Genetic Variability for Physiological Characters and their Relationships with Seed Yield and its Components in Indian Mustard [Brassica juncea (L.) Czern. & Coss.] under Drought

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Evaluation of Indigenous and Exotic Germplasm of Indian Mustard [*Brassica juncea* (L.) Czernj & Cosson] for Morpho-Physiological and Quality Characters

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Sixty germplasm accessions from India (27), Australia (25) and China (8) along with 5 check varieties (Bio-902, Bio-772, PCR-7, Rohini and Varuna) of Indian mustard [*Brassica juncea (*L.) Czernj. & Cosson] were grown in an augmented block design with four blocks during *rabi* season of 2007-08 to assess magnitude and nature of variability for morpho-physiological and quality characters. Significant mean sum of squares due to genotypes indicated presence of substantial variability for all the morpho-physiological characters investigated except protein content. Seed yield/plant, secondary branches/plant, biological yield, 1000-seed weight, specific leaf weight at 50% and full flowering, leaf area index at full flowering and total dry matter at 50% flowering had high genetic variability and could be exploited through selection. The Chinese accessions were very late in maturity due to fact that they were selected under long day conditions and no seed could be harvested. Indian varieties such as GM-2, RGN-13, JM-3, VSL-5 and Australian accession JM-018 showed very low saturated fatty acids (<2 %). Oleic acid content in the indigenous germplasm was low varying from 6.0-18.1 % and all had high erucic acid in the range of 31.5% (Basanti)-52.5% (GM-2). The Australian accessions exhibited low erucic acid (< 2 %) and high oleic acid (29.7-58.4%). Glucosinolate content in the indigenous germplasm ranged from 75-136.6 µ moles/g defatted seed meal. In the Australian accessions it was up to 30 µ moles/g defatted seed meal. Several potentials accessions were identified from exotic as well as indigenous germplasm for utilization in the breeding programme.

Key Words: *Brassicca juncea*, Genetic resources, Indian mustard, Physiological characters, Promising accessions, Quality characters, Variability

Introduction

Genetic resources of any crop species constitute the reservoirs of new and valuable genes, which could be of immense help in the varietal improvement programme. The precise evaluation of genetic resources of rapeseed-mustard would play a pivotal role in breeding programme by continuously providing new genes for the target character(s). Valuable donors for various biotic, abiotic stresses, agro-morphological characters have been identified among the indigenous Indian mustard germplasm (Chauhan *et al.*, 2005, 2006; Misra *et al.*, 2007). However, limited variability existed for oil and seed meal quality characters. Exotic Indian mustard germplasm from Canada, China and Australia were reported to possess low erucic, high oleic and low glucosinolate content (Chauhan and Kumar, 2007). Furthermore, use of exotic germplasm along with indigenous ones often results in the increased genetic variability for desired selection. Khalf *et al.* (1984) reported that there was greater variability for yield in populations where exotic genotypes contributed to the crosses. The present investigation attempts to assess the magnitude and nature of variability, heritability and genetic advance in indigenous and exotic germplasm of Indian mustard for morpho-physiological, oil and seed meal quality characters.

Materials and Methods

The materials for the present investigation comprised 60 germplasm accessions from India (27), Australia (25), China (8) and 5 check varieties of Indian mustard [*Brassica juncea* (L.) Czernj & Cosson]. There were grown in an augmented block design (Federer, 1956) with 4 blocks during *rabi* season of 2007-2008. In each block, 15 accessions and 5 checks were grown. There were 3 rows of 5 m length for each accession in a block. The row spacing was 30 cm and plant spacing within a row was maintained at 10 cm by thinning. A fertilizer dose of 40: 40: 20 kg/ha (N: P_2O_5: K_2O) was applied at the time of sowing and 40 kg N/ha was applied after first irrigation (36 days after sowing). Second irrigation was

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applied at 72 days after sowing. Standard plant protection measures were adopted as and when required.

