



DEVELOPMENT AND EVALUATION OF DOUBLE LOW QUALITY LINES IN INDIAN MUSTARD (*Brassica juncea* L. Czern & Coss)

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SUMMARY

Indian mustard (*Brassica juncea* L. Czern & Coss) is an important source of edible oil and meal in India. However, traditional Indian mustard varieties accumulate high amount of erucic acid and glucosinolate in their seeds. These quantitatively inherited anti-nutritional factors drastically reduce the utility of mustard derived seed oil and meal for consumption purposes. The present study aimed at development and evaluation of double low quality lines (seed oil with < 2% erucic acid and glucosinolate content < 30 μ moles/gram defatted seed meal) in Indian mustard. A total of 11 double low lines were selected from among a pool of 1200 lines in F₇ generation. Amongst the selected lines, 2 lines BPRQ-2-1-5 and BPRQ-2-2-11 were found to be highly promising in terms of oil quality and yield performance. Analysis of variance for selected lines indicated significant differences for plant height, shoot length, number of branches, number of siliquae, number of seeds/siliqua, seed yield/plant, 1000 seed weight, oil content, fatty acid composition and glucosinolate content, indicating sufficient variability for effective selection. High heritability estimates (broad sense) were observed for oleic acid (93.0%), oil (92.9%), linoleic acid (91.4%) content and 1000-seed weight (91.3%). Genetic advance ranged from 1.0% (seed yield/plant) to 46.0% (oleic acid content). Oleic acid content showed high heritability (93.0%). A negative and significant correlation was obtained between oleic and erucic acid, indicating the possibility of reducing the erucic acid content by enhancing the level of oleic acid by an increased breeding intensity utilizing potential donors. Sequence-tagged microsatellite site (STMS) analysis of selected double low lines along with canola quality genotype 'Heera' as well as a high yielding popular variety 'Kranti' clearly distinguished quality and non-quality lines. In conclusion, present study reports the development of 2 highly promising double low lines of Indian mustard which may be released as a variety after multi-location testing or used as potential donors for transferring double low quality traits in high yielding varieties.

Keywords: *Brassica*, erucic acid, glucosinolate, double low, yield performance, STMS analysis

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INTRODUCTION

Indian mustard (*Brassica juncea* L. Czern & Coss) is the second largest oilseed crop in India after soybean. It accounts for nearly 30% of the total oilseeds and 27% to edible oil pool of the country (Sutariya *et al.*, 2011). The oil obtained from the seeds of Indian mustard is mainly used

for edible purposes. The suitability of seed oils for human consumption is determined largely by its fatty acid composition. Seed oils having higher proportion of 16 and 18 carbons unsaturated fatty acids, particularly monounsaturated fatty acids (MUFAs) are considered suitable for use as edible oil (Simopoulos, 2008; Ramos *et al.*, 2009). Indian

mustard seed contains 35-45% oil having 92-98% triacylglycerol of fatty acids (C16-C22). It contains lowest saturated fat and possesses more proportion of linoleic (C18:2) and linolenic (C18:3) acid which are not synthesized by the human body. Linolenic acid is an essential dietary fatty acid, but undesirable in edible oil because of prone to auto-oxidation resulting in off-flavours and reduced shelf life of the oil. Erucic acid (C22:1) comprises nearly 50% of total fatty acid which is undesirable for human consumption as they are reported to impair myocardial conductance and increase blood cholesterol. Therefore, reduction in linolenic and erucic acids is one of the important objectives in quality amelioration of Indian mustard seed oil.

