Full Length Research Paper

Seed oil diversity of Ethiopian linseed (*Linum usitatissimum* L.) landraces accessions and some exotic cultivars

Mulusew Fikere[1]*, Firew Mekbib[2] and Adugna Wakjira[3]

[1]Oromia Agricultural Research Institute (OARI), Sinana Agricultural Research Center, P.O.Box 208, Bale Robe, Ethiopia.
[2]Department of Plant Science, Faculty of Agriculture and Environmental Science, Haramaya University, P.O.Box 138 Dire Dawa, Ethiopia.
[3]Ethiopian Institute of Agricultural Research (EIAR), P.O.Box 2003, Ethiopia.

Keeping in view, lack of adequate information on genetic diversity in the Ethiopian linseed (*Linum ustatisimum* L.) landraces on one hand and its immense importance in the agricultural systems on the other hand in Ethiopia, 49 accessions collected from five regions of Ethiopia together with fifteen exotic cultivars were used in this study with the objective of investigating biochemical diversity between and within germplasms. The variation among and between Ethiopian linseed landrace accession and exotic cultivars based on grand mean responses revealed that, higher oil content (39.8%) was recorded by exotic cultivar [PI-52335] and the lowest was by Acc. 219333 from Oromia with 30.63%. The highest palmitic proportion (7.06% of all fatty acids) was observed in an Ethiopian accession [Acc. 237494] collected from the province Tigray. Maximum and minimum stearic acid was found in accessions collected from the Oromia Region with 6.21 and 4.74% from Acc. 13545 and 13756, respectively. Maximum oleic acid (21.4%) was also observed in Acc. 13545 that was collected from the Oromia region. Crude protein, crude fat and iodine value were analyzed and discussed. The diversity of Ethiopian linseed landraces will keep serving as a reservoir for future genetic improvements. The first three principal components accounted for more than 73.3% of the total variation. The genotypes were grouped into five clusters for which Mahalanobis' D^2 statistics was calculated. Maximum distance was observed between cluster 1 and 4 (D^2 = 32.79) and the minimum (D^2 = 5.08), between clusters 2 and 1 and also between 3 and 2. A correlation analysis revealed the presence of associations among seed oil traits. It could also be concluded that there was ample variation among Ethiopian linseed landraces and exotic cultivars, implying opportunities for genetic improvements by plant breeding.

Key words: Genetic diversity, fatty acid, flax, seed oil content.

INTRODUCTION

Linseed (2n = 30) is an important oilseed and fiber crop which belongs to the family Linaceae. It is believed that this crop species may have originated from *Linum* *angustifolium* Huds. (2n = 30) native to the Mediterranean region (Cooke, 1903; Legesse, 2010). It is one of the widely grown and economically important oil crops for

*Corresponding author. E-mail: mulufiker@gmail.com. Tel: +251913120681.

industrial use. Linseed is an annual field crop that is largely grown in temperate climates (Mansby et al., 2000) and cool tropics including the highlands (>2500 m above sea level) of Ethiopia. It is the second most important oil crop in the highlands of Ethiopia in terms of area and production (Adefris et al., 1992; Adugna, 2000). Linseed requires cool temperatures during its growing period to produce good yields. The mean temperature can range from 10 to 30°C although the crop grows best within 21 and 22°C.

In Ethiopia, linseed has been cultivated for two primary purposes, seed and oil use. It has traditionally been used for food and as a cash crop since ancient times (Seegler, 1983). It is now grown primarily for food and to generate revenue, either in local markets or by export. For food, the seeds are usually roasted, ground, mixed with spices and water, and served with various local breads. It is also consumed in soups, with porridges and cooked potatoes, etc. Limited amounts are also pressed locally for its edible oil, which is often blended with other high quality vegetable oils.

As a result, linseed is currently becoming popular worldwide for its functional food products. Detailed studies were not done to generate adequate information on Ethiopian linseed that is required by the current and future breeding programmes. Ethiopia is considered as one of the center of diversity for linseed (Seegler, 1983; Adugna and Adefris, 1995), and the wide range of agro-climatic conditions in the country (MoA, 1998; Adugna, 2000) may have contributed to its diversification. Information on the extent of genetic diversity of Ethiopian linseed landraces and some exotic cultivars based on biochemicals characters is meager. The present study was envisaged to assess the diversity of seed oil characteristics in Ethiopian linseed landraces and some exotic cultivars.

MATERIALS AND METHODS

Plant materials

A total of 64 linseed genotypes were used, forty-nine of which were landrace accessions collected from five different administrative regions of Ethiopia (Amhara, Oromia, Southern Nations, Nationalities and Peoples (SNNP), Tigray and Somali region) and obtained from the Institute of Biodiversity Conservation (IBC), Addis Ababa, Ethiopia, along with fifteen exotic cultivars (Tables 1 and 2). The germplasms were grown at Sinana Agricultural Research Center experimental site in 2011/2012 in lattice square design with two replications. A total of five randomly selected plants per replication were used in the analysis. The average minimum and maximum annual temperature of the study area was about 9.4 and 21.2°C, respectively. In the past 11 years, the total annual rainfall distribution ranged from 534.9 to 1003.4 mm, while the average annual rainfall was about 752.4 mm. The soil was clay in texture with slightly acidic pH (SARC, 2005).

