Proceedings of the Pakistan Academy of Sciences:
B. Life and Environmental Sciences 58(3): 55-63 (2021)

ISSN (Print): 2518-4261; ISSN (Online): 2518-427X
http://doi.org/10.53560/PPASB (58-3)662

Pakistan Academy of Sciences

Research Article

Characterization of Selected Soybean Germplasm through Fatty Acid and Oil Composition using Near-Infrared Spectroscopy

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Abstract: Soybean belongs to legumes and is said to be one of the best sources of oil and fats. The objective of this research was to identify specific and individual accession among the fifty soybean accessions collected from Plants Genetics Research Institute (PGRI) Islamabad for oil contents and fatty acids profile using near-infrared reflectance spectroscopy. The amount of oil content in 50 soybean accessions was recorded in the range of 13.816-23.40 % among which accessions 017421, 17423, 17430, 17435, and 17459 were found with the highest percentage of 23.4 %, 22.80 %, 22.214 %, 22.105 %, and 22.08 % respectively. The values of fatty acid content in these accessions for palmitic acid (16:0), stearic acid (18:0), oleic acid (18:1), linoleic acid (18:2) and linolenic acid (18:3) ranged between 10.077-18.48%, 1.95-5.88 %, 18.204-30.12 %, 22.756-53.879 % and 4.714-9.96 % respectively. Soybean accessions observed higher for oil content are recommended for cultivation; where it is grown for oil production. Based on essential fatty acids contents, accessions 017418 and 017420 are recommended for a future breeding program to improve human health. Due to the variability identified in the present research, it is critical to select single Soybean accession having all the traits for further breeding programs until a specific trait is chosen.

Keywords: Soybean; Glycine Max (L).; Oil; fatty acid; NIRS; ANOVA.

1. INTRODUCTION

Soybean *Glycine max* L. is recognized as a "miracle" bean plant and 4th most important crop of the 20th century, because of its multiple uses, least expensive source of best quality protein, phytochemicals, and dietary fiber [1–4]. The crop also plays a vital role in the economy as well as an important source of protein used by a large population of the world and constituent for various biochemicals including isoflavone which makes it unique among the other legumes [5, 6]. It supplies about 20 % of fats and oil to the world which has a pivotal role in the production of various chemicals like soap, medicines, paints, varnishes, and lubricants [7, 8]. Moreover, its help in the prevention of diabetes, cardiovascular diseases, and weight loss cannot be neglected [9, 10]. Soybean was first introduced to Pakistan in 1969 and is grown during the spring and autumn seasons for the production of oil [11, 12]. The vast areas of Sindh, Khyber Pakhtunkhwa and Punjab provinces of Pakistan were found ideal for the cultivation of soybean [13]. Chemically, a fatty acid is known as an organic acid that carries both acidic and methyl groups to each end [13]. Fatty acids are typically categorized based on their omega (ω) groups like ω, 3, 6, 7, and 9, their specific double bonds location and are therefore very important to human health except ω7 [15]. It is reported that the fatty acid composition of the oil is dependent on the climate of the area where the crop is grown [16]. Cooler the climates higher will be its linoleic acid and n-6 polyunsaturated fatty acid (PUFA) concentrations while, if the climate is hot it will produce monounsaturated fatty acid (MUFA) and oleic acid in higher concentrations

Received: June 2021; Accepted: September 2021
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and will therefore have different properties to affect human health [16]. With the advancement of electronics, near-infrared reflectance spectroscopy (NIRS) is said to be one of the best tools for the estimation and evaluation of food products due to its fast and non-destructive properties [18].

