.264	0.117
SB						-0.030	0.027	-0.174	-0.179	0.287	0.352	0.548	-0.250	-0.214	0.065	0.174	0.079
MSL							-0.103	0.173	0.191	0.397	0.342	0.283	0.360	0.177	-0.032	-0.403	-0.007
DM								-0.075	-0.001	0.155	0.048	0.122	-0.017	-0.110	0.038	0.062	0.220
SBL									0.358	-0.067	0.085	0.026	0.187	0.140	0.006	-0.186	-0.128
SL										-0.003	0.076	0.035	0.215	0.075	0.158	-0.165	0.032
SMS											0.269	0.361	-0.086	-0.091	0.083	0.034	0.205
SW/P												0.811	0.223	0.484	0.040	-0.186	0.056
TW													0.087	-0.083	0.013	-0.073	0.088
1000SW														0.274	0.018	-0.390	-0.056
HI															0.037	-0.212	-0.048
SS																0.017	0.207
PC																	-0.002
OC																	

Table 4. Cluster wise mean of different characters along with over all mean of all the characters

Cluster name	IF	50%F	PH	PB	SB	MSL	DM	SBL	SL	SMS	SW/P	TW	1000SW	HI	SS	PC	OC
1	57.06	76.37	165.27	7.13	12.45	45.39	144.31	0.67	3.41	38.25	9.02	39.92	3.38	22.84	15.16	22.52	40.43
2	48.76	58.97	144.12	6.28	10.74	52.69	138.58	0.79	3.85	36.96	7.88	37.20	3.69	21.23	14.08	22.04	38.82
3	48.26	57.71	138.20	5.57	8.33	53.74	141.96	0.66	3.38	35.95	7.09	29.25	4.01	24.31	12.39	21.80	38.89
4	53.00	61.00	151.90	6.60	9.90	48.60	146.00	0.73	3.41	36.20	8.00	36.50	3.62	21.71	10.78	21.63	20.71
5	54.74	70.74	157.71	6.48	8.75	44.20	142.67	0.63	3.33	37.17	4.93	28.13	3.20	17.81	14.03	22.69	39.80
6	50.12	60.08	147.21	5.35	7.06	53.18	143.41	0.86	4.09	35.63	7.29	30.22	4.50	24.23	15.70	21.65	39.82
7	50.64	61.55	160.73	6.41	11.46	57.07	143.09	0.74	3.61	41.96	13.42	54.47	4.08	24.95	14.07	21.74	39.33
8	75.52	91.10	165.85	6.98	12.90	41.51	144.81	0.67	3.51	38.42	5.84	32.77	3.05	18.42	14.29	22.96	40.04
9	47.51	56.00	115.24	5.18	7.15	44.09	137.73	0.74	3.59	28.99	5.27	21.96	3.67	24.18	13.81	22.27	38.01
Over all mean	52.89	65.12	148.00	6.11	9.67	49.49	141.93	0.72	3.60	36.48	7.50	33.61	3.74	22.42	14.09	22.16	39.25

Table 5. Description of quantitative characters for each cluster

Cluster name	IF	50%F	PH	PB	SB	MSL	DM	SBL	SL	SMS	SW/P	TW	1000SW	HI	SS	PC	OC
1	L	L	L	L	L	M	L	M	M	L	L	L	M	L	L	L	L
2	M	M	M	L	L	L	M	L	L	L	L	L	M	M	M	M	M
3	M	M	M	M	M	L	L	M	M	M	M	M	L	L	M	M	M
4	L	M	L	L	L	M	VL	L	M	M	L	L	M	M	S	M	S
5	L	L	L	L	M	M	L	M	M	L	M	M	M	M	M	L	L
6	M	M	M	M	M	L	L	L	VL	M	M	M	VL	L	L	M	L
7	M	M	L	L	L	L	L	L	L	L	VL	VL	L	L	M	M	L
8	VL	VL	L	L	L	S	L	M	M	L	M	M	M	M	L	L	L
9	M	M	S	M	M	M	S	L	M	S	M	S	M	L	M	L	M

> Over all mean =L; < Over all mean = M; <Overall mean - SD = S; > Over all mean + SD = VL

(a) ***Characteristics of different clusters***

Cluster # 1 has maximum plant height, a high total plant weight and has the highest oil content amongst all clusters but has a low harvest index. Cluster # 2 is early maturing and is comparable to cluster # 1 in oil content but again has a low harvest index. Cluster # 3 is similar to cluster # 2 but has a lower total weight but an improved harvest index. Cluster # 4 has the lowest oil content while cluster # 5 has the lowest harvest index. Cluster # 6 & cluster # 8 are much better than the above-mentioned clusters but have greater plant height and a high total plant weight. Cluster # 7 has the maximum total plant weight and rest of the traits comparable to cluster # 6 and 8. Cluster # 9 is the best cluster when all traits are compared as it has the lowest plant height, earliest maturing, has lowest total plant weight, high harvest index and high oil content (Mishra *et al.*, 1991).

