



ISSN (E): 2277-7695
 ISSN (P): 2349-8242
 NAAS Rating: 5.23
 TPI 2022; SP-11(9): 795-799
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www.thepharmajournal.com
 Received: 28-06-2022
 Accepted: 30-07-2022

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Population structure analysis for white rust and economically important traits in Indian mustard

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Abstract

Ninety genotypes of *Brassica juncea* belonging to European and Indian gene pool were evaluated for five economically important traits such as white rust resistance, grain yield/plant (g), oil content (%), erucic acid (%) and glucosinolates ($\mu\text{mole/g}$). Three genotypes (Donskaja IV, Domo and Cutlass) showed no infection and they were considered as immune against white rust. Maximum grain yield/plant was recorded in the genotypes DRMR-2017 (19.07 g) followed by J807/1/6 (18.36 g) and Vardan (17.72 g). Maximum oil content was found in PGR 12573 and EC 287711 (44.16%) followed by Stoke (44.01%) and Jubilejnaja (43.54%). Minimum erucic acid (12.18%) and glucosinolate content (28.92 $\mu\text{mole/g}$) content was found in genotypes Donskaja IV and Heera, respectively. The genotypes were grouped into different clusters using R-Studio software. The population was divided mainly in two main clusters and five clusters indicating significant genetic variation in the germplasm. The maximum cluster distance (83.27) was observed between cluster 3 and cluster 5 followed by cluster 2 and cluster 5 (29.20) and between cluster 1 and cluster 3 (57.72). It indicated maximum genetic diversity between these pair of clusters; and possibility of developing high yielding, better quality and white rust resistant genotypes through hybridization between genotypes belonging to contrasting clusters.

Keywords: Population structure, white rust, economically, traits

Introduction

Brassica juncea is one of the important oilseed crop belonging to the family *Brassicaceae* (also known as *Cruciferae*). It is commonly known as Indian or Brown Mustard. *Brassica juncea* is a natural allopolyploid (AABB) with $2n=36$ that contains genomes of two diploid species, *Brassica rapa* (AA), $2n=20$ and *Brassica nigra* (BB), $2n=16$. Nuclear and organelle phylogenetic studies by Kang *et al.* (2021) [5] established that *B. juncea* had most likely a single origin in West Asia, 8,000–14,000 years ago, via natural interspecific hybridization. It is one of the earliest cultivated species of the family *Brassicaceae* in the agricultural history. Indian mustard or brown mustard is an economically important oilseed crop, which is grown in almost all parts of India and used as edible oils, vegetables, condiments and fodder. Brassica oil is the third-most important vegetable oil after only soya bean and oil palm. In India it grown under low moisture regimes during the winter/*Rabi* season and is the primary source of edible oil (Sharma *et al.*, 2014) [15]. During 2021, India produced 8.5 million tones of rape-seed mustard from 8.16 lakh hectare area (DRMR, 2022). Despite being the third largest producer (11.3%) of oilseed Brassica after Canada and China in the world, India meets 57 percent of the domestic edible oil requirements through imports and is ranked 7th largest importer of edible oils in the world. Production and productivity in *Brassica species* is affected globally by a number of biotic and abiotic stresses. White rust disease caused by an oomycete fungus *Albugo candida* (Pers.) Kuntze is a serious disease which leads to huge losses (Behra, 2016) [16]. Both the vegetative and reproductive phases of the plants are affected by this fungal pathogen.

Brassica juncea consists of two diverse gene pools, the Indian and the East European gene pool (exotic). The East European gene pool shows more diversity at the molecular level and has more yield potential while the Indian gene pool has narrow genetic diversity with low yield potential (Yadava *et al.*, 2012; Dhaka *et al.*, 2017) [14, 3]. Hence, the present study was conducted to estimate the genetic diversity among the ninety diverse germplasm of Indian and European gene pool of mustard in respect of five characters such as white rust resistance, grain yield/plant, oil content, erucic acid and glucosinolates, as well as grouping the genotypes into different cluters based on these traits for exploiting them in breeding programmes.