The application of NIRS for the chemical characterizations of food products has been confirmed previously by many researchers i.e. Lopez (2013) and his co-workers have successfully analyzed potato products for their fats and moistures [19]. Another study also used NIRS measurement to find out the concentration of fatty acid content using 262 winter oilseed rap while, keeping the calibration for this experiment in such a way that it will only calculate the amount of linoleic acid, oleic acid, and linolenic acid with different composition during screening [20]. Topas, pactol, screw 4, screw 6, and silvo of rapeseed cultivars were also used for total tocopherol, glucosinolate, phenolic content, amino acids, and fatty acids profile among which significant variation was found in fatty acids contents [21]. The contents of fatty acids as published in 2010 by El-Beltagi and Mohamed in their research article revealed the percentage of linolenic acid from 8.83 %-10.32 %, linoleic acid 10.52-13.74 %, erucic acid 0.15 % - 0.9 %, and oleic acid 56.31 % - 85.24 % respectively [22]. The oils of corn, peanut, and cotton revealed a greater value of linoleic acids and oleic acid. Oleic acid will also effectively enhance soybean oil uses in cosmetics, medical and industrial goods like biodiesel and lubricants [23]. The essential fatty acids (EFAs) are used by our body for the development of well-built cell membranes, the proper growth and working of the brain and nervous system, and for the making of hormone-like chemicals called eicosanoids (thromboxanes, leukotrienes, prostaglandins). These substances control various body functions such as hypertension, the viscosity of blood, inflammation, immune mechanism, and vasoconstriction [23, 24].

As the previous studies revealed that the composition of fatty acids and the variability found in their concentrations play a key role in terms of human health and industries, therefore, the present study was proposed to examine the fatty acid composition of soybean and to select those specific accessions which will be best fit for future breeding program, health care management and in industries.

2. MATERIALS AND METHODS

Fifty (50) accessions of Soybean *Glycine max* (L.) were collected from NARC, Islamabad Pakistan for oil and fatty acids contents analysis. NIRS reflectance spectra, expressed in the form of log (1/R), were collected from 5gm of seed sample using NIRS model 6500 spectrometer. NIRS spectra were measured for each and individual seed samples with the help of a spinning cup that has a diameter of 3.8 cm and depth of 0.9 cm, for the collection of radiation reflected from the overall surface of the seeds. The instrument evaluates diffuse reflectance within the range of 400 nm-2500 nm at 2 nm resolutions. Twenty-five monochromatic scans were averaged from each seed sample and simultaneous data was recorded for oil content, stearic acid, linolenic acid, palmitic acid, linoleic acid, and oleic acid as described by Choung *et al.* [25], Kovalenko *et al.* [26], Roberts *et al.* [27] with a little modification. ANOVA with LSD was performed on the obtained data using SPSS version 21.

3. RESULTS AND DISCUSSION

Significant results were obtained using analysis of variance with LSD values ranging from 0.2170 to 0.6148 respectively (Table 1). The lowest LSD value was found in Olic Acid while the highest LSD value was found in Stearic Acid.

Percent oil contents were determined for 50 soybean accessions and the results obtained were found with different values for oil contents as mentioned in Table 2 and Fig 1. The results depicted that accession 017421 gave the highest oil contents of 23 % which was followed by accessions 17423, 17430, 17435, and 17459 with the values of 22 % while, the lowest oil contents were obtained by the accession 017445 (13.816 %) (Table 2 and Fig 1). It has been reported that soybean is grown for its oil production and those having their values greater than 22 % are said to be better therefore, the accessions having more or equal values within this range should be selected for oil production. Esmaeili *et al.* [28], Chowdhury *et al.* [29] Messina [30] reported similar findings for oil contents as obtained in this study.

Table 1. Analysis of variance (ANOVA) and least significant difference (LSD) in 50 Accessions of *Glycine max* L.