Laboratory procedures

Laboratory work of the present study was conducted at JIJE

analytical service laboratory and parts of the samples were analyzed at Holeta Agricultural Research Center in oilseeds and nutrition laboratories. Data were recorded on oil content, crude protein, crude fat, iodine value, fatty acid composition such as linoleic, stearic, palmitic, linolenic and oleic were analyzed using the following procurers.

Oil content determination

The nuclear magnetic resonance (NMR) analyzer was used to measure the oil content of seeds by measuring the liquid proton (H$^+$) content of the seed. The principle of NMR is that the oilseed placed in the NMR, which possess magnetic field and the nuclear H atom in oilseeds absorb radio-frequency energy of specific frequency and exhibit resonance due to the magnetic field. Then, the oil content was measured using the procedure of Röbbelen et al. (1989).

Crude protein determination

Protein content was determined by the AOAC Official 984.13 Method. Copper Catalyst Kjeldahl Method.

Crude fat determination

AOAC Official Method 920.39 extraction procedure was applied and determined by the Soxhlet extraction method with diethyl ether method. In this extraction procedure, the entire Soxhlet extraction units were used. Extractions of about 2 g seed samples were carried out with anhydrous ether. Thimbles with porosity permitting rapid passage of ether were used. The extraction period varied from 4 h at condensation rate of 5 to 6 drops/s to 16 h at 2 to 3 drops/s. Extract was dried for 30 min at 100°C, cooled and weighed.

Iodine value determination

Iodine value was determined using AOAC Official Method 993.20 Iodine Value of Fats and Oils Wij's (Cyclohexane-Acetic Acid Solvent) Method First Action 1993 IUPAC-AOCS-AOAC Method).

Fatty acid composition determination

Fatty acid composition was determined using near infrared reflectance spectroscopy (NIRS) using the procedure of Röbbelen et al. (1989). Major fatty acid composition such as linoleic, stearic, palmitic, linolenic and oleic were considered in this study. Each parameter was measured as percentage of total fatty acid and was performed on 3 g of samples using Foss NIRS 5000 in the 1108-2492 ranges with an 8 nm steps. The spectrum of each sample was taken by scanning (Win scan version 1.5, 2000, Intrasoft international, L.L.C.).

Statistical analysis

The results obtained from analysis were expressed as means and range values. Mean separation was carried out and reported at 0.01 and 0.05 levels of significance. In addition, the correlations between biochemical parameters were computed at P<0.05 and P<0.01 using SAS statistical program.

Table 1. Geographical origin of Ethiopian linseed (*L. usitatissimum* L.) landraces.

Acc.	Region	Latitude	Longitude	Altitude	S/N	Acc.	Region	Latitude	Longitude	Altitude
10061	SNNP	06-12-00-N	37-37-00-E	2400	26	13758	SNNP	07-10-00-N	36-25-00-E	1790
10064	SNNP	05-49-00-N	36-36-00-E	1410	27	211477	SNNP	05-20-00-N	37-25-00-E	1780
10067	Amhara	11-25-00-N	37-12-00-E	1990	28	212512	Amhara	09-57-00-N	38-54-00-E	1580
10075	Oromia	09-11-00-N	42-23-00-E	1860	29	212747	Amhara	37-15-00-N	11-11-00-E	2340
10079	Oromia	08-52-00-N	40-42-00-E	1950	30	212752	Amhara	38-14-00-N	11-46-00-E	3000
10096	SNNP	08-23-00-N	38-39-00-E	2000	31	216812	Somali	ND	ND	ND
10114	SNNP	07-07-00-N	38-32-00-E	1800	32	219333	Oromia	04-50-00-N	38-05-00-E	1880
13504	SNNP	07-16-00-N	37-44-00-E	2580	33	219969	Tigray	14-02-00-N	38-43-00-E	1800
13519	Oromia	08-57-00-N	37-32-00-E	2420	34	229804	Amhara	10-29-00-N	38-18-00-E	2680
13520	Oromia	09-48-00-N	38-36-00-E	3110	35	230569	Somali	ND	ND	ND
13526	Oromia	07-13-00-N	39-50-00-E	2610	36	230816	Somali	09-29-00-N	42-40-00-E	1980
13529	Oromia	07-11-00-N	38-38-00-E	2160	37	230822	Oromia	09-25-00-N	41-37-00-E	2270
13545	Oromia	08-13-00-N	39-55-00-E	3090	38	231457	Somali	ND	ND	ND
13558	Amhara	11-17-00-N	36-51-00-E	2130	39	233993	Tigray	14-12-00-N	38-52-00-E	2110
13644	Oromia	08-46-00-N	36-30-00-E	2420	40	234002	Tigray	14-05-00-N	38-58-00-E	1860
13657	SNNP	05-58-00-N	37-18-00-E	2000	41	234006	Tigray	14-23-00-N	38-06-00-E	1820
13680	Amhara	12-46-00-N	37-37-00-E	2950	42	235163	Tigray	12-52-00-N	39-32-00-E	2500
13688	Amhara	10-45-00-N	39-59-00-E	2130	43	235177	Tigray	13-40-00-N	39-14-00-E	1780
13698	Amhara	10-34-00-N	39-24-00-E	2635	44	237494	Tigray	13-30-00-N	39-28-00-E	2150
13713	Amhara	10-53-00-N	39-19-00-E	3170	45	238276	Tigray	14-30-00-N	39-51-00-E	2930
13727	Amhara	11-14-00-N	39-41-00-E	2400	46	240666	SNNP	ND	ND	2500
13753	Oromia	08-38-00-N	34-57-00-E	1680	47	243807	Amhara	11-51-00-N	39-24-00-E	3090
13754	Oromia	08-16-00-N	35-07-00-E	1680	48	243810	Tigray	13-38-00-N	39-07-00-E	2580
13756	Oromia	08-21-00-N	36-21-00-E	1900	49	244809	SNNP	ND	ND	2892
13757	Oromia	07-33-00-N	36-37-00-E	1860						