The meal remaining as a by-product after extraction of oil is another valuable commodity obtained from Indian mustard. It possesses about 40.0% protein (dry weight after oil extraction) with a favourable composition of amino acids, including comparatively high contents of the essential amino acids, methionine and cysteine (Downey and Bell, 1990). In addition, it is also rich in minerals (particularly Ca, Mg and P) and contains vitamin B₄ and E. Presently, Indian mustard seed meal is largely used as animal feed but can also be utilized for production of value-added products (Bala and Singh, 2012). However, in comparison to other popular sources such as soybean, it contains high amount of glucosinolate, which lessen its feed value (Yoshie-Stark *et al.*, 2008; Wanasundara, 2011). The amelioration of nutritional qualities of seed meal of Indian mustard by reducing the amount of glucosinolate, therefore, can be of high economic value. Considering the adverse effects of diet rich in erucic acid and glucosinolate, international efforts for developing rapeseed-mustard strains with double low characteristics were initiated as early as the 1950s. However, most of the research was restricted to *B. napus* and not a single variety has been developed till date in Indian mustard, possessing double low traits. Therefore, this study was designed to develop, evaluate and identify some of the promising double low lines from amongst a pool of advanced breeding lines, to be used as potential donors for transferring double low quality traits in high yielding varieties.

MATERIALS AND METHODS

Plant material

The material was selected from 1200 single plants of F₄ generation derived from 3 crosses [Varuna × NUDHYJ-3; EC-564648 × (PCR-7 × NUDHYJ-3); PCR-7 × NUDHYJ-3] involving double low donors and high yielding varieties. These 1200 plants were screened for erucic acid and glucosinolate content in seed oil and meal, respectively. Near double low plants were advanced to next generation through selfing. Finally, 11 lines derived from the cross (Varuna × NUDHYJ-3), consistently found to be double low were selected from F₇ generation. Selected lines were evaluated during *rabi* 2012-13 in randomized complete block design (RCCBD) with 3 replications. Each progeny and check variety was sown in 5 rows plot of 5 m length spaced 30 cm apart with plant to plant spacing of 10 cm achieved by thinning after 15-20 days of sowing. Recommended package of practices for raising a healthy crop was followed. Ten plants were randomly selected from each plot to record data on plant height (cm), main shoot length (cm), number of primary branches/plant, number of siliquae on main shoot, number of siliquae/plant, number of seeds/siliqua, 1000 seed weight (g), oil content (%), seed yield/plant (g), seed yield/plot of 2.7 m² (kg/ha), oil yield (kg/ha), glucosinolate content (μmoles/gram of defatted seed meal), oleic acid (%), linoleic acid (%), linolenic acid (%) and erucic acid (%). The detail of the breeding methodology adopted in the present study is indicated in Figure 1.

Determination of oil content and seed meal preparation

The seeds were thoroughly ground in a pestle and mortar and 10.0 g triplicates of ground seeds were extracted with hexane for 24 h in a Soxhlet apparatus (AOAC, 1997). Subsequently, hexane was removed from the oil by rotary evaporator under reduced pressure and the weights of the residual oils were calculated. The seed meal remaining after the extraction of total oil was preserved for estimation of glucosinolates.