To study different growth parameters, plant samples from 30 cm running length were harvested above ground level at 50% flowering and full flowering stages. The shoot and leaves were separated and leaf area was recorded by leaf area meter (LICOR: LI 3000 A). Shoot and leaves were dried in an oven ($65^o \pm 2^o$ C) for at least 72 h till constant weight was achieved. The samples were weighed and total dry matter (TDM) of leaves, shoot as well as shoot + leaves was expressed in g/m^2. Fourth fully expanded leaf from the top was chosen from three randomly selected plants in each block for recording photosynthesis and transpiration by photosynthesis system (CIRAS-2). On the same leaves chlorophyll meter readings (SCMR) were recorded by SPAD chlorophyll meter (Konica Minolta: SPAD-502). Leaf area index (LAI) and specific leaf weight (SLW) were computed using leaf area and dry weight of leaves (Radford, 1967). Transpiration quotient (TQ) was measured as ratio of transpiration to photosynthesis.

Days to maturity (DM) were recorded on plot basis. At the time of harvest, 10 randomly competitive plants were taken from the middle row to record plant height (PH, cm), primary branches/plant (PB, no.), secondary branches/plant(SB, no.), main shoot length (MSL, cm), siliquae on main shoot (SMS, no.), siliqua length (SL, cm),seeds/siliqua (SS, no.), 1000-seed weight (SW, g), seed yield/plant (SY, g), biological yield/plant (BY, g) and harvest index (HI, %). Oil (OC) and protein content (PC) were recorded on a composite sample of 10 plants taken for recording observations on seed yield and related characters using NIRS (Kumar *et al*., 2003). Two plants were selfed in each accession and seeds were harvested separately. The fatty acid profile was analysed using selfed seeds (Paquot and Hautfenne, 1987) and open-pollinated seeds of the same plant was used for glucosinolate analysis (Kumar *et al*., 2004).

The mean data for agro-morphological and physiological characters were subjected for analysis of variance of an augmented block design as suggested by Federer (1956). The data were analyzed using software *SPAD* (IASRI, New Delhi). Genotypic and phenotypic coefficients of variation, heritability (in broad-sense) and genetic advance expressed as percentage of mean were computed following Johnson *et al.* (1955), Hanson *(*1963) and Lush (1949), respectively.

Results and Discussion

The ANOVA indicated significant differences among the checks for DM and BY and highly significant differences for PH, SB, MSL, SMS, SL, SW, SY and OC. The analysis of variance also revealed highly significant differences among the test genotypes for DM, PH, SB, MSL, SMS, SL, SW, SY, HI and OC. The mean sum of squares due to genotypes for BY was significant. The mean sum of squares due to checks *vs* test genotypes were highly significant for PH, SB, MSL, SL, SW, OC and PC. The differences among the checks *vs* test genotypes were significant for BY and SY (Table 1).

Table 1. Analysis of variance for seed yield and morphological characters in Indian mustard (recorded on 35 entries and 5 checks)

Source of variation	DF	Mean sum of squares						
		DM	PH	SB	MSL	SMS	SL	SW
Block	3	1.517	126.102*	4.305	24.994	13.012	0.011	0.157*
Entries	59	3.798**	278.764**	20.792**	84.637**	49.041**	0.243**	0.557**
Checks	4	3.700*	842.47**	15.733**	156.534**	125.213**	0.120**	5.709**
Test genotypes	54	3.863**	238.067**	21.140**	65.945**	43.962**	0.203**	1.000**
Checks vs test genotypes	1	0.875	99.458**	23.25**	750.320**	3.413	0.740**	13.348**
Error	12	0.933	29.283	2.256	20.357	10.656	0.025	0.039
Critical difference (CD)		–	13.95	3.87	11.63	–	0.41	0.51
Genotypes vs checks		–	**19.56**	**5.43**	**16.31**	–	**0.57**	**0.72**
Checks vs checks		1.49	8.34	2.31	6.95	5.03	0.25	0.31
		2.08	**11.69**	**3.24**	**9.75**	**7.05**	**0.34**	**0.43**
Genotypes within same block		2.98	16.68	4.63	13.90	10.06	0.49	0.61
		4.17	**23.38**	**6.49**	**19.49**	**14.10**	**0.69**	**0.86**
Genotypes in different blocks		3.26	18.27	5.07	15.23	11.02	0.54	0.67
		4.57	**25.61**	**7.10**	21.35	**15.45**	**0.75**	**0.94**

Cont ....