Source: Institute of Biodiversity Conservation (IBC); ND = no data.

Genetic divergence and clustering analysis

Records on all traits were pre-standardized to means of zero and variances of unity before clustering to avoid bias due to differences in measurement scales (Manly, 1986). Clustering of accessions was performed by average linkage method and principal component analysis using the SAS software (SAS Institute, 2001). Points where local peaks of the pseudo F statistic joining with small values of the pseudo t^2 statistic followed by a larger pseudo t^2 for the next cluster fusion and were examined to decide the number of clusters. Genetic distances between clusters as standardized Mahalanobis's D^2 statistics were calculated as:

$$D^2_{ij} = (x_i - x_j)' \, cov^{-1} \, (x_i - x_j)$$

Where, D^2_{ij} = the distance between cases i and j; xi and xj = vectors of the values of the variables for cases i and j and cov^{-1} = the pooled within groups variance-covariance matrix. The D^2 values obtained for pairs of clusters were considered as the calculated values of Chi-square (X^2) and were tested for significance both at 1 and 5% probability levels against the tabulated value of X^2 for 'P' degree of freedom, where P is the number of characters considered (Singh and Chaudhary, 1985). The dendrogram was built based on Ward's agglomerative hierarchical classification with Euclidian distance as a measure of dissimilarity (Ward, 1963) and, subsequently, the results were comple-mented with a pattern analysis based on classification and

Table 2. Passport data of linseed (*Linum usitatissimum* L.) landraces and exotic cultivars used in the study.

List of exotic cultivars*	Backgraound	Source/Origin
R12 D33C X CI 1525/P1/S1	Exotic	Canada/USA
CI 1525 X R12-N27G/P1/S1	Exotic	Canada/USA
CI 1525 X CDC 1747/P1/S1	Exotic	Canada/USA
CI 1525 X R12 D33C/P1/S1	Exotic	Canada/USA
R12- N27G X CI 1525/P1/S1	Exotic	Canada/USA
CDC 1747 X CI 1652/SPS/1	Exotic	Canada/USA
CI 1652 X R12 D33C/SPS	Exotic	Canada/USA
CI 1652 X R12-N27G/SPS1	Exotic	Canada/USA
R12-N100 X CI 1525/SPS1	Exotic	Canada/USA
OMEGA X CI 1525/B/3/M	Exotic	USA/Canada
CI 1652 X OMEGA/B/53/M	Exotic	Canada/USA
CI 1652 X CDC 1747/SPS1	Exotic	Canada/USA
OMEGA X CI 1525/B/1	Exotic	USA/Canada
PI – 523353	Exotic	USA
CI 2698 X P136611	Exotic	Canada /USA

*Source: Holeta Agricultural Research Center.

ordination by scatter plots of the first two principal components using the MINITAB statistical package.

RESULTS AND DISCUSSIONS

The mean, minimum and maximum values for biochemical traits are given in Table 3. There was ample variation on biochemical traits among and between the Ethiopian linseed landraces and exotic cultivars. Higher oil content (39.8%) was recorded by exotic cultivars (PI-523353), while the lowest was obtained from Acc. 219333 (30.63%) acquired from Oromia. The highest palmitic content (7.06%) was observed from the Ethiopian accession [Acc. 237494], collected from Tigray against the minimum (5.99%) that was from exotic cultivars [Omega x CI-1525]. On the other hand, both maximum and minimum (6.21% and 4.74%) stearic acids were scored by accessions 13545 and 13756, respectively that were collected from Oromia region. Similarly, the highest oleic acid was registered by Acc. 13545 (21.4%) and was collected from Oromia ARs against the minimum (15.86%), which was attained from the exotic cultivar, PI-523353. Likewise, the maximum value (14.38%) of linoleic acid was attained by accession collected from SNNP (Acc. 13758), while the minimum (13.21%) was obtained from exotic cultivar [R12 D33cx CI-1525].

