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A comparative study of physicochemical properties, antioxidant and enzyme inhibition activities of oils extracted from seeds of seven new sunflower (*Helianthus annuus* L.) lines

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Abstract: This study was aimed to evaluate the physicochemical properties and chemical profile of seeds oil obtained from new seven sunflower (*Helianthus annuus* L.) lines as well as their antioxidant and enzyme inhibition activity. The seeds powder was extracted by maceration in n-hexane. The oil content of the seven lines was ranged from 20.04% to 36.65% and was either yellow or pale yellow in color. No significant variations were observed on the refractive index (1.46 unit) of the oil. Oils of the seven lines were significantly ($p < 0.05$) different in their saponification values (32.13–282.66 mg KOH/g oil), peroxide values (1.76–13.26 mg KOH/g-oil), acid values (0.016–1.766 mg KOH/g oil) and free fatty acids content (6.26–72.23 mg KOH/g-oil). The chemical profile of the oil revealed that line APO42 contained the highest amount of monounsaturated fatty acids (55.9%). All the seven lines contained a considerable amount of linoleic acid (27.5%– 42.5%), and it represented the major compound in lines BOH3 (42.5%) and H1733 (39.8%). The variation was remarkable in their oleic acid content where the highest amount was observed in lines APO42 (55.4%) and APO43 (42.2%), respectively. Lines BOH3, APO43, APO41, and H1733 exerted the best total antioxidant activity in addition to their capacity to reduce Cu^{2+} and Fe^{3+}, while lines H1733 and APO43 had a metal chelating activity as well. All oils showed weak acetylcholinesterase, butyrylcholinesterase, α-glucosidase and α-amylase inhibitory activities. Only four lines showed considerable enzyme inhibitory activity against tyrosinase enzymes. Multivariance analysis suggested that linoleic acid participated in the observed biological activity of the oils. In conclusion, these new lines might contribute to the nutritional and phytotherapeutic properties of sunflower oil in addition to other industrial applications.

Key words: Sunflower oil, chemical profile, antioxidant activity, enzyme inhibition activity

1. Introduction

Sunflower (*Helianthus annuus* L.), belonging to the Asteraceae family, is an oilseed crop cultivated worldwide for the production of high-quality edible oil. Sunflower seed oil is ranked the fourth in the international oilseed market after palm, soybean, and rapeseed (Adeleke and Babalola, 2020). Two types of sunflower seeds are commercially cultivated: the oilseed (sunfoil) type, which is rich in oil content, and the non-oilseed seed, which is used for confectionary purposes (Giada and Mancini-Filho, 2009; Eryilmaz and Yesilyurt, 2016). In traditional medicine, the oil is commonly used to cure many diseases; among which, there is heart disease, bronchial, laryngeal, and pulmonary infections, coughs and colds, and whooping cough (Bashir et al., 2015).

Sunflower seed oil was found to exert several biological activities like healing properties, gastric protection, being antiinflammatory, antimicrobial, antioxidant, antidiabetic, and antihypertensive, having antitumor activities, reducing both total cholesterol and low-density lipoprotein (LDL) cholesterol (Guo et al., 2017). The seed oil contains saturated and unsaturated fatty acids, phenolic compounds, flavonoids, and vitamins. The oil is rich in polyunsaturated fatty acids (approximately 31.0%) especially linoleic acid (55%–70%) and generally, commercially available sunflower phenotypes contain less amount of oleic acid (20–25%) (Premnath et al., 2016).

Sunflower is considered one of the promising crops introduced in Sudan. Commercial production of sunflower in Sudan was started in the late 1980s with

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the introduction of Hysun-33 hybrid from Australia and PAN-7351 hybrid from South Africa. Then, two open-pollinated sunflower varieties namely, Damazin-1 and Damazin-2 were produced. However, due to many production constraints, the cultivation of sunflower has failed to expand in the country. This enforced breeding programs in Sudan to produce new local sunflower hybrids were adapted to Sudan conditions in order to boosting sunflower production and productivity in the country (Mohamed, 2010).

Moreover, the demand for functional foods is steadily growing, as it plays a beneficial effect on human health. For example, a food with balanced polyunsaturated fatty acids composition influences diverse aspects of immunity and metabolism. The high content of linoleic and oleic acids in sunflower oil makes this oil an ideal food with physiologically preventative and/ or health-enhancing effects (Franco et al., 2018). In fact, plant breeding programs are currently applying biotechnological approaches to obtain improved sunflower varieties with maximum nutraceutical properties that significantly help in developing products with high nutritional and beneficial health effects in addition to satisfying the global demand for chemical industries (Aremu et al., 2016). Several sunflower lines, well adapted to the agro-climatic condition of Sudan, were produced at the Department of Agronomy, Faculty of Agriculture, University of Khartoum, Sudan. The germplasm of these new lines has a wide variation in characters such as yield, seed number, plant height, earliness and susceptibility to biotic and a biotic stresses. Moreover, the demand for functional foods is steadily growing, as it plays a beneficial effect on human health (Sergio et al., 2020). Hence, the present study is a continuation for evaluation of the beneficial properties of the seeds of these seven new sunflower lines and was designed to determine the physicochemical characteristics, fatty acid profile of their oil. In addition, the antioxidant and inhibitory properties of the oil against key enzymes (amylase, glucosidase, tyrosinase, acetyl- and butyryl-cholinesterases) involved in the pathogenesis of diseases such as diabetes, skin hyperpigmentation, and neurological diseases were evaluated.

2. Materials and methods

2.1. Plant materials

Seeds from seven sunflower new lines, namely APO41, APO42, APO43, APO44, APO45, BOH3, and H1733 were kindly provided by Dr. Abd El Wahab H. Abdalla, Department of Agronomy, Faculty of Agriculture, University of Khartoum, Sudan. These lines were developed from the random mating population through continued selfing. Seeds were obtained from plant materials grown at shambat (lat 15° 40′ N and long, 32° 32′ E). The mean maximum and minimum temperatures were in the range of 34.4 °C and 21.5 °C, respectively during the growing season, and the average rainfall is 18.8 mm/annum. The soil of the area is heaving loamy soil.

2.2. Preparation of the oil

The fine powder (20 g) of seeds was extracted by maceration in n-hexane (400 mL) using a shaker apparatus, for about 24 h at room temperature, filtered, and then the solvent was evaporated. The resultant dry extract from each sample was weighed and stored at 4°C, in amber-colored glass container until used.

2.3. Physicochemical properties of oil

Refractive index, free fatty acids, peroxide, acid, and saponification values were evaluated following the standard method described by AOCS (Official Methods and Recommended Practices of the AOCS, 2004).

2.4. Fatty acids analysis

Fatty acids present in oils were converted to fatty acid methyl esters as described by Liu (1994).