Table 1. Cont ....

Source of variation	DF	Mean sum of squares				
		BY	SY	HI	OC	PC
Block	3	573.366*	30.855**	20.787	0.181	0.703*
Entries	56	349.157*	25.665**	32.735**	1.104**	0.448*
Checks	4	489.699*	20.048**	14.538	1.305**	0.327
Test genotypes	51	327.964*	25.966**	34.680**	1.052**	0.365
Checks vs test genotypes	1	867.827*	32.745*	6.306	2.916**	5.152**
Error	12	128.437	5.47	6.735	0.293	0.163
Critical difference (CD)		29.22	6.03	-	1.40	1.04
Genotypes vs checks		**40.97**	**8.46**	-	**1.96**	**1.46**
Checks vs checks		17.46	3.60	-	0.84	0.62
		24.48	**5.05**	-	**1. 17**	**0.87**
Genotypes with in same block		34.92	7.21	8.00	1.67	1.25
		48.96	**10.11**	**11.21**	**2.34**	**1.75**
Genotypes in different blocks		38.26	7.90	8.76	1.83	1.37
		53.64	**11.07**	**12.28**	**2.57**	**1.91**

*, ** Significant at 5% and 1% probability level, respectively, Values in light and bold face indicate CD at 5% and 1% probability level, respectively.

The checks also showed highly significant differences for SLW and SCMR and significant differences for TQ at 50% flowering. The genotypic differences were highly significant for SLW at 50% and full flowering, total dry matter (TDM) at full flowering. There were significant differences for LAI at full flowering and TQ at 50% and full flowering and SCMR at 50% flowering. The genotypes had significant differences for TQ at full flowering (Table 2). The test genotypes had significant to highly significant differences from the checks for all the physiological characters except TQ at full flowering, TDM and SCMR at 50% flowering. The significant mean sum of squares indicated presence of substantial variability in the experimental materials for all the morpho-physiological characters investigated except protein content.

The experimental materials showed high genotypic and phenotypic variability for seed yield/plant, secondary branches/plant, biological yield/plant, 1000-seed weight, specific leaf weight at 50% and full flowering, LAI at full flowering and TDM/plant at 50% flowering. The harvest index, main-shoot length, siliquae on main shoot, siliqua length and transpiration quotient at full flowering also exhibited moderate genetic variability (Table 3). The findings of the present investigation confirmed the earlier reports of high variability for 1000-seed weight, siliquae on main shoot, seed yield/plant (Meena *et al.*, 2006; Patel *et al.*, 2006), total biomass and harvest index (Singh *et al.*, 2006); secondary branches/plant (Meena *et al.*, 2006). Low variability as observed in the present investigation for days to maturity, plant height and oil content were also recorded by Misra *et al.* (2007). But Sikarwar *et al.* (2000) observed low variability for primary and secondary branches/plant and Meena *et al.* (2000) reported high variability for days to maturity. Variable trend of genetic variability in the present study and the earlier reports could be due to differential genetic background of the experimental materials and/or genotype x environmental (g x e) interactions. No published report dealing with genotypic variation for physiological characters in Indian mustard was available to support or contradict the findings of the present investigation. Presence of moderate to high genetic variability in morpho-physiological characters recorded in the present investigation could be exploited through selection.

The Chinese accessions were very late in maturity, flowering was initiated in mid-February and seeds could not be harvested. This could be due to fact that they were selected under long day conditions in China. As a matter of fact some of the accessions like Berry (EC597329), Loiret (EC597326) and Ekla (EC597327) appeared to be introduced from Canada to China. In earlier studies, similar trend of maturity and segregation

Table 2. Analysis of variance for some physiological characters in Indian mustard (recorded on 35 entries including 5 checks)