Similarly, the highest linolenic acid (57.61%) was obtained from exotic cultivar [PI-523353], whereas the least (50.93%) was obtained by accession 13545 collected from Oromia AR. Maximum crude fat (35%) was secured by accession collected from Amhara ARs against the minimum (30%) and was obtained from Exotic cultivars. Crude protein varied from 18 to 23.3%, an accession

collected from Tigray takes the minimum and the maximum and was exotic cultivar. These findings were in the range with that of Khan et al. (2010) who reported crude protein of 22.37 to 27.24%. This variability in palmitic, stearic, oleic, crude protein, crude fat and iodine values implies that, the Ethiopian linseed accessions can serve as valuable gene pool for breeding. In the experiment, the range for oil content was 30.6 to 39.8%, palmitic 6.0 to 7.1%, stearic 4.74 to 6.21%, oleic 15.86 to 21.4%, linoliec 13.21 to 14.38% and linolenic 50.93 to 57.61%. This result partly agrees with Rubilar et al. (2010) who reported findings of Soxhlet ether extraction procedure as follows: oleic acid (24.1%), linoliec acid (14.7) and linolenic acid (49.3%) but he emphasized that the values can vary based on the extraction methods.

The other important biochemical parameter is iodine value and it is a measure of the saturation of the fatty acids. When the iodine value scale goes up, more double bonds or more unsaturation level occurs in the oils. Hence, evaluation of iodine value can provide a lot of information on its shelf life (the lower the iodine values, the longer the oil keeps). The highest iodine value was recorded by Acc. 13504 collected from SNNP (196.05 Wij's g/100 g) against the lowest (156.4 Wij's g/100 g) that was obtained from Oromia ARs. The current study reveals that the Ethiopian linseed accessions can be exploited for multipurpose quality traits such as iodine values and various fatty acid profiles.

Clustering groups based on biochemical traits

Genetic diversity plays an important role in plant breeding because hybrids between lines of diverse origin generally

Table 3. Mean, maximum and minimum values of biochemical parameters of Ethiopian linseed landrace accessions collected from Administrative regions of Ethiopia (ARs) and exotic cultivars.

Origin	Range	OC (%)	Palmitic	Stearic (%)	Oleic (%)	Linoleic (%)	Linolenic (%)	CF (%)	IV Wij's g/100 g	CP (%)
Amhara (n=12)	Min.	32.5	6.3	5.2	18.1	13.6	51.9	30.83	164.14	19.60
	Max.	37.8	6.9	5.8	20.7	14.2	55.2	43.13	196.05	22.60
	Mean	34.7	6.6	5.5	19.6	13.9	53.1	35.46	176.64	21.10
Oromia (n=12)	Min.	30.6	6.0	4.7	17.4	13.5	50.9	14.83	156.16	16.70
	Max.	37.7	6.9	6.2	21.3	14.3	56.4	41.23	182.92	21.80
	Mean	34.3	6.5	5.4	19.6	13.9	53.2	32.93	173.84	19.25
SNNP (n=5)	Min.	30.9	6.4	5.0	18.1	13.7	51.6	22.10	167.25	18.00
	Max.	35.3	7.0	5.6	20.9	14.3	54.8	37.10	196.05	23.30
	Mean	33.3	6.7	5.3	19.7	13.9	52.9	31.56	176.17	20.65
Somali (n=2)	Min.	33.0	6.5	5.4	19.2	13.8	52.7	31.72	156.16	19.97
	Max.	35.2	6.8	5.6	19.8	14.1	53.5	36.90	181.22	20.00
	Mean	34.2	6.6	5.5	19.5	13.9	53.0	33.14	170.03	19.99
Tigray (n=9)	Min.	34.5	6.2	5.3	16.7	13.7	51.6	30.73	170.98	18.00
	Max.	37.0	7.0	6.1	20.4	14.2	56.6	32.21	175.08	19.40
	Mean	35.5	6.5	5.7	19.2	14.0	53.3	31.36	173.96	18.70
Exotic (n=15)	Min.	35.2	5.9	4.9	15.8	13.2	52.2	29.45	169.67	22.56
	Max.	39.8	6.8	5.9	20.1	14.2	57.6	34.77	183.52	24.00
	Mean	38.4	6.3	5.4	17.5	13.6	55.4	30.65	172.86	23.28
Overall	Min	30.63	5.99	4.74	15.86	13.21	50.93	14.00	156.00	16.70
	Max	39.80	7.06	6.21	21.40	14.38	57.61	43.00	196.05	24.00
Landraces $\overline{x}$		34.4	6.6	5.5	19.6	14.0	53.2	28.45	180.43	19.06
Exotic $\overline{x}$		38.4	6.3	5.4	17.6	13.7	55.5	39.03	169.91	21.05
Grand $\overline{x}$		35.4	6.5	5.5	19.1	13.9	53.7	32.78	179.98	22.56