2.5. GC/MS analysis

The chemical profile of oil seeds of the seven lines of sunflower were determined by gas chromatography-mass spectrometry system (GC-MS) according to Hemmati and co-workers (2020) method. Analyses were performed using QP2010-Shimadzu equipment operating in the EI mode at 70 eV. An SLB5 column DB-5 ms (30 m, 0.25 mm film thickness) was employed with a 36 min temperature program of 60–320 at 10 °C/min followed by a 10 min hold at 320 °C. The injector temperature was 250 °C, the flow rate of the carrier gas (helium) was 1 mL/min, and the split ratio was 1:50. The interval of the scan m/z was between 35 and 900. The identity of different compounds was achieved by comparing the measured data with the NIST08.LIB database.

2.6. Antioxidant activity

The antioxidant potential including the total antioxidant activity, scavenging 2,2-diphenyl-1-picrylhydrazyl (DPPH) and 2,2'-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid (ABTS) radicals, ferric and cupric ion reducing antioxidant power (FRAP and CUPRAC), and ferrous chelating properties of each oil were evaluated via colorimetric assays described by Zengin et al. (2015) and Mohammed et al (2020). Different concentrations of the tested oils were prepared in methanol and then the samples were used in above-mentioned assays. In CUPRAC and ferrous chelating assays, we prepared a blank for each concentration without $CuCl_2$ and ferrozine, respectively. Afterwards, the absorbance of the blank was subtracted from that of the sample in the assays.

2.7. Enzyme inhibition activity

Colorimetric methods were also adopted to evaluate the enzyme inhibition property of the studied sunflower seed

oils. The enzymes used were acetylcholinesterase (AChE), butyrylcholinesterase (BuCh), tyrosinase, α-glucosidase and α-amylase. Different concentrations of the tested oils were prepared in methanol and then the samples were used in the enzyme inhibition assays. We prepared a blank for each concentration without enzymes in the assays and the absorbance of the blank was subtracted from that of the sample. All experimental details were given in our previous papers (Zengin et al., 2015; Mohammed et al., 2020).

2.8. Statistical analysis

Results of physicochemical properties, antioxidant and enzyme inhibitory activity were presented as the mean ± standard deviation (SD), and significant differences ($p < 0.05$) were determined by One-way ANOVA with post-hoc Tukey HSD, using SPSS 17 software. The multivariate analysis was done with SIMCA 10.0 software (Umetrics, Umeå, Sweden). The chemical profile of sunflower lines and bioactivities datasets were analyzed using Partial Least Square (PLS) analysis model to highlight the difference in chemical compounds and bioactivities between the species parts.

3. Results

3.1. Identity and quality of the oil of seven sunflower lines

The physicochemical characteristics of the seed oil extracted from the seven lines of sunflower are presented in Table 1. The oils, extracted by maceration with n-hexane, had a yield ranging from 20.04% to 36.65% with the highest content obtained from line APO43. However, these values were less than the commercially available sunflower varieties, which contained 39% to 49% oil in seed. Also, they were lower in oil content than those obtained from the Brazilian cultivars, which was in the range from 38 to 48% (Porto et al., 2008) but within the range (29.5% - 50.2%) of some sunflower genotypes obtained by Carvalho et al. (2009). Different biotic and abiotic factors among which are genetic diversity, climate conditions and agricultural conditions may be responsible for these variations in oil contents of sunflower seeds (Carvalho et al., 2009). Oils of the seven lines had oil color either yellow or pale yellow and refractive index value (1.46 unit) in accordance with the recommended (standard) physicochemical characteristics of edible oils as given by the Food and Agriculture Organization (FAO) and the World Health Organization (WHO/FAO) (1993). Acid values ranged between 0.016 to 1.766 mg KOH/g oil.

Lower peroxide values indicated high quality of the oils. Peroxide values of the seven sunflower lines ranged between 1.76 to 13.26 mg KOH/g-oil. The lowest peroxide values were recorded by lines APO41 and APO42 (1.76 and 2.86 mg KOH/g-oil, respectively). These values were higher than those obtained from other sunflower varieties (0.16 mg KOH/g-oil) by Oliveira et al. (2019). The seven lines showed significant ($p < 0.05$) differences in their free fatty acids content. It ranged from 6.26 (for APO45) to 72.23 mg KOH/g-oil (for H1733). These values were higher than those obtained from oils extracted from other sunflower lines reported by Tabasum et al. (2012). Also, they exerted significant ($p < 0.05$) variation in their saponification values (32.13–282.66 mg KOH/g). The highest value (282.66 mg KOH/g) was obtained from line APO41 followed respectively by APO42 (281.23 mg KOH/g), APO43 (272.36 mg KOH/g) and APO45 (195.66 mg KOH/g) respectively. These results were higher than those reported from other sunflower genotypes obtained by Sadoudi et al. (2014) (186.13–192.6 mg KOH/g).

3.2. Chemical profile of seed oils from the seven lines of sunflower

Analysis by GC/MS was carried out to determine the chemical profile of the oils extracted from the seven lines of sunflower. Results are presented in Table 2. About 14 to 18 compounds were identified in all lines excepted line APO45 where a total of 27 compounds were identified.

Table 1. Physicochemical characterization of crude oil from the seeds of the seven sunflower lines.

Lines	Yield (%)	Colour	Refractive Index	Saponification value[a]	Free fatty acid[a]	Peroxide value[a]	Acid value[a]
APO41	34.05	Pale yellow	1.46 ± 0.05[a]	282.66 ± 0.06[a]	42.13 ± 0.05[c]	1.76 ± 0.05[c]	0.086 ± 0.05[b]
APO42	22.90	Yellow	1.46 ± 0.05[a]	281.23 ± 0.06[a]	9.94 ± 0.05[f]	2.86 ± 0.05[e]	0.016 ± 0.05[e]
APO43	36.65	Yellow	1.47 ± 0.05[a]	272.36 ± 0.06[b]	12.73 ± 0.05[e]	4.76 ± 0.05[d]	0.026 ± 0.05[de]
APO44	32.55	Yellow	1.47 ± 0.05[a]	32.13 ± 0.06[f]	17.66 ± 0.06[d]	4.76 ± 0.05[d]	0.036 ± 0.05[ce]
APO45	25.14	Yellow	1.46 ± 0.05[a]	195.66 ± 0.06[c]	6.26 ± 0.12[g]	6.16 ± 0.05[b]	0.016 ± 0.05[fe]
BOH3	20.04	Pale yellow	1.46 ± 0.05[a]	100.63 ± 0.12[d]	53.16 ± 0.06[b]	6.13 ± 0.05[b]	0.066 ± 0.05[bc]
H1733	24.86	Yellow	1.46 ± 0.05[a]	97.66 ± 0.06[e]	72.23 ± 0.12[a]	13.26 ± 0.05[a]	1.766 ± 0.06[a]

[a], values expressed as mg KOH/g-oil. In each column different superscript letters indicate significant differences ($p < 0.05$).

