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Agro-Morphological Traits and Microsatellite Markers Based Genetic Diversity in Indian Genotypes of Linseed (*Linum usitatissimum* L.)

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ABSTRACT

Linseed is an important oilseed and fibre crop predominantly grown in India. The aim of the present research was to evaluate genetic diversity and patterns of relationships among the 58 genotypes through 10 morphological traits and 12 polymorphic microsatellite (SSR) markers. Euclidean analysis of agro-morphological traits grouped the 58 genotypes into four clusters of which cluster I was the largest with 20 accessions while clusters II and IV were most genetically diverse due to maximum inter-cluster distance. Principal component analysis revealed three traits accounted for more than 86% of the total variation. A total of 41 alleles were amplified with 12 SSRs having an average of 5.71 alleles per primer locus. The Polymorphic Information Content (PIC) varied between 0.18 to 0.78. Based on Jaccard's similarity coefficient, the genetic distance varied from 0.07 to 0.89 with an average of 0.54±0.10. The genotypes RKY-14, KL-213, LC-185 and Kartika were found to be the most divergent among all the genotypes studied on the basis of genetic distance. The most diverse genotypes identified in this study can be used in breeding programs to broaden the genetic base of the linseed germplasm.

Keywords: Diversity, Linseed, Microsatellite markers, Principal component analysis.

INTRODUCTION

Linseed (*Linum usitatissimum* L., 2n= 2x= 30) is a self-pollinating crop which belongs to the Lineaceae family. It is cultivated over 2.6 million ha in more than 50 countries producing 614,000 metric tons in the year 2013-14 (FAOSTAT, 2014). Canada is the world's largest producer of linseed which accounts for almost 80% of the worldwide trade in linseed followed by China, United States and India (FAOSTAT, 2014). It is commercially grown as a source of stem fiber and seed oil (Cloutier *et al.*, 2012). The linseed oil primarily goes into industries for the manufacture of paints, varnish, oil cloth, linoleum and pad ink. The flax fibre is widely used and serves as valuable raw material for textiles, thread/rope and packaging materials; the straw and short fibre is used for pulp to produce special papers for cigarettes, currency notes and artwork; and the wooden part serves as biomass energy or litter in cattle farming (Rowland, 1998; Mackiewicz-Talarczyk *et al*., 2008). Linseed derivatives such as whole flax seed, flax meal and milled flax have been reported as a functional or nutraceutical food due to its health promoting or disease preventive properties

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(Fitzpatrick, 2007). Lignans is one of the major compounds in linseed hull which acts as an important antioxidant agent. Flax seed provides 800 times higher lignans than the seeds of other plants except sesame (Jhala and Hall, 2010). The amino acid pattern of linseed protein resembles that the pattern of soybean protein, which is considered to be one of the most nutritious plant proteins (Rabetafika *et al*., 2011)

The evaluation of genetic diversity is useful for the selection of the most efficient genotypes (Khodadadi *et al.,* 2011). The knowledge regarding its nature and extent of genetic variability available in the germplasm and the correlation among various characters are important requirements for planning a successful breeding program for the linseed crop (Dikshit and Shivaraj, 2015). To evaluate the diversity in germplasm, various morphological, biochemical and molecular markers can be utilized. The molecular markers reflect the true expression of genotype, while morphological traits encompassed the expression of genotype, environment and their interaction (Adugna *et al.*, 2006). In linseed, various types of molecular markers such as RAPD, AFLP, ISSR and SSR have been used to estimate the genetic variation and relationships in different sets of germplasm (Wiesnerova and Wiesner, 2004; Diederichsen and Fu, 2006; Fu, 2006, Uysal *et al*., 2010; Deng *et al*., 2011; Rajwade *et al.,* 2010; Chandrawati *et al.,* 2014). Apart from molecular markers based diversity studies, limited reports are also available to describe the extent of genetic variability in linseed based on morphological traits (Verma, 1996; Begum *et al*., 2007; Khan *et al*., 2013; Ali *et al.,* 2014; Dikshit and Shivaraj, 2015; Worku *et al*., 2015). The complementation of morphological based diversity with molecular marker could provide more information about the genetic variability but diversity is best estimated if agro-morphological and molecular marker studies are used together (Panahi *et al.,* 2013; Mansour *et al.,* 2015). Therefore, the present investigation was carried out to assess the genetic diversity of 58 Indian genotypes of linseed based on 10 agro-morphological traits along with SSR markers.

