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Genetic variability and divergence analysis for seed yield, its component characters in linseed (*Linum usitatissimum* L.) over environments in north western Himalayas

Satish Paul and Arpna Kumari

Abstract
Genetic variability and divergence studies were conducted among 18 genotypes of Linseed. Phenotypic coefficient of variation (PCV) was higher than genotypic coefficient of variation (GCV) for all the observed characters. Based on the present study, high heritability coupled with high genetic advance was observed for seed yield per plot and straw yield per plot. The result suggested the importance of additive gene action for their inheritance and improvement could be brought about by phenotypic selection. The cluster analysis grouped the genotypes into 2, 3 and 3 clusters in Env. I, Env. II and pooled over the environments, respectively indicating that clustering pattern was different in Env. I. Crosses involving parents belonging to most divergent clusters would be expected to manifest maximum heterosis and release of desirable recombinants in segregating generations. Therefore, in Env. I, parents should be selected from cluster combination from clusters I and II, in Env. II and in pooled over the environments from clusters I and III. The character 1000-seed weight contributed maximum towards total genetic divergence in Env. I, Env. II and in pooled over the environment among 18 genotypes under study.

Keywords: Linseed, genetic variability, genetic divergence, cluster analysis

Introduction
Linseed (*Linum usitatissimum* L.) also known as 'Alsi' is one of the most important oilseed crop ranking next to rapeseed and mustard in the area as well as production. According to Vavilov [1] linseed/flax has two centres of origin *viz.*, the oil type originated in south west Asia and fibre type originated in Mediterranean region. Fibre flax is grown mainly in Northern Europe, Russia and China, but linseed is the primary type grown in Canada, USA, Argentina and India as well as Russia and China [2]. The current worldwide acreage is only 24.86 lakh ha, whereby the European Union (EU) member countries contributed 10 per cent of the total annual production of 20.55 lakh tonnes seed with the productivity of 827 kg/ha. India is an important linseed growing country in the world ranking second in area 2.96 lakh ha (16%) after Canada, with annual production of 1.49 lakh tonnes, whereas, in terms of productivity India (502 Kg/ha) is far behind to USA (3389.2 Kg/ha), Egypt (1333.3 Kg/ha), Canada (1197.3 Kg/ha), China (1029.4 Kg/ha) and Russia (786.8 Kg/ha) [3]. Linseed being an important oilseed crop has a very low average productivity in India as well as in Himachal Pradesh because of many reasons. Such as, narrow genetic base of the present varieties (developed through continuous selection of the existing ones), common heritage, cultivation in marginal lands and due to biotic and abiotic stresses. As we know that the genetic diversity is pre-requisite for genetic improvement of any crop. The further development of superior varieties largely depends on the wide genetic base of diverse parents and the breeding approaches being followed. There is need to look for diverse genotypes so that genetic improvement over existing linseed varieties can be achieved.
Genetic diversity being so important for effective breeding programmes can be quantified using multivariate analysis. The D^2-statistics measures the degree of diversification and determines the relative proportion of each component character to the total divergence. Initial diversity assessments in flax were carried out using morphological parameters [4]. Flax germplasm collections contain thousands of accessions of *L. usitatissimum* and related species, of which, subsets were assessed for the extent of genetic diversity for morphological characteristics [5-7]. Thus, the study was undertaken to evaluate both local and indigenous

linseed genotypes through D^2 analysis to measure the nature and magnitude of genetic diversity for yield and yield related traits [8].

2. Materials and Methods

2.1 Plant material and experimental site

An experiment was conducted with 18 (local and indigenous) genotypes of Linseed (Table 1) at the experimental farm of the Department of Crop Improvement, CSKHPKV, Palampur, during the two *Rabi* seasons *viz.*, 2011-12 and 2012-13. The experimental site is located at 1290.8 m amsl and at $32^0$8' N latitude and $76^0$3' E longitude. Agro-climatically, the location represents the mid-hill zone of Himachal Pradesh (Zone-II) and is characterized by humid sub-temperate climate with high rainfall (2,693 mm). The soils are clay loam to silty clay loam in texture. The reaction of soil is acidic with pH ranging from 5.0 to 5.6.