Table 2. Chemical profile of the oil extracted from the seeds of the seven sunflower lines.

No.	RT	Compound	Formula	MW	Area (%)						
					APO41	APO42	APO43	APO44	APO45	BOH3	H1733
		Saturated fatty acids									
1	17.653	Myristic acid	$C_{14}H28O_2$	228	0.1	0.1	0.1	-	0.1	0.1	0.1
2	19.767	Palmitic acid	$C_{16}H_{32}O_2$	256	9.7^{a}	7.3^{bc}	8.4^{b}	10.1^{a}	10.0^{a}	4.8^{d}	6.6^{c}
3	20.750	Stearic acid	$C_{18}H_{36}O_2$	284	-	0.1	-	-	0.1	-	-
4	20.747	Margaric acid	$C_{17}H_{34}O_2$	270	0.1	-	-	-	-	0.1	-
5	21.687	Stearic acid	$C_{18}H_{36}O_2$	284	5.7^{b}	5.6^{b}	4.2^{c}	3.8^{d}	6.6^{a}	5.5^{b}	4.7^{c}
6	23.450	Arachidic acid	$C_{20}H_{42}O_2$	326	0.4	0.4	0.3	0.3	0.5	0.4	0.3
7	24.680	9,10-Dihydroxystearic acid	$C_{18}H_{36}O_4$	316	0.6	-	-	-	-	-	-
8	25.077	Behenic acid	$C_{23}H_{44}O_2$	340	0.9	0.7	0.7	0.6	0.8	0.8	0.7
9	26.347	Capric acid	$C_{10}H_{20}O_2$	172	-	-	-	-	0.2	-	-
10	26.583	Lignoceric acid	$C_{24}H_{48}O_2$	368	0.3	0.2	0.2	0.3	0.3	0.2	0.2
11	30.533	Melissic acid	$C_{30}H_{60}O_2$	466	-	-	-	-	0.1	-	-
		Monounsaturated fatty acids									
12	19.547	Palmitoleic acid	$C_{16}H_{30}O_2$	268	0.2	0.2	0.2	0.4	0.4	0.1	-
13	20.513	Petroselinic acid	$C_{18}H_{34}O_2$	282	0.1	-	0.3	0.4	-	-	-
14	20.517	cis-10-Heptadecenoic acid	$C_{17}H_{32}O_2$	268	-	-	-	-	0.1	-	-
15	21.473	Oleic acid	$C_{18}H_{34}O_2$	282	-	55.4^{a}	42.2^{b}	38.6^{c}	-	37.6^{c}	32.2^{d}
16	21.500	Elaidic acid	$C_{18}H_{34}O_2$	282	46.4^{a}	-	1.2^{c}	-	40.7^{b}	-	0.8^{c}
17	21.500	cis-Vaccenic acid	$C_{18}H_{34}O_2$	282	-	-	-	1.3^{b}	-	0.6^{c}	3.2^{a}
18	21.777	trans-2-Dodecenoic acid	$C_{12}H_{22}O_2$	198	-	0.1	-	-	-	-	-
19	23.223	cis-11-Eicosenoic acid	$C_{20}H38O_2$	310	0.5	0.2	-	-	-	0.5	-
20	24.270	8-Octadecenoic acid	$C_{18}H_{34}O_2$	282	-	-	0.2	-	-	-	-
21	24.453	7-Hexadecenoic acid	$C_{16}H_{30}O_2$	268	0.2	-	-	-	-	0.2	-
		Polyunsaturated fatty acids									
22	21.390	Linoleic acid	$C_{18}H_{34}O_2$	280	33.6^{d}	27.5^{e}	37.1^{c}	39.2^{b}	32.4^{d}	42.5^{a}	39.8^{b}
23	22.753	gamma-Linolenic acid	$C_{18}H_{30}O_2$	292	0.3	-	0.2	-	0.1	0.1	-
24	22.867	Adrenic acid	$C_{22}H_{36}O_2$	332	-	-	-	-	0.1	-	-
		Sterols									
25	29.930	Campesterol	$C_{28}H_{48}O$	400	-	-	-	-	0.1	-	-
26	30.080	Stigmasterol	$C_{29}H_{48}O$	412	-	-	-	-	0.1	-	-
27	30.460	gamma.-Sitosterol	$C_{29}H_{50}O$	414	0.2	0.4	0.3	0.3	0.9	0.1	0.7
		Hydrocarbons									
28	24.040	Tetracosane.	$C_{24}H_{50}$	338	-	-	-	-	0.4	-	-
29	24.613	1,E-6,Z-11-Hexadecatriene	$C_{16}H_{28}$	220	0.1	0.3	-	-	-	-	0.6
30	24.613	Cyclododecyne	$C_{12}H_{20}$	164	-	-	0.6	0.7	-	-	-
31	24.660	1,19-Eicosadiene	$C_{20}H_{38}$	278	-	1.4	-	-	-	-	-

Table 2. (Continued).

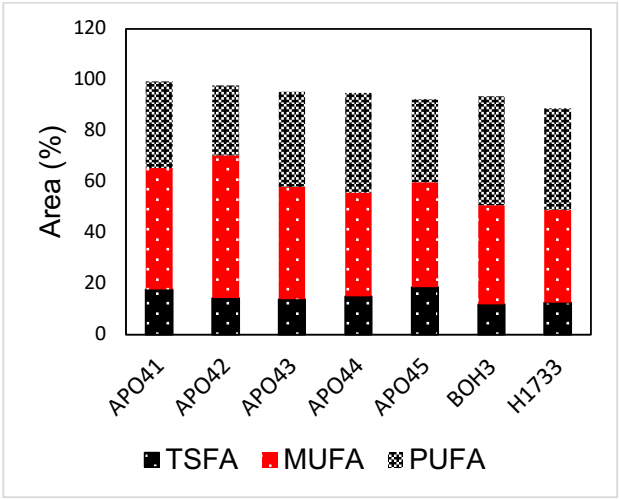
32	24.840	Eicosane.	$C_{20}H_{42}$	282	-	-	-	-	1.5	-	-
33	27.070	Hexatriacontane.	$C_{36}H_{74}$	506	-	-	-	-	0.1	-	-
34	27.763	Nonacosane	$C_{29}H_{60}$	408	-	-	-	-	0.1	-	-
35	29.080	Hentriacontane	$C_{31}H_{64}$	436	-	-	-	-	0.1	-	-
		Others									
36	22.763	Citric acid, tributyl ester	$C_{18}H_{32}O_7$	360	-	-	0.3	-	0.2	0.1	-
37	23.190	Dipalmitin	$C_{35}H_{68}O_5$	568	-	-	-	-	-	-	0.8
38	23.667	2,6,6,10-Tetramethyl-undeca-8,10-diene-3,7-dione	$C_{15}H_{24}O_2$	236	-	-	0.2	-	-	-	-
39	23.733	Phytol	$C_{20}H_{40}O$	296	-	-	-	0.2	-	-	-
40	24.223	Glycerol 1-monolinolate	$C_{21}H_{38}O_5$	370	-	-	-	-	-	-	0.3
41	24.270	alpha.-Monoolein.	$C_{21}H_{40}O_4$	356	-	0.1	-	-	-	-	-
42	24.657	Butanoic acid 2,3-dihydroxypropyl ester	$C_2H_{14}O_4$	162	-	-	2.4	2.1	-	-	-
43	24.663	Cyclopentadecanone, 2-hydroxy-.	$C_{15}H_{28}O_2$	240	-	-	-	-	1.5	-	-
44	29.277	alpha.-Tocopherol	$C_{29}H_{50}O_2$	430	-	0.2	-	-	0.3	-	-
45	30.963	Handianol	$C_{30}H_{50}O$	426	-	-	-	-	0.1	-	-