MATERIALS AND METHODS

Plant Materials and Field Experiment

The seed materials used in the present investigation includes 58 genotypes of linseed obtained from All India Co-ordinated Research Program on Linseed (AICRP on Linseed), Chandra Shekhar Azad University of Agriculture and Technology (CSAUA&T), Kanpur, Uttar Pradesh, India. All the genotypes were grown in a field at CSIR-National Botanical Research Institute, Lucknow during the crop season 2012–13 and 2013-14. All the entries were planted in Randomized Complete Block Design (RCBD) with 3 replications. Four rows of 2 meters with spacing of 45 cm between rows and 15 cm between plants were adopted with two non-experimental border rows. The recommended cultural practices were followed to raise them as healthy crops during both years. Ten competitive plants in each replication were randomly tagged to record various morpho-metric traits. The major traits include: Days Of 50% Flowering (DOF), Plant Height (PH), number of Branches/Plant (BP), number of Capsules/Plant (CP), Capsule Weight/Plant (CWP), Seed Weight/Plant (SWP), Husk Weight/plant (HW), Test Weigh (TW), number of Seeds/Capsule (SPC) and Oil Content (OC). The oil content was measured through minispec Time Domain-Nuclear Magnetic Resonance (TD-NMR) analyzer (Bruker Corporation, USA).

Morphological Data Analyses

The mean values of each trait were used for statistical analysis and subjected to analysis of variance using WINDOSTAT software (www.windostat.org). Principal component analysis (Sneath and Sokal,

1973) was performed using correlation matrix to define the pattern of variation between genotypes. Further hierarchical clustering was done to bring out the pattern of similarity using ward's minimum variance method, which produced a dendrogram showing a successive fusion of individuals and ultimately culminating into a single cluster (Ward, 1963). The relationships among the clusters were assessed by estimating the inter-cluster distances using Mahalanobis Distance (D^2) statistics (Rao, 1952).

SSR Amplification

Total genomic DNA was extracted from the fresh and young leaves collected from randomly selected individual plants of each genotype before flowering using DNeasy® Plant Mini Kit (Qiagen, Hilden, Germany) following manufacturer's instructions. The quality of DNA was checked in 0.8% agarose gel and quantified using a nanodrop spectrophotometer ND1000 (Nanodrop Technologies, DE, USA). Finally, the DNA was normalized to 10 ng μl^{-1} for PCR amplification. A set of 12 previously reported nuclear SSRs (Cloutier *et al.,* 2012) was selected based on wide genomic distribution and high *PIC* value for SSR profiling of 58 linseed genotypes. The primers were synthesized having an additional 18 base (5'-TGTAAAACGACGGCCAGT-3') tag added at 5' end of the all forward primers as M13 tail following the Schuelke (2000). The PCR was carried out in 10 µl reaction volume containing 10 ng of genomic DNA, 1× PCR master mix (Fermentas Inc, USA), 0.1 µl (5 pmol μl^{-1}) of forward primer (tailed with M13 tag), 0.3 µl (5 pmol μl^{-1}) each of both normal reverse and M13 tag (labeled with either 6-FAM NED, VIC and PET) using Applied Biosystems Veriti PCR machine. The amplification was carried out with an initial denaturation of 5 minutes at 95^0C, followed by 35 cycles for denaturation for 30 seconds at 94^0C, annealing for 45 seconds at 48–52^0C (primer specific) and extension for 30 seconds at 72^0C. Subsequently, 10 cycles of denaturation for 30 seconds at 94^0C, annealing for 45 seconds at 53^0C, extension for 45 seconds at 72^0C followed by final extension for 15 minutes at 72^0C was performed. Finally, PCR products were run in 1.5% agarose gel and checked, and then the post PCR multiplex sets was prepared based on fluorescence labeled primers. For the post PCR multiplexing, 1 µl of 6-FAM and 2 µl of each VIC, NED and PET labeled PCR product representing different SSRs were combined with 13 µl of water. 1 µl of this mixed product was added to 10 µl Hi-Di formamide containing 0.25 µl GeneScan TM 600 LIZ ® as internal size standard, denatured for 5 minutes at 95^0C, quick chilled on ice for 5 minutes and loaded on ABI 3730xl DNA Analyzer. The fragment analysis was performed by GeneMapper ver. 4.0 software (Applied Biosystems, Foster City, CA, USA)

SSR Data Analysis

The allelic data of polymorphic SSRs were subjected to statistical analysis using PowerMarker (Liu and Muse, 2005) in order to calculate the observed Heterozygosity (Ho), the gene diversity or expected Heterozygosity (He), major allele frequency and Polymorphic Information Content (PIC) value. To understand the genetic relationship pair-wise genetic dissimilarities among all 58 genotypes were calculated according to Jaccard's coefficient using DARwin 5.0.128 software (Perrier *et al*., 2003). The calculated dissimilarity matrix was then used to construct a Neighbor-Joining (NJ) tree with a 1,000 replicate bootstrap test.