S. No.	Traits	Sum of square	Degree of freedom	Mean Square	F value	P value	LSD value
1	Oil contents	1214.67	49	24.7092	376.66	0.0000	0.415
2	Palmitic Acid	826.907	49	16.8757	884.17	0.0000	0.2239
3	Stearic Acid	110.030	49	2.2455	15.60	0.0000	0.6148
4	Oleic Acid	1427.65	49	29.1356	1624.00	0.0000	0.2170
5	Linoleic Acid	5199.35	49	106.109	2027.05	0.0000	0.3707
6	Linolenic Acid	222.841	49	4.5478	64.62	0.0000	0.4298

Table 2. Contents of fatty acids composition in 50 *Glycine max* L. based on NIRS

Accession	Oil contents %	Palmitic Acid C16:0 (%)	Stearic Acid C18:0 (%)	Oleic Acid C18:1%	Linoleic Acid C18:2 (%)	Linolenic Acid C18:3 (%)
17415	17.100	13.500	4.500	22.050	42.300	7.380
17416	20.880	16.800	4.800	20.700	41.130	6.930
17417	21.720	17.040	5.040	20.790	42.660	5.850
17418	16.200	13.140	4.140	21.365	53.879	7.200
17419	15.300	13.500	4.500	22.487	50.123	8.880
17420	18.720	10.258	3.060	26.357	49.654	9.960
17421	23.400	15.120	3.120	22.860	41.400	7.920
17422	21.600	17.760	4.800	21.600	42.300	7.560
17423	22.800	17.160	5.880	21.780	43.650	5.760
17424	17.640	12.330	2.790	30.120	44.235	9.840
17425	16.020	13.680	4.320	22.456	46.568	8.760
17426	18.000	13.140	3.870	22.547	50.326	9.120
17427	21.240	15.720	3.600	20.340	41.040	7.290
17428	20.400	18.480	4.800	22.500	41.400	6.480
17429	20.400	18.000	5.760	20.700	42.210	6.300
17430	22.080	15.600	4.680	22.230	40.770	7.110
17431	15.441	11.784	3.820	18.773	37.953	6.502
17432	14.791	10.565	3.088	20.155	38.685	6.177
17433	15.360	10.646	2.926	19.749	37.222	4.714
17434	14.060	11.784	3.657	19.992	39.010	5.689
17435	22.214	14.087	5.635	25.573	45.698	8.019
17436	15.035	11.053	3.738	26.006	47.889	9.211
17437	15.116	11.378	3.413	25.681	49.195	6.718
17438	15.441	12.028	1.950	27.307	49.521	8.019
17439	14.385	11.297	2.926	26.657	48.762	7.694
17440	14.629	12.272	2.438	27.090	48.762	9.319
17441	14.791	10.403	3.657	25.356	50.062	6.935
17442	15.197	11.378	2.438	27.090	48.256	8.019
17444	15.929	11.134	2.519	27.198	49.412	8.886
17445	13.816	12.191	4.064	24.578	48.99	8.019
17446	16.904	10.890	2.763	25.898	50.279	8.994
17447	15.441	12.191	4.064	19.911	38.197	6.664
17448	18.855	15.170	4.334	18.692	37.140	6.258
17449	20.588	15.495	5.310	19.667	39.416	5.201

Accession	Oil contents %	Palmitic Acid C16:0 (%)	Stearic Acid C18:0 (%)	Oleic Acid C18:1%	Linoleic Acid C18:2 (%)	Linolenic Acid C18:3 (%)
17450	19.180	14.195	3.251	18.367	37.059	6.583
17451	18.421	16.687	4.334	20.318	37.384	5.851
17452	16.660	11.540	2.519	19.667	37.384	5.201
17453	14.304	10.077	4.389	18.204	37.547	6.420
17454	15.766	11.053	4.064	19.180	37.791	5.201
17455	14.629	10.565	2.438	18.692	37.872	5.283
17456	19.722	15.929	5.526	19.342	22.756	6.502
17457	21.455	14.954	3.034	19.505	37.465	6.827
17458	19.071	15.170	4.334	20.236	38.603	5.526
17459	22.105	16.146	4.009	18.692	37.384	6.827

The 50 Soybean accessions were also examined for palmitic acid content and found variation in their results as indicated in table 2 and Fig 2. The accessions 017428, 017429, 017422, and 017423 were found highest for palmitic acid content with the values of 18.48 %, 18 %, 17.76 %, and 17.16 % respectively while; the least among them was obtained in the accession 017453 with the value of 10.077 %. Accessions with higher palmitic acid are called more desirable as compared to those having lower palmitic acid contents. The same results (18 % - 19.5 %) for palmitic acid were also reported by Erawati *et al.* [31] Asghar and Majeed [32].