Materials and Methods

The field trails were carried out at Experimental Farm, School of Biotechnology, SKUAST-Jammu during *Rabi* seasons from 2019 to 2021. The experiment was conducted in augmented design with ninety diverse genotypes of Indian and East European gene pool (Table 1). White rust resistant (Heera and Donskaja) and susceptible (RSPR-01 and Varuna) genotypes were used as checks and repeated in all the four blocks of 86 genotypes. The data was recorded for five characters such as white rust reaction, grain yield/plant (g), oil content (%), erucic acid (%) and glucosinolates ($\mu\text{mole/g}$). Disease scoring was done according to the method proposed

by Conn *et al.* (1990). Oil content (%) was estimated by Infratech™ 1241 whole Grain Analyzer (Foss Alle, Denmark) based on the principle of near-infrared transmittance technology in wavelength range of 570-1050 nm. Erucic acid (%) was estimated by ^{13}C NMR using JEOL ECA-400 MHz NMR spectrometer at a frequency of 100.40 MHz fitted with a 5-mm-i.d. dual probe (Ming-Kok *et al.*, 2017). Glucosinolate content ($\mu\text{mole/g}$) was estimated by spectrophotometric estimation using methanolic extract. The data was analysed using R-Studio software which grouped genotypes into different clusters.

Table 1: Details of the genotypes used in the study

Genotypes	Country of origin	Genotypes	Country of origin	Genotypes	Country of origin
Neosypajuscajasia 2	Russian Federation	Jubilejnaja	Russian Federation	German accession No. 114	Germany
Volgogradskaja1891	Russian Federation	Skorospelka 2	Russian Federation	Zem 1	Australia
Skorospelka	Russian Federation	PGR 12568	Russian Federation	Skorospieka II	Russia
VNIIMK 351	Russian Federation	74/5	United Kingdom	EC 287711	Sweden
VNIIMK 405	Russian Federation	PGR 12573	Russian Federation	EC 491584	Sweden
Zeltosemiannaja 230	Russian Federation	PGR 12574	Canada	EC 206712	France
Zaria	Russian Federation	Stepniacka	Russian Federation	EC 699059	Spain
Donskaja 4	Russian Federation	J807/1/6	United Kingdom	EC 699038 –I	Spain
PGR 3330	Finland	J/807/12/1	United Kingdom	EC 699038-II	Spain
PGR 3383	Romania	J/817/2	United Kingdom	B.Junceea from Turkey	Turkey
Commercial Brown Mustard	Canada	J/824	United Kingdom	PGR 12585	Pakistan
Lethbridge 22A	Canada	J/824/6	United Kingdom	PAK 85387	Pakistan
Blaze	Canada	PGR 12586	China	PAK 85483	Pakistan
Domo	Canada	Cutlass	Canada	PAK 85506	Pakistan
AC Vulcan	Canada	Scimitar	Canada	PAK 85590	Pakistan
Yanagawa Shirokuki	Japan	SRS 319	Czechoslovakia	PAK 85667	Pakistan
Miike Akachirimem	Japan	I-49-24	Turkey	Toria Mitha	Pakistan
Sendai Bashouna	Japan	M.Br.4	France	Desi Saram	Pakistan
Ooba Takana	Japan	63-0134-68	Spain	Sharsham	Pakistan
Burgonde	Canada	64-1398-69	Spain	PAK 85393	Pakistan
Ekla	Canada	Bass	Unknown	Raya (L.23)	Pakistan
Primus	Canada	R 871	Romania	SB-12-P4	India
Stoke	Canada	Kafiav N Zagora	Bulgaria	RLM-240	India

Genotypes	Country of origin	Genotypes	Country of origin	Genotypes	Country of origin
RLC 1021	India	RSPR-01	India	RSPR-69	India
Krishna	India	Urvashi	India	Pusa Bold	India
Rohini	India	DRMR-2017	India	Heera	Europe
Vardan	India	Pusa Mehak	India	DMR-J-31	India
IB 1479	India	Pusa Karishma	India	RSPR-03	India
IB 1436	India	Varuna	India	RH-749	India
Pusa Tarak	India	Kranti	India	RB-55	India

Results and Discussion

The data recorded with respect to five economically recorded traits is presented in Table 2. Out of 90 accessions, three genotypes (Donskaja IV, Domo and Cutlass) showed no infection and they were considered as immune against white rust. Fifteen genotypes (Skorospelka, VN11MK 351, Zeltosemiannaja 230, AC Vulcan, Yanagawa Shirokuki, Miike Akachirimem, Sendai Bashouna, J807/1/6, J/807/1/6, J/824/6, I-49-24, M.Br.4, Kafiav N Zagora, German accession No. 114, Zem 1, Heera) were considered as highly resistant with disease severity of 5 percent and were given white rust score 1. They belonged to East European gene pool. Another twenty nine genotypes from East European gene pool were resistant with disease severity of 5-10 percent and they were given a score of 3. Twenty four genotypes were considered as moderately resistant with disease severity 11-25 percent and they were given white rust score of 5. Nineteen accessions from Indian gene pool were considered as

susceptible with disease severity 26-50 percent and they were given white rust score of 7.