NB: OC = Oil content (%); CF = crude fat content (%), CP = crude protein content (%); IV = iodine value (Wij's g/100 g).

display a greater heterosis than those between closely related strains. The ward's technique of clustering produced a more understandable portrayal of the 49 landrace accessions and 15 exotic cultivars by grouping them into five clusters, whereby different members within a cluster were assumed to be more closely related in terms of the trait under consideration with each other than those members in different clusters. Similarly, members in clusters with non-significant distance were assumed to have closer relationship with each other than they were with those in signi-

Table 4. Percentage, cumulative variances and Eigen vectors on the first three principal components for nine biochemical characters in 49 linseed landrace accessions and 15 exotic cultivars.

Parameter	PC1	PC2	PC3
Eigen values	4.05	1.55	1.001
% Variance	45	17.2	11.1
Cumulative	45	62.1	73.3
Trait			
Oil content (%)	0.428	-0.120	0.127
Palmitic (%)	-0.434	0.007	0.023
Stearic (%)	-0.129	-0.656	0.340
Oleic (%)	-0.451	-0.172	-0.068
Linoleic (%)	-0.341	0.334	-0.154
Linolenic (%)	0.468	0.169	0.037
Crude protein (%)	0.069	-0.221	-0.900
Crude fat (%)	-0.259	0.139	0.137
Iodine value (Wji's g/100 g)	-0.040	0.563	0.095

Table 5. Grouping and clustering pattern of 49 Ethiopian linseed landrace accession and 15 exotic cultivars into different diversity classes over five clusters based on biochemical characters.

Cluster	Number of accessions in the cluster	Accessions included in the cluster	Origin*
C1	14	212747, 229804, CI 1652 X R12-N27G/SPS1, CI 1652 X CDC 1747/SPS1, CI 1652 X OMEGA/B/53/M, PI – 523353, R12 D33C X CI 1525/P1/S1, 13519, 13644, 230822, 10061, 10114, 233993, 237494	SNNP [2], Oromia [3], Amhara [2], Exotic [5]Tigray [2]
C2	25	10064, 10075, 10079, 13504, 13520, 13526, 13529, 212512, 212752, 216812, 219333, 219969, 230569, 235163, 235177, 238276, 240666, 243807, CI 1525 X CDC 1747/P1/S1, CI 1525 X R12 D33C/P1/S1, CI 1525 X R12-N27G/P1/S1, CI 1652 X R12 D33C/SPS, CI 2698 X P136611 , OMEGA X CI 1525/B/1, OMEGA X CI 1525/B/3/M	Oromia [6], Amhara [3], SNNP [3], Somali [2], Tigray [4], Exotic [7]
C3	13	10067, 10096, 13657, 13680, 13688, 13698, 13753, 13758, 230816, 231457, CDC 1747 X CI 1652/SPS/1, R12- N27G X CI 1525/P1/S1, R12-N100 X CI 1525/SPS1	Amhara [4], Oromia [2], Exotic [3] and SNNP [2], Somali [2]
C4	9	13545, 13558, 13713, 13727, 13754, 13756, 13757, 211477, 234006	Amhara [3] and Tigray [1], SNNP [1] Oromia [4]
C5	3	234002, 243810, 244809	Tigray [2] and SNNP [1]

*Number of accessions in the cluster/origin.

significantly distant clusters (Table 7). Grouping of the accessions and exotic cultivars into different diversity classes based on biochemical characters and the mean of genetic divergence in some of the biochemical traits of the five clusters are shown in Tables 5 and 6, respect-tively. Figure 1 shows the dendrogram of geno-types in their respective cluster groups based on biochemical parameters. The detail account of the characteristics of each cluster is presented below.

Cluster 1

This consisted of a total of fourteen accessions together with two exotic cultivars. Out of which two of them were from SNNP, Tigray and Amhara and five exotic cultivars. The major characteristics of germplasm grouped under this category had above average oil content (%), palmitic and linolenic acids. For example, exotic cultivar (CI 1652 X CDC 1747/SPS1) had high linolenic acid. Likewise, the

Table 6. Cluster mean of forty-nine linseed landrace accessions and fifteen exotic cultivars based on nine biochemical characters.

Biochemical trait	C1	C2	C3	C4	C5	GM
Oil content (%)	39.26	34.24	36.07	38.50	33.92	35.37
Palmitic (%)	6.61	6.63	6.55	6.32	6.12	6.54
Stearic (%)	5.56	5.48	5.62	5.35	5.48	5.51
Oleic (%)	20.22	19.46	18.98	17.41	17.24	19.13
Linoleic (%)	13.89	13.97	13.93	13.70	13.61	13.89
Linolenic (%)	56.17	53.34	53.73	55.54	52.60	53.70
Crude protein (%)	19.25	20.26	19.25	21.1	23.26	20.77
Crude fat (%)	34.76	32.82	32.95	30.78	30.69	32.39
Iodine Value Wij's/100 g)	177.10	170.82	178.52	172.84	174.32	174.29

Table 7. Pair wise generalized squared distances (D^2) among forty nine linseed landraces and 15 exotic cultivars in five clusters based on biochemical characters.

