

Research Article

Biochemical Characterization of Seed Oil of Tunisian Sunflower (*Helianthus annuus* L.) Accessions with Special Reference to Its Fatty Acid Composition and Oil Content

Taoufik Hosni ¹, Zouhaier Abbes,¹ Leila Abaza,² Sana Medimagh,¹ Hamadi Ben Salah,¹ and Mohamed Kharrat¹

¹Field Crops Laboratory, National Institute for Agricultural Research of Tunisia (INRAT), Carthage University, Hedi Karray Street, 1004, El Menzah, Ariana, Tunisia

²Laboratory of Olive Biotechnology, Centre of Biotechnology of Borj-Cedria (CBBC), B.P. 901, 2050 Hammam Lif, Tunisia

Correspondence should be addressed to Taoufik Hosni; taoufik.hosni@fsb.ucar.tn

Received 21 January 2022; Revised 2 March 2022; Accepted 8 March 2022; Published 23 March 2022

Academic Editor: Ammar AL-Farga

Copyright © 2022 Taoufik Hosni et al. This is an open access article distributed under the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Sunflower (*Helianthus annuus*) is a major oilseed crop, cultivated throughout the world, and the nutritional quality of its edible oil ranks among the best vegetable oils in agricultural product. In Tunisia, there is a lack of study on biochemical characterization of sunflower germplasm oil. The present study was conducted to analyze oil content and fatty acid composition of 22 local and introduced sunflower accessions. Results revealed significant variation among studied accessions for all measured biochemical traits. The average oil content of the *H. annuus* accessions was 53.2%, ranging from 35.33% to 59.67%. The results of this study also indicated that unsaturated acids, particularly oleic acid and linoleic acid, were the most abundant fatty acids in oils. Fatty acid compositions of sunflower oils showed diversity depending on the accession. The first two components of the principal component analysis (PCA) contributed 45.7% of the total variability. Cluster analysis based on PCA separated the accessions into four clear groups, which were not grouped according to their geographical origin. Moreover, the classification of the evaluated sunflower accessions using clustering by Euclidean distance revealed four main groups. Linoleic acid had significant and negative correlations with some saturated acids (palmitic, stearic, and arachidic acids). These data can be useful for selecting sunflower accessions and the development of varieties with improved oil quality.

1. Introduction

Sunflower cultivated (*Helianthus annuus* L.) is an annual allogamous plant belonging to the *Asteraceae* family [1]. *H. annuus* is one of the main oilseed crops ranked third in production after soybean and rapeseed throughout the world [2]. In 2021, the sunflower production was 56.97 million tons in approximately 28.27 million ha in the world [2].

A wide range of sunflower varieties are produced worldwide. Three basic types of sunflowers are known, namely, oilseed, confectionery, and ornamental type. Nowadays, sunflower is primarily grown for its edible oil, due to its high content of unsaturated fatty acids such as oleic

and linoleic acids, together with the occurrence of the relatively high content of bioactive compounds such as tocopherols and phytosterols [3].

Overall, in cultivated oil-type sunflower varieties, the oil content ranged from 36% to 50% [4]. It was observed that the oil yield was affected by genotypes and environmental conditions [5]. Standard type sunflower contains about 15% saturated and 85% unsaturated fatty acids [6, 7]. Previous research showed that genetic and environmental factors affected the fatty acids composition of sunflower seeds [8, 9]. Additionally, the plant species and the processing analysis used during sunflower production influenced the fatty acids content of oil [10]. In fact, many factors affect the fatty acids composition and oil content of sunflower seeds and increase

diversity among sunflower oils. This variability can be useful for the selection of sunflower in breeding programs.

Breeding efforts in sunflower are widely used for improving traits, such as oil content and fatty acid composition, and mainly for obtaining commercial oil [11, 12]. Mutagenesis technology represents a powerful tool for developing variation in the fatty acid composition of sunflower oil [13, 14]. Using mutagenesis of dry seeds through X-rays irradiation, two different sunflower mutants were developed, having high oleic acid contents [15]. Mutagenesis was effective for developing mutants with increased levels of oleic acid (more than 90%) by treatment with dimethyl sulfate [16]. Molecular genetics technique is one of the best of current knowledge to improve the seed oil contents and modify the fatty acids composition [17, 18]. In this context, QTLs (quantitative trait locus) were identified on the various linkage groups, explaining the high genetic variability for the seed oil content in sunflower [19]. The marker-assisted selection was used to detect high-oleic genotypes of sunflower [20].

Presently, the sunflower cultivated is one of the most leading oilseed crops in Tunisia. It ranked first in 2019 before soybean and rapeseed, with an average annual production of about 4877 tons [21]. Despite many benefits of fatty acids composition as an important indicator for vegetable oil quality, in Tunisia, few studies were carried out on breeding for these components in oilseeds, especially on sunflower. Therefore, this study aimed to characterize and evaluate the variability of fatty acids and oil contents in sunflower from different geographical locations of Tunisia.

2. Materials and Methods

2.1. Plant Materials. As indicated in Table 1, the experimental material for the present study included twenty-two accessions of sunflower (*Helianthus annuus* L.). Nineteen of them were local accessions collected from different localities from Northern Tunisia, and three introduced lines were provided by the National Plant Germplasm System (NPGS, USA) and the Plant Gene Resources of Canada (PGRC, Canada).

2.2. Experimental Design. Field experiment was conducted at Beja region in northwest Tunisia, during the growing season from March to July 2016 to evaluate the seed quality (content oil and fatty acids composition) of twenty-two sunflower accessions. The experiment was arranged as a randomized complete block with three replications. Plants were sown in clay soil in rows planted 0.8 m apart, with the seeds placed 0.6 m apart along the row. There were no fertilizers applications during the vegetation cycle or supplementary irrigation. For oil quality evaluation, 5 sunflower heads/accessions were harvested at physiological maturity, and seeds were then dried.