Source of variation	Mean sum of squares							
	DF	LAI	SLW		TDM	TQ		SCMR
		Full flowering	50% flowering	Full flowering	50% flowering	50% flowering	Full flowering	50% flowering
Block	3	0.113	0.834	7.827**	3. 673	0.006**	0.0029	4.266
Entries	34	0.983*	4.453**	2.963**	52.848**	0.006	0.0027*	6.447**
Checks	4	0.744	4.314**	0.874	21.107	0.006*	0.0009	26.008**
Test genotypes	29	0.993*	4.594**	3.026**	58.803**	0.005*	0.0030*	3.870*
Checks vs test genotypes	1	3.236**	1.676*	8.143**	16.079	0.003**	0.0049	2.889
Error	12	0.373	0.304	0.850	11.177	0.001	0.0011	1.484
Critical difference (CD)		1.57	1.42	2.37	8.61	0.033	–	–
Genotypes *vs* checks		**2.20**	**1.99**	**3.33**	**12.08**	**0.047**	–	–
		–	0.85	–	–	0.020	–	1.87
Check vs check		–	**1.19**	–	–	**0.028**	–	**2.63**
		1.88	1.70	2.84	10.30	0.040	0.010	3.75
Genotypes within same block		**2.64**	**2.38**	**3.98**	**14.44**	**0.056**	**0.14**	**5.26**
		2.06	1.86	3.11	11.28	0.044	0.11	4.11
Genotypes in different blocks		**2.89**	**2.61**	**4.36**	**15.82**	**0.062**	**0.16**	**5.76**

*, ** Significant at 5 % and 1 % probability level, respectively, Values in light and bold face indicate CD at 5 % and 1 % probability level, respectively.

Table 3. Range, mean, phenotypic (PCV) and genotypic (GCV) coefficient of variability for morpho-physiological characters in Indian mustard

Character	Range	Mean ± SEM	PCV (%)	GCV (%)
Days to maturity	133.6 - 143.6	141.0 ± 1.0	1.4	1.2
Plant height (cm)	126.0 - 200.4	165.0 ± 5.4	9.4	8.8
Secondary branches/plant (no.)	3.0 - 28.8	11.9 ± 1.5	38.8	35.5
Main shoot length (cm)	37.2 - 75.8	62.2 ± 4.5	13.0	11.2
Siliquae on main shoot (no.)	33.3 - 64.1	45.9 ± 3.3	14.4	12.6
Siliqua length (cm)	2.2 - 4.4	3.6 ± 0.2	12.7	12.3
1000- seed weight (g)	1.8 - 6.4	3.8 ± 0.2	26.2	25.7
Biological yield / plant (g)	11.3 - 101.0	56.4 ± 11.3	32.0	26.1
Seed yield/plant (g)	2.7 - 24.9	15.0 ± 2.3	33.9	31.0
Harvest index (%)	15.2 - 39.1	27.0 ± 2.6	21.8	19.4
Oil content (%)	36.1 - 41.2	38.8 ± 0.5	2.6	2.2
LAI - Full flowering	0.70 – 4.8	2.33 ± 0.61	41.4	29.3
SLW (mg/cm^2) - 50% flowering	1.5 – 12.8	3.73 ± 0.55	57.4	53.2
Full flowering	3.2 – 10.4	6.40 ± 0.92	27.2	24.4
TDM/plant (g) 50% flowering	4.8- 35.8	13.51 ± 3.34	56.8	49.4
TQ (m moles/µ mole) -50% flowering	0.23-0.38	0.34 ± 0.01	6.5	5.4
Full flowering	0.27- 0.56	0.37 ± 0.03	14.7	11.7
SCMR - 50% flowering	44.24 – 52.10	47.23 ± 1.21	4.2	3.3

Table 4. Promising Accessions identified for seed yield and physiological characters