The data have standard deviation in the range of 0.01 - 0.20; Different superscript letters in the same raw indicate significant difference ($p < 0.05$). Values without superscript letters are significantly not different.

Oils were dominated by saturated, monounsaturated, and polyunsaturated fatty acids. Other compounds, among them α-tocopherol was only detected in lines APO42 and APO45. Campesterol (0.1%), stigmasterol (0.1%), and γ-sitosterol (0.9%) were identified in line APO45. The presence of phytosterols in sunflower seeds was found to have a remarkable effect in reducing the cholesterol level and risk of colon cancer as well as increasing the body immunity (Smith et al., 2015).

Lines BOH3 (42.6%) and H1733 (39.8%) contained slightly higher amount of polyunsaturated fatty acid compounds while other lines had more monounsaturated fatty acids (Fig. 1-a). Interestingly, line APO42 contained 2-fold higher content in monounsaturated fatty acids than polyunsaturated ones. Also, lines APO41, APO45 and APO43 showed 1.4-, 1.3- and 1.2-fold higher monounsaturated fatty acids than polyunsaturated ones while the content in monounsaturated fatty acids (40.7%) in line APO44 was slightly higher than the polyunsaturated fatty acids (39.2 %) (Figure 1-a).

Four fatty acids namely, palmitic, stearic, oleic, and linoleic acids are known as important constituents of sunflower oil (Baydar and Erbaş, 2005). However, the distribution of these four fatty acids in the investigated lines is summarized in Figure 1-b. It was clear that all the seven lines contained a considerable amount of linoleic acid, and it represented the major compound in lines APO44 (39.2%), BOH3 (42.5%), and H1733 (39.8%). Although it was in low concentration, the content in palmitic and stearic acids was comparable in the majority of the lines. However, the variation was remarkable in their oleic acid content where the highest amount was observed in line APO42 (55.4%) followed by lines APO43 (42.2%), APO44 (38.6%), BOH3 (37.6%), and H1733 (32.2%), respectively. Merwe and co-workers found that an increase in temperature during seed development leads to an increase of oleic acid content (Merwe et al. 2015). They also noted a significant negative correlation between oleic and linoleic acid percentage where a phenotype with low oleic acid would essentially be high in linoleic acid. This observation was also noted in the present study except for line APO44 where its content in these two fatty acids was not largely different. Furthermore, lines APO41 and APO45 were completely devoid of oleic acid, instead, they were dominated by elaidic acid (trans form of oleic acid) (46.4% and 40.7% respectively). Overall, the percentage content in major fatty acids varied according to sunflower phenotypes (Fayyaz and Ahmad, 2003; Merwe et al., 2015). Additionally, the presence of relatively low saturated fatty acids and a high amount of unsaturated fatty acids in all the seven investigated lines is an advantage. Dietary with reduced saturated fatty acids and a moderate increase in



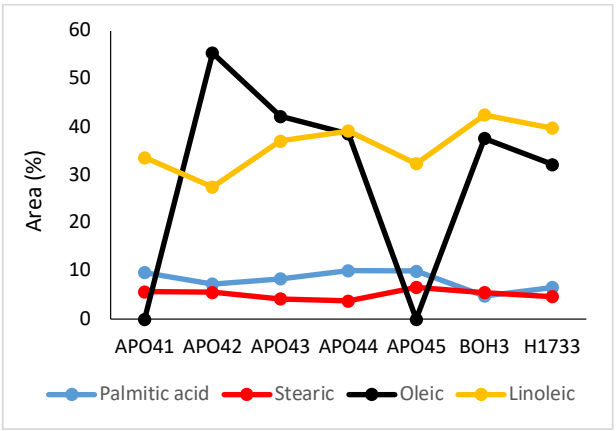


Figure 1. a: Distribution of total fatty acids content in the oil of the seeds of the seven sunflower lines. TSFA, total saturated fatty acids; MUFA, monounsaturated fatty acids; PUFA, polyunsaturated fatty acids. 1-b: Distribution of the major fatty acids in the oil of the seven lines of sunflower.

mono- and poly-unsaturated fatty acids are recommended in human nutrition in order to prevent many diseases, especially cardiovascular ones (Nakić et al., 2006). In fact, studies had shown that sunflower oil rich in oleic acid had a positive impact on cardiovascular diseases risk factors including glucose metabolism, the status of lipid profile, and levels of blood pressure (Huth et al., 2015; Vijayakumar et al., 2016). Furthermore, oleic acid has been suggested to be associated with a low risk of breast cancer. Evidence based on the studies of southern European populations, whose nutritional habits to eat food rich in oleic acid, showed oleic acid to be protective (Simonsen et al., 1998). This was further supported by the study of Menendez and co-workers who concluded that the gene (Her-2/neu (erbB-2)) expression, which participated in the development of breast cancer, could be intimidated by oleic acid (Menendez et al, 2005).

Additionally, an oil rich in oleic acid positively contributed to its stability, as it resists the oxidative degradation caused by exposure to high temperatures (Belingheri et al., 2015). Thus, a high oleic acid sunflower oil is more preferable for cooking including frying, refining and storage process than that with lower oleic acid content (Marmesat et al., 2012). On the other hand, an increased level of elaidic acid was suggested to be associated with a variety of cardiovascular diseases (Sun et al., 2007). Furthermore, a recent study demonstrated that elaidic acid enhances the metastasis of colorectal cancer cells (Ohmori et al., 2017).