According to stearic acid content analyzed in the present study, the highest percentage of stearic acid was found in the accession 017423 (5.88 %) followed by accessions 017429 (5.760 %), 017435 (5.635 %), 017456 (5.526 %) while, its least content was obtained in accessions 017438, 017455 and 017442 ranging from 1.95%-2.44 % respectively (Table 2 and Fig 3). These results are nearly similar to the results described previously [2, 32]. As the stearic acid of soybean has no Side effect on the level of blood cholesterol and hence may not increase threats for cardiovascular disease [33]. It is hence revealed that the accessions with higher stearic acid contents are said to be more desirable as compared to those with lower content of stearic acid for human development.

In the present study fifty soybean accessions, the results for oleic acid contents were found varied between 18.204 % - 30.12 % respectively, among which the highest (30.12 %) was found in accession# 017424 followed by the accessions 017438 (27.31), 017444 (27.2 %), 017440 and 017442 (27.10 %) as described in Table 2 and Fig 4. The lowest content of oleic acid was found in the accession 017453 (18.20 %) followed by 017459, 017455, and 017448 with the values of 18.69 % (Table 2; Fig 4). Our results are in accordance with those obtained by Prabakaran *et al.* [2] and El-Beltagi and Mohamed [21].

Similarly, contents of linoleic acid in these 50 accessions of soybean were found with different concentrations ranging from 22.756 % - 53. 879 % among which the highest was found in the accession 017418 with the values of 53.87 % followed by the accessions 017462 (51.22 %), 017426 (50.23 %), and 017446 (50.27 %) while, the lowest contents of linoleic acid were found in accessions 017456 (22.756 %) followed by 017450 (37.059 %) (Table 2 and Fig 5). The results obtained for linoleic acid during this study are in agreement with the results reported previously [21, 30, 34].

Linoleic acid has the potential of precursor activities with many positive benefits for health i.e. to improved cognitive function and to reduce cardio vascular diseases [35–37]. Linolenic acid is vital meant for appropriate composition, function and structure of several body systems for example eyes and nervous system [24]. Table 2 and Fig 6 indicates that highest value (9.960 %) of linolenic acid was found in accession # 017420 followed by accessions 017424 (9.840 %), 017440 (9.319 %) and 017436 (9.319 %), as the minimum content for linolenic acid was found in accessions 017433 (4.714 %), followed by 017449, 017452 and 017454 with the same value of 5.201 % respectively. These highest values presented in our study are much