Character	Best Check	Accessions				
Days to maturity (< 139 days)	PCR-7 (139)	Kranti	JM-06011 (EC-597316)	JM-06004 (EC-597312)	JM- 06010 (EC-597315)	JM- 06002 (EC-597310)
Plant height (< 165 cm)	Varuna (164.5)	JM-06014 (EC-597319)	JM- 06013 (EC-597318)	JM- 06012 (EC-597317)	JM- 06018 (EC-597321)	JM- 06011 (EC-597316)
Secondary branches/plant (>12.8)	Varuna (12.8)	JM- 06002 (EC-597310)	JM- 06003 (EC-597311)	JM- 06019 (EC-597322)	Ashirwad	RGN- 13
Main shoot length (>71.5 cm)	Varuna (71.5)	JM- 1	VSL - 5	Ashirwad	JR- 049 (EC-552584)	Geeta
Siliquae on main shoot (>52.0)	Bio-772 (52.0)	Geeta	JM- 06002 (EC-597310)	JM- 06010 (EC-597315)	VSL- 5	JM- 1
Siliqua length (>4.0 cm)	Varuna and Bio-772 (4.0)	Laxmi	JM- 1	CS –54	Basanti	JM- 009 (EC-552582)
1000-seed weight (> 6.6 g)	Bio-902 (6.6)	Urvashi				
Biological yield/plant (>72.6 g)	Varuna (72.6)	JM- 06010 (EC-597315)	JM- 06003 (EC-597311)	JM- 1	JM- 06002 (EC-597310)	GM- 2
Seed yield/plant (>18.7 g)	Varuna (18.7)	JM- 06003 (EC-597311)	JM- 06010 (EC-597315)	JR- 042 (EC-552583)	GM- 2	Ashirwad
Harvest index (>29.3%)	Bio 902 (29.3)	Pusa mahak	JM- 06014 (EC-597319)	JN- 004 (EC-552573)	JM- 016 (EC-552579)	JM- 3
Oil content (>39.3%)	Rohini (39.3)	Kranti	CS- 52	JN- 004 (EC-552573)	JM- 033 (EC-552578)	JM- 032 (EC-552577)
LAI full flowering (>2.3)	Rohini (2.3)	VSL- 5	Swaran Jyoti	CS-62	JM- 2	JM- 06006 (EC-597313)
SLW (mg/cm^2) 50% flowering (>5. 3)	Bio-902 (5.3)	Datonghuangy oucai (EC-597337)	Ekla (EC-597327)	Berry (EC-597329)	RH-13 (EC-597330)	Qianxianjiecai (EC-597338)
Full flowering (>7.3)	Varuna (7.3)	Berry (EC-597329))	Qianxianjiecai (EC-597338)	Loiret (EC-597326)	RH-13 (EC-597330)	Ekla (EC-597327)
TDM/plant (g) 50 % flowering (>15.9 g)	Varuna (15.9)	Qianxianjiecai (EC 597338)	Berry (EC-597329)	Datonghuangy-oucai (EC-597337)	Loiret (EC-597326)	Ekela (EC-597327)
TQ (mmoles / µmole) 50 % flowering (< 0.34)	Bio-902 (0. 34)	JM-3	NDYR-8	JM-1	VSL-5	Pusa mahak
Full flowering (< 0.36)	PCR-7 (0.36)	VSL-5	JM-1	RH-13 (EC-597330)	Vasundhra	CS-52
SCMR 50% flowering (>50.8)	Bio-902 (50.8)	VSL-5				

* Within parenthesis is the actual mean value (adjusted).

for various morphological traits in 30 exotic rapeseed-mustard strains from Sweden was observed because of breeding and selection under long day conditions. The Chinese accessions were extremely tall, had excellent vigour, produced few secondary branches and showed compact plant type. They could be evaluated for certain physiological characters only. The Australian accessions were also slightly late in maturity but comparable to indigenous accessions. Based on the mean performance, the top five promising accessions from indigenous as well as exotic germplasm each for different morpho-physiological characters were identified (Table 4).

The indigenous accessions predominnantly had long main shoot, long siliqua, high seed weight, oil content

and leaf area index at full flowering. The Australian accessions appeared to be good sources of short plant stature, secondary branches/plant, siliquae on main shoot, biological yield/plant as well as seed yield/plant. Pusa Mahak and three Australian accessions exhibited high harvest index. The Chinese accessions seemed promising for specific leaf weight and total dry matter/ plant. The accessions- Qianxianjiecai (EC597338), Berry (EC597329), Loiret (EC597326), RH-13 (EC597330), Datonghuangyoucai (EC597337) and Ekla (EC597327) showed thicker leaves as indicated by high SLW. Since SLW has been reported to be associated with water use efficiency in groundnut (Nageswara Rao *et al.*, 1993) it would be quite interesting to study these accessions in detail for drought tolerance in terms of osmotic adjustment, water use efficiency and root characters. Further, Indian varieties, VSL-5, JM-1, Vasundhra, CS-52 and RH-13 (EC597330) from China showed low transpiration quotient, *i.e.*, high water use efficiency.