Cluster	Cluster 1	Cluster 2	Cluster 3	Cluster 4
Cluster 2	5.08ns	-		
Cluster 3	11.22ns	5.08ns	-	
Cluster 4	32.79**	23.18**	13.70ns	-
Cluster 5	26.81**	18.47*	10.51ns	6.60ns

X^2: **Significant at P≤0.01 and *Significant at P≤0.05, ns = non-significant.

highest oil content was recorded in this cluster group by the exotic cultivars, PI–23353. Stearic and oleic acids were the most relevant traits for distinguishing this cluster from the rest.

Cluster 2

Over 39% of the studied materials were grouped under this category. Six from Oromia, three from Amhara and SNNP, two from Somali and four of them were from Tigray and the remaining seven materials were exotic cultivars. The peculiar feature of this cluster group is: Acc. 243807 from Amhara was observed with higher oil content and linoleic acid. Similarly, CI 1525 x R12 D33C/P1/S1 (exotic cultivar) showed higher palmitic, stearic and linoleic acids than the germplasm included in this study. Oil content and oleic acid were the most relevant traits that distinguished this cluster from the rest.

Cluster 3

Nine accessions and three exotic cultivars were included in this cluster, of which four were from Amhara, two from SNNP and Somali, one from Oromia and the remaining three are exotic ones. The common characteristics of these genotypes were that almost all of them possessed intermediate palmitic, oil content an stearic acid as compared to the previous clusters. Oleic and linolenic acids were the most contributing traits that create variability of this cluster from the rest.

Custer 4

Nine accessions were included in this clustering group. The major characteristics in this group were higher oleic acid by Acc. 13558 obtained from Amhara. Similarly, Acc.13545 obtained from Oromia showed higher oleic acid. Acc. 13754 had higher palmitic and Acc. 13756 possessed higher linolenic acid.

Cluster 5

In this group, only 3 accessions where included, of which two were from Tigray and the remaining one is from SNNP. The peculiar feature of this cluster was high crude protein, oil content of Acc. 243810 and higher linoleic and stearic acids of Acc. 244809 from SNNP.

Principal component analysis

Principal component analysis (PCA) showed that the first three (PC1-PC3) of the nine biochemical traits accounted for 73.3% of the total variation, PC1 alone with the largest contribution of nearly 45% (Table 4). The first two principal components (PC1 and PC2) contributed about 62.1% of the total variation. Therefore, characters with relatively larger absolute values of eigenvector weights in PC1 had the largest contribution to the differentiation of the genotypes into clusters as it is normally assumed that characters with larger absolute values closer to unity
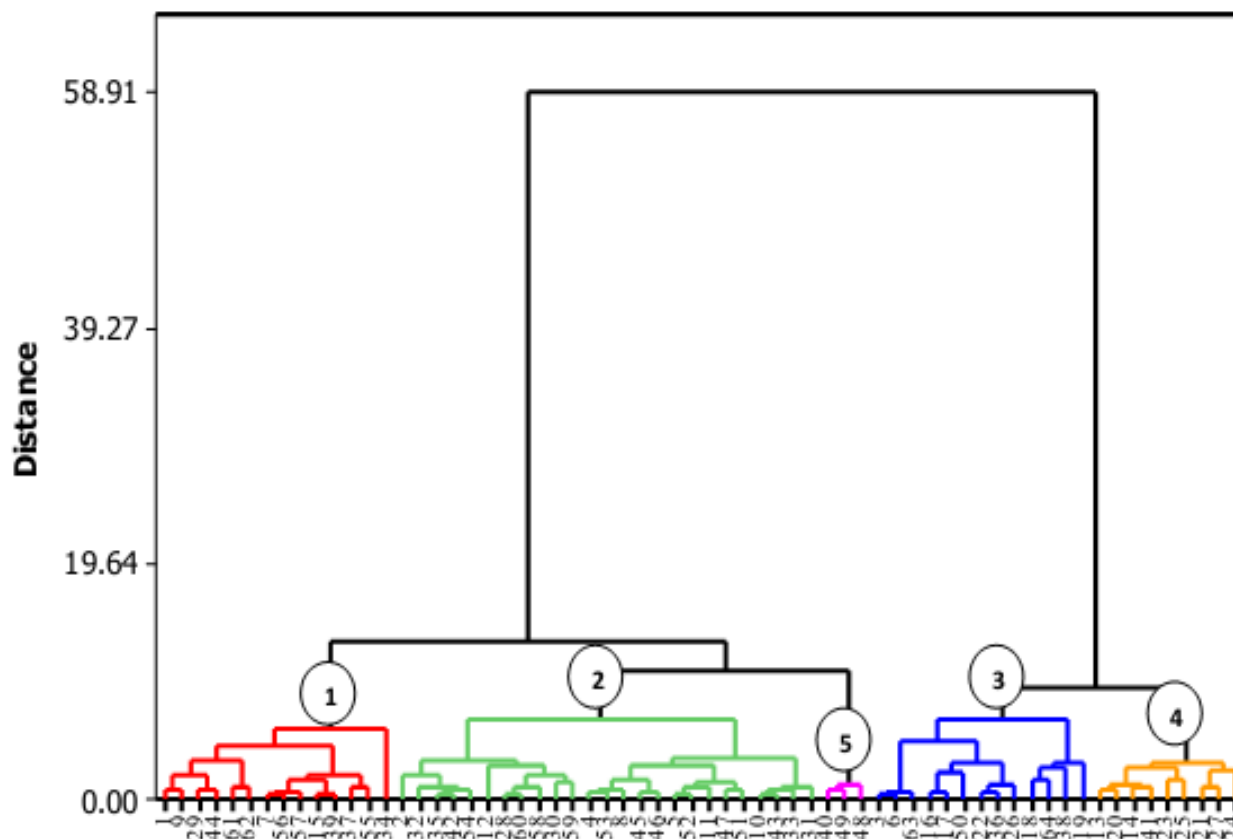
Figure 1. Dendrogram of forty-nine Ethiopian linseed landrace accessions and fifteen exotic cultivars developed by Ward's agglomerative hierarchical classification method based on Euclidian distance using mean of nine biochemical characters.