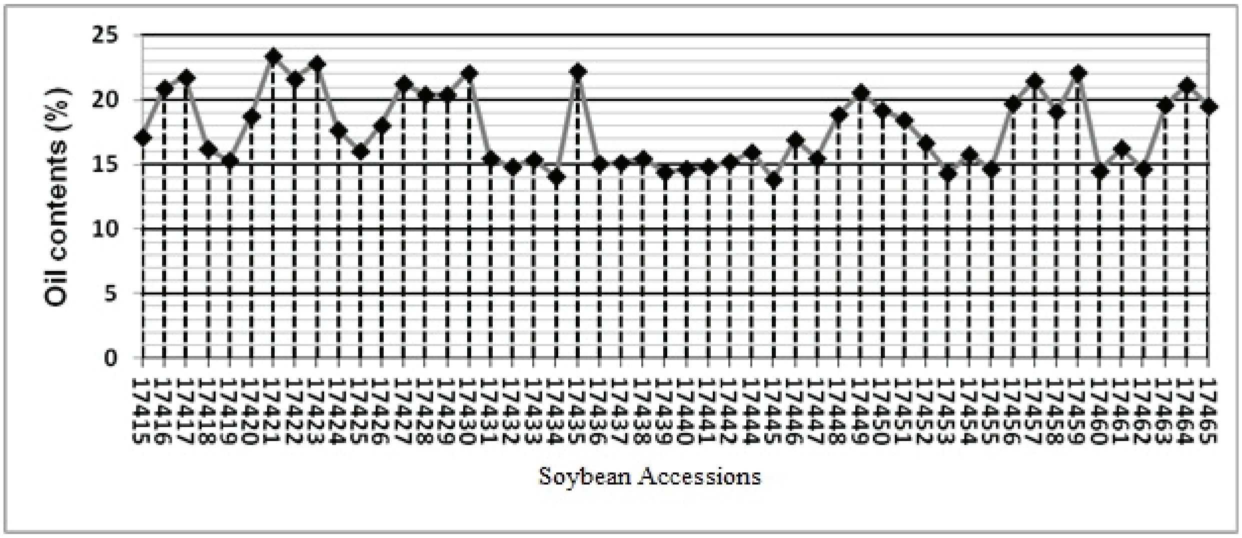

Fig. 1. The concentration of oil content in 50 accessions of soybean

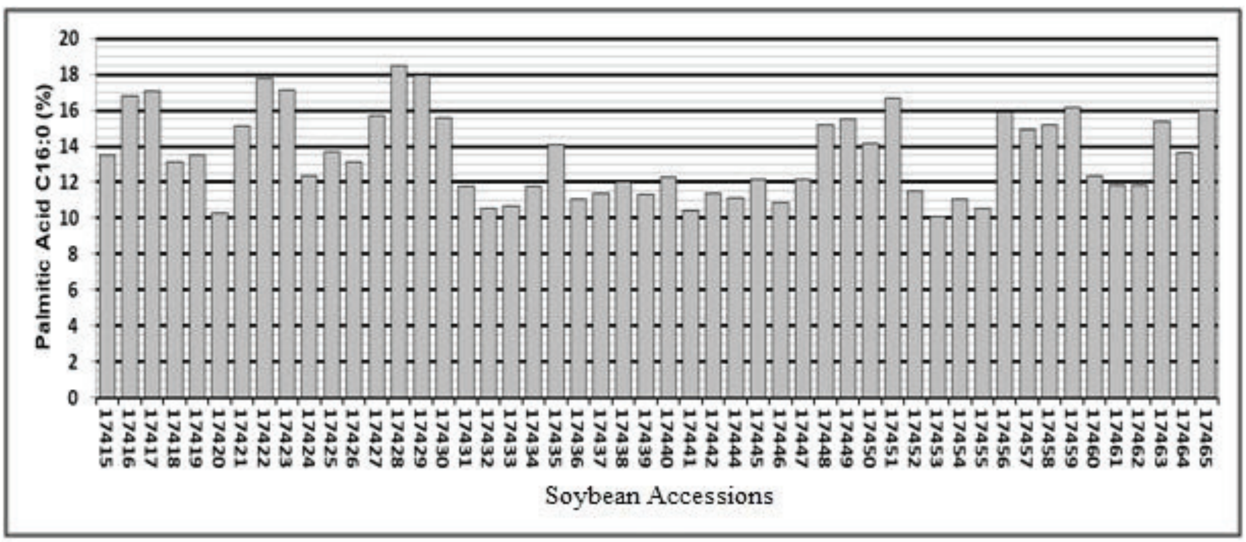

Fig. 2. The concentration of Palmatic acid in 50 Soybean accessions.