RESULTS

Diversity Based on Morphological Data

The analysis based on 10 morphological traits revealed significant genetic variability

and a simultaneous testing of significance based on Wilk's (Lambda) criterion for pooled effect of all characters also showed significant differences among the population (χ2= 570, df= 4249.81**). The hierarchical clustering (Wards minimum variance) grouped all the 58 genotypes into 4 clusters based on their morphological similarities (Table 1, Figure 1). The number of genotypes in each cluster varied from 11 to 20. The cluster I was found to be the largest having 20 genotypes followed by clusters II and III having 14 and 13 genotypes respectively. The cluster IV was the smallest having 11 genotypes. The intra and inter-cluster distances revealed that cluster II had a maximum intra cluster distance (253.27) followed by cluster III (239.85), cluster IV (199.77) and cluster I (177.73) (Supplementary Table 1). The inter cluster distance was varied from 353.12 (between clusters I and II) to 1600.66 (between clusters II and IV). The cluster mean value showed that there was a maximum seed weight/plant, test weight and oil content were showed by cluster IV. The cluster IV also showed the highest mean value for capsules/plant, capsule weight/plant, the second highest value for seeds per capsule and the smallest plant height with early flowering (Table 2). The cluster II showed the highest mean value for plant height, number of branches/plant, and number of seeds/capsule and delayed flowering time. The genotypes grouped in the cluster I had the second highest value for oil content, seed weight/plant and plant height. The cluster III had moderate to low values for majority of the traits except husk weight/plant and test weight.

The Principal Component Analysis (PCA) was done by considering all the ten variables simultaneously in order to assess the pattern of variation factors responsible for differentiation of genotypes. The first three Principal Components (PCs) accounted for more than 86% of the total variation (Table 3). The first principal component accounted for 68.10% of the total variation due to the days of 50% flowering and the plant height which had the maximum and positive weight on this component and all other traits had a negative weight of the PC_1. The PC_2 exhibited about 11% of total variation and was positively associated with days of 50% flowering, plant height, test weight and oil content. The oil content had the highest positive weight on PC2 also followed by test weight and plant height. The PC_3 accounted for 8% of total variation was mainly due to the test weight/plant which had the highest positive weight on this component also followed by capsule weight/plant and capsules/plant.

Diversity Based on SSR Data

The genetic diversity of the 58 genotypes of linseed was also evaluated by 12 polymorphic SSRs. These SSRs produced a total of 41 alleles across the genotypes (Supplementary Table 2). The number of alleles varied from 2 (LU3148, LU3251) to

Table 1. Distribution of 58 genotype of linseed in 4 clusters based on their 10 quantitative traits.

Cluster	Number of genotypes	Name of genotypes
Cluster I	20	Shikha, Meera, Sweta, RLU-6, Sheela, LC-54, Hira, Kiran, RL-914, Himani, C-429, Chambal, Shekhar, Pusa -3, Jawahar-7, Gaurav, LC-2063, Kartika, Neela, Surabhi,
Cluster II	14	Rashmi, EC-1392, BAU 616A, Sharda, Him Alsi-1, Mukta, Nagarkot,LC-2023, Parvati, Baner, K-2, Pusa -2, S-36, KL 213
Cluster III	13	Neelum, JRF 4, Garima, R-552, Suyog, Jawahar-23, Mau Azad Alsi -1, Him Alsi-2, Indra Alsi- 32, NL-97, Deepika, Jawahar -1, Jawahar-17,
Cluster IV	11	Laxmi -27, GS 41, Padmini, Shubhra, GS 234, Janki, T-397, IC 15888, LC-185, JLS-9, RKY -14,