within the first principal component influence the clustering more than those with lower absolute values closer to zero (Chahal and Gosal, 2002). Accordingly, most of biochemical characters individually contributed small effects (±0.007 to 0.9) to the variation in PC1-PC3 and, therefore, the differentiation of the accessions into different clusters was rather dictated by the cumulative effects of a number of characters.

A character with relatively greater positive weights of eigenvectors in PC1 includes oil content and linolenic acid while crude protein showed positive and small PC1 values. Crude fat, iodine value, stearic acid and linoleic acid showed small and negative value in the first principal component, indicating that these characters contributed the least for the differentiation of the population. Breeding efforts may need to simultaneously focus on genetic manipulation of these characters in order to increase the performance of selected characters so as to contribute to the enhancement of production and productivity of linseed.

Correlations among seed oil traits

Oil content was positive and significantly associated only with linolenic content (r = 0.73) but negatively and significantly correlated with palmitic, oleic and linoleic acids (r

= -0.75, r = -0.74 and r = -0.59), respectively. Palmitic acid had positive and strong correlation with oleic acid (r = 0.67), linoleic acid (r = 0.58), and crude fat (r = 0.42). Similarly, oleic acid was positively and strongly associated with linoleic acid (r = 0.49) and crude fat (r = 0.37) but reversely associated with linolenic acid (r = -0.97). Linolenic acid was also negatively and strongly correlated with linoleic acid (r = -0.55). On the other hand, crude protein and iodine value shows negative and non-significant association with other quality traits (Table 8). These findings are partly in agreement with a report by Kenaschuk (1975) and Adugna (2004). They indicated that, oleic and linolenic acids were strongly and nega-tively correlated to tested varieties. This inverse relation-ship implied the synthesis of linolenic acid in linseed in the following sequence: Oleic acid → linoleic acid → linolenic acid. They further indicated the presence of additive gene action at one location and non-allelic at another location. That is to say, identical genotypes can exhibit different types of gene actions when grown in different environments (Kenaschuk, 1975; Adugna, 2004).

Conclusion

Biochemical analysis has shown the highest oil content (39.8%) for exotic cultivars [PI-523353] against the lowest,

Table 8. Correlation coefficients among biochemical traits in Ethiopian linseed landrace accessions and exotic cultivars.

	OC	Palmitic	Stearic	Oleic	Linoleic	Linolenic	CF	IV
Palmitic	-0.75**							
Stearic	-0.01ns	0.19ns						
Oleic	-0.74**	0.67**	0.38**					
Linoleic	-0.59**	0.58**	-0.19ns	0.49**				
Linolenic	0.73**	-0.77**	-0.42**	-0.97**	-0.55**			
CF	-0.34**	0.42**	0.06ns	0.37**	0.28*	-0.38**		
IV	-0.09ns	0.01ns	-0.31ns	0.02ns	0.15ns	-0.02ns	0.16ns	
CP	0.10ns	-0.15ns	-0.03ns	-0.01ns	-0.14ns	0.03ns	-0.08ns	-0.13ns

OC = Oil content (%); CF = crude fat content (%), CP = crude protein content (%); IV = iodine value (Wij's g/100 g).

30.63% for Acc. 219333 collected from Oromia. PI-523353 was also observed to produce the lowest oleic fatty acid (15.86%). In contrast, the highest linolenic (57.61%) was obtained from the same exotic cultivar. The highest palmitic acid (7.06%) was from Acc. 237494, one of the Ethiopian accessions collected from Tigray Region. Moreover, maximum stearic acid (6.21%) was recorded from Acc.13545, an accession collected from Oromia Region, and the highest oleic acid (21.4%) was from Acc. 13545, another collection from Oromia. The results revealed the presence of enormous genetic variations among the Ethiopian linseed landraces and exotic cultivars for majority of their characters.