2.3. Seed Oil Determination Using Soxhlet Extraction. Oil contents were determined in three samples per accession. The sunflower oils were obtained by chemical extraction. For each accession, 10 g of decorticated seeds were crushed and used to extract the oil with hexane solvent for 4 h by Soxhlet type extractor. The oil extract was evaporated under low pressure in a rotary evaporator at 70°C until the solvent was completely removed. The weight of oil was then recorded, and the oil content was determined using the following formula:

$$\text{oil content (\%)} = \frac{(\text{weight of flask with oil} - \text{weight of empty flask})}{\text{weight of ground seeds}} \times 100. \quad (1)$$

2.4. Fatty Acids Content Determination. Fatty acids were first converted to their methyl esters. 0.1 g of oil samples was mixed with 3 mL of hexane and 0.5 mL of 2 N methanolic potassium hydroxide. The mixtures were well shaken and allowed to settle. The top layer of hexane was then separated, and 1 mL was injected into a gas chromatography (GC) (HP 4890 D, Hewlett-Packard Company, Wilmington, DE, USA). The column used was a capillary column (Supelco-wax: 30 × 0.53 m; 0.25 μm, Agilent Technologies, USA) with nitrogen as the carrier gas at 1 mL min⁻¹. The detector was a flame ionization detector (FID). The temperature of the injector, the detector, and the oven was maintained at a temperature of 230, 250, and 210°C, respectively. Fatty acids composition was determined by identifying and calculating relative peak areas percent by the ChemStation software. Fatty acids analyses were repeated three times for each accession.

2.5. Statistical Analysis. All statistical analyses were performed using SPSS software version 21 (IBM, Armonk, NY, USA). Results were expressed as means ± standard errors. The averages were compared using Duncan's test at a 5% significance level. Genetic variability among sunflower accessions based on biochemical data was examined using the clustering analysis with Euclidean distance matrix and the principal components analysis (PCA). Correlations analysis between biochemical traits was performed by applying Pearson's correlation coefficient.

3. Results

3.1. Oil Content. The seed oil content of the 22 accessions of sunflower is presented in Table 2. There were statistically significant ($P < 0.05$) differences among accessions for oil content. The mean oil content of the sunflower accessions

TABLE 1: List of the sunflower (*Helianthus annuus* L.) accessions studied and their origins.

Accessions no.	Codes of NPGS and PGRC	Origins	Latitudes (N)	Longitudes (E)	Altitudes (m)
TL1	—	Tunisia	36° 53' 46.405''	9° 26' 43.522''	258
TL2	—	Tunisia	36° 53' 03.856''	9° 26' 00.820''	263
TL5	—	Tunisia	36° 51' 37.225''	9° 18' 15.015''	335
TL7	—	Tunisia	36° 44' 50.068''	9° 04' 30.182''	341
TL8	—	Tunisia	36° 45' 59.276''	9° 03' 44.647''	351
TL9	—	Tunisia	36° 44' 50.068''	9° 04' 30.182''	341
TL11	—	Tunisia	36° 50' 33.992''	9° 12' 36.437''	370
TL13	PI 257641	Former Soviet Union	—	—	—
TL17	PI 607925	United States	—	—	—
TL18	CN 37370	Canada	—	—	—
TL19	—	Tunisia	36° 53' 46.405''	9° 26' 43.522''	258
TL20	—	Tunisia	36° 45' 47.657''	9° 11' 43.281''	225
TL22	—	Tunisia	36° 51' 46.655''	9° 22' 58.578''	370
TL23	—	Tunisia	36° 47' 06.360''	9° 19' 24.550''	307
TL25	—	Tunisia	36° 46' 39.454''	9° 08' 09.588''	266
TL26	—	Tunisia	36° 44' 50.068''	9° 04' 30.182''	341
TL27	—	Tunisia	36° 43' 15.577''	9° 05' 12.526''	315
TL29	—	Tunisia	36° 48' 37.964''	9° 11' 07.264''	270
TL30	—	Tunisia	36° 48' 37.964''	9° 11' 07.264''	270
TL31	—	Tunisia	36° 50' 33.997''	9° 12' 36.499''	374
TL32	—	Tunisia	36° 50' 33.992''	9° 12' 36.437''	370
TL33	—	Tunisia	36° 47' 27.886''	9° 16' 47.552''	360

TABLE 2: Mean values of oil content and relative fatty acid composition of 22 sunflower accessions.