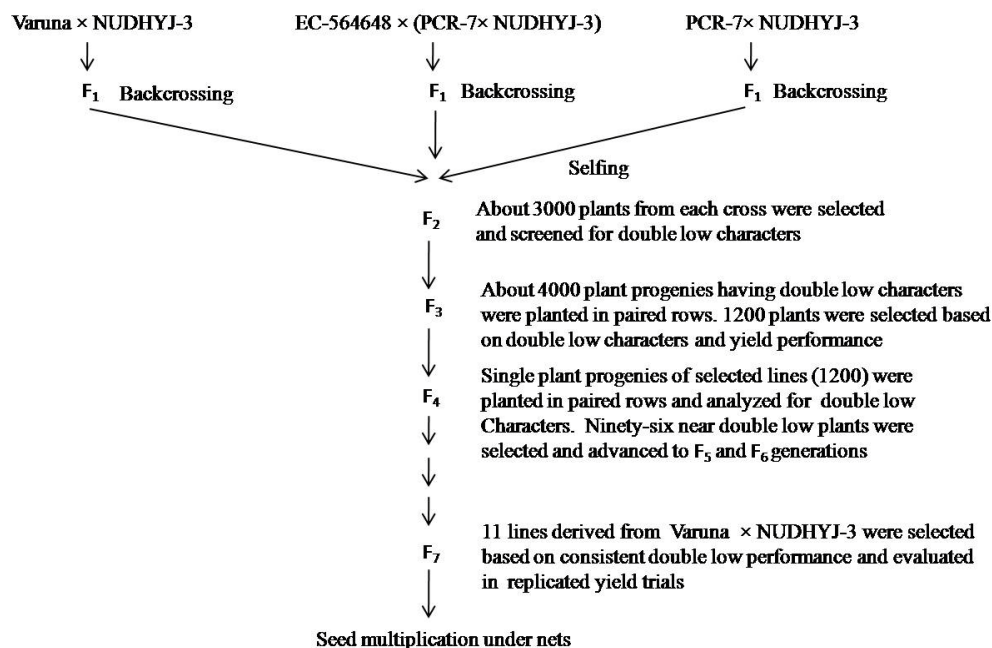


Figure 1. Schematic diagram of the breeding methodology adopted in the study.

Estimation of glucosinolates

Total glucosinolate content in the seed meal was estimated by complex formation between glucosinolates and sodium tetrachloropalladate solution. The intensity of the color produced was measured using spectrophotometer at 405 nm (Kumar *et al.*, 2004).

Fatty acid analysis by Gas Liquid Chromatography (GLC)

Methyl esters of oil samples were prepared by transesterification according to the method described by Sarinet *et al.*, (2009) with slight modifications. 1.0 µl of the methyl ester sample was injected into SP 2300 + 2310 SS column. A Nucon model 5765 gas chromatograph equipped with flame ionization detector (FID) was used. The oven, injector and detector temperature were 240 °C, 230 °C and 250 °C, respectively. The carrier gas was nitrogen, at flow rate of 40-50 ml/min. Peaks of fatty acid methyl esters were identified by comparing their retention time with that of the known standards run under similar separation conditions. Individual fatty

acids were expressed as % of the total fatty acids.

Statistical analysis

Mean data were subjected to analysis of variance (ANOVA) as suggested by Panse and Sukhatme (1978). Genetic parameters and simple correlations in all possible combinations were worked out as per standard procedure. Heritability in broad sense was estimated according to Hanson *et al.*, (1956). Genetic advance was estimated according to the formula given by Johnson *et al.*, (1955).

DNA extraction and STMS analysis

Genomic DNA was isolated from fresh and young leaves using the CTAB method (Murray and Thompson, 1980) with slight modifications. A total of 66 STMS primers were used for PCR amplification (Singh *et al.*, 2011, 2012). Amplification reactions were carried out in a volume of 20 µl containing 10 mM Tris HCl (pH 8.3), 50 mM KCl, 1.5 mM MgCl₂, 200 mM each dNTP, 0.4 µM forward and reverse primers, 1.0 unit DNA polymerase (DreamTaq) and 30 ng of

template DNA. Amplifications were carried out in a Veriti 96-well Fast Thermal Cycler (PE Applied Biosystems, USA). Thermal cycler was programmed to 1 cycle of 5 min at 94 °C for initial strand separation. This was followed by 35 cycles of 45 s at 94 °C for denaturation, 40 s of 58 °C for annealing and 1 min at 72 °C for primer extension. Finally, 1 cycle of 7 min at 72 °C was used for the final extension, followed by soaking at 4 °C. The reproducibility of the amplification products was checked twice for each primer.

