Oxidative Stability of Flax and Hemp Oils

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ABSTRACT: Oxidative stability of flax and hemp oils, and of flax and hemp oils stripped of their minor components, was evaluated in the dark at 60°C and under fluorescent light at 27°C. Several analytical methods were used to assess the oxidative stability of oils. Oil extracts were also investigated for their scavenging of 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical and for their total phenolic contents. The results indicate that bioactive constituents of these edible oils play a major role in their oxidative stability. However, the FA composition of the oils and their total content of tocopherols as well as the type of pigments present contribute to their stability. Nonstripped flax and hemp oils were more stable than their corresponding stripped counterparts. Furthermore, nonstripped hemp oil had a higher oxidative stability than nonstripped flax oil as evidenced by scavenging of DPPH radical and consideration of total phenolic contents.

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KEY WORDS: Conjugated dienes, 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical, flax oil, headspace analysis, hemp oil, high-performance liquid chromatography, oxidative stability, photo-oxidation, thiobarbituric acid-reactive substances (TBARS), tocopherol content.

The oils from seeds of flax (*Linum usitatissimum* L.) and hemp (*Cannabis sativa*) are rich in α-linolenic acid (ALA) and linoleic acid (LA). Moreover, hemp oil has a reasonable amount of γ-linolenic acid (GLA). The stability of oils depends on various factors, but mainly on the oil's FA composition, the content of natural antioxidants, and the presence of oxygen, as well as different storage and packaging conditions (1). Edible oils, in general, consist mainly of TAG (95%). Non-TAG or unsaponifiable matter makes up the remaining 5%. These minor components are naturally occurring compounds with antioxidative properties that help protect oils against oxidative deterioration and hence play an important role in their oxidative stability (OS) (2,3). The minor components of vegetable oils are primarily phospholipids, tocols, phenolic compounds, pigments (carotenoids, chlorophylls), sterols, and FFA, as well as MAG and DAG (2). Several classes of these components might be present in each oil and contribute to its OS (2).

A number of techniques, including determination of primary and secondary oxidation products as well as sensory analysis, are commonly used to monitor oxidation of foods and predict their shelf life (4,5). PV and conjugated dienes (CD) are often used to measure primary oxidation products, i.e., hydroperoxides, whereas TBARS and headspace volatiles are among methods used for monitoring secondary products of oxidation (6).

Little is known about the OS of flax and hemp oils, and virtually nothing is known about the OS of these oils when stripped of their minor components. It is generally accepted that the OS of edible oils is dictated by their degree of unsaturation and the content and profile of their minor components; higher OS of oils means the presence of a higher level of natural free radical scavengers and phenolic compounds.

To examine these hypotheses regarding the OS of flax and hemp oils, this research was carried out using both native nonstripped oils and their stripped counterparts. The minor components in nonstripped and stripped oils, such as pigments (carotenoids and chlorophylls) and antioxidants (tocopherols and phenolics), were also examined. Finally, free radical-scavenging properties of extracts from flax and hemp oils were tested.

MATERIALS AND METHODS

Materials. Cold-pressed flax oil was obtained from Herbal Select (Guelph, Ontario, Canada). Cold-pressed hemp oil was obtained from Hemp Oil Canada Inc. (Ste. Agathe, Manitoba, Canada). 1,1-Diphenyl-2-picrylhydrazyl (DPPH), Folin-Ciocalteu reagent, 2-thiobarbituric acid (2-TBA), gallic acid (3,4,5-trihydroxybenzoic acid), silicic acid powder (mesh size: 100–200, acid-washed), and α-tocopherol were obtained from Sigma Chemical Co. (St. Louis, MO). Activated charcoal was acquired from BDH Inc. (Toronto, Ontario, Canada). Compressed air, hydrogen, and ultra-high-purity (UHP) helium were obtained from Canadian Liquid Air Ltd. (St. John's, Newfoundland, Canada). Hexane, methanol, sulfuric acid, iso-octane, and 1-butanol were purchased from Fisher Scientific Company (Nepean, Ontario, Canada).