Furthermore, the health effects of linoleic acid are controversial. Farvid and coworkers reported that data analysis from 310,602 subjects showed that the group with high dietary linoleic acid intake represented a reduced risk of coronary heart diseases up to 15% when compared to the group with lower linoleic acid intake (Farvid et al., 2014). In contrast, Chowdhury and co-workers (Chowdhury et al., 2014) and Ramsden and co-workers (Ramsden et al., 2013) did not observe the beneficial effect of linoleic acid as dietary supplements with regard to heart diseases. Although some studies had associated dietary linoleic acid with cancer development (Zock and Katan, 1998; Sauer et al., 2007), others suggested that only under certain conditions it can be carcinogenic (Ip et al., 1985). Nevertheless, in 2014, the Academy of Nutrition and Dietetics summarized reports of the Dietary Guidelines for America, American Heart Association and the WHO emphasizing the intake of linoleic acid should not exceed 10% of energy, and it can improve cardiovascular health (Vannice and Rasmussen, 2014).

3.3. Antioxidant activity of the oil

Extracts derived from plants contain antioxidant agents that are capable to neutralize the harmful effects generated by reactive oxygen species, and they are believed to have insignificant side-effects (Guo et al., 2017). Six complementary in vitro assays have been employed in order to understand the different mechanisms of natural antioxidants present in the seed oil of the seven new sunflower lines. Oils exerted different antioxidant capacity depending to the assay used (Table 3). They did not exhibit any antiradicals activity against both DPPH and ABTS radicles. However, all the seven lines showed considerable metal reducing capacity; however, lines APO43 (53.31 mg TE/g), BOH3 (52.42 mg TE/g) and APO41 (50.68 mg TE/g) showed the highest Cu^{2+} reducing activity with no significant difference ($p \geq 0.05$). Line H1733 (43.45 mg TE/g) exerted significantly ($p < 0.05$) the highest Fe^{3+} reducing capacity followed by lines BOH3 (29.90

mg TE/g), APO43 (27.17 mg TE/g), and APO41 (26.11 mg TE/g) with no significant difference ($p \geq 0.05$). Only 3 lines, namely H1733, APO45, and APO43 showed iron-chelating activities (12.13, 11.42, and 10.10 mg EDTAE/g, with no significant difference ($p \geq 0.05$)). The highest total antioxidant activity was obtained from lines BOH3 (0.97 mmol TE/g) and APO43 (0.87 mmol TE/g). Previous antioxidant activity studies on sunflower seeds were carried out on the striped seed cotyledon by Guo et al. (2017). They found that water extract exerted higher antioxidant activity than the ethanolic one from the FRAP, DPPH, and oxygen radical absorbance capacity assays. Furthermore, it was reported that the antioxidant activity of the seed could be due to the presence of enzymatic antioxidants like catalase, glutathione reductase, guaiacol peroxidase and glutathione dehydrogenase, phenolic compounds including flavonoids, phenolic acids and tocopherols, carotenoids, L-ascorbic acid, and peptides (Guo et al., 2017).

3.4. Enzyme inhibition activity of the oil

The enzyme inhibition capacity of the oil extracted from the seven sunflower seeds was evaluated against AChE, BChE, tyrosinase, alpha-amylase, and alpha-glucosidase. Results showed that all oils exerted weak or no enzyme inhibition activity against all tested enzymes except tyrosinase (Table 4). Line APO43 (10.62 mg KAE/g) revealed some significant ($p < 0.05$) level of inhibitory activity against tyrosinase followed by lines BOH3 (6.81 mg KAE/g) and APO41 (5.18 mg KAE/g) with no significant difference ($p \geq 0.05$), and line APO42 showed the least activity (2.55±0.34

Table 3. Antioxidant activity of the oil extracted from the seeds of the seven sunflower lines.

Line	DPPH (mg TE*/g)	ABTS (mg TE/g)	CUPRAC (mg TE/g)	FRAP (mg TE/g)	MCA (mg EDTAE**/g)	PBD (mmol TE/g)
APO41	na	na	50.68±**1.01**^{a}	26.11±**0.81**^{bc}	na	0.77±0.01^{c}
APO42	na	na	45.35±**1.61**^{b}	21.79±**0.70**^{cd}	na	0.60±0.04^{f}
APO43	na	na	53.31±**0.74**^{a}	27.17±**0.46**^{b}	10.10±**0.0**^{9}a	0.87±0.02^{b}
APO44	na	na	48.72±**1.00**^{ab}	19.98±**0.92**^{d}	na	0.69±0.01^{de}
APO45	na	na	44.49±**1.33**^{b}	18.78±**0.10**^{d}	11.42±**1.0**^{8}a	0.63±0.01^{ef}
BOH3	na	na	52.42±**2.97**^{a}	29.90±**0.86**^{b}	na	0.97±0.01^{a}
H1733	na	na	49.37±**2.72**^{ab}	43.45±**4.12**^{a}	12.13±**1.7**^{6}a	0.73±0.07^{cd}

DPPH: 2,2-diphenyl-1-picrylhydrazyl, ABTS: 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid), CUPRAC: cupric ion reducing antioxidant capacity, FRAP: reducing power (ferric reducing antioxidant power, MCA: metal chelating activity, PBD: Phosphomolybdenum .

* TEs, trolox equivalents.

** EDTAEs, disodium edetate equivalents.

Different superscript letters in the same column indicate significant difference ($p < 0.05$).

Table 4. Enzyme inhibition activity of the oil extracted from the seeds of the seven sunflower lines.