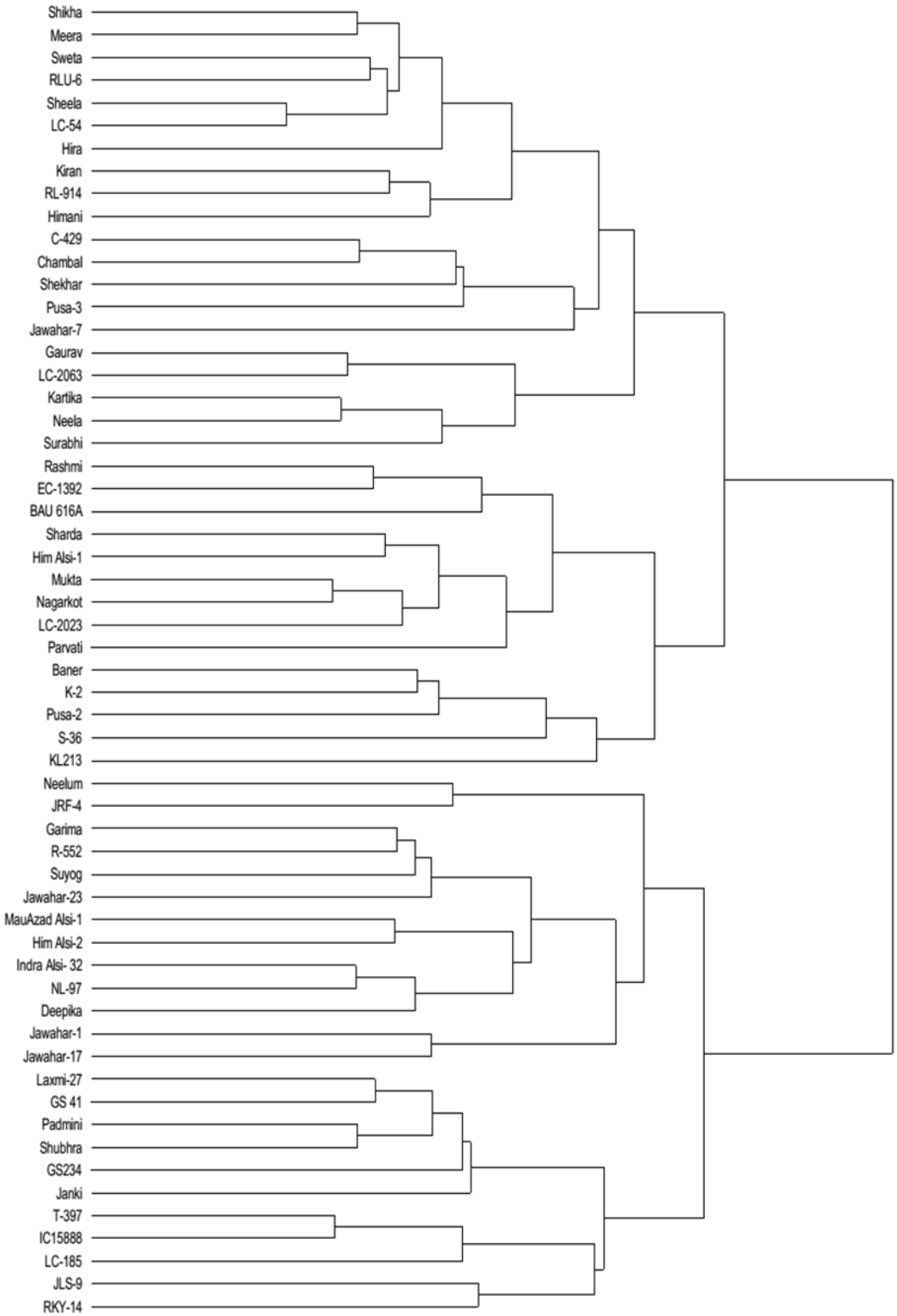

Figure 1. Dendrogram of 58 Linseed genotypes derived from the Wards minimum variance cluster analysis using Mahalanobis distances.

Table 2. Cluster means and standard errors of the means of different traits in linseed.

Cluster	Traits									
	DOF[a]	PH[b]	BP[c]	CP[d]	CWP[e]	SWP[f]	HW[g]	TW[h]	SPC[i]	OC[j]
Cluster I	90.725±1.48	91.47±2.60	3.92±.24	96.95±3.96	7.90±.26	5.89±.19	2.01±.12	6.61±.24	7.05±.13	41.29±.39
Cluster II	100.48±1.68	110.50±3.31	4.31±.33	105.338±8.75	7.29±.41	5.14±.34	2.16±.14	6.06±.24	7.47±.26	38.07±.96
Cluster III	72.05±1.40	85.17±2.3	3.25±.25	91.63±6.35	7.93±.46	5.73±.37	2.21±.14	7.23±.43	7.177±.20	40.27±.72
Cluster IV	59.11±1.70	72.72±2.62	3.68±.24	110.02±9.06	8.43±.60	6.35±.47	2.09±.14	7.35±.41	7.142±.19	42.56±.30

[a] Days Of 50% Flowering, [b] Plant Height, [c] number of Branches/Plant, [d] number of Capsules/Plant, [e] Capsule Weight/Plant, [f] Seed Weight/Plant, [g] Husk Weight/plant, [h] Test Weigh, [i] number of Seeds/Capsule, and [j] Oil Content.