Methods. (i) Preparation oils stripped of their minor components. Flax and hemp oils were stripped of their minor components essentially according to the method of Khan and Shahidi (7) with minor modifications. A chromatographic column (3.4 cm i.d. × 40 cm height) was connected to an aspirator vacuum pump and packed sequentially with two adsorbents. The first layer in the column consisted of 50 g of activated silicic acid, the second layer was 50 g of activated charcoal, and the top layer was another 50 g of activated silicic acid. All adsorbents were suspended in *n*-hexane. Before introduction of the solvent, the silicic acid (100 g) was activated as described by Min (8) by washing three times with a total of 3 L of distilled water. After each treatment, the silicic acid was left to set-

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tle for 30 min, then the suspended silicic acid was decanted. Finally, the silicic acid was washed with methanol and the supernatant decanted. The remaining methanol was removed by filtering through a Büchner funnel under vacuum, and the semidried material was activated at 200°C for 22 h.

Oil (50 g) was diluted with an equal volume of *n*-hexane and passed through the chromatographic column. The solvent in the eluent (stripped oil) was evaporated under vacuum at 30°C, and traces of the solvent were removed by flushing with nitrogen. Stripped oils (stripped hemp oil, SHO; stripped flax oil, SFO) were obtained and transferred into 10-mL bottles, flushed with nitrogen, and kept at –70°C for subsequent studies.

(ii) Preparation of samples for accelerated oxidation tests. Stripped and nonstripped oil samples (0.5 g in 2-mL vials) were used to study their OS in the dark on heating, and in the light (photo-oxidation). For accelerated oxidation at 60°C, the sample containers were placed in a forced air oven (Model 2; Precision Scientific Co., Chicago, IL). For photo-oxidation studies the samples were placed in a box (70 cm length × 35 cm width × 25 cm height) equipped with two 40-W cool-white fluorescent lights that were suspended approximately 10 cm above the surface of the oil containers. The remaining open space was covered with aluminum foil. The fluorescent radiation was at a level of 2650 lux, and the temperature inside the container was 27 ± 1°C. Oil samples were removed from the oven after 1, 3, 5, and 7 d, and from the light box after 4, 8, 12, and 24 h, flushed with nitrogen, covered with Parafilm (American Can Co., Greenwich, CT) and kept at –70°C for OS tests.

(iii) OS tests. The OS of stripped and nonstripped oils was evaluated by determining CD, TBARS, and headspace volatiles.

(iv) Determination of CD. CD contents of the oil samples were determined by the IUPAC method (9). Oil samples (0.02–0.04 g) were weighed into 25-mL volumetric flasks, dissolved in iso-octane (2,2,4-trimethylpentane), and made up to the mark with the same solvent. The contents were mixed thoroughly and the absorbance was read at 234 nm in a 10-mm Hellma quartz cell using a Hewlett-Packard 8452A diode array spectrophotometer (Agilent, Palo Alto, CA). Pure iso-octane was used as the blank. CD were calculated according to the following equation: $CD = A/(C \times d)$, where A = absorbance of the solution at 234 nm, C = concentration of the solution in g/100 mL solution, and d = length of the cell in cm.

(v) Determination of TBARS. Oil samples (0.05–0.20 g) were analyzed for their contents of TBARS according to the AOCS (10) method. The samples were accurately weighed into 25-mL volumetric flasks, dissolved in a small volume of 1-butanol, and made up to the mark with the same solvent. A 5.0-mL portion of this mixture was transferred into a dry test tube, then fresh 2-TBA reagent (5 mL of a solution of 200 mg 2-TBA in 100 mL 1-butanol) were added to it. The contents were mixed and heated in a water bath at 95°C for 2 h. The intensity of the resultant colored complex was measured at 532 nm using a Hewlett-Packard 8452A diode array spectrophotometer. The TBARS values were calculated by multiplying the absorbance readings by a factor of 0.415, determined from a standard line prepared using 1,1,3,3- tetramethoxypropane as a precursor of malonaldehyde (MA) (*7*).