2.2 Experimental design and layout

At two environments, experimental layout was a randomized complete block design with two replications. Row to row and plant to plant spacings were kept 25 cm and 5 cm during both the seasons. A pre-sowing irrigation was given to ensure proper germination. The experimental field was well prepared and FYM was added before sowing. The recommended dose of fertilizer (50 Kg N, 40 Kg P^2O^5 and 20 Kg K^2O/ha) was applied. Half dose of nitrogen and full dose of phosphorous and potash was applied as basal and the remaining half nitrogen was top dressed after 2 months of sowing. Irrigation was given whenever required and regular weeding was done to keep the trial free from weeds.

2.3 Recording of observations

The observations were recorded on thirteen morphological and yield related characters, *viz*., days to 50 per cent flowering, days to 75 per cent maturity, plant height, technical height, number of primary branches per plant, number of secondary branches per plant, capsules per plant, seeds per capsule, 1000-seed weight, seed yield per plot, biological yield per plot, straw yield per plot and harvest index. The data on morphological and yield related characters were recorded on randomly selected five competitive plants except days to 50 per cent flowering and days to 75 per cent maturity, seed yield per plot, biological yield per plot and straw yield per plot which were recorded on plot basis.

2.4 Statistical analysis

In order to assess and quantify the genetic variability among the genotypes for the characters under study, the phenotypic coefficient of variation (PCV), genotypic coefficient of variation (GCV), heritability and genetic advance were estimated following standard statistical procedures [9, 10]. The genetic divergence among genotypes was computed by means of Mahalanobis D^2 technique [8]. The difference between the genotypes for the set of characters was tested and the genotypes were grouped into clusters following Tocher's method [11].

3. Results and Discussion

3.1 Estimates of parameters of variability

Analysis of variance revealed that highly significant differences were found for all the traits studied except for seeds per capsules in both the seasons [12]. The estimates of PCV were higher than their corresponding GCV for all characters studied which indicated that apparent variation is not only due to genotypes but, also due to the influence of environment (Table 2). Therefore, caution has to be exercised in making selection for these characters on the basis of phenotype alone as environmental variation is unpredictable in nature as earlier reported by [13]. In Env. I, a wide range of variability was observed for all the characters studied (Table 2). The moderate estimates of PCV and GCV were observed for number of secondary branches per plant, seed yield per plot, biological yield per plot and straw yield per plot. Heritability estimates were high for days to 50 per cent flowering, days to 75 per cent maturity, 1000-seed weight and seed yield per plot. The high expected genetic advance expressed as percentage of mean was observed for seed yield per plot. Based on the present study, high heritability coupled with high genetic advance was observed for seed yield per plot (Table 2). The result suggested the importance of additive gene action for their inheritance and improvement could be brought about by phenotypic selection. Similarly, Mishra and Yadav [14], Singh [15] also reported high GCV for seed yield per plant and branches per plant and Savita [16] reported high PCV and GCV for seed yield per plant, oil yield per plant and primary branches per plant. On the other hand, in Env. II, moderate estimates of PCV and GCV were recorded for 1000-seed weight, seed yield per plot, biological yield per plot, straw yield per plot and harvest index (Table 2). Heritability estimates were high for days to 50 per cent flowering, days to 75 per cent maturity, capsules per plant, 1000-seed weight, seed yield per plot, biological yield per plot and straw yield per plot. Similar results were observed by different workers *viz*., Payasi *et al.* [17] and Kant *et al.* [18] for seed yield per plant and Singh [15] for days to 50 per cent flowering. High genetic advance expressed as percentage of mean was observed for straw yield per plot. Based on the above results in Env. II, high heritability coupled with high genetic advance was observed only for straw yield per plot. Kant *et al.* [18] reported high heritability coupled with moderate genetic advance for seed yield per plant, capsules per plant and primary branches per plant. In pooled over the environments, moderate PCV was recorded for number of primary branches per plant, number of secondary branches per plant and 1000-seed weight (Table 2). Estimates of GCV were moderate for seed yield per plot, biological yield per plot and straw yield per plot. Heritability estimates were high for days to 50 per cent flowering, days to 75 per cent maturity, 1000-seed weight, seed yield per plot, and straw yield per plot. Expected genetic advance, expressed as percentage of mean was moderate for 1000-seed weight.