7 (LU2332) with an average of 3.42 alleles per primer. The polymorphism information content (PIC) ranged from 0.18 (LU 3148) to 0.78(LU 2332) with an average of 0.39 alleles per SSRs. The majority of the SSRs (50%) showed 3 alleles across the genotypes. The *PIC* for each SSR varied from 0.18 (Lu3148) to 0.78 (Lu2332) with an average of 0.39. All the SSRs had a low to moderate *PIC* value except SSR Lu2332. Observed heterozygosity ranged from 0.00 (Lu485, Lu3217, Lu2155) to 0.64 (Lu3251) with an average of 0.16**.** The gene diversity was observed ranging from 0.20 (Lu3148) to 0.81 (Lu2332) with an average of 0.44. The allelic data of SSRs were used to calculate a pair-wise genetic distance based on Jaccard's similarity coefficient which varied from 0.07 to 0.89 with an average 0.54±0.10. The maximum genetic similarity (93%) was noticed between genotypes Jawahar-17 and Jawahar-23 and the same percentage of genetic similarity was also found in S36 and Pusa-2, followed by a genetic diversity of 88% which was noticed between Kartika and Indira Alsi-32 and a genetic similarity of 87% between Rashmi and Parvati and between Jawahar-7 and Pusa 3. The minimum, maximum and average genetic dissimilarity of each genotype with others (Table 4) showed that maximum differences were observed in Parvati, followed by Kartika, Jawahar-17 and Jawahar-32. On the other hand the lowest differences were found between JRF-4 and Sweta (89%) followed by Baner and RKY-14 (88%). The genotypes C-429, S-36 and Pusa-2 had the lowest range of pairwise genetic similarity coefficient compared to all other genotypes varying from 0.07 to 0.67 with an average of 0.43±0.13 (Table 4). Considering the genetic among distance the genotypes RKY-14, KL-213, LC-185 and Kartika were found to be the most divergent among all the genotypes studied. The neighbor joining clustering classified all the genotypes into three major clusters named as clusters I, II, and III (Figure 2). The cluster III was found to be the largest having 29 genotypes (50%) followed by cluster II having 15 genotypes

Table 3. Loadings of the first three principal components (PCs) in 58 accessions of linseed.

Traits	PCI	PCII	PCIII
Days of 50%flowering	0.91	0.06	0.20
Plant Height	0.29	0.20	-0.27
Branches/plant	-0.08	-0.04	-0.07
Capsules/ plant	-0.06	-0.63	0.34
Capsule weight/plant	-0.13	-0.07	0.42
Seed weight/plant	-0.03	-0.04	0.10
Husk weight/plant	-0.03	-0.01	-0.02
Test weight/plant	-0.10	0.50	0.73
Seeds/capsule	-0.03	-0.15	-0.13
Oil content	-0.22	0.54	-0.16
Components			
Eigen value(Root)	3711.06	575.10	443.66
% Var. Exp.	68.10	10.55	8.14
Cum. Var. Exp.	68.10	78.65	86.79