Cluster analysis

Differences in the DNA banding patterns were qualitatively scored from gel photographs for presence (1) and absence (0) of bands assuming that each band represents a unique genetic locus. Homology of bands among samples was based on the distance of migration in gel. Scoring was done for clear, unambiguous amplicons and their sizes were determined by comparing with 100 bp DNA ladder. Based on the presence or absence of amplicons, a binary 1/0 data matrix was created and used to calculate Jaccard's similarity coefficient (Jaccard, 1908). Cluster analysis was carried out among the lines based on Jaccard's similarity coefficients using UPGMA (Sneath and Sokal, 1973) and SAHN-clustering algorithm. All the analyses were performed using NTSYS-pc, version 2.02e (Applied Biostatistics) software. The confidence limits of the UPGMA based dendrogram was determined by bootstrap analysis. One thousand bootstrap replicates were computed and bootstrap of 50% majority rule consensus tree was constructed using the Win Boot software (Yap and Nelson, 1996).

RESULTS AND DISCUSSION

A total of 1200 single plants of F₄ generation derived from 3 crosses involving double low donors and high yielding varieties were screened for double low quality as well as yield contributing traits. Ninety-six near double low plants were advanced to F₆ generations. Finally, 11 lines derived from the cross (Varuna

× NUDHYJ-3), consistently found to be double low with better yield performance were selected from F₇ generation and considered for further analysis. The analysis of variance of the selected lines revealed highly significant difference among the genotypes for all the traits studied, indicating sufficient amount of genetic variability in the material. This suggests that selection of superior genotypes having high yield potential along with double low quality traits may be practiced in the breeding material generated in the present study.

Estimates of mean and range for all the characters exhibited wide range of variation (Table 1). The most pronounced range was obtained for seed yield/plot, number of siliquae/plant, oil yield, glucosinolate (µmoles/gram of defatted seed meal) and erucic acid content. On other hand, characters like number of primary branches/plant, 1000 seed weight, number of seeds/siliqua and oil content exhibited narrow range of variation. Significant differences were also observed between the genotypes for seed yield/plot which ranged from 17.4 g (BPRQ-2-2-10) to 24.4 g (Kranti). However, seed yield/plot for these lines ranged from 1274 to 2001 kg/ha. The plant height was highest for Pusa Mustard - 21 (204.9 cm) and lowest for BPRQ-2-1-7 (176.8 cm). Thousand seed weight ranged from 2.2 g (BPRQ-2-2-11) to 5.5 g (Kranti) (Table 2). Significant estimates for oil quality traits were observed among the different lines. The lowest oil content was obtained in Pusa Mustard - 21 (38.9%) while it was highest for BPRQ-2-1-8 (42.5%). Data for glucosinolate content ranged from 12.3 (BPRQ-2-2-5) to 74.20 (Kranti) µmoles/gram defatted seed meal (Table 2). Oleic acid content in the studied genotypes ranged from 12.1% (Kranti) to 41.8% (Pusa Mustard - 21). Similarly, linoleic and linolenic acid content in the seed oil of different genotypes also exhibited wide variation. Linoleic acid content ranged from 15.7% (Kranti) to 41.0% (BPRQ-2-1-6) while linolenic acid varied from 12.5% (Pusa Mustard - 21) to 28.7% (BPRQ-2-2-2). BPR-Q-2-1-5 (1884 kg/ha) followed by BPRQ-2-2-11 (1851 kg/ha) recorded maximum yield among all the selected quality lines.

Table 1. Mean, standard error, range, coefficient of phenotypic (PCV) and genotypic variance (GCV), heritability in broad sense (h^2_b) and genetic advance for morpho-quality traits in Indian mustard.