3.2 Genetic divergence studies

3.2.1 Grouping of genotypes into clusters

On the basis of D^2 values for all possible pairs, 18 genotypes of Linseed were grouped into various clusters using Tocher's procedure and dendrograms were made. The composition of clusters so constructed both for Env. I and Env. II as well as pooled over the environments are presented in Tables 3 and 4 and Fig. 1, 2 and 3. The cluster analysis revealed that all the genotypes could be grouped into 2, 3 and 3 clusters in Env. I, Env. II and pooled over the environments, respectively indicating that clustering pattern was different in Env. I. Different clustering pattern were also reported on linseed by some earlier workers [19-24]. The results suggested that the genotypes from same geographical locations fell into different clusters as well as genotypes from different geographical

locations fell into same cluster. Indicating that clustering of populations did not follow their geographic distribution. Similar observations have been reported by [25-27].

3.2.2 Average intra and inter-cluster distances

Average intra- and inter-cluster distances for Env. I, Env. II and pooled over the environments are presented in Table 5. In Env. I, the intra-cluster distance was comparable in cluster I while for clusters II, it was zero, because it was constituted by single genotype. In Env. II, the intra-cluster distance was comparable in all the clusters *i.e.* cluster I, cluster II and Cluster III. Since the intra-cluster distances were more, therefore the chances of developing good segregants by hybridization among parents within clusters would be more, therefore it is logical to attempt crosses between the genotypes falling in same cluster based on intra-cluster distances. In the analysis of genetic divergence pooled over the environments, the intra-cluster distance was comparable for cluster I while for clusters II and III, intra-cluster divergence was zero (Table 5).

In Env. I, maximum genetic divergence was recorded between clusters I and II. On the other hand in Env. II, the maximum inter-cluster divergence was observed between clusters I and III followed by clusters II and III and between clusters I and II. The analysis of genetic divergence pooled over the environments, highest inter-cluster divergence was recorded between I and III followed by clusters II and III and between clusters II and III (Table 5). This clearly indicates that the genotypes included in these clusters are having sufficient genetic diversity and selection of parents from diverse clusters could be used in hybridization programme for improving seed yield. Crosses involving parents belonging to most divergent clusters would be expected to manifest maximum heterosis and release of desirable recombinants in segregating generations. In a similar study on inter and intra-cluster distances, Fulkar *et al.* [20] reported maximum inter cluster distance between cluster II and X, Tadesse *et al.* [27] reported between cluster I and IV, cluster I and III and minimum for cluster VIII and IX and IX and X. Whereas, Srivastava *et al.* [21] reported maximum inter-cluster distance between cluster I and VI based on inter-cluster distance and per se performance of genotypes. Indicating that the genotypes from divergent clusters can be intercrossed to obtain high heterotic response and also to recover desirable transgressive segregants as discussed earlier.