The grain yield/plant varied from 9.05 to 19.07 g. Maximum grain yield/plant was recorded in the genotype DRMR-2017 (19.07 g) followed by J807/1/6 (18.36 g) and Vardan (17.72 g). The oil content varied from 31.90 to 44.16 percent. Maximum oil content was found in PGR 12573 and EC 287711 (44.16%) followed by Stoke (44.01%) and Jubilejnaja (43.54%). The erucic acid content varied from 12.18 to 45.61 percent. Minimum erucic acid was found in Donskaja IV (12.18%) followed with slightly higher erucic acid content in Volgogradskaja 1891 (12.69%) and PGR 12573 (14.29%). The glucosinolate content varied from 28.92 to 120.41 $\mu\text{mole/gm}$. Minimum glucosinolate content was found in Heera (28.92 $\mu\text{mole/g}$).

Multivariate cluster analysis (Fig.1) divided the whole set of 90 genotypes mainly into two clusters. One (main cluster I) predominantly represented European gene pool while the

other (main cluster II) predominantly represented Indian gene pool (Table 3). The Main cluster I (Cluster 1) had maximum of 67 genotypes. Main cluster II was further sub-divided into 3 clusters (2, 3, 4 and 5). Cluster 2 had 17 genotypes while cluster 3 and 4 had 3 and 2 genotypes, respectively. Heera was the only genotype in cluster 5.

An analysis of inter cluster distance (Table 4) was done to know about diversity among the clusters formed. The maximum cluster distance (83.27) was observed between cluster 3 and cluster 5 followed by cluster 2 and cluster 5 (29.20) and between cluster 1 and cluster 3 (57.72). It indicated maximum genetic diversity between these pair of

clusters; and possibility of developing high yielding, better quality and white rust resistant genotypes through hybridization between genotypes belonging to contrasting clusters. Sinha and Singh (2004) [11] and Thul *et al.* (2004) [12], Monalisa *et al.* (2005) [9], Malik *et al.* (2006) [8], Ahmad *et al.* (2009) [1], Kumar *et al.* (2013) [6], Kumari *et al.* (2018) [7], Rout *et al.*, (2019) [10], Chaturvedi *et al.*, (2021) [2], Gupta *et al.* (2021) [4] and Vanukuri *et al.* (2022) [13] also carried out genetic divergence studies in *Brassica juncea* and grouped mustard genotypes different clusters for ascertaining genetic variability in the germplasm and identification of suitable parents for utilization in breeding programmes.

Table 2: Comparative performance of *Brassica juncea* germplasm for economically important traits