In the present investigations the saturated fatty acids in the genotypes were in the desirable range (< 7%) and some Indian varieties such as GM-2, RGN-13, JM-3, VSL-5 and Australian accession JM-018 showed very low saturated fatty acids (< 2 %). An Australian accession, JM-06011 had slightly higher amount (10.1%) of saturated fatty acids. Oleic acid is very important for increased shelf life as it reduces photo-oxidation as well as for reducing the blood cholesterol (Grundy, 1986). Oleic acid content in the indigenous germplasm was low varying from 6.0-18.1% and all had high erucic acid in the range of 31.5% (Basanti)-52.5% (GM-2). All the Australian accessions exhibited low erucic acid (<2%) and high oleic acid (29.7-58.4%). The investigated genotypes showed wide variation for linoleic acid. This fatty acid was lower in indigenous accessions (12.5-28.8%) than that of exotics (7.8-49.7%). Preferred ratio of oleic: linoleic acid in edible oil should be at least 2.0, Australian accessions JM-06001 and JM-06002 had very high ratio (5.0-6.0) but in Indian accessions such ratio was only 0.75. Similarly, linolenic acid also showed considerable variability in the germplasm investigated and exotics had higher amount (11.9-29.3%) as compared to indigenous ones (9.0-23.0%). Similar pattern of fatty acids had been reported earlier in Indian mustard varieties and exotic germplasm from Canada (Chauhan *et al.*, 2006, 2007). Since oleic acid is the precursor for the synthesis of eicosenoic and erucic acid through chain elongation and also of linoleic and linolenic acid through desaturation pathway (Jonsson, 1977) and any reduction in erucic acid in oil would simultaneously increase oleic, linoleic and linolenic acid. Therefore, high amount of erucic acid in Indian varieties obviously caused reduction in oleic acid while, low erucic acid in Australian accessions resulted in concomitant high level of oleic acid. Glucosinolate content in the indigenous germplasm ranged from 75-136.6 µ moles/g defatted seed meal. In the Australian accessions, it was up to 30 µ moles/g defatted seed meal. These accessions could serve as valuable sources of low glucosinloate in the Indian breeding programme. Indigenous germplasm and varieties of Indian mustard have also been reported to have high glucosinolate content (Chauhan *et al*., 2007). Potential donors for different morpho-physiological characters, desirable fatty acid profile and low glucosinolate content were identified (Tables 4 and 5).

The present gene pool of Indian mustard investigated proved to be quite valuable and several potentials accessions were identified from exotic as well as indigenous germplasm for possible utilization in the breeding programme. The Chinese accessions appeared

Table 5. Promising accessions identified for desirable fatty acid and glucosinolate content

Character	Accessions				
Palmitic + stearic acid (< 2.0 %)	GM-2	RGN-13	JM-3	JM-018 (EC552580)	VSL-5
Oleic acid (> 37%)	JM-06002 (EC597310)	JM-06001 (EC597309)	JN-033 (EC552578)	JM-06020 (EC597323)	JR-042 (EC552583)
Linoleic acid (15-20%)	JM-1	Arawali	CS-52	CS-54	NDYR-8
Linolenic acid (5-10 %)	VSL-5	Ashirwad	GM-2	Laxmi	JM-3
Eurcic acid (< 2 %)	JM-06002 (EC597310)	JM-06004 (EC597312)	JM-016 (EC552579)	JN-010 (EC552574)	JM-06020 (EC597323)
Glucosinolate content (< 25µmoles/g defatted seed meal)	JM-06001 (EC597309)	JM-018 (EC552580)	JM-016 (EC 552579)	JR- 042 (EC 552583)	JM-0009 (EC552582)

quite different but very late and needs to be grown for at least one/two seasons more for identifying better adapted segregating plants under Indian conditions. All the Australian accessions would serve as an important reservoir of gene(s) for low erucic acid, high oleic acid, high oleic:linoleic acid ratio and low glucosinolate content. It is also expected that utilization of this gene pool would lead to broadening of the genetic base of Indian cultivars because of its divergent nature.

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