(b) ***Methodology to select core collections from different classified groups***

Core accessions of different clusters were selected with the following description. Mean ± SD of each character of different clusters were calculated and taken into consideration for selecting the core accessions of each cluster. All the characters in different cluster were studied and the character, which is more than Mean ± SD were marked rank first. All the accessions that ranked first in a minimum of four characters were selected as core accession. This way, out of 385 accessions only 116 accessions were selected as core one, which is 30.13 % (Table 6) (Mishra *et al.*, 1991). Selection of core collection of the germplasm was adopted with the help of discriminant function analysis as well as with the help of Mean and S.D. It was observed that core collection selected in either of the above methods is identical. Hence, in present study Mean and S.D. were considered appropriate to select representative/core accession to maintain the germplasm, because discriminant function analysis was little difficult and need specific software for the purpose. In the present study 30.13% accessions of germplasm were selected as core collection to retain the same correlation structure of the characters as found in the base collections. In the present study, when only 10 % optimum sample size was considered to retain as core collection, it vacillated correlation structure between the characters of core and base collections.

Table 6. Representative varieties of different clusters along with (%) of total no. of Accessions in each cluster

Cluster no.	Total no. of accessions in a cluster	Representative varieties	Total no. of representative accessions	(%) of total no. of accessions in a cluster
1	52	UP II-100, P-15, P-72, UP II-14, UP II-9, UP II-84, UP II-10, UP I-74, P-48, P-68, P-71, P-65, P-20, JGM-01-04, P-74, P-23	16	30.77
2	59	RNS-72, UP II-92, UP II-105, UP I-69, UP II-21, P-II-12 UP I-31, RH 115, P-37, RH 118, UP I-24, 330-2KP 2, UP-I-68 (BJ), UP I-73, NPJ-82, RK-01-3, RGN-74	17	28.81
3	70	UP II-39, P-63, PR-2001, UP I-70, UP II-93, HUJM-01-02, UP I-71, UP I-23, UP I-50, P-42, RH-27, 508 4, SKM-9927, UP I-48, UP I-42, RM-51, UP II-49, UP I-20, UP I-28, UP II-3, UP II-118, P-60, UP I-81, UP II-91	24	34.29
4	2	Nil	Nil	Nil
5	46	UP II-38, P-62, UP II-99, UP II-95, UP II-88, RGN-79, RGN-89, UP II-129, UP II-20, P-53, UP II-43, P-56, P-10, NUDH-YJ-I, UP II-18, P-106, P-16, P-82	18	39.13
6	51	UP II-85, UP II-131, HUJM-101, PR-2005, UP II-89, RN 510, DHR-9701, UP I-13, UP II-121, RH 9615, NDRS-2001, YET 17, UP I-3, YRN-6, UP-I-11, P-38, SKM-9816, HP-2	18	35.29
7	33	NDR-8209, UP I-34, RGN 55, UP I-8, P-36, RH –0117, Bio-169-96, Bio 902, PR 2001-65, JMWR-13	10	30.30
8	31	P-99, UP II-16, P-102, P-107, P-41, P-57, P-49, P-54	8	25.81
9	41	UP II-67, UP II-31, Bio -772 R, UP I-27, RNT-1	5	12.20
Over all	385		116	30.13

(c) *Method for selection of accession for breeding programme*

Germplasm accession between the groups from core collection can be selected for hybridization programme, considering the Euclidean square distance (Table 7). It reveals that breeding can be performed after selecting the specific characters suitable to the breeder as per requirement of the breeding programme from the clusters which have maximum Euclidean square distance such as cluster # 9 and # 6 having maximum Euclidean square distance (122.84) followed by cluster # 9 and # 3 (117.41); # 9 and #1 (114.38); # 9 and # 7(113.43); # 9 and # 8 (107.91); # 9 and #4 (104.03); #9 and # 2(88.72); # 9 and #5 (82.78); # 8 and #2 (40.76); # 2 and # 1 (38.09); # 8 and # 3 (34.64), respectively (Mishra *et al.*, 1998; Reddy *et al.*, 2003).

(d) *Future aspects for improving production and productivity to sustain yield*

In all the clusters except in cluster # 9 the photosynthates that are being produced by the green parts of the plant are being mobilized to different plant sinks where they are being utilized to increase plant height and total plant weight. But the harvestable sink (seeds) remains deprived of those essential photosynthates, which form the major constituents that sustain the quality and yield of the crop.

Hence, there is a need that all the five representative varieties of cluster # 9 be screened thoroughly for those genes that are responsible to block the unnecessary translocation of the valuable photosynthates to the non harvestable sinks, so that they can be partitioned unidirectionally towards the harvestable seeds and thus, improve the harvest index and the oil content .

Table 7. Euclidean square distance matrix for 9 clusters

Clusters	1	2	3	4	5	6	7	8	9
1	0.00	38.09	28.32	20.71	22.39	12.71	22.30	10.40	114.38
2		0.00	11.65	20.10	11.39	29.64	14.98	40.76	88.72
3			0.00	12.42	10.40	15.74	8.61	34.64	117.41
4				0.00	4.61	10.85	7.63	20.55	104.03
5					0.00	13.33	8.71	18.11	82.78
6						0.00	10.54	16.98	122.84
7							0.00	24.37	113.43
8								0.00	107.91
9									0.00

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