Characters	Mean \pm SEM	CV (%)	Range	PCV (%)	GCV (%)	Heritability h^2 (%)	Genetic advance as % of mean 1 %
Plant height (cm)	192.62 \pm 6.33	5.69	185.13-204.86	4.69	3.35	50.9	6.31
Main shoot length (cm)	59.48 \pm 3.69	10.76	55.86- 64.93	7.27	3.78	27.10	5.20
Number of primary branches	5.93 \pm 0.35	10.49	5.46- 6.66	6.64	2.73	16.9	2.97
Number of siliquae on main shoot	52.56 \pm 5.53	18.22	43.63-61.03	14.42	9.93	47.10	18.00
Number of siliquae/plant	635.11 \pm 78.73	19.47	454.33- 929.33	20.12	15.85	62.10	32.98
Number of seeds/siliqua	13.53 \pm 0.56	7.18	12.59- 15.20	4.74	2.30	23.50	2.94
1000 seed weight (g)	2.77 \pm 0.18	11.56	2.16- 5.51	22.57	21.56	91.30	44.38
Oil content (%)	41.61 \pm 0.29	2.24	38.88- 42.19	2.10	1.97	88.40	4.90
Seed yield/plant (g)	17.86 \pm 2.69	16.09	15.33-24.42	12.36	8.61	4.85	1.01
Seed yield/plot (2.7m ²) (kg/ha)	1652.66 \pm 13.51	14.16	1274- 2001	14.09	14.07	89.70	37.09
Oil yield (kg/ha)	687.26 \pm 3.51	2.88	533.80- 837.35	13.92	13.91	92.90	39.70
Glucosinolates (μ moles/gram)	26.68 \pm 0.36	2.38	12.34- 74.20	75.21	75.20	91.23	32.49
Oleic acid (%)	34.96 \pm 0.59	2.95	12.1- 41.8	20.98	20.91	92.96	46.02
Linoleic acid (%)	34.47 \pm 0.49	2.49	15.7- 40.95	18.47	18.41	91.42	38.46
Linolenic acid (%)	19.77 \pm 0.06	2.57	12.5-28.67	21.36	21.35	86.90	36.36
Erucic acid (%)	4.35 \pm 0.03	2.48	0.46-42.4	62.56	52.15	75.11	21.14

Table 2. Morphological characterization of double low lines in Indian mustard.

Genotype	Plant height (cm)	Main shoot length (cm)	Number of primary branches/plant	Number of siliquae on main shoot	Number of siliquae/plant	Number of seeds/siliqua	1000 seed weight (g)	Oil content (%)	Seed yield/plant (g)	Seed yield/plot (2.7m ²) (kg/ha)	Oil yield (kg/ha)	Glucosinolates (µmoles/gram of defatted seed meal)	Oleic acid (%)	Linoleic acid (%)	Linolenic acid (%)	Erucic acid (%)
BPRQ-2-1-4	191.6	60.7	6.2	52.4	547.0	12.6	2.3	41.7	17.4	1453	605.5	14.0	37.6	37.0	19.1	1.9
BPRQ-2-1-5	196.9	62.7	6.1	60.5	822.9	13.1	2.8	42.2	23.5	1884	795.0	15.7	38.4	32.8	22.8	1.1
BPRQ-2-1-6	201.9	58.3	5.7	43.6	843.6	13.3	2.6	42.1	19.1	1770	744.5	21.4	37.2	41.0	17.3	0.5
BPRQ-2-1-7	176.8	57.2	5.5	55.9	804.8	14.0	2.7	41.6	18.7	1501	624.6	19.6	35.7	37.8	21.0	0.7
BPRQ-2-1-8	187.8	63.6	5.7	61.0	860.5	13.6	2.4	42.5	19.6	1784	757.5	26.2	32.6	37.6	25.3	0.7
BPRQ-2-2-2	189.8	64.9	6.5	58.4	735.9	13.6	2.5	41.8	17.9	1466	612.7	25.1	36.4	28.8	28.7	1.2
BPRQ-2-2-3	185.1	56.3	5.9	51.7	454.3	13.4	2.5	41.5	15.3	1575	652.9	13.9	39.8	33.9	17.1	0.6
BPRQ-2-2-5	204.2	56.4	6.7	57.2	823.1	15.2	2.5	41.3	18.1	1681	694.0	12.3	37.3	36.4	18.8	0.9
BPRQ-2-2-9	193.3	49.8	5.5	56.5	529.3	13.2	2.5	41.8	17.6	1343	561.6	13.6	33.3	37.0	22.1	2.0
BPRQ-2-2-11	200.9	55.9	5.7	60.9	855.8	13.4	2.2	42.0	18.6	1851	776.6	22.1	38.3	38.2	16.1	1.0
BPRQ-2-2-10	191.9	61.4	6.3	60.4	472.2	14.1	2.5	41.9	17.4	1274	533.8	20.9	37.3	36.5	17.2	1.9
Kranti	179.0	64.5	5.6	50.6	929.3	13.3	5.5	41.8	24.4	2001	837.4	74.2	12.1	15.7	19.2	42.4
Pusa mustard - 21	204.9	61.6	5.8	60.9	899.1	13.1	3.8	38.9	23.9	1902	739.4	66.7	41.8	35.5	12.5	1.9
CD (P=0.05)	18.5	10.8	1.0	16.1	229.8	1.6	0.5	0.9	7.9	39.4	10.3	1.1	1.7	1.5	0.2	0.1