Principal component (PC) analysis showed that the first three of the nine biochemical traits accounted for 73.3% of the total variation among the genotypes, PC1 alone with the largest contribution of nearly 45%. The first two principal components contributed about 62.2% of the total variation. The genetic distance of Mahalanobis D^2 statistics grouped the 49 Ethiopian landraces into five clusters. There was highly significant difference between the clusters. The pair-wise square distance between biochemical characters revealed that maximum distance was noted between cluster 1 and 4 (D^2 = 32.79). These findings with wider distances suggest that the linseed germplasm could be a good source of parents for genetic improvement through hybridization and selection schemes. The varying characters of the superior accessions have implications for further work. Thus, the variation for the different characters found in Ethiopian linseed landraces and exotic cultivars in this study could be exploited and used in future linseed improvement programs.

REFERENCES

Adefris TW, Getinet A, Tesfaye G (1992). Linseed breeding in Ethiopia. In: Oilseeds Research and Development in Ethiopia. Proc. of the First National Oilseeds Workshop 3-5 Dec. 1991. IAR, Addis Ababa, Ethiopia, pp. 41-50.
Adugna W, Adefris TW (1995). Agronomic performance of linseed regenerants at two locations in Ethiopia. In: *Sebil.* Proceedings of the 7th Annual Conference of the Crop Science Society of Ethiopia (CSSE), 27-28 April 1995, Addis Ababa, Ethiopia, pp.9-21.
Adugna W (2000). Assessment of tissue culture drived regenerants of linseed (*Linum usitatissimum* L.*)* in Ethiopia. M.S.c. Thesis. Department of plant breeding, faculty of Agriculture, University of the Free State, Bloemfontein, South Africa.
Blakeney M (2002). Protection of Plant Varieties and Farmers' Rights. 24 Eur. Intell. Prop. Rev. pp. 9-19.
Cooke T (1903). Flora of the Presidency of Bombay. Red Lion Court, Fleet Street, London.
Chahal GS, Gosal SS (2002). Principles and procedures of plant breeding: biotechnological and conventional approaches. Narosa Publishing House, New Delhi.
Gaston KJ (1998). Species–range size distributions: products of speciation, extinction and transformation. Philosophical Transactions of the Royal Society, London, B 353:219–230.
Kenaschuk EC (1975). Flax breeding and genetics. In: .J.T. Harapiak (eds.), oilseed and pulse crops in western Canada. Modern press, Saskatoon, Canada. pp. 201- 221.
Khan ML, Sharif M, Sarwar S, Ameen M (2010). Chemical composition of different varieties of linseed. Pak. Vet J. 30(2):79-82.
Kresovich S, Williams JGK, McFerson JR, Routman EJ, Schaal BA (1992). Characterization of genetic identities and relationships of *Brassica oleracea* L. *via* random amplified polymorphic DNA assay.Theor. Appl. Genet. 85:190-196.
Kumar PR (1999). Rapeseed mustard research in India: 21st century strategies. 10th International Rapeseed Congress, Canberra, Australia.
Legesse Burako (2010). Genetic Diversity Study of Linseed Genotypes on Acidic Soil at Bedi Trial Site, Central Highland of Ethiopia, MSc.Thesis. Addis Ababa University, Addis Ababa.
Manly BFJ (1986). Multivariate statistical methods: A primer. Chapman and Hall. London.
Mansby E, Díaz O, von Bothmer R (2000). Preliminary study of genetic diversity in Swedish flax (*Linum usitatissimum*). Genet. Res. Crop Evol. 47:417-424.

MINITAB (1996). Minitab for windows release 11:12.

MoA (1998). Agro-Ecological Zones of Ethiopia, Addis Ababa, Ethiopia.

Robbelen G, Downey RK, Ashri A (1989). Oil Crops of the World, McGrawHill, New York, pp.157-183.

Rubilar M, Gutiérrez C, Verdugo M, Shene C (2010). Scenario flaxseed as a source of functional ingredients J. Soil Sci. Plant Nutri. 10(3):373–377.

SARC (2005). Annual report on seed spices crop research. Horticultural crop Research Division, Sinana Agricultural Research Center. OARI.

SAS Institute (2001). SAS software. SAS Institute INC., Cary. NC. USA.

Seegler CJP (1983). Oil plants in Ethiopia, their economy and agriculture. M. H. Thijssen, Zewde B, Beshir A, de Boef WS (2008). (eds) farmers seed and varieties: supporting informal seed supply in Ethiopia. Wagningen, Wagenningen International.

Singh RK, Chaudhary BD (1985). Biometrical Methods in Quantitative Genetics Analysis. Kalyanin publishers, New Delhi-Ludhiana.

Ward JH (1963). Hierarchical grouping to optimize an objective function. J. Am. Statistical Assoc. 58:236-244.