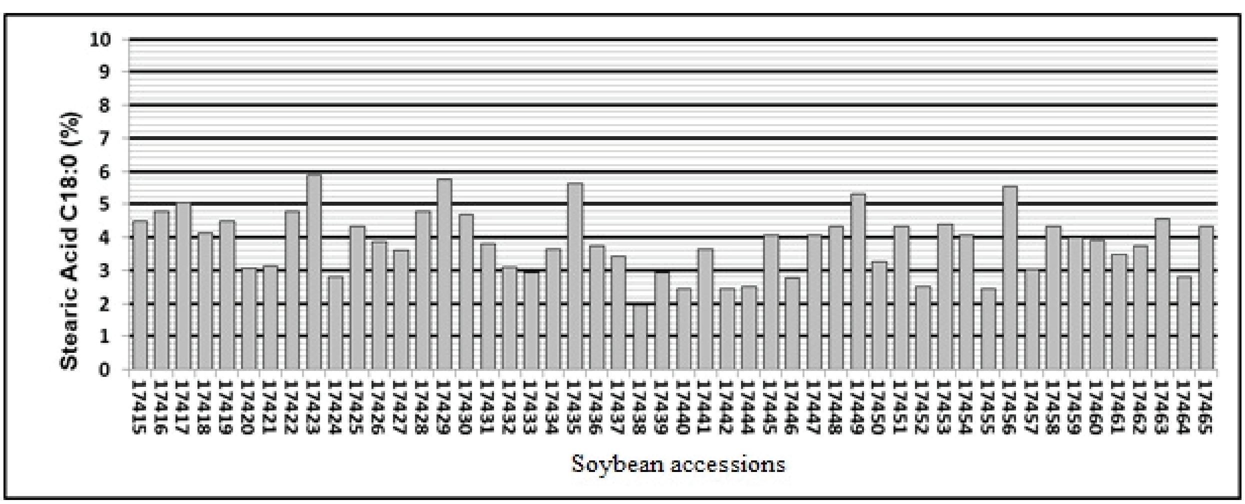

Fig. 3. The concentrations of Stearic acid in 50 Soybean accessions.

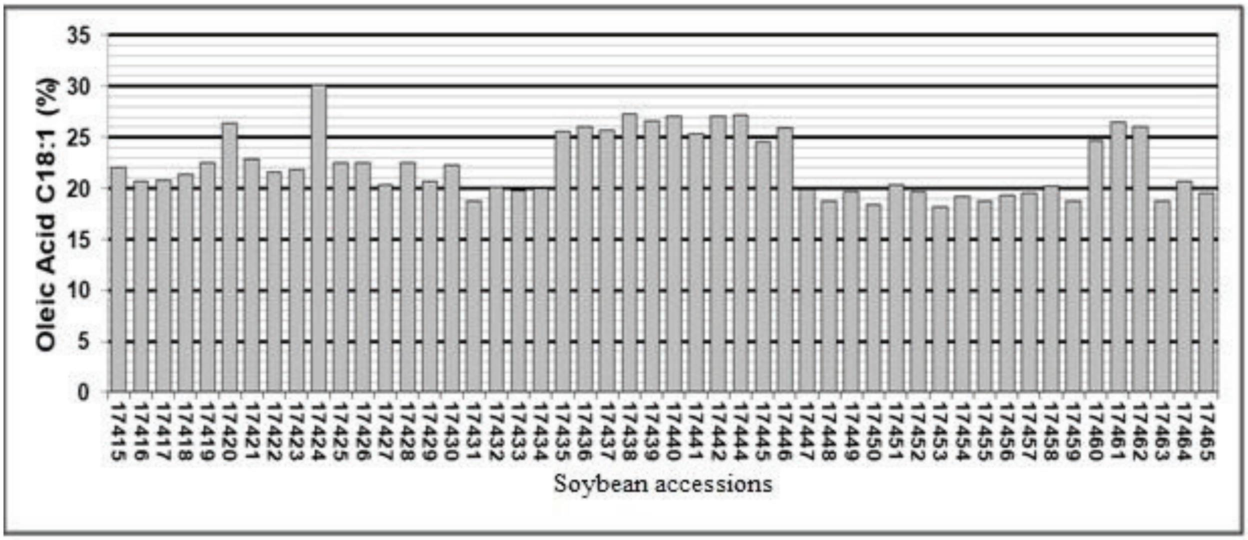

Fig. 4. The concentrations of Oleic acid in 50 Soybean accessions.