BPRQ-2-1-5 recorded 15.7 μ moles/gram glucosinolates along with very low content of erucic acid (1.1%). Similarly, the glucosinolate and erucic acid content in another line BPRQ-2-2-11 were recorded to be 22.1 μ moles/gram defatted seed meal and 0.95%, respectively.

The glucosinolate and erucic acid content for these 2 lines were found to be much lower as compared to quality check Pusa Mustard - 21 (66.7 μ moles/gram glucosinolate and 1.9% erucic acid). On the basis of quality data and yield potential, these 2 lines hold promise for multi-location testing as no double low variety of Indian mustard has been bred for general cultivation in India.

The estimates of genotypic and phenotypic coefficients of variation were considerably high for glucosinolate and erucic acid content, 1000-seed weight, oleic, linoleic and linolenic acid content (%) and number of siliquae/plant. High genotypic coefficient of variation for oleic and linoleic acid contents provide good opportunity for selecting desirable levels of these fatty acids. High genotypic and phenotypic coefficients of variation for 1000 seed weight, oleic, linoleic and linolenic acid content (%) and number of siliquae/plant is also reported by Kumar *et al.*, (2013) in Indian mustard. High heritability estimates (broad sense) were observed for oleic acid content (93.0%), oil yield (92.9%), linoleic acid content (91.4%) and 1000-seed weight (91.3%). Results indicated that these characters are less influenced by the environmental factor and direct selection for these characters would be effective for further improvement. The high genetic advance was observed for oleic acid (46.0%) and for 1000-seed weight (44.4%), while the other characters showed moderate to low genetic advance.

According to Hanson *et al.* (1956), heritability estimates along with genetic advance are of more value than the former alone in predicting effect of selection. Further, Panse (1957) has reported that high genetic advance might be expected when heritability is mainly due to additive gene effect. In the present study, heritability (broad sense) of oleic acid (93.0%) with maximum genetic advance (46.0 %) was observed, which might be due to heritability with additive gene impact and therefore

selection may be effective (Table 3). These results are in agreement with the results obtained by Chauhan *et al.* (2002) and Kumar *et al.*, (2013) for oleic acid (%).

Genotypic and phenotypic correlations were estimated for yield and quality traits in all possible combinations. Erucic acid showed negative and highly significant correlations with oleic acid (Table 3). The negative relationship of erucic with oleic acid has also been reported earlier (Zhou and Liu, 1987; Singh *et al.*, 2001; Meena, 2006 and Kumar, 2013). Increase in the level of oleic acid might also result in high oil yield as both characters were found to be positively correlated. Similarly, reduction in the erucic acid is also expected as a negative and significant correlation has been obtained between oleic and erucic acid content (Table 3).

Oil content showed negative significant correlation with seed yield per plant while a positive and highly significant correlation was obtained between oleic acid and oil yield ($r = 0.745^{**}$). Moreover, erucic acid showed negative highly significant relationship with oil yield ($r = -0.845^{**}$). On the basis of the present investigation, it would be appropriate to enhance the level of oleic acid which shows large variability and negative relationship with erucic acid, through hybridization among varieties/donors having high level of oleic and low level of erucic acid as well as high oil yield.