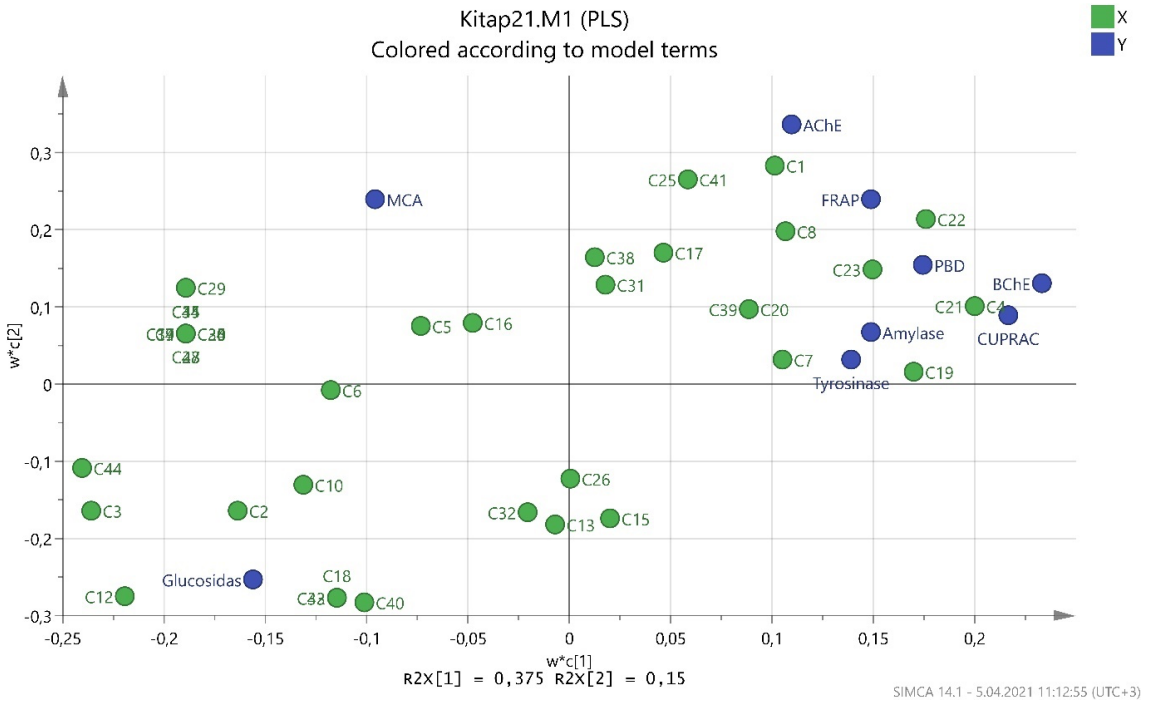
Line	AchE inhibition (mg GALAE*/g)	BchE inhibition (mg GALAE/g)	Tyrosinase inhibition (mg KAE**/g)	α-amylase inhibition (mmol ACAE***/g)	α-glucosidase inhibition (mmol ACAE/g)
APO41	0.76±**0.02**^{abc}	0.90±**0.01**^{ab}	5.18±**0.68**^{b}	0.50±**0.01**^{a}	na
APO42	0.65±**0.04**^{d}	0.76±**0.04**^{bc}	2.55±**0.34**^{c}	0.10±**0.01**^{e}	0.44±**0.01**^{a}
APO43	0.79±**0.01**^{ab}	0.88±**0.03**^{ab}	10.62±**1.63**^{a}	0.25±**0.01**^{c}	0.44±**0.01**^{a}
APO44	0.67±**0.01**^{cd}	0.65±**0.06**^{cd}	na	0.13±**0.01**^{d}	0.43±**0.01**^{a}
APO45	0.72±**0.04**^{bcd}	0.52±**0.07**^{bcd}	na	0.07±**0.01**^{f}	0.44±**0.01**^{a}
BOH3	0.77±**0.07**^{abc}	0.87±**0.10**^{abc}	6.81±**1.37**^{b}	0.39±**0.01**^{b}	na
H1733	0.83±**0.03**^{a}	0.95±**0.02**^{a}	na	0.05±**0.01**^{f}	na

* GALAEs, galanthamine equivalents.

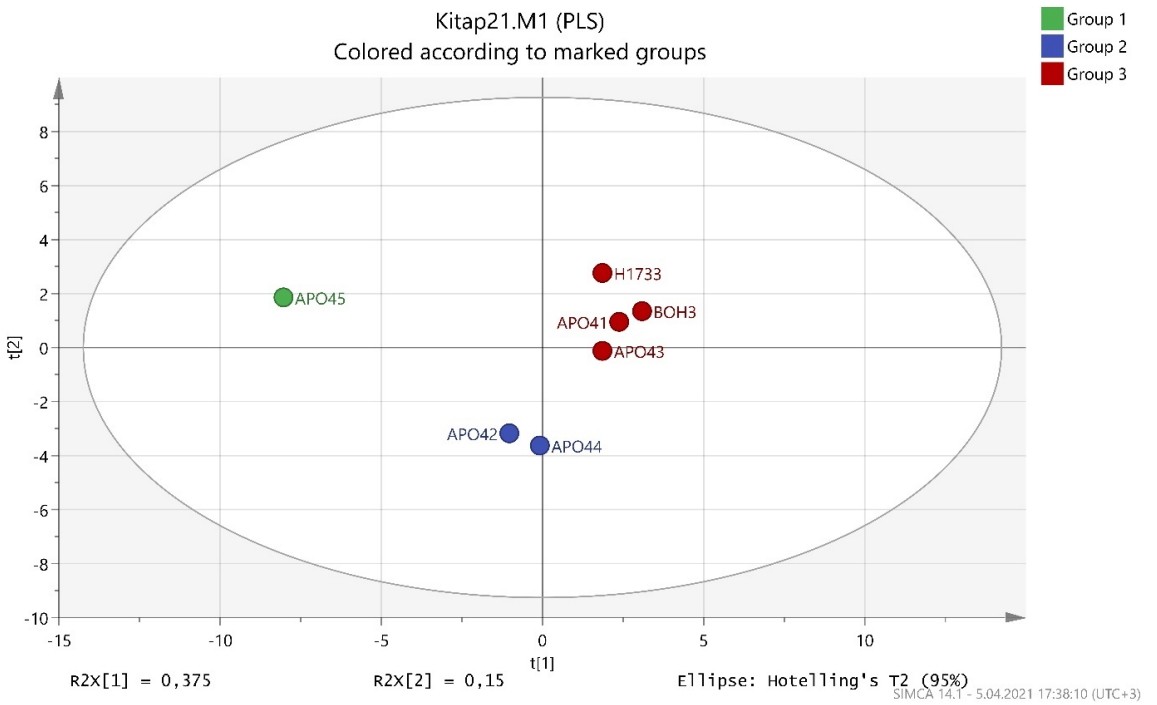
** KAEs, kojic acid equivalents.

*** ACEs, acarbose equivalents.

Different superscript letters in the same column indicate significant difference ($p < 0.05$); na, not active.



(a)



(b)

Figure 2. (a) Relationship between chemical compounds and biological activities through PLS analysis. (b) Cluster analysis based on chemical compounds and biological activities through PLS analysis (for the compound number see Table 2).

mg KAE/g). Other lines did not exert any inhibition against tyrosinase. It's worth to mention that this was the first detailed evaluation of the enzyme inhibitory capacity of the sunflower oil. By comparing these results with previous ones obtained from the seed methanolic extract of the same sunflower lines, it was clear that the oil exerted low enzyme inhibitory potential (Abdalla et al. 2021). Moreover, a previous study was performed on the acetone extract of the seed to evaluate its property as α-amylase and α-glucosidase inhibitors (Sonkamble et al., 2018). Their results showed that the highest α-amylase (60.42±0.6%) and α-glucosidase (83.22 ± 0.18 %) inhibitory activity was obtained at a concentration of 0.01 mg of acetone extract.