3.2.3 Cluster means and contribution of individual character towards divergence

Based on cluster means in Env. I, cluster I was characterized by genotypes had highest cluster mean values for technical height, number of primary branches per plant, number of secondary branches per plant, capsules per plant, seeds per capsule, 1000-seed weight, harvest index along with earliest in days to 50 per cent flowering (Table 6). The genotypes from cluster II had shortest plant height along with earliest in days to 75 per cent maturity coupled with highest mean values for seed yield per plot, biological yield per plot and straw yield per plot (Table 6). In Env. II, the highest cluster mean was recorded by cluster I for number of primary branches per plant and 1000-seed weight along with the lowest cluster mean for days to 50 per cent flowering. The genotypes from cluster II had shortest plant height coupled with highest cluster mean values for number of secondary branches per plant and seeds per capsule. Cluster III had the genotypes with highest cluster mean values for technical height, capsules per plant, seed yield per plot, biological yield per plot, straw yield per plot and harvest index along with earliest in days to 75 per cent maturity. In pooled over the environments, cluster I was represented by the genotypes having maximum cluster mean for 1000-seed weight along with earliest in days to 50 per cent flowering (Table 6). The genotypes from cluster II had shortest plant height coupled with highest cluster mean values for number of primary branches per plant, number of secondary branches per plant, capsules per plant, seeds per capsule, straw yield per plot and harvest index. Cluster III consisted of genotypes with highest mean values for technical height, seed yield per plot and biological yield per plot along with earliest in days to 75 per cent maturity (Table 6).

The contribution of individual characters to divergence has been worked out in terms of number of times it appeared first (Table 7). The character 1000-seed weight contributed maximum towards total genetic divergence in Env. I (77.78 %), Env. II (8.58 %) and in pooled over the environment (88.89 %) among 18 genotypes under study. Srivastava *et al.* [21] in their study on genetic divergence reported that seed yield per plant contributed maximum towards genetic divergence followed by number of capsules per plant and days to flowering.

Table 1: List of linseed genotypes and their parentage used in the study

S. No.	Genotype	Parentage	Category on the basis of use
1.	KL-225	K2 × EC-23239	Dual-purpose
2.	KL-226	Aoyagi × JRF-2	Dual-purpose
3.	KL-227	Flak-1 × Janaki (DP)	Dual-purpose
4.	KL-228	Polf-22 × KL-31	Dual-purpose
5.	KL-229	Polf-16 × KL-31	Dual-purpose
6.	KL-230	Aoyagi × RL-33-4	Dual-purpose
7.	KL-231	Polf-16 × KL-1	Dual-purpose
8.	KL-232	Polf-38 × Janaki	Dual-purpose
9.	KL-233	Flax purple × Gaurav	Dual-purpose
10.	KL-234	Polf-22 × Jeewan	Dual-purpose
11.	KL-235	Polf-22 × Aoyagi	Dual-purpose
12.	KL-236	Jeewan × Janaki	Dual-purpose
13.	KL-237	Aoyagi × Jeewan	Dual-purpose
14.	KL-238	Aoyagi × Nagarkot	Dual-purpose
15.	KL-239	Polf-27 × RL-33-4	Dual-purpose
16.	Nagarkot	New River × LC-216	Dual-purpose
17.	Jeewan	Sumit × LC-216	Dual-purpose
18.	Him Alsi-2	Ec-21741 × LC-216	Dual-purpose

Dual-purpose flax: used for both fibre and oil extraction

Table 2: Estimates of different parameters of variability for various characters in Env. I, Env. II and pooled over the environments