Accessions	Palmitic acid (C16:0) (%)	Stearic acid (C18:0) (%)	Oleic acid (C18:1) (%)	Linoleic acid (C18:2) (%)	Arachidic acid (C20:0) (%)	Gadoleic acid (C20:1) (%)	Behenic acid (C22:0) (%)	Oil content (%)
TL1	7.41ab ± 0.43	4.96b ± 0.05	29.9bd ± 1.87	50.62g ± 6.08	0.92ac ± 0.40	2.01af ± 1.16	1.31af ± 0.74	52be ± 1.53
TL2	13.5c ± 4.05	4.07a ± 0.13	25.95ab ± 0.42	36.92bd ± 1.21	2.14ag ± 0.35	0.58a ± 0.33	2.96fg ± 0.89	50.67bc ± 1.20
TL5	7.16ab ± 0.11	6.91ef ± 0.41	30.68bd ± 0.45	40.48ce ± 0.29	5.57ij ± 0.30	4.11f ± 0.11	2.3df ± 0.30	55.67ce ± 1.45
TL7	7.26ab ± 0.26	5.01b ± 0.01	30.89be ± 0.49	41.59df ± 1.12	5.98j ± 0.02	1.15ac ± 1.15	0.79ae ± 0.79	51bd ± 2.08
TL8	12.99c ± 0.01	6.97ef ± 0.03	29.91bd ± 0.09	30.23ab ± 0.23	4.11gj ± 0.11	2.0af ± 0.00	4.11g ± 0.11	58.67de ± 0.88
TL9	6.67a ± 0.76	6.07cd ± 0.50	32.28bf ± 2.56	41.9df ± 4.20	4.52hj ± 2.58	3.15cf ± 1.74	2.17df ± 1.20	55.33ce ± 2.33
TL11	6.57a ± 0.06	6.06cd ± 0.06	36.17df ± 0.17	34.1ac ± 0.10	4.04f ± 0.04	2.93bf ± 0.07	1.97cf ± 0.03	49.67bc ± 1.33
TL13	10.02b ± 0.54	5.87cd ± 0.58	31.21be ± 2.40	46.25eg ± 6.54	0.12a ± 0.07	1.42ad ± 0.74	2.62eg ± 1.39	35.33a ± 3.76
TL17	8.23ab ± 0.14	4.09a ± 0.09	34.01cf ± 0.01	30.2ab ± 0.20	12.27k ± 0.27	1.55ad ± 0.05	2.97fg ± 0.03	57.33ce ± 1.67
TL18	8.97ab ± 0.03	5.03b ± 0.03	20a ± 10	50.05g ± 0.05	1.02ad ± 0.02	1.12ac ± 0.02	1.29af ± 0.01	50bc ± 0.00
TL19	7.97ab ± 0.26	6.07cd ± 0.32	33.67bf ± 2.05	44.07dg ± 1.70	1.96ae ± 0.87	3.54df ± 1.74	1.62af ± 0.47	56.67ce ± 2.03
TL20	7.98ab ± 0.02	4.05a ± 0.05	26.97ac ± 0.03	46.23eg ± 0.23	2.48bg ± 0.02	8.09g ± 0.09	1.93bf ± 0.07	50.67bc ± 3.84
TL22	7.97ab ± 0.03	6.08cd ± 0.08	26.93ac ± 0.07	29.9ab ± 0.10	2.08af ± 0.08	4f ± 0.00	0a ± 0.00	46.67b ± 2.19
TL23	7.93ab ± 0.67	4.07a ± 0.07	37.2df ± 0.20	49.95g ± 0.05	0.2a ± 0.00	0.12a ± 0.02	0.1ab ± 0.00	55.67bc ± 3.33
TL25	7.51ab ± 0.01	6.49de ± 0.01	30bd ± 0.00	40.48ce ± 0.02	2.99dh ± 0.01	3.98ef ± 0.02	8.87 h ± 0.13	55.33ce ± 1.86
TL26	9.32ab ± 0.09	4.07a ± 0.07	34.22cf ± 0.11	44.01dg ± 0.01	2.75ch ± 0.14	3bf ± 0.00	2.34df ± 0.09	56ce ± 1.73
TL27	6.23a ± 0.15	4a ± 0.00	39.95f ± 0.05	48.41fg ± 0.24	0.22a ± 0.02	0.30a ± 0.00	0.2ac ± 0.01	59.67e ± 1.33
TL29	7.51ab ± 0.01	5.81c ± 0.01	37.16df ± 0.16	47.48eg ± 0.02	0.47ab ± 0.00	0.71ab ± 0.01	0.5ad ± 0.00	56ce ± 2.08
TL30	8.02ab ± 0.02	4.93b ± 0.07	26.72ac ± 0.02	36.92bd ± 0.08	3.41eh ± 0.01	3.07cf ± 0.07	0a ± 0.00	55.33ce ± 2.33
TL31	8.96ab ± 0.04	7.97g ± 0.03	38.80ef ± 0.00	38.47cd ± 0.03	2.03af ± 0.03	1.91af ± 0.01	1.91bf ± 0.01	55.33ce ± 4.18
TL32	21.56d ± 0.06	7.50fg ± 0.00	35.46df ± 0.04	28.87a ± 0.13	1.82ae ± 0.02	2.29af ± 0.01	0.62ad ± 0.02	54.33ce ± 0.88
TL33	7.02ab ± 0.14	4.63ab ± 0.21	34.98df ± 0.90	47.9fg ± 2.06	1.14ad ± 0.57	1.68ae ± 0.97	1.77af ± 0.99	53be ± 2.52
Mean	8.94	5.49	31.96	41.14	2.83	2.4	1.93	53.2

Means with different letters in the same column differ significantly at 5% as a probability level.

was 53.2%, varying from a low of 35.33 ± 3.76% in TL13 to a high of 59.67 ± 1.33% in TL27.

3.2. Fatty Acid Composition. As shown in Table 2, there were statistically significant differences ($P < 0.05$) among the examined sunflower accessions in terms of all fatty acid

content. Analysis of fatty acid composition indicated that linoleic (C18:2) and oleic (C18:1) acids represented the major component of total fatty acids (with an average of 41.14% and 31.96%, respectively). Linoleic acid content ranged from 28.87 ± 0.13% in TL32 accession to 50.62 ± 6.08% in TL1 accession. Oleic acid content varied from 20 ±

10% in TL18 accession to $39.95 \pm 0.05\%$ in TL27 accession. Palmitic acid (C16:0) content ranged from $6.23 \pm 0.15\%$ in TL27 to $21.56 \pm 0.06\%$ in TL32 with an average of 8.94%. The highest stearic acid (C18:0) content was observed in TL31 accession as $7.97 \pm 0.03\%$, while the lowest stearic acid content was obtained from TL27 accession as $4 \pm 0.00\%$. Several minor fatty acids were present in the oil of these sunflower accessions including arachidic, gadoleic, and behenic acids. The average arachidic acid (C20:0) concentration was 2.83%, varying from a high of $12.27 \pm 0.27\%$ in TL17 to a low of $0.92 \pm 0.40\%$ in TL1. Gadoleic acid (C20:1) averaged 2.4% and was between $0.12 \pm 0.02\%$ in TL23 accession and $8.09 \pm 0.09\%$ in TL20 accession. The mean behenic acid (C22:0) concentration of these accessions was 1.93%, and TL22 and TL30 accessions had the lowest with $0 \pm 0.00\%$, while the highest behenic acid percentage was observed in TL25 accession with $8.87 \pm 0.13\%$.