Genotypes	GY/P (g)	WRS (0-9)	OC (%)	EA (%)	GC (μ mole/g)	Genotypes	GY/P (g)	WRS (0-9)	OC (%)	EA (%)	GC (μ mole/g)
Neosypajuscajasia 2	10.31	3	38.86	18.82	51.32	Primus	14.06	3	41.79	16.29	50.13
Volgogradskaja1891	13.79	3	39.63	12.69	54.88	Stoke	13.13	3	44.01	22.42	48.94
Skorospelka	13.99	1	41.81	16.73	48.94	Jubilejnaja	10.35	3	43.54	28.92	52.50
VNIIMK 351	9.85	1	38.42	15.98	50.13	Skorospelka 2	15.24	3	43.29	18.64	51.32
VNIIMK 405	11.38	3	41.67	15.64	54.88	PGR 12568	11.75	5	40.14	27.94	58.45
Zeltosemiannaja 230	12.01	1	40.23	18.29	59.64	74/5	13.8	5	42.30	17.64	57.26
Zaria	13.99	3	41.24	29.84	47.75	PGR 12573	14.14	3	44.16	14.29	58.64
Donskaja 4	14.97	0	40.33	12.18	52.83	PGR 12574	15.81	3	39.26	22.14	55.58
PGR 3330	9.05	3	39.20	16.64	53.20	Stepniacka	13.56	3	38.16	15.69	54.88
PGR 3383	13.31	3	40.09	16.29	58.45	J807/1/6	18.36	1	40.82	21.19	58.83
Commercial Brown Mustard	13.00	3	38.59	19.16	59.01	J/807/12/1	16.1	3	40.45	25.94	59.02
Lethbridge 22A	10.75	3	41.29	18.29	54.39	J/817/2	11.52	3	42.14	32.19	42.64
Blaze	9.96	3	42.46	27.04	57.96	J/824	9.66	3	42.29	42.69	49.92
Domo	15.6	0	35.28	16.79	50.13	J/824/6	11.06	1	41.10	30.49	53.64
AC Vulcan	16.16	1	37.14	36.19	51.32	PGR 12586	14.34	3	39.45	19.52	59.64
Yanagawa Shirokuki	16.27	1	42.29	42.01	52.50	Cutlass	17.37	0	39.52	25.10	48.14
Miike Akachirimem	17.08	1	41.42	20.29	56.06	Scimitar	15.50	3	41.08	27.92	55.64
Sendai Bashouna	13.34	1	40.16	20.96	47.75	SRS 319	17.71	3	37.54	17.46	49.24
Ooba Takana	9.66	5	41.29	14.64	48.94	I-49-24	11.34	1	39.89	19.33	56.69
Burgonde	12.57	5	40.64	22.29	53.69	M.Br.4	17.41	1	35.69	21.52	49.24
Ekla	13.91	3	42.13	28.64	52.51	63-0134-68	12.89	3	40.19	22.12	53.09
Genotypes	GY/P (g)	WRS (0-9)	OC (%)	EA (%)	GC (μ mole/g)	Genotypes	GY/P (g)	WRS (0-9)	OC (%)	EA (%)	GC (μ mole/g)
64-1398-69	13.25	5	41.20	36.24	58.93	Raya (L.23)	16.72	7	41.16	39.28	60.64
Bass	9.26	3	36.29	21.14	53.64	SB-12-P4	13.82	5	37.97	40.64	89.64
R 871	15.71	3	38.16	15.93	55.34	RLM-240	11.13	5	39.42	35.93	89.24
Kafiav N Zagora	14.61	1	37.29	16.14	59.61	RLC 1021	16.65	7	36.29	38.49	95.64
German accession No. 114	16.57	1	38.54	23.69	49.24	Krishna	16.56	7	35.16	35.29	90.21
Zem 1	9.35	1	42.14	27.24	45.64	Rohini	14.90	7	37.29	34.18	92.14
Skorospieka II	9.68	3	39.24	35.14	49.24	Vardan	17.72	5	38.14	29.59	95.64
EC 287711	12.86	5	44.16	34.36	50.09	IB 1479	15.80	5	39.29	45.61	100.29
EC 491584	12.56	5	38.56	39.59	52.16	IB 1436	13.65	5	38.98	31.49	98.16
EC 206712	12.36	5	39.29	37.61	48.29	Pusa Tarak	14.50	7	40.23	39.26	99.24
EC 699059	12.99	5	38.19	34.66	55.69	RSPR-01	15.29	7	39.27	34.64	84.19
EC 699038 -I	13.20	3	40.16	28.21	58.12	Urvashi	16.90	5	41.40	43.19	89.32
EC 699038-II	12.56	3	42.55	31.15	59.84	DRMR-2017	19.07	7	40.92	42.21	92.12
B.Juncea from Turkey	10.15	7	39.71	27.64	55.29	Pusa Mehak	17.14	7	39.28	44.64	102.14
PGR 12585	10.04	7	40.99	15.93	65.61	Pusa Karishma	14.22	5	31.90	21.29	100.96
PAK 85387	9.84	5	40.11	36.64	64.12	Varuna	12.25	7	34.23	33.24	118.29
PAK 85483	12.57	5	39.50	31.29	62.64	Kranti	12.13	5	35.69	30.64	115.29
PAK 85506	12.91	7	40.35	38.45	68.24	RSPR-69	12.74	5	38.03	40.29	104.24
PAK 85590	14.45	7	40.96	19.29	63.04	Pusa Bold	14.84	7	39.57	34.64	108.64
PAK 85667	16.00	7	35.29	26.24	65.29	Heera	11.26	1	38.29	16.64	28.92
Toria Mitha	16.13	7	34.15	35.14	60.06	DMR-J-31	13.73	5	42.62	26.94	89.29
Desi Saram	16.06	5	38.68	32.64	58.98	RSPR-03	15.46	5	39.87	29.64	90.21
Sharsham	17.14	5	39.37	27.29	63.64	RH-749	13.60	7	37.54	24.22	120.41
PAK 85393	14.70	7	35.29	45.16	61.94	RB-55	13.22	7	41.09	20.24	102.21

GY/P: Grain yield/plant; WRS: White rust score; OC: Oil content; EA: Erucic acid; GC: Glucosinolate content