Characters	Env. I				Env. II				Pooled over the environments			
	PCV (%)	GCV (%)	h^2_{bs} (%)	Genetic advance (%) of mean	PCV (%)	GCV (%)	h^2_{bs} (%)	Genetic advance (%) of mean	PCV (%)	GCV (%)	h^2_{bs} (%)	Genetic advance (%) of mean
Days to 50% flowering	1.22	1.15	88.64	2.23	1.73	1.59	84.61	3.02	0.90	1.05	76.82	1.42
Days to 75% maturity	0.91	0.76	69.64	1.30	1.12	1.12	99.95	2.32	0.57	0.58	74.28	0.87
Plant height (cm)	7.03	5.42	59.53	8.62	8.28	6.20	56.06	9.57	3.42	5.18	49.28	3.47
Technical height (cm)	12.27	8.01	42.67	10.78	10.73	7.83	53.26	11.77	5.12	5.97	34.70	3.66
No. of primary branches/ plant	9.72	6.31	42.18	8.44	6.28	0.48	5.90	7.65	13.12	2.85	15.08	4.08
No. of secondary branches/ plant	18.91	12.02	40.42	15.75	9.38	1.38	2.18	0.42	18.97	5.28	16.24	6.35
Capsules/ plant	14.09	9.75	47.95	13.91	7.44	5.88	62.48	9.57	5.84	4.29	27.76	3.34
Seeds/ capsule	7.12	0.97	1.85	0.27	5.62	3.49	38.50	4.45	9.24	1.77	8.51	1.62
1000-seed weight (g)	7.53	7.51	99.61	15.46	10.59	10.59	99.94	21.81	11.53	8.72	99.79	23.69
Seed yield/ plot (g)	20.39	18.24	79.95	33.59	18.92	15.93	70.85	27.62	2.12	12.30	60.17	2.63
Biological yield/ plot (g)	22.96	16.02	48.72	23.04	16.44	14.67	79.69	26.99	1.13	11.67	52.89	1.23
Straw yield/ plot (g)	22.77	16.16	50.40	23.64	21.93	20.22	84.98	38.39	1.68	14.74	62.39	2.16
Harvest index (%)	18.55	7.47	16.21	6.19	16.22	11.37	49.08	16.40	7.62	8.19	25.28	3.97

Table 3: Clustering pattern of 18 Linseed genotypes on the basis of D^2-analysis in Env. I

Cluster number	Number of Genotypes	
I	17	KL-233, KL-239, KL-232, KL-230, Jeevan, KL-226, KL-236, KL-235, KL-237, KL-227, KL-238, KL-231, KL-228, KL-234, Nagarkot, Himalsi-2, KL-225
II	1	KL-229

Table 4: Clustering patterns of 18 Linseed genotypes on the basis of D^2-analysis in Env. II and pooled over the environments

Cluster number	Env. II		Pooled over the environments	
	Number of Genotypes		Number of Genotypes	
I	11	KL-233, KL-235, KL-234, KL-230, Himalsi-2, KL-226, KL-232, KL-228, KL-239, KL-238, KL-236	16	KL-230, KL-233, KL-228, KL-235, Nagarkot, KL-232, KL-226, KL-239, KL-236, Jeevan, KL-238, KL-237, KL-231, KL-234, Himalsi-2, KL-225
II	5	Nagarkot, Jeevan, KL-231, KL-237, KL-225	1	KL-227
III	2	KL-227, KL-229	1	KL-229

Table 5: Average intra- and inter-cluster distances in Env. I, Env. II and pooled over the environments

Env. I			Env. II				Pooled over the environments			
Clusters	I	II	Clusters	I	II	III	Clusters	I	II	III
I	11.20 (3.35)	37.43 (6.12)	I	12.55 (3.54)	23.06 (4.80)	82.97 (9.11)	I	11.84 (3.44)	28.48 (5.34)	55.14 (7.43)
II		0.00 (0.00)	II		11.20 (3.35)	62.95 (7.93)	II		0.00 (0.00)	27.32 (5.23)
			III			16.59 (4.07)	III			0.00 (0.00)

Values in bold figures are intra-cluster distances; Values in parenthesis are $\sqrt{D^2} = D$ values

Table 6: Cluster means for different characters Env. I, Env. II and pooled over the environments