3.3. Principal Component Analysis (PCA). The importance of each biochemical trait in explaining the observed variability was assessed through principal component analysis (PCA). The results of PCA showed that the first three principal components accounted for 63.4% of the total variability (Table 3). The first principal component (PC1) absorbed 26.3% of the total variation; it was related mostly to stearic acid, linoleic acid, and arachidic acid. The second principal component (PC2) explained 19.4% of the total variability, and it was defined by the strong correlation to oleic acid, gadoleic acid, and oil content. The third principal component (PC3) contributed around 17.7% of the variability present among the accessions for the traits used in this study, and it was correlated with palmitic acid, arachidic acid, and oil content. The major correlated variability of sunflower accessions was shown by axes 1 and 2 which explained 45.7% of the total variability and revealed four groups (Figure 1). Cluster I contained TL1, TL23, TL27, TL29, and TL33; cluster II was composed of TL8, TL17, TL31, and TL32; cluster III included TL13, TL18, and TL20; and cluster IV consisted of TL2, TL5, TL7, TL9, TL11, TL19, TL22, TL25, TL26, and TL30.

3.4. Hierarchical Classification Using Matrix of Euclidean Distances. The dendrogram of the matrix of Euclidean distances (Figure 2), which lies between the studied twenty-two sunflower accessions based on biochemical traits, revealed four main groups, indicating a considerable variability genetic. Group I included the twelve sunflower accessions: TL5, TL7, TL9, TL11, TL19, TL23, TL25, TL26, TL27, TL29, TL31, and TL33. The sunflower accessions, TL1, TL18, and TL20, were clustered in Group II. Group III combined the two sunflower accessions: TL8 and TL17. Group IV contained the three sunflower accessions: TL2, TL22, and TL30 whereas, the sunflower accessions TL13 and TL32 were not clustered into the four mentioned groups.

3.5. Interrelations among the Biochemical Traits. The analysis of simple correlation coefficients (Table 4) showed

TABLE 3: Eigenvectors, eigenvalues, total and cumulative variability of the first three principal components.

Principal components (axes)	PC1	PC2	PC3
Eigenvalues	2.104	1.551	1.415
Variability (%)	26.30	19.40	17.70
Cumulative (%)	26.30	45.70	63.40
Traits	Eigenvectors		
Palmitic acid (C16:0) (%)	0.495	0.198	-0.664
Stearic acid (C18:0) (%)	0.607	0.168	-0.397
Oleic acid (C18:1) (%)	-0.120	0.816	0.098
Linoleic acid (C18:2) (%)	-0.881	-0.141	0.076
Arachidic acid (C20:0) (%)	0.578	0.036	0.625
Gadoleic acid (C20:1) (%)	0.345	-0.589	0.241
Behenic acid (C22:0) (%)	0.475	-0.280	0.267
Oil content (%)	0.146	0.609	0.529

that significant ($P < 0.05$) and negative correlations were found between linoleic acid (C18:2), palmitic acid (C16:0), and stearic acid (C18:0) (-0.488; -0.439, respectively). Arachidic acid (C20:0) showed significant ($P < 0.01$) and negative correlation with linoleic acid (C18:2) (-0.552). The results showed also that there were no significant correlations between other biochemical traits.

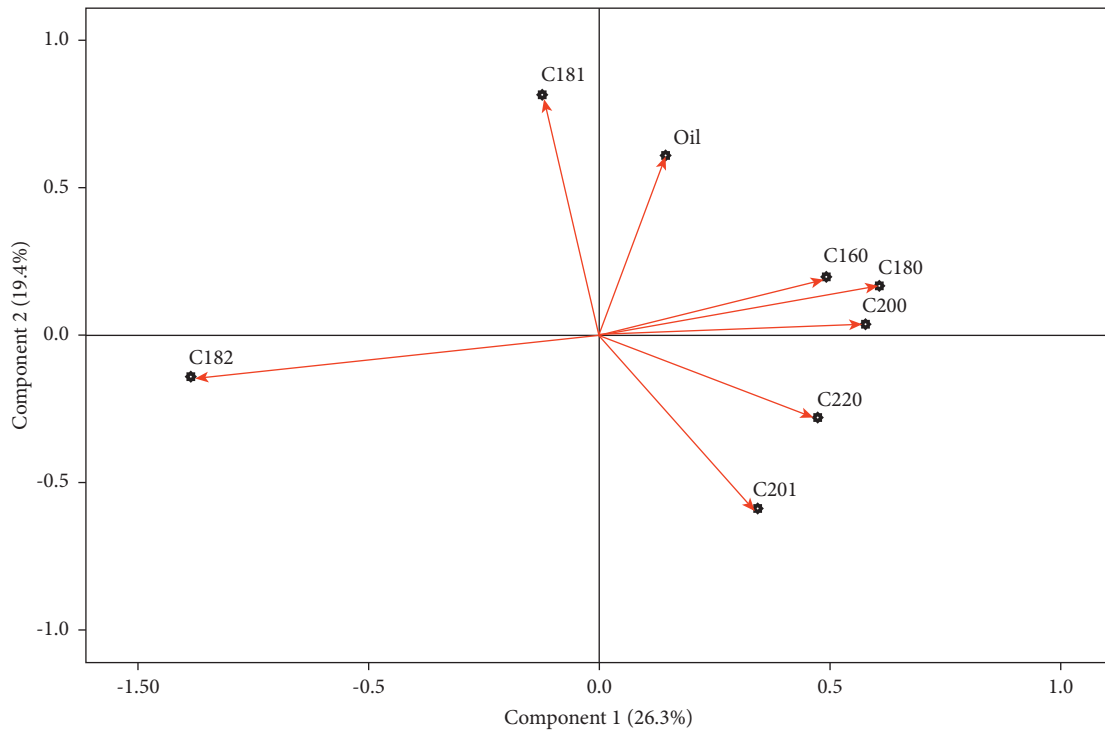
4. Discussion

Based on biochemical traits, oil content and fatty acid composition were analyzed, using some sunflower accessions cultivated in Tunisia and three accessions introduced from abroad.