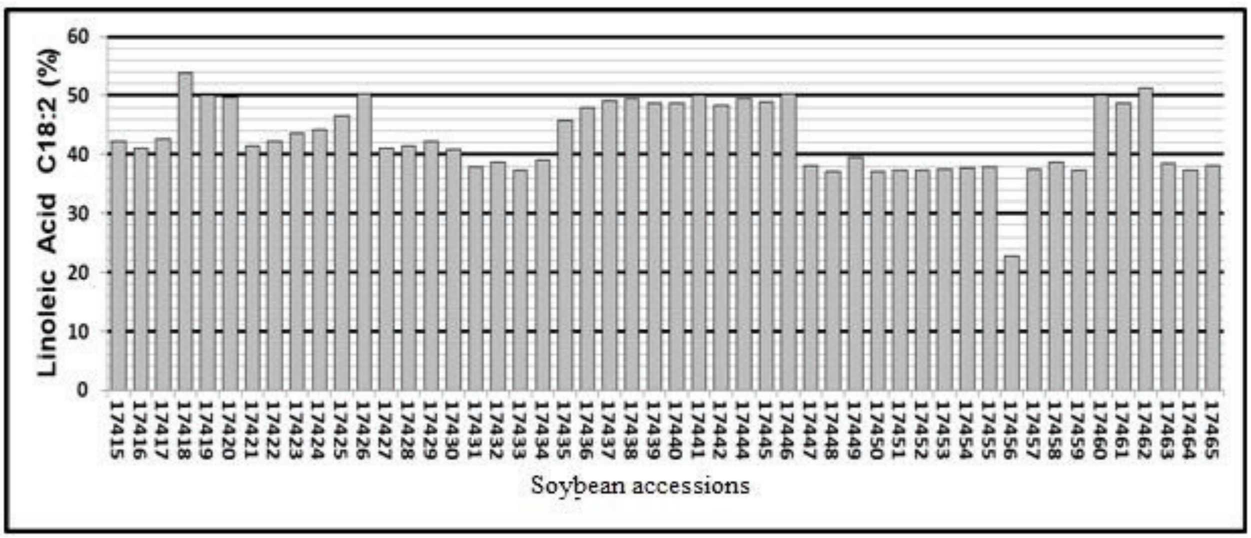

Fig. 5. The concentrations of Linoleic acid in 50 Soybean accessions.

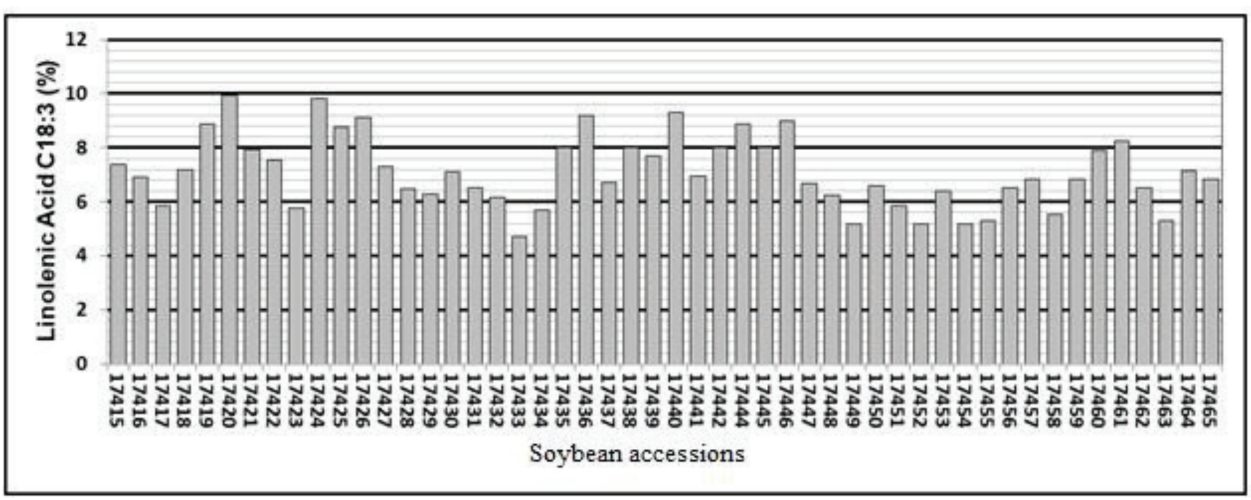

Fig. 6. The concentration of Linolenic acid in 50 Soybean accessions.