Clusters / Characters	Env. I					Env. II						Pooled over the environments					
	I	II	Mean	Minimum	Maximum	I	II	III	Mean	Minimum	Maximum	I	II	III	Mean	Minimum	Maximum
Days to 50 % flowering	141.71	145.33	143.52	141.71	145.33	155.03	156.00	156.83	155.95	155.03	156.83	148.58	149.00	150.33	149.30	148.58	150.33
Days to 75% maturity	196.31	196.00	196.16	196.00	196.31	214.91	215.60	212.00	214.17	212.00	215.60	205.71	204.33	204.00	204.68	204.00	205.71
Plant height	54.38	51.97	53.18	51.97	54.38	73.43	71.20	73.15	72.59	71.20	73.43	63.63	61.02	64.13	62.93	61.02	64.13
Technical height	32.54	29.00	30.77	29.00	32.54	45.79	43.97	47.03	45.60	43.97	47.03	38.74	39.72	40.33	39.60	38.74	40.33
No. of primary branches/ plant	3.98	3.73	3.86	3.73	3.98	4.56	4.51	4.47	4.51	4.47	4.56	4.26	4.27	4.10	4.21	4.10	4.27
No. of secondary branches/ plant	3.58	2.87	3.23	2.87	3.58	3.57	3.76	3.67	3.67	3.57	3.76	3.59	3.90	3.23	3.57	3.23	3.90
Capsules/ plant	19.41	18.67	19.04	18.67	19.41	27.10	28.01	29.53	28.21	27.10	29.53	23.39	25.17	23.53	24.03	23.39	25.17
Seeds/ capsule	6.82	6.67	6.75	6.67	6.82	7.31	7.51	7.47	7.43	7.31	7.51	7.10	7.13	7.00	7.08	7.00	7.13
1000-seed weight	6.88	5.18	6.03	5.18	6.88	7.18	6.65	5.00	6.28	5.00	7.18	6.95	5.97	4.98	5.97	4.98	6.95
Seed yield/ plot	251.08	263.33	257.21	251.08	263.33	419.39	435.33	498.33	451.02	419.39	498.33	337.34	376.67	385.00	366.34	337.34	385.00
Biological yield/ plot	966.47	1050.00	1008.24	966.47	1050	1531.82	1496.67	1691.67	1573.39	1496.67	1691.67	1245.73	1266.67	1400.00	1304.13	1245.73	1400.00
Straw yield/ plot	522.94	586.67	554.81	522.94	586.67	765.15	706.67	900.00	790.61	706.67	900.00	630.73	761.67	760.00	717.47	630.73	761.67
Harvest index	26.76	25.73	26.25	25.73	26.76	27.63	29.36	29.45	28.81	27.63	29.45	27.33	30.49	27.34	28.39	27.33	30.49

Table 7: Contribution of individual characters to the divergence among 18 genotypes of Linseed in Env. I, Env. II and pooled over the environments

Characters	Env. I		Env. II		Pooled over the environments	
	Times ranked Ist	Contribution (%)	Times ranked Ist	Contribution (%)	Times ranked Ist	Contribution (%)
Days to 50% flowering	9	5.88	1	0.65	1	0.65
Days to 75% maturity	1	0.65	12	7.84	5	3.27
Plant height	0	0.00	0	0.00	0	0.00
Technical height	5	3.27	0	0.00	1	0.65
No. of primary branches/ plant	6	3.92	0	0.00	0	0.00
No. of secondary branches/ plant	0	0.00	0	0.00	0	0.00
Capsules/ plant	4	2.61	0	0.00	0	0.00
Seed/ capsule	1	0.65	0	0.00	1	0.65
1000-seed weight	119	77.78**	134	87.58**	136	88.89**
Seed yield/ plot	5	3.27	1	0.65	4	2.61
Biological yield/ plot	0	0.00	5	3.27	2	1.31
Straw yield/ plot	0	0.00	0	0.00	2	1.31
Harvest index	3	1.96	0	0.00	1	0.65