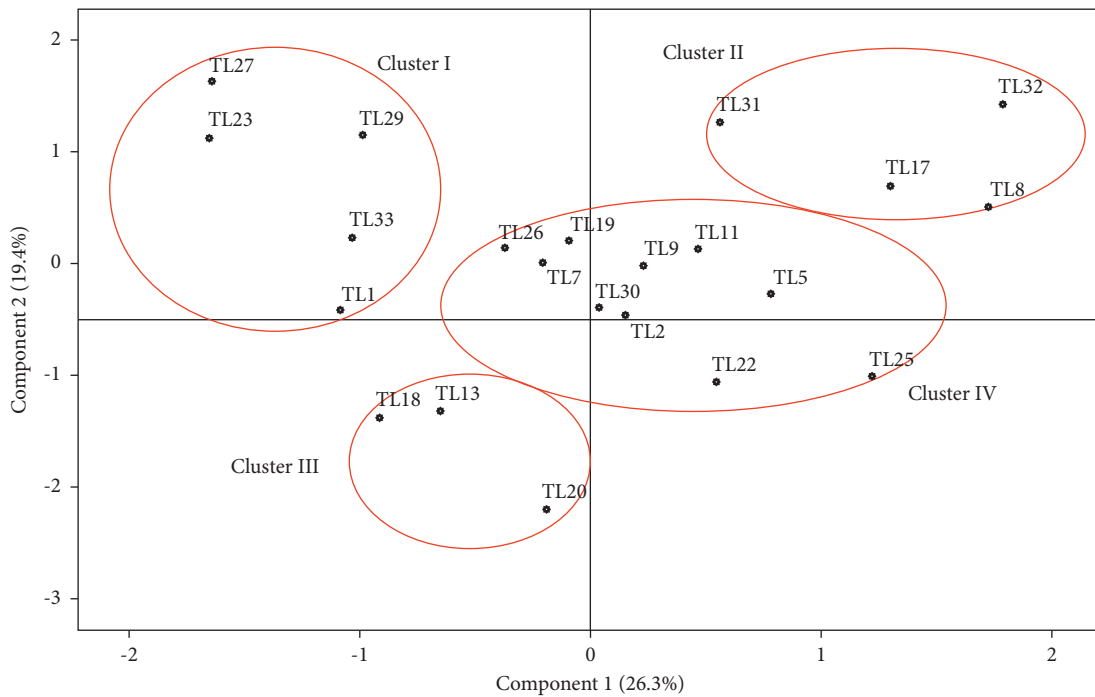
The oil content of the seed was significantly affected by the accession and ranged from 35.33% in TL13 to 59.67% in TL27. These results agreed with the findings of [22]. It was found that the oil content of *H. annuus* was dependent on the genotype. In contrast, several authors studied the influence of environmental factors on the oil content. In this context, the oil yield of the Egyptian hybrids H (A9×RF6) and H (A9×RH8) was affected by both irrigation and salinity treatments [23]. The oil concentration in seeds of Tunisian sunflower was influenced by *Orobanche cumana* causing a decrease in seed oil yield in susceptible sunflower [24].

As consequence of the evaluation of the fatty acid content in sunflower, it was observed that seed oil was characterized by the presence of a high proportion of unsaturated fatty acids than saturated fatty acids. The prevalence of the unsaturated fraction in this oil was mainly due to the abundance of linoleic acid (C18:2) and oleic acid (C18:1), which exceeded 75% of the total fatty acids for the majority of sunflower accessions. The predominance of unsaturated fatty acids attributes to a nutritional advantage of sunflower oil and confers it a more therapeutic advantage. According to [25], a good quality oil of sunflower is associated with its balanced composition of unsaturated fatty acids. It was observed that food rich in unsaturated fatty acids reduced the risk of heart attacks and cardiovascular disease and led to a decrease in serum cholesterol levels [26].

In this study, the percentage of oleic acid and linoleic acid may be considered interesting and distinct among all the other biochemical traits. Thus, this trait may be used to



(a)



(b)

FIGURE 1: Two-dimensional plot of the principal component analysis (PCA) of the twenty-two accessions of *Helianthus annuus* based on biochemical traits along the first two principal axes. (a) Projection of the biochemical traits in the layout generated by PC1 and PC2. (b) Projection of the sunflower accessions in the layout generated by PC1 and PC2.

select accessions having seeds oil for improved diet. These findings are in accordance with the previous study of [27], who observed that oleic and linoleic fatty acids as the dominant acids in oils of various plant species. These acids may be useful for breeders to improve oil quality.

Furthermore, statistical analysis of the fatty acid contents showed significant genotypic effects ($P < 0.05$) for the biochemical parameters studied. This might be due to the genotypic characteristics of the sunflower accession. Several previous studies available on this topic showed that the ratio