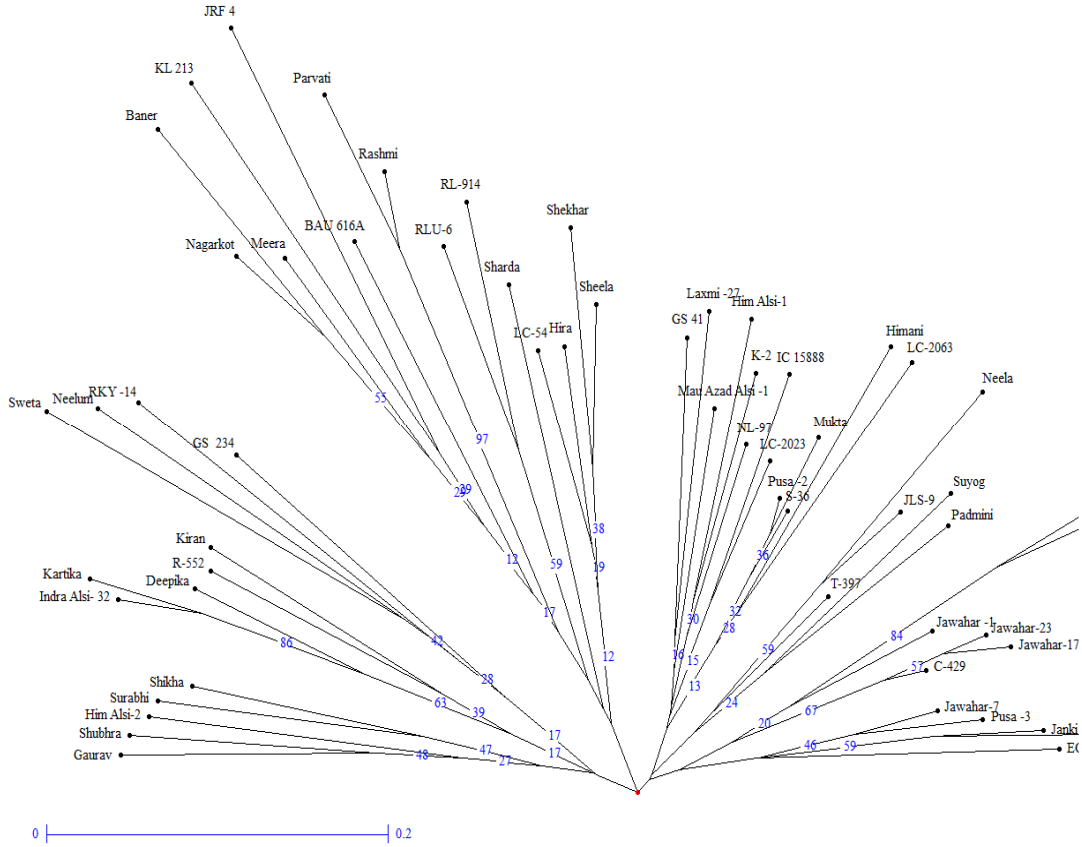

Figure2. Genetic relatedness among 58 genotypes of linseed based on Neighbor-Joining clustering. The scale at the bottom is for NJ distances.

(26%) and cluster I with 14 genotypes (24%). The cluster III could be further subdivided into three sub-clusters namely IIIa, IIIb, and IIIc with 13, 5 and 11 genotypes respectively. The other two clusters could not be subdivided further.

Table 4. Minimum, maximum and mean of the Jaccard's similarity coefficient of 58 genotypes of linseed.

Varieties	Minimum	Maximum	Mean± SD
Shikha	0.26	0.71	0.47±0.10
Neelum	0.13	0.56	0.37±0.09
Rashmi	0.17	0.87	0.40±0.11
Parvati	0.13	0.87	0.35±0.10
Sweta	0.11	0.55	0.34±0.09
Sheela	0.3	0.79	0.49±0.11
Garima	0.22	0.79	0.42±0.11
Shekhar	0.26	0.79	0.45±0.12
Laxmi -27	0.22	0.69	0.48±0.11
Padmini	0.3	0.72	0.50±0.09
T-397	0.32	0.8	0.58±0.12
Shubhra	0.29	0.71	0.44±0.09
Gaurav	0.26	0.71	0.43±0.09
Sharda	0.25	0.69	0.46±0.08
Mau Azad Alsi -1	0.3	0.76	0.53±0.12
Hira	0.19	0.85	0.50±0.13
Mukta	0.27	0.92	0.53±0.14
C-429	0.26	0.93	0.56±0.14
S-36	0.33	0.93	0.58±0.13
Pusa -2	0.32	0.93	0.57±0.14
Pusa -3	0.32	0.88	0.53±0.12
Jawahar -1	0.27	0.81	0.55±0.13
Jawahar-7	0.32	0.88	0.56±0.13
Jawahar-17	0.21	0.93	0.51±0.14
Jawahar-23	0.22	0.93	0.53±0.14
JLS-9	0.27	0.8	0.52±0.11
Suyog	0.23	0.75	0.49±0.12
Kartika	0.15	0.88	0.41±0.12
Indra Alsi- 32	0.2	0.88	0.43±0.11
Deepika	0.22	0.78	0.46±0.11
Kiran	0.19	0.7	0.46±0.11
R-552	0.25	0.7	0.46±0.10
RL-914	0.22	0.75	0.42±0.09
RLU-6	0.22	0.75	0.44±0.10
Meera	0.22	0.69	0.42±0.10
Chambal	0.17	0.8	0.42±0.12
LC-185	0.13	0.8	0.39±0.13
LC-54	0.24	0.79	0.50±0.12
LC-2023	0.24	0.85	0.55±0.15
LC-2063	0.24	0.75	0.47±0.11
NL-97	0.23	0.8	0.55±0.14
Neela	0.17	0.8	0.45±0.12
Baner	0.12	0.79	0.32±0.13
K-2	0.22	0.8	0.51±0.13
Him Alsi-1	0.24	0.72	0.49±0.10
Him Alsi-2	0.23	0.73	0.45±0.10
Surabhi	0.17	0.71	0.45±0.10
Janki	0.29	0.79	0.50±0.12
Himani	0.26	0.75	0.47±0.11
Nagarkot	0.21	0.79	0.40±0.11
GS 41	0.21	0.77	0.50±0.13
IC 15888	0.24	0.79	0.50±0.12
GS 234	0.17	0.65	0.44±0.11
JRF 4	0.11	0.56	0.30±0.10
BAU 616A	0.17	0.67	0.42±0.11
EC-1392	0.24	0.71	0.49±0.11
KL 213	0.13	0.56	0.31±0.09
RKY -14	0.12	0.55	0.38±0.09