(v) Headspace analysis of volatiles. A PerkinElmer 8500 gas chromatograph equipped with an HS-6 headspace sampler (PerkinElmer Co., Montréal, Canada) was used for the analysis of the volatiles contents in oil samples (11). The column used to separate the volatiles was a SUPELCOWAX-10 fused-silica capillary (30 m × 0.32 mm i.d., 0.10 μm film: Supelco Canada Ltd., Mississauga, Ontario, Canada). Helium (UHP) was the carrier gas, used at an inlet column pressure of 20 psig with a split ratio of 7:1. The injector and FID temperatures were 280°C. The oven temperature was maintained at 40°C for 5 min, then increased to 115°C at 10°C/min, and subsequently ramped to 200°C at 30°C/min and held there for 5 min.

Oil samples (0.2 g) were transferred into glass vials that were capped with butyl septa, crimped, and analyzed. Vials were preheated in the HS-6 magazine assembly at 90°C for a 45-min equilibrium period. Pressurization time was 6 s, and the volume of the vapor phase drawn was approximately 1.5 mL. Individual volatile compounds were tentatively identified by comparing the relative retention times of GC peaks with those of commercially available standards. Quantitative determination of the dominant aldehyde, hexanal, and/or propanal was accomplished using 1% 2-heptanone (in stripped corn oil) as an internal standard. Formation of volatiles was monitored as a measure of oxidation of oil samples.

(vi) Preparation of antioxidant extract. A measured amount of oil sample (20 g) was diluted with hexane (1:10, wt/vol) and extracted three times with methanol (10:2, vol/vol, hexane/methanol) at room temperature. The methanol extract was washed with hexane (1:1, vol/vol), and the methanol was completely removed under vacuum. The extract (2 g) was redissolved in 10 mL of methanol, flushed with nitrogen, and kept at –70°C for further analysis.

(vii) Determination of total phenolic content. The total phenolic content was determined following the procedure of Singleton and Rossi (12) with minor modifications. One milliliter of Folin-Ciocalteu reagent was added to 50-mL centrifuge tubes containing 1 mL of the extracts (0.2 g/mL). Contents were mixed thoroughly and 20% (wt/vol) sodium carbonate (1.5 mL) was added. The final volume was made up to 10 mL with distilled water and mixed again. The absorbance of the mixture after standing for 2 h at room temperature was measured at 765 nm and used to calculate the phenolic contents using a standard curve prepared with gallic acid. Total extracted phenolics were expressed as mg of gallic acid equivalents (GAE) per mL of extract.

(viii) DPPH radical-scavenging activity. One milliliter of freshly prepared DPPH radical solution (0.125 mM) was added to 1 mL of the extract (0.2 g/mL) prepared as just described and mixed well to start the radical-antioxidant reaction. The absorbance at 517 nm was determined against a blank of pure methanol after 0, 2, 4, 6, 8, 10, 15, and 20 min of reaction and used to estimate the remaining radical levels according to the standard curve. The reference antioxidant

used was α-tocopherol. The percent inhibition was calculated according to Lee *et al.* (13) using the following equation:

$$\% \text{ inhibition} = [(\text{absorbance of control} - \text{absorbance of test sample})/\text{absorbance of control}] \times 100 \quad [1]$$

Chemical and instrumental analysis. (i) Analysis of FA composition. FA composition of the oils, as their FAME, was determined according to the method described by Wanasundara and Shahidi (14) by using a Hewlett-Packard 5890 II gas chromatograph (Agilent) equipped with a 30 m × 0.25 µm, 0.25 µm film thickness SUPELCOWAX-10 column (SP 2330; Supelco Canada Ltd., Oakville, Ontario, Canada). The injector and FID temperatures were both set at 270°C. The oven temperature was initially 220°C for 10.25 min and then increased to 240°C at 30°C/min and held there for 9 min. Helium (UHP) was used as the carrier gas. The FAME were identified by comparing their retention times with those of an authentic standard mixture (GLC-461; Nu-Chek-Prep, Elysian, MN). Results were presented as weight percentages.

(ii) Measurement of pigments. Pigments present in the stripped and nonstripped oil samples were determined qualitatively by measuring the absorbance at 430–460 nm for carotenoids and 550–710 for chlorophylls and their derivatives (15). The oil sample was mixed with hexane [1:12 (vol/vol) for hemp oil and 1:5 (vol/vol) for flax oil] and transferred into Hellma glass cells; the absorbance was read using Spectronic Genesys™ 2 and Genesys™ 5 spectrophotometers and by recording the absorption spectra between 430 and 710 nm.