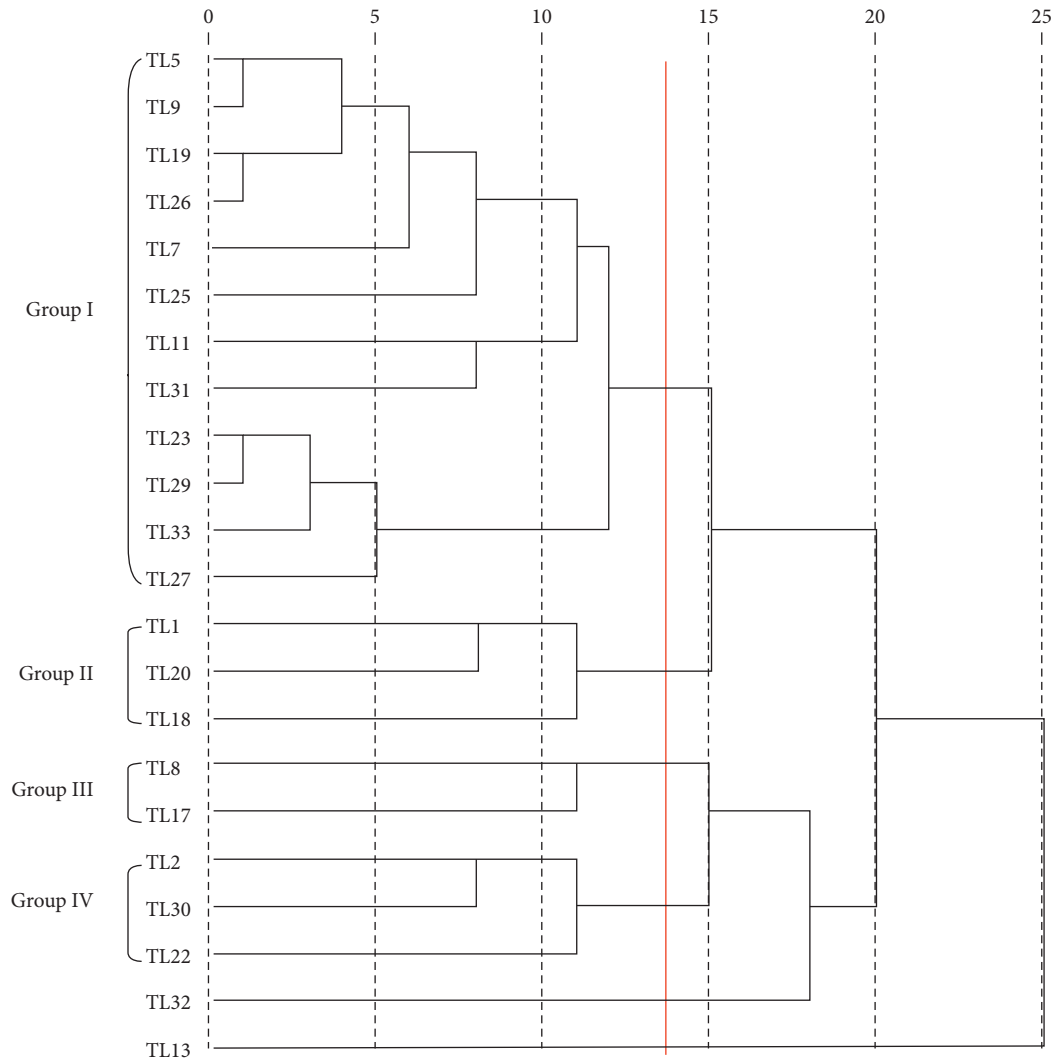


FIGURE 2: Classification of the studied sunflower accessions based on biochemical traits using clustering by Euclidean distance.

TABLE 4: Pearson's correlation coefficients between biochemical traits.

	C16:0	C18:0	C18:1	C18:2	C20:0	C20:1	C22:0	Oil content
C16:0	1							
C18:0	0.328	1						
C18:1	-0.062	0.137	1					
C18:2	-0.488*	-0.439*	0.040	1				
C20:0	-0.089	-0.025	-0.048	-0.552**	1			
C20:1	-0.113	0.136	-0.307	-0.151	0.147	1		
C22:0	0.012	0.221	-0.148	-0.160	0.243	0.228	1	
Oil content	-0.041	-0.006	0.368	-0.088	0.259	-0.062	0.049	1

P value * and ** were significant at 0.05 and 0.01 probability levels, respectively.

between fatty acids could be used as a marker of varietal discrimination and classification of oils [28–30]. Sixty-four genotypes of sunflower were studied using high oleic acid trait, and the biochemical characterization was efficient in detecting genetic variability among genotypes [31]. In contrast, some studies showed that the fatty acid composition of sunflower oil was affected by several abiotic factors such as temperature and water regime [32, 33].

Principal component analysis (PCA) is a powerful tool in analyzing relationships and genetic variability among plant accessions [34]. Our results indicated that the first two components of the PCA express 45.7% of the total variation. The PCA provided a significant insight and separated the sunflower germplasm into four distinct groups. Cluster I was characterized by high content of oleic and linoleic acids and low content of other fatty acids. Cluster II was represented

by the majority of accessions which defined by high content of oleic acid and moderate content of linoleic acid. Cluster III was the smallest cluster and formed by two accessions introduced from abroad (TL13 and TL18) and one accession cultivated in Tunisia (TL20). They were characterized by high content of linoleic acid. Cluster IV was the largest group and included the remaining accessions which were cultivated in Tunisia. The rate of oleic acid and linoleic acid of these accessions varied from 25.95% to 36.17% and from 29.9% to 44%, respectively. The current results showed that each group includes accessions from different geographical origins. The accessions arrangement based on the PCA cluster does not depend on the geographical dispersion. This situation can be due to a considerable gene flow among these studied accessions and the existence of seed exchanges among farmers of different locations of Tunisia [35].

The classification of the studied sunflower accessions based on biochemical traits using clustering by Euclidean distance revealed four different groups. This classification was not in agreement with their geographical distribution. These data were in agreement with previous analysis of [36], and they postulated that fatty-acid contents were similar for North American wild populations, Australian naturalized populations, and improved cultivars of sunflower. Furthermore, the accessions TL13 and TL32 originated from the former Soviet Union and Tunisia, respectively, which clustered out of the four mentioned groups. This reflects a clear genetic gap between these accessions and the rest of the studied ones. In order to design an effective breeding program for any crop, knowledge of correlations among different traits is very important [37]. It was determined in this study that significant and negative correlations were noted between linoleic acid and some saturated acids (palmitic acid, stearic acid, and arachidic acid). In this regard, the quality of sunflower oil is associated with an increase in linoleic acid content and a decrease in these saturated acids and vice versa. Consequently, a breeder may consider these biochemical traits while performing genetic improvement in sunflower.