STMS analysis

A total of 66 STMS primer pairs were tested on 11 double low quality lines and 2 check entries, Heera (quality) and Kranti (non-quality). Out of the 40 primer pairs, which showed successful amplification, 21 (31.3%) amplified monomorphic while 19 (28.4%) amplified polymorphic DNA bands. The cluster analysis based on STMS markers discriminated well between the quality and non-quality lines and gave 2 major groups i.e. I, II of which cluster I was found to be multi-genotypic with 3 sub-groups (IIA, IIB and IIC). Cluster I contained the double low quality lines grouped with Heera. It is expected as NUDHYJ-3 which was used as quality donor is itself derived from Heera. Non-quality line 'Kranti' was entirely separated from cluster I and is included in cluster II (Figure 2).

Table 3. Phenotypic (rp) and genotypic (rg) correlation coefficients between different morphological and quality traits of Indian mustard.

Characters		Plant height (cm)	Main shoot length (cm)	No. of primary branches	No. of siliquae on main shoot	No. of siliquae/plant	No. of seeds/siliqua	1000 seed weight (g)	Seed yield/plant (g)	Seed yield/plot (2.7m ²) (kg/ha)	Oil yield (kg/ha)	Glucosinolate (µmoles/g)	Oleic acid (%)	Linoleic acid (%)	Linolenic acid (%)	Erucic acid (%)	Oil content (%)
Plant height (cm)	rp	1.000	0.149	0.053	0.495	0.530*	0.375	0.035	0.265	0.349	0.338	-0.059	-0.089	0.439	-0.145	0.001	-0.243
	rg	1.000	0.256	0.122	0.611*	0.598*	0.391	0.023	0.311	0.421	0.429	-0.045	-0.099	0.544*	-0.255	0.002	-0.261
Main shoot length (cm)	rp		1.000	0.533*	0.501*	0.166	0.410	0.002	0.505*	-0.251	-0.234	0.400	0.004	-0.366	0.343	0.164	-0.190
	rg		1.000	0.569*	0.615*	0.187	0.462	0.001	0.574*	-0.314	-0.220	0.645**	0.006	-0.523*	0.456	0.301	-0.214
Number of primary branches	rp			1.000	0.372	-0.126	0.771**	0.097	0.562*	-0.217	-0.220	-0.043	-0.048	-0.288	0.279	0.085	-0.459
	rg			1.000	0.459	-0.321	0.801**	0.145	0.654*	-0.356	-0.321	-0.125	-0.023	-0.346	0.214	0.112	-0.514*
Number of siliqua on main shoot	rp				1.000	0.497*	0.345	0.366	0.346	0.001	0.001	0.359	-0.162	0.022	0.018	0.201	0.163
	rg				1.000	0.514*	0.399	0.247	0.402	0.002	0.003	0.312	-0.213	0.156	0.041	0.256	0.215
Number of siliqua/plant	rp					1.000	0.119	-0.081	0.210	0.491	0.508*	0.136	0.091	0.243	-0.101	-0.159	0.119
	rg					1.000	0.089	0.126	0.477	0.563*	0.481	0.241	0.098	0.314	-0.412	-0.106	0.186
Number of seeds/siliqua	rp						1.000	-0.226	0.560*	-0.038	-0.040	-0.262	-0.111	0.147	-0.020	0.083	-0.383
	rg						1.000	-0.331	0.612	-0.014	-0.081	-0.301	-0.146	0.216	-0.060	0.096	-0.405
1000-seed weight (g)	rp							1.000	0.282	-0.324	-0.363	0.274	-0.160	-0.489	0.474	0.176	0.084
	rg							1.000	0.356	-0.401	-0.412	0.144	-0.126	-0.701*	0.506*	0.189	0.096
Seed yield/plant (g)	rp								1.000	-0.059	-0.076	-0.067	0.102	-0.282	0.355	-0.066	-0.448
	rg								1.000	-0.081	-0.062	-0.082	0.174	-0.365	0.451	-0.056	-0.614*
Seed yield/plot (2.7m ²) (kg/ha)	rp									1.000	0.995**	-0.183	0.676**	0.320	-0.320	-0.745**	-0.069
	rg									1.000	0.856**	-0.214	0.811**	0.456	-0.301	-0.731**	-0.092
Oil yield (kg/ha)	rp										1.000	-0.140	0.653**	0.324	-0.317	-0.721**	-0.050
	rg										1.000	-0.158	0.745**	0.412	-0.412	-0.845**	-0.061
Glucosinolates (µmoles/gram)	rp											1.000	-0.299	-0.329	0.423	0.409	0.412
	rg											1.000	-0.312	-0.301	0.548*	0.619*	0.388
Oleic acid (%)	rp												1.000	-0.069	-0.262	-0.906**	-0.285
	rg												1.000	-0.063	-0.415	-0.762**	-0.412
Linoleic acid (%)	rp													1.000	-0.851**	0.102	-0.038
	rg													1.000	-0.956**	0.312	-0.021
Linolenic acid (%)	rp														1.000	0.177	0.029
	rg														1.000	0.189	0.031
Erucic acid (%)	rp															1.000	0.198
	rg															1.000	0.274
Oil Content (%)	rp																1.000
	rg																1.000