DISCUSSION

The analysis of genetic diversity at both phenotypic and molecular level is essential for any crop improvement program as it plays an important role in optimizing the breeding strategies. In linseed, the significant genetic variation in morphological traits and at molecular level has been reported (Begum *et al*., 2007; Khan *et al*., 2013; Dikshit and Shivaraj, 2015). In the present study, 58 genotypes of linseed were grown in two successive years (2012-2013 and 2013-2014), that showed an appreciable level of genetic variability for various morphological traits. Similar to the present findings, significant genetic variability was also reported by Begum *et al*. (2007) while studying morphological trait based genetic variation in 36 linseed genotypes. All the 36 genotypes were grouped into five clusters. Cluster mean value for plant height varied from 42.0 cm to 96.0 and seed yield/plant from 1.49 to 4.54g. Dikshit and Shivaraj (2015) also observed morphological variation in linseed collected from different states of India. They noticed Shannon diversity index varying from 0.23 to 0.70 and phenotypic coefficient of variation ranged from 6.0 to 37.1%. They reported morphological variation for plant height from 23.2 cm to 45.9 cm, number of capsules/plant from 26.6 to 86.3 and oil content from 31.9 to 41.5%. Worku *et al.* (2015) have evaluated large numbers of Ethiopian linseed germplasm based on morphological traits and reported that the days to flowering and days to maturity varied from 37.0 to 86.0 days and 88.0 to 159.0 days respectively. Soto-Cerda *et al.* (2013) evaluated 407 flax accessions using SSR markers and developed association mapping panel. They reported an average of 5.32 alleles per markers, higher than that observed in the present investigation probably due to the large number of genotypes and markers used.

Based on Euclidean analysis of agro-morphological traits, the genotypes grouped in clusters II and IV would be the most genetically diverse due to the maximum inter cluster distance between them while the accessions within cluster I are least genetically diverse due to their minimum intra cluster distance. The genotypes present in cluster II could be used as a parent in hybridization based genetic improvement program for increasing number of branches/plant and number of capsules/plant as it has the highest cluster mean for both the traits. The genotypes included in cluster IV could be used for increasing oil content and test weight as it has the highest cluster mean for the aforesaid characters. Therefore, grouping a large number of linseed genotypes into few homogenous clusters facilitates the selection of diverse parents for crossing program. Khan *et al.* (2013) clustered 55 genotypes into thirteen clusters in which cluster I and II genotypes were found to be most suitable for hybridization. Adugna *et al.* (2006) grouped sixty genotypes into 18 clusters across which the genotypes of four clusters were observed to be highly genetically diverse. Dikshit *et al*. (2015) evaluated 111 accessions for genetic diversity and reported that clustering pattern revealed considerable amount of diversity in the linseed germplasm.

Principal component analysis helps to identify the most relevant characters by explaining the total variation in the original set of variables with few of the components as possible, and it reduces the complexity of the problem. We observed that the PC_1 accounted for 68.10% of the total variation which had a maximum and a positive weight on this component. The traits with the largest impact on the components showed the highest rate of variation and hence can be used for grouping genotypes effectively. Similar findings were also reported in rice (Hoque *et al*., 2015), potato (Lohani *et al*., 2012) and wheat (Khodadadi *et al*., 2011).