5. Conclusions

The biochemical characterization of edible vegetable oils is important for understanding the mechanism of oil's function in human diet and health. In this regard, the present study was conducted with an aim to evaluate biochemical variability in 22 accessions of sunflower, using oil content and fatty acid composition. We observed a variation in oil content of different sunflower accessions. Sunflower oils had the best fatty acid profile, showing the predominant unsaturated acids as oleic and linoleic acids. Fatty acid compositions of sunflower oils may show variability according to the accession. The importance of biochemical traits in explaining the observed variability was assessed through the PCA method and clustering by Euclidean distance. Thus, these two methods of cluster analysis allowed the 22 accessions studied to be divided into four major groups. From the research that has been carried out, it is possible that this considerable biochemical variability can be used in breeding

programs focusing on seed oil quality in sunflower germplasm.

Data Availability

All the data used to support the result of this research are available from the corresponding author upon request.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

Authors' Contributions

Taoufik Hosni carried out the experiments, contributed to the analysis of the results, and participated in drafting and revising the manuscript. Zouhaier Abbes discussed the results and participated in revising the manuscript. Leila Abaza, Sana Medimagh, Hamadi Ben Salah, and Mohamed Kharrat read and approved the final version of the manuscript.

Acknowledgments

The authors would like to express gratitude to Ms. Salma Nait Mohamed from the Laboratory of Olive Biotechnology at Centre of Biotechnology of Borj-Cedria (CBBC) for the help in GC analysis. This research was kindly financed by the Ministry of Agriculture, Hydraulic Resources and Fisheries and the Ministry of High Education and Scientific Research.

References

- [1] J. L. Panero and V. A. Funk, "Toward a phylogenetic sub-familial classification for the compositae (asteraceae)," *Proceedings of the Biological Society of Washington*, vol. 115, no. 4, pp. 909–922, 2002.
- [2] USDA, *United States Department of Agriculture): Oilseeds: World Markets and Trade*, USDA, Washington, DC, USA, 2021.
- [3] A. Ayerdi-Gotor, M. Berger, F. Lablette, S. Centis, J. Dayde, and A. Calmon, "Comparative analysis of fatty acids, tocopherols and phytosterols content in sunflower cultivars (*Helianthus annuus*) from a three-year multi-local study," *Phyton*, vol. 84, no. 1, pp. 14–25, 2015.
- [4] S. Rauf, N. Jamil, S. A. Tariq, M. Khan, M. Kausar, and Y. Kaya, "Progress in modification of sunflower oil to expand its industrial value," *Journal of the Science of Food and Agriculture*, vol. 97, no. 7, pp. 1997–2006, 2017.
- [5] C. Ferfuaia, M. Turi, and G. P. Vannozzi, "Variability of seed fatty acid composition to growing degree-days in high oleic acid sunflower genotypes," *Helia*, vol. 38, no. 62, pp. 61–78, 2015.
- [6] M. R. Akkaya, "Fatty acid compositions of sunflowers (*Helianthus annuus* L.) grown in east Mediterranean region," *Rivista Italiana Delle Sostanze Grasse*, vol. XCV, no. 4, pp. 239–247, 2018.
- [7] V. Gavrilova, T. Shelenga, E. Porokhvinova et al., "The diversity of fatty acid composition in traditional and rare oil crops cultivated in Russia," *Biological Communications*, vol. 65, no. 1, pp. 68–81, 2020.