* and ** Significant at 5% and 1% level of significance, respectively.

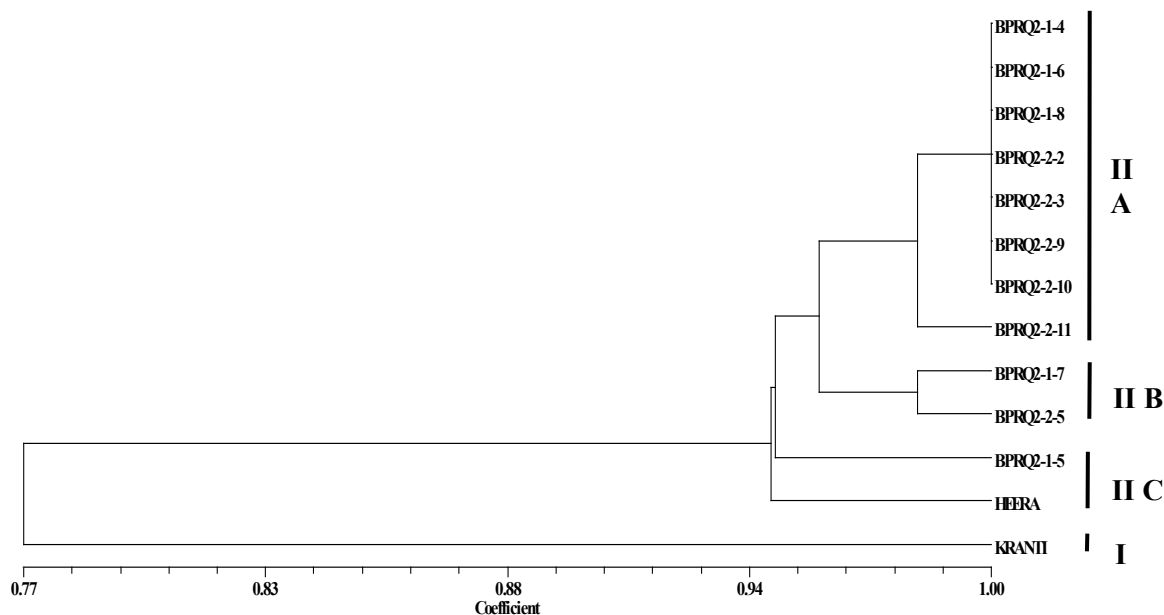


Figure 2. UPGMA dendrogram showing genetic relationship among double low quality lines based on Jaccard's similarity coefficients using STMS markers.

Therefore, the genetic relationships among the quality lines in this study were consistent and in complete agreement with the available pedigree data.

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