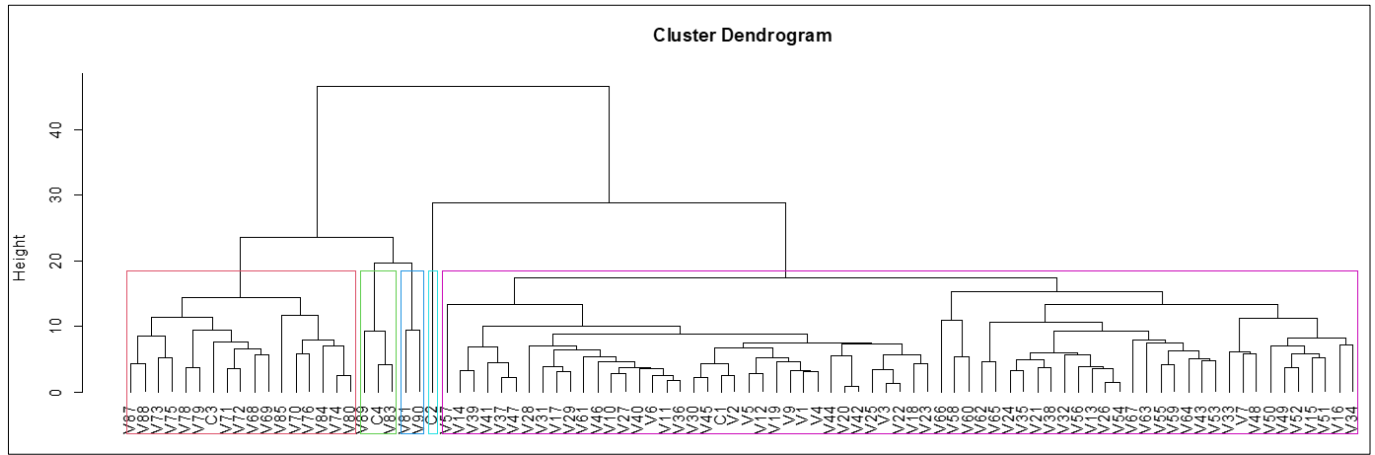


Fig 1: Cluster diagram showing distribution of different genotypes

Table 3: Distribution of ninety genotypes of *Brassica juncea* in five clusters

No. of clusters	No. of genotypes	List of germplasm
Cluster 1	67	Burgonde, 63-0134-68, PGR 12574, Miike Akachirimen, I-49-24, Lethbridge 22A, 74/5, Commercial Brown Mustard, PGR 12586, Zeltosemiannaja 230, PGR 3383, VNIIMK 405, Stepniacka, R 871, Kafiov N Zagora, PGR 3330, Neosypajuscjasia 2, Volgogradskaja 189/191, Skorospelka 2, Primus, PGR 12573, Donskaja 4, Skorospelka, VNIIMK 351, Bass, J807/1/6, Domo, SRS 319, Sendai Bashouna, Ooba Takana, Stoke, German accession No. 114, M.Br.4, PAK 85590, J/807/12/1, Cutlass, Scimitar, B.Juncea from Turkey, EC 699038 –I, PGR 12568, Blaze, Ekla, Jubilejnaja, J/824/6, Zaria, Zem 1, EC 699038-II, Sharsham, Desi Saram, PAK 85483, PGR 12585, PAK 85667, EC 699059, EC 287711, 64-1398-69, Skorospieka II, AC Vulcan, Toria Mitha, J/817/2, EC 206712, EC 491584, PAK 85387, Raya (L.23), Yanagawa Shirokuki, J/824, PAK 85393, PAK 85506
Cluster 2	17	IB 1479, Pusa Mehak, Pusa Tarak, RSPR-69, RLC 1021, DRMR-2017, SB-12-P4, Urvashi, Rohini, Krishna, RLM-240, IB 1436, Vardan, RSPR-03, RSPR-01, DMR-J-31, Pusa Bold
Cluster 3	3	Varuna, Kranti, RH-749
Cluster 4	2	RB-55, Pusa Karishma
Cluster 5	1	Heera

Table 4: Genetic distances between different clusters of *Brassica juncea* genotypes

Cluster	Cluster 1	Cluster 2	Cluster 3	Cluster 4	Cluster 5
Cluster 1	12.60				
Cluster 2	43.13	11.83			
Cluster 3	57.72	23.51	14.99		
Cluster 4	21.39	37.81	56.54	13.47	
Cluster 5	28.23	69.20	83.27	39.47	0.00

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