- [8] P. M. Rosa, R. Antoniassi, S. C. Freitas et al., "Chemical composition of Brazilian sunflower varieties," *Helia*, vol. 32, no. 50, pp. 145–155, 2009.
- [9] C. Santonoceto, U. Anastasi, E. Riggi, and V. Abbate, "Accumulation dynamics of dry matter, oil and major fatty acids in sunflower seeds in relation to genotype and water regime," *Italian Journal of Agronomy*, vol. 7, no. 1, pp. 3–14, 2002.
- [10] B. S. Adeleke and O. O. Babalola, "Oilseed crop sunflower (*Helianthus annuus*) as a source of food: nutritional and health benefits," *Food Sciences and Nutrition*, vol. 8, no. 9, pp. 4666–4684, 2020.
- [11] J. M. Fernández-Martínez, B. Pérez-Vich, L. Velasco, and J. Domínguez, "Breeding for specialty oil types in sunflower," *Helia*, vol. 30, no. 46, pp. 75–84, 2007.
- [12] J. J. Salas, M. A. Bootello, E. Martínez-Force, M. Venegas Calerón, and R. Garcés, "High stearic sunflower oil: latest advances and applications," *Oilseeds and Fats Crops and Lipids*, vol. 28, no. 35, pp. 1–8, 2021.
- [13] G. J. Seiler, "Wild *Helianthus annuus*, a potential source of reduced palmitic and stearic fatty acids in sunflower oil," *Helia*, vol. 27, no. 40, pp. 55–62, 2004.
- [14] V. Fernández-Moya, E. Martínez-Force, and R. Garcés, "Oils from improved high stearic acid sunflower seeds," *Journal of Agricultural and Food Chemistry*, vol. 53, no. 13, pp. 5326–5330, 2005.
- [15] J. M. Fernández-Martínez, M. Mancha, J. Osorio, and R. Garcés, "Sunflower mutant containing high levels of palmitic acid in high oleic background," *Euphytica*, vol. 97, no. 1, pp. 113–116, 1997.
- [16] D. Škorić, S. Jocić, Z. Sakač, and N. Lečić, "Genetic possibilities for altering sunflower oil quality to obtain novel oils," *Canadian Journal of Physiology and Pharmacology*, vol. 86, no. 4, pp. 215–221, 2008.
- [17] L. Mokrani, L. Gentzbittel, F. Azanza, L. Fitamant, G. Al-Chaarani, and A. Sarrafi, "Mapping and analysis of quantitative trait loci for grain oil content and agronomic traits using AFLP and SSR in sunflower (*Helianthus annuus* L.)," *Theoretical and Applied Genetics*, vol. 106, no. 1, pp. 149–156, 2002.
- [18] F. Vear, "Changes in sunflower breeding over the last fifty years," *OCL*, vol. 23, no. 2, p. D202, 2016.
- [19] A. J. León, F. H. Andrade, and M. Lee, "Genetic analysis of seed-oil concentration across generations and environments in sunflower," *Crop Science*, vol. 43, no. 1, pp. 135–140, 2003.
- [20] A. Dimitrijević, I. Imerovski, D. Miladinović et al., "Oleic acid variation and marker-assisted detection of pervenets mutation in high- and low-oleic sunflower cross," *Crop Breeding and Applied Biotechnology*, vol. 17, no. 3, pp. 229–235, 2017.
- [21] FAOSTAT, *Prod Stat: Crops, FAO Statistical Database (Faostat)*, Food and Agriculture Organization of the United Nations (FAO), Rome, Italy, 2019.
- [22] T. F. Abro, P. K. Oad, J. K. Sootaher et al., "Genetic variability and character association between grain yield and oil content traits in sunflower (*Helianthus annuus* L.)," *International Journal of Biology and Biotechnology*, vol. 17, no. 4, pp. 701–706, 2020.
- [23] H. T. Ebeed, N. M. Hassan, M. M. Keshta, and O. S. Hassanin, "Comparative analysis of seed yield and biochemical attributes in different sunflower genotypes under different levels of irrigation and salinity," *Egyptian Journal of Botany*, vol. 59, no. 2, pp. 339–355, 2019.
- [24] T. Hosni, Z. Abbas, L. Abaza, S. Medimagh, H. Ben Salah, and M. Kharrat, "Effect of broomrape (*Orobanche cumana* Wallr.) on some agro-morphological and biochemical traits of Tunisian and some reference sunflower (*Helianthus annuus* L.) accessions," *Journal of Plant Diseases and Protection*, vol. 127, no. 6, pp. 831–841, 2020.
- [25] J. Roche, A. Bouniols, Z. Mouloungui, T. Barranco, and M. Cerny, "Management of environmental crop conditions to produce useful sunflower oil components," *European Journal of Lipid Science and Technology*, vol. 108, no. 4, pp. 287–297, 2006.
- [26] M. Gallardo-Velázquez, T. G. Osorio-Revilla, E. Castañeda-Pérez, and K. Uribe-Hernández, "Characterization of Mexican fishes according to fatty acid profile and fat nutritional indices," *International Journal of Food Properties*, vol. 19, no. 6, pp. 1401–1412, 2015.
- [27] D. Kazlauskienė, G. Kasparavičienė, P. Nenortienė et al., "Determination of fatty acid composition and antioxidant activity in vegetable oils," *Chemija*, vol. 32, no. 1, pp. 17–27, 2021.
- [28] B. Perez-Vich, R. Garcés, and J. M. Fernandez-Martinez, "Inheritance of high palmitic acid content in the sunflower mutant CAS-12 and its relationship with high oleic content," *Plant Breeding*, vol. 121, no. 1, pp. 49–56, 2002.
- [29] J. J. Salas, A. J. Moreno-Pérez, E. Martínez-Force, and R. Garcés, "Characterization of the glycerolipid composition of a high-palmitoleic acid sunflower mutant," *European Journal of Lipid Science and Technology*, vol. 109, no. 6, pp. 591–599, 2007.
- [30] S. Baud and L. Lepiniec, "Physiological and developmental regulation of seed oil production," *Progress in Lipid Research*, vol. 49, no. 3, pp. 235–249, 2010.
- [31] I. S. Tilak, B. Kisan, I. S. Goud et al., "Biochemical and molecular characterization of parents and its crosses for high oleic acid content in sunflower (*Helianthus annuus* L.)," *International Journal of Current Microbiology and Applied Sciences*, vol. 7, no. 4, pp. 2000–2020, 2018.
- [32] V. Fernández-Moya, E. Martínez-Force, and R. Garcés, "Temperature effect on a high stearic acid sunflower mutant," *Phytochemistry*, vol. 59, no. 1, pp. 33–37, 2002.
- [33] A. Ebrahimi, P. Maury, M. Berger et al., "QTL mapping of seed-quality traits in sunflower recombinant inbred lines under different water regimes," *Genome*, vol. 51, no. 8, pp. 599–615, 2008.
- [34] A. L. Price, N. J. Patterson, R. M. Plenge, M. E. Weinblatt, N. A. Shadick, and D. Reich, "Principal components analysis corrects for stratification in genome-wide association studies," *Nature Genetics*, vol. 38, no. 8, pp. 904–909, 2006.
- [35] F. Rebaa, G. Abid, M. Aouida et al., "Genetic variability in Tunisian populations of faba bean (*Vicia faba* L. var. major) assessed by morphological and SSR markers," *Physiology and Molecular Biology of Plants*, vol. 23, no. 2, pp. 397–409, 2017.
- [36] G. J. Seiler, T. Gulya, G. Kong, S. Thompson, and J. Mitchell, "Oil concentration and fatty-acid profile of naturalized *Helianthus annuus* populations from Australia," *Genetic Resources and Crop Evolution*, vol. 65, no. 8, pp. 2215–2229, 2018.
- [37] M. S. Mazid, M. Y. Rafii, M. M. Hanafi, H. A. Rahim, M. Shabanimofrad, and M. A. Latif, "Agro-morphological characterization and assessment of variability, heritability, genetic advance and divergence in bacterial blight resistant rice genotypes," *South African Journal of Botany*, vol. 86, pp. 15–22, 2013.