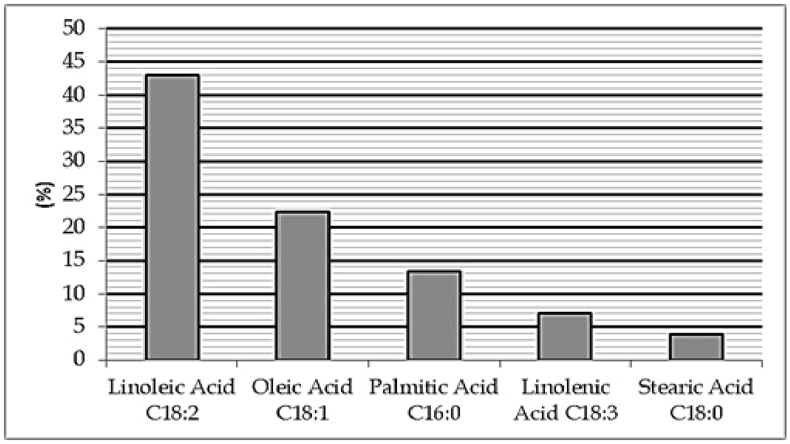

Fig. 7. Average concentrations of the five fatty acids contents in 50 Soybean accessions

related to those observed by [21, 30].

When the average values of the present results were calculated for the five major fatty acid components i.e. 13.4 % palmitic acid (16:0), 3.9 % stearic acid (18:0), 22.3 % oleic acid (18:1), 43 % linoleic acid (18:2) and 7.1 % linolenic acid (18:3) (Fig 7), it was found that these average values are nearly identical to those reported by the other researchers [38, 39].

4. CONCLUSION AND RECOMMENDATIONS

Accessions of soybean having a high amount of essential fatty acid and oil are considered more advantageous in contrast to those with a lesser amount of fatty acid and oil. In the present study, none of the accessions was found consistent for oil and fatty acid composition. The composition of oil in accessions exceeding 22 % is regarded as best because it is mainly cultivated for the production of oil therefore we recommend accessions 017421, 17423, 17435, 17430, and 17459 for the improvement of oil production. Palmitic acid contents are an important trait of soybean as their low contents are desirable for human consumption while higher contents are required for lubrication and other mechanical purposes. In the current research work, the highest palmitic acid content was observed within two accessions 017428 and 017429 and hence can be recommended for lubricant production while, the accessions 17453, 17420, 17441, and 17432 for human consumption. Stearic acid found in the fatty acids of soybeans is important for the prevention of cardiovascular disorders in humans as its high level has no negative effects on blood and LDL cholesterol levels. In the present study accessions, 017417, 017423, 01729, 017435, 017449, and 017456 are found with the highest stearic acid content and are therefore suitable for breeding to improve stearic acid production. Oleic acid is called an important fatty acid that helps to improve the shelf life of oil hence, accessions 017424, 017438, 017444, 017440, and 017442 and may be selected to improve the shelf life of soybean oil. Linoleic acid concentration is very important in both lower and high concentrations because its lower concentration is helpful for flavor stability while its high concentration is important for the human diet therefore its recommendations depend on its need. Similarly, the accessions 017420, 017424, 017440, and 017436 were found with high concentrations for linolenic acid and are therefore recommended for the high yield of linolenic acid to overcome brain and eye diseases.

5. ACKNOWLEDGEMENTS

The authors are highly thankful to Nuclear Institute for Food and Agriculture (NIFA), Peshawar for providing all facilities for the experiment and laboratory analysis. The authors are also thankful to Plant Genetic Resources Program, National Agriculture Research Center Islamabad (NARC) for providing 50 soybean accessions.

6. CONFLICT OF INTEREST

The authors declared no conflict of interest.

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