3.5. Statistical evaluation

To determine the connection between the tested oils, multivariate analysis was performed for the tested oils. Firstly, the possible relation between the chemical components in the tested oils and biological activities was investigated. Figure 2a showed the interpretation of the relationships between chemical components and biological activities. Clearly, most of the biological activities (reducing power, phosphomolybdenum, AChE, BChE, amylase, and tyrosinase) were closely linked to several compounds, especially linoleic acid. However, the metal chelating ability was not directly linked to the chemical components. In this sense, the observed metal chelating ability might be due to the presence of other chelator agents such as peptides or sulfides. As another activity, observed glucosidase inhibitory effects may be caused by several compounds such as palmitoleic acid, trans-2-dodecenoic acid, 1,19-eicosadiene, and phytol. However, these approaches must be confirmed by further studies on the biological activities of the above-mentioned compounds. Based on the chemical profile and biological activities of the tested oil, they were grouped in three clusters. One group included sunflower oil lines, namely H1733, APO41, BOH3, and APO43. APO42 and PO44 were classified as another group. The last group just included APO45 (Figure 2b).

4. Conclusion

In this study, oils of the seven new sunflower lines possessed physicochemical, chemical profile and antioxidant and enzyme inhibition that may be of interest for food and nonfood applications.

Lines APO42, APO43, and APO44 could be the best choice for food and nutrition applications due to their high oleic acid content and considerable antioxidant activity. Lines APO43 and BOH3 showed considerable enzyme inhibitory activity against tyrosinase enzymes suggesting their beneficial application in the cosmetic industry as a skin lighting agent. Lines APO41, APO42, APO43, and APO45 that had high saponification values could be suitable for soap and shampoos fabrication.

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References

Abdalla AAA, Yagi S, Abdallah AH, Abdalla M, Sinan KI et al. (2021). Phenolic profile, antioxidant and enzyme inhibition properties of seed methanolic extract of seven new sunflower lines: From fields to industrial applications. Process Biochemistry 111 (2): 53–61. doi: 10.1016/j.procbio.2021.10.014

Adeleke BS, Babalola OO (2020). Oilseed crop sunflower (*Helianthus annuus*) as a source of food: Nutritional and health benefits. Food Science and Nutrition 8 (9): 4666–4684. doi: 10.1002/fsn3.1783

AOCS. American Oil Chemists' Society. Official Methods and Recommended Practices of the AOCS. Champaign, Illinois, USA: AOCS Press; 2004.

Aremu BR, Omotayo AO, Babalola OO (2016). Biotechnology in agriculture: risks and opportunities for the rural poor in semi-arid-tropics. Journal of Human Ecology 56 (1-2): 55–59. doi:10.1080/09709274.2016.11907037

Bashir T, Mashwani ZUR, Zahara K, Haider S, Tabassum S et al(2015). Chemistry, pharmacology and ethnomedicinal uses of *Helianthus annuus* (sunflower): A Review. Pure and Applied Biology 4 (2): 226–235. doi:10.19045/bspab.2015.42011

Baydar H, Erbaş S (2005). Influence of seed development and seed position on oil, fatty acids and total tocopherol contents in sunflower (*Helianthus annuus* L.). Turkish Journal of Agriculture and Forestry 29 (3): 179–186

Belingheri C, Giussani B, Rodriguez-Estrada MT, Ferrillo A, Vittadini E (2015). Oxidative stability of high-oleic sunflower oil in a porous starch carrier. Food Chemistry 166: 346–351. doi: 10.1016/j.foodchem.2014.06.029

Carvalho MG, de Silva VC, Rocha Jr JG da (2009). A new derivative of dihydroochnaflavone isolated from *Luxemburgia* species (Ochnaceae) and the complete ^{1}H and ^{13}C NMR chemical shifts assignments. Revista Brasileira de Farmacognosia 19 (1a): 33–35. doi:10.1590/S0102-695X2009000100008

Chowdhury R, Warnakula S, Kunutsor S, Crowe F, Ward HA et al (2014). Association of dietary, circulating, and supplement fatty acids with coronary risk. Annals of Internal Medicine 160 (6): 398–406. doi: 10.7326/M13-1788

Eryilmaz T, Yesilyurt MK (2016). Influence of blending ratio on the physicochemical properties of safflower oil methyl ester-safflower oil, safflower oil methyl ester-diesel and safflower oil-diesel. Renewable Energy 95: 233–247. doi: 10.1016/j.renene.2016.04.009

FAO and WHO. (1993). Fats and oils in human nutrition. Report of a joint FAO/WHO expert consultation, Organized by the Food and Agriculture Organization of the United Nations and the World Health Organization Rome, Italy, 19-26 October 1993. pp.147,

Farvid MS, Ding M, Pan A, Sun Q, Chiuve SE et al (2014). Dietary linoleic acid and risk of coronary heart disease: a systematic review and meta-analysis of prospective cohort studies. Circulation 130 (18): 1568–1578. doi: 10.1161/CIRCULATIONAHA.114.010236

Fayyaz-ul-Hasan, Ahmad RA (2003). Effects of seasonal variations on oil and fatty acid profile of sunflower. Helia 26: 159–166.

Franco R, Iseppi L, Taverna M (2018). Sunflower oil functional properties for specialty food. Nutrition and Food Science International Journal 5(4): 555668. doi: 10.19080/NFSIJ.2018.05.555668

Giada MDLR, Mancini-Filho J (2009). Antioxidant capacity of the striped sunflower (*Helianthus annuus* L.) seed extracts evaluated by three in vitro methods. International Journal of Food Sciences and Nutrition 60 (5): 395–401. doi: 10.1080/09637480701755262

Guo S, Ge Y, Na Jom K (2017). A review of phytochemistry, metabolite changes, and medicinal uses of the common sunflower seed and sprouts (*Helianthus annuus* L.). Chemistry Central Journal 11 (1): 95. doi: 10.1186/s13065-017-0328-7

Hemmati N, Azizi M, Spina R, Dupire F, Arouei H et al (2020). Accumulation of ajmalicine and vinblastine in cell cultures is enhanced by endophytic fungi of *Catharanthus roseus* cv. Icy Pink. Industrial Crops and Products 158: 112776. doi: 10.1016/j.indcrop.2020.112776

Huth PJ, Fulgoni VL, Larson BT (2015). A systematic review of high-oleic vegetable oil substitutions for other fats and oils on cardiovascular disease risk factors: implications for novel high-oleic soybean oils. Advances in Nutrition 6 (6): 674–693. doi: 10.3945/an.115.008979

Ip C, Carter CA, Ip MM (1985). Requirement of essential fatty acid for mammary tumorigenesis in the rat. Cancer Research 45 (5): 1997–2001.