The application of DNA based marker technology in the assessment of genetic diversity improves the efficiency of parental selection for breeding programs (Falconer and Mackay, 1996). In the present study, the

genetic diversity was also evaluated through the microsatellite markers along with the morphological traits for more reliable identification of diverse genotypes. A total of 41 alleles were identified by 12 polymorphic SSR with an average of 3.42 alleles per SSR among the 58 genotypes. The markers showed an average *PIC* of 0.39, which confirms that SSR markers used in this study were highly informative. Roose-Amsaleg *et al*. (2006) reported an average of 3.32 allele per locus through 28 SSR among 93 accessions of linseed. In contrary, Deng *et al.* (2011) reported a total of 129 alleles with an average of 3.39 alleles per locus among 8 flax genotypes by means of using 38 SSR primers while the *PIC* value varied from 0.11 to 0.88 with an average of 0.42. Pali *et al*. (2014) observed 61 alleles with an average of 2.17 among 4 flax genotypes using 28 SSRs and their *PIC* 0.37 to 0.75 with an average of 0.41. A pair-wise genetic similarity matrix based neighbor joining tree was constructed to understand the extent of genetic diversity among the Indian genotypes of linseed. In the present set of genetic material high level of genetic diversity was noticed (Average similarity coefficient: 0.54±0.10). Four genotypes named as JRF-4, Sweta, Baner and RKY-14 was highly diverse among all. Out of these four diverse varieties were grouped together in two clusters (clusters I and II). Among the four diverse genotypes, JRF-4 (30%), KL-213 (31%), Baner (32%) and Sweta (34%) also showed differences in oil content and thus they could be utilized as parental lines for developing mapping population and tagging oil content related QTLs. As analyzed by UPGMA dendogram the above 4 genotypes are also present in different clusters which reveals convergent relationship between phenotypic and molecular level.

The present investigation was an attempt to evaluate the diversity through both morphological traits and SSR markers in linseed which showed a high level of variability among 58 Indian linseed genotypes. The most diverse genotypes i.e. JRF-4, Sweta, Baner and RKY-14 can provide a broad spectrum of variability in segregating populations and may be used as parents for future hybridization programs to develop desirable linseed cultivars. Furthermore, the wider phenotypic and molecular variability observed represents a good indication for the importance of linseed breeding programs.

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خصوصیات کشاورزی- مورفولوژیکی و مارکرهای ریزماهواره ای براساس تنوع ژنتیکی در ژنوتیپ های هندی بزرک (*Linum usitatissimum* L.)

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چکیده

بزرک دانه های روغنی و محصول فیبر دار مهم در هند می باشد. هدف این مطالعه، بررسی تنوع ژنتیکی و الگوهای روابطی بین ۵۸ ژنوتیپ از طریق ۱۰ صفت مورفولوژیک و ۱۲ ریزماهواره (SSR) نشانگر بود. تجزیه و تحلیل اقلیدسی صفات زراعی و مورفولوژیک ، ۵۸ ژنوتیپ را به چهار خوشه تقسیم کرد به طوریکه که خوشه Iبزرگترین با ۲۰ الحاقیه بود و خوشه II و IV با توجه به حداکثر فاصله درون خوشه، متنوع ترین طبقه بندی شدند. تجزیه و تحلیل مولفه های اصلی نشان داد سه صفت برای بیش از ۸۶ درصد از تنوع کل حساب شده است. در مجموع ۴۱ آلل تکثیر شده با ۱۲ نشانگر SSRبه طور میانگین ۵.۷۱ الل در هر لوکوس پرایمر دارد. محتوای اطلاعات چندشکلی (PIC) بین ۰.۱۸–۰.۷۸ متفاوت بود. بر اساس ضریب تشابه جاکارد، فاصله ژنتیکی ۰.۰۷–۰.۸۹ با میانگین ۰.۱۰ ± ۰.۵۴ در نوسان بود. بر اساس فاصله ژنتیکی ژنوتیپ های RkY-14، KL-213، LC-185 و Kartika متفاوت ترین در میان تمام ژنوتیپ های مورد مطالعه پیدا شد. متنوع ترین ارقام مشخص شده در این مطالعه می تواند به عنوان برنامه های اصلاحی در گسترش پایه ژنتیکی ژرم پلاسم بزرک استفاده می شود.