** Maximum values

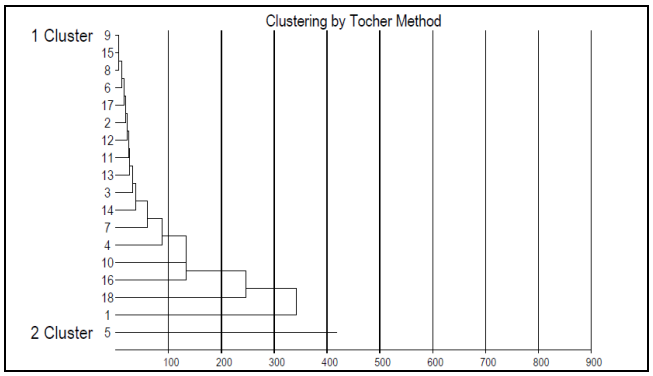

Fig 1

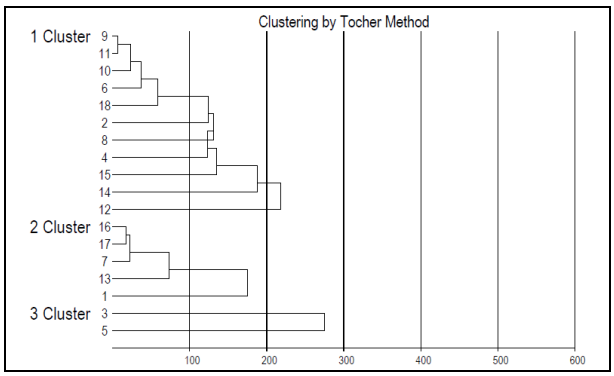

Fig 2

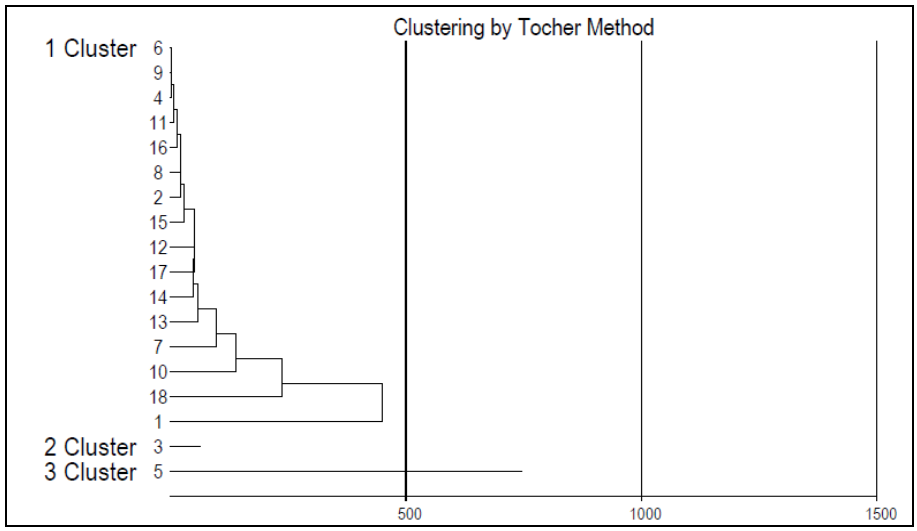

Fig 3

Fig 1, 2 3: Dendrograms showing grouping of 18 Linseed genotypes generated using D^2 cluster analysis (Tocher's method) in Env. I, Env. II and pooled over the environments

4. Conclusion

This study will provide a more concrete picture of the extent and distribution of genetic diversity in the flax gene pool and help to facilitate germplasm management and enhance the utilization of flax germplasm in flax breeding. The overall results indicated that a considerable diversity exists in the set of accessions analysed in this investigation. Considering the importance of diversity in germplasm improvement and that a greater combining ability is expected in crosses among genetically diverse parents, the genotype belonging to different groups identified during the present study will constitute promising parents for hybridization in linseed improvement programme.

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