Liu KS (1994). Preparation of fatty acid methyl esters for gas-chromatographic analysis of lipids in biological materials. Journal of the American Oil Chemists' Society 71: 1179–1187. doi: 10.1007/BF02540534

Marmesat S, Morales A, Velasco J, Carmen Dobarganes M (2012). Influence of fatty acid composition on chemical changes in blends of sunflower oils during thermoxidation and frying. Food Chemistry 135 (4): 2333–2339. doi: 10.1016/j.foodchem.2012.06.128

Menendez JA, Vellon L, Colomer R, Lupu R (2005). Oleic acid, the main monounsaturated fatty acid of olive oil, suppresses Her-2/neu (erbB-2) expression and synergistically enhances the growth inhibitory effects of trastuzumab (Herceptin) in breast cancer cells with Her-2/neu oncogene amplification. Annals of Oncology 16 (3): 359–371. doi: 10.1093/annonc/mdi090

Merwe R, van der Labuschagne MT, Herselman L, Hugo A (2015). Effect of heat stress on seed yield components and oil composition in high- and mid-oleic sunflower hybrids. South African Journal of Plant and Soil 32 (3): 121–128. doi: 10.1080/02571862.2015.1018354

Mohamed MY (2010). Development and Stability of some Sudanese sunflower hybrids under irrigated conditions. Helia 33: 135–144. DOI: 10.2298/HEL1052135M

Mohammed ABA, Yagi S, Tzanova T, Schohn H, Abdelgadir H et al. (2020). Chemical profile, antiproliferative, antioxidant and enzyme inhibition activities of *Ocimum basilicum* L. and *Pulicaria undulata* (L.) C.A. Mey. grown in Sudan. South African Journal of Botany 132: 403–409. doi: 10.1016/j.sajb.2020.06.006

Nakić SN, Rade D, Škevin D, Štrucelj D, Mokrovčak Ž et al. (2006). Chemical characteristics of oils from naked and husk seeds of *Cucurbita pepo* L. European Journal of Lipid Science and Technology 108 (11): 936–943. doi: 10.1002/ejlt.200600161

Ohmori H, Fujii K, Kadochi Y, Mori S, Nishiguchi Y et al. (2017). Elaidic acid, a trans-fatty acid, enhances the metastasis of colorectal cancer cells. Pathobiology: Journal of Immunopathology, Molecular and Cellular Biology 84 (3): 144–151. doi: 10.1159/000449205

Oliveira V da S, Oliveira RL, Goes RHTB, Silva TM, Silva LF et al. (2019). Physicochemical composition, fatty acid profile and sensory attributes of the meat of young Nellore bulls fed sunflower cake from the biodiesel industry. Livestock Science 227: 97–104.

Porto WS, Carvalho CGPde, Pinto RJB, Oliveira MFde, Oliveira ACBde (2008). Evaluation of sunflower cultivars for central Brazil. Scientia Agricola 65 (2): 139–144. doi: 10.1590/S0103-90162008000200005

Premnath A, Narayana M, Ramakrishnan C, Kuppusamy S, Chockalingam V (2016). Mapping quantitative trait loci controlling oil content, oleic acid and linoleic acid content in sunflower (*Helianthus annuus* L.). Molecular Breeding 36: 106. doi: 10.1007/s11032-016-0527-2

Ramsden CE, Zamora D, Leelarthaepin B, Majchrzak-Hong SF, Faurot KR et al. (2013). Use of dietary linoleic acid for secondary prevention of coronary heart disease and death: evaluation of recovered data from the Sydney Diet Heart Study and updated meta-analysis. British Medical Journal 346: e8707. doi: 10.1136/bmj.e8707

Sadoudi R, Ammouche A, Ali AD (2014). Thermal oxidative alteration of sunflower oil. African Journal of. Food Science 8 (3): 116–121. doi: 10.5897/AJFS12.112

Sauer LA, Blask DE, Dauchy RT (2007). Dietary factors and growth and metabolism in experimental tumors. Journal of Nutritional Biochemistry 18 (10): 637–649. doi: 10.1016/j.jnutbio.2006

Sergio L, Boari F, Pieralice M, Linsalata V, Cantore V et al . (2020). Bioactive phenolics and antioxidant capacity of some edible wild greens as affected by different cooking treatments. Foods 9 (9): 1320. doi: 10.3390/foods9091320

Simonsen NR, Fernandez-Crehuet Navajas J, Martin-Moreno JM, Strain JJ, Huttunen JK et al. (1998). Tissue stores of individual monounsaturated fatty acids and breast cancer: the EURAMIC study. American Journal of Clinical Nutrition 68 (1): 134–141. doi: 10.1093/ajcn/68.1.134

Smith LF, Patterson J, Walker LT, Verghese M (2015). Chemopreventive potential of sunflower seeds in a human colon cancer cell line. International Journal of Cancer Research 12 (1): 40–50. doi:10.3923/ijcr.2016.40.50

Sonkamble VV, Wagh NS, Kamble LH (2018). Inhibition of α-amylase and α-glucosidase by (6RS)-22-hydroxy-23,24,25,26,27-pentanor-vitamin-D3-6,19-sulfur dioxide-adduct, manoalide and 5β-cholestane-3α,7α,12α,24,25,26-hexol isolated from acetone extract of *Helianthus annuus* L. seeds. International Journal of Pharmacy and Pharmaceutical Sciences 10 (5): 39–49. doi : 10.22159/ijpps.2018v10i5.25309

Sun Q, Ma J, Campos H, Hankinson SE, Manson JE et al. (2007). A prospective study of trans fatty acids in erythrocytes and risk of coronary heart disease. Circulation 115(14): 1858–1865. doi: 10.1161/CIRCULATIONAHA.106.679985

Tabasum S, Asghar S, Ashraf SN, Ahmad HB, Akhtar N et al. (2012). Physicochemical characterization and frying quality of canola and sunflower oil samples. Journal of the Chemical Society of Pakistan 34 (3):513-517.

Vannice G, Rasmussen H (2014). Position of the academy of nutrition and dietetics: dietary fatty acids for healthy adults. Journal of the Academy of Nutrition and Dietetics 114 (1): 136–153. doi: 10.1016/j.jand.2013.11.001

Vijayakumar M, Vasudevan DM, Sundaram KR, Krishnan S, Vaidyanathan K et al (2016). A randomized study of coconut oil versus sunflower oil on cardiovascular risk factors in patients with stable coronary heart disease. Indian Heart Journal 68 (4): 498–506. doi: 10.1016/j.ihj.2015.10.384

WHO/FAO (1993). World Health Organization & Food and Agriculture Organization of the United Nations. (‽ 1993)‽ . Fats and oils in human nutrition: report of a joint expert consultation, Rome, Italy.

Zengin G, Sarikurkcu C, Gunes E, Uysal A, Ceylan R et al. (2015). Two *Ganoderma* species: profiling of phenolic compounds by HPLC-DAD, antioxidant, antimicrobial and inhibitory activities on key enzymes linked to diabetes mellitus, Alzheimer's disease and skin disorders. Food and Function 6: 2794–2802. doi: 10.1039/c5fo00665a

Zock PL, Katan MB (1998). Linoleic acid intake and cancer risk: a review and meta-analysis. American Journal of Clinical Nutrition 68: 142–153. doi: 10.1093/ajcn/68.1.142