(iii) Determination of tocopherols by HPLC. For γ- and δ-tocopherols analysis, a Shimadzu (Kyoto, Japan) high-performance liquid chromatograph (HPLC) equipped with two LC-10AD pumps, an SPD-M10A diode array detector, and an SCL AA system controller was used. The separation conditions were as follows: pre-packed Luna Silica(2) column (25 cm, 4.6 mm in diameter, 5 µm particles; Phenomenex Aschaffenburg, Germany); mobile phase 4% dioxane in hexane, flow rate 1.5 mL/ min, injection volume 20 µL and the detector was set at 295 nm. A Shimadzu (HPLC) system was used for α-tocopherol analysis (LC 10AD pumps, RF-535 fluorescence detector, C-R4A Chromatopac). The conditions of separation were as follows: pre-packed Luna Silica(2) column (25 cm × 4.6 mm in diameter, 5 µm particles; Phenomenex), mobile phase 0.5% isopropanol in hexane, flow rate 1mL/min; injection volume 20 µL. The detector was set for excitation at 290 nm and emission at 330 nm. Stripped and nonstripped oil samples (1.0 g) were dissolved in 10 mL of mobile phase, passed through 0.45 µm filters, and injected onto the HPLC column (Hoffmann-La Roche Ltd., Basel, Switzerland).

Statistical analysis. All the experiments were performed in triplicate and the results reported as means ± SD. Normality was examined using SigmaStat for Windows Version 2.0 (Jandel Corporation, San Rafael, CA). ANOVA and Tukey's standardized test were performed at a level of $P < 0.05$ to assess the significance of differences among mean values.

RESULTS AND DISCUSSION

Analysis of nonstripped and stripped oil samples. (i) Chemical characteristics of nonstripped and stripped flax and hemp oils. The chemical characteristics of nonstripped and stripped flax and hemp oils are summarized in Table 1. The original nonstripped hemp oil (NHO) contained more ($P < 0.05$) primary oxidation products than the original nonstripped flax oil (NFO). Similarly, the original samples of NHO and NFO contained more ($P < 0.05$) primary and secondary oxidation products than their stripped counterparts, i.e., stripped flax oil (SFO) and

TABLE 1
Chemical Characteristics of Nonstripped and Stripped Flax and Hemp Oils[a]

Characteristics	NFO	SFO	NHO	SHO
Oxidative status				
Conjugated dienes	1.65 ± 0.00^{c}	1.09 ± 0.05^{a,b}	1.95 ± 0.15^{d}	1.09 ± 0.02^{a,b}
TBARS (µmol/g)	6.01 ± 0.04^{c,d}	4.54 ± 0.11^{b}	6.14 ± 0.05^{c,d}	3.02 ± 0.04^{a}
Tocopherols (mg/kg)				
α	40^{a}	0	0	0
γ	800^{a}	0	900^{b}	0
δ	0	0	70^{a}	0
Total	840^{a}	0	970^{b}	0
Pigments[b,c] (absorbance)				
430 nm	0.94 ± 0.00^{c}	0.024 ± 0.00^{b}	0.67 ± 0.00^{d}	0.01 ± 0.00^{a}
460 nm	0.81 ± 0.00^{d}	0^{a,b}	0.35 ± 0.00^{c}	0^{a,b}
550 nm	0.14 ± 0.00^{d}	0^{a,b}	0.06 ± 0.00^{c}	0^{a,b}
620 nm	0.11 ± 0.00^{d}	0$^{a.b}$	0.07 ± 0.00^{c}	0^{a,b}
670 nm	0.11 ± 0.00^{c}	0^{a,b}	0.37 ± 0.01^{d}	0^{a,b}

[a]Values are means of three determinations ± SD. Values in each row with different superscripts are different ($P < 0.05$) from one another. NFO, nonstripped flax oil; NHO, nonstripped hemp oil; SFO, stripped flax oil; SHO, stripped hemp oil.
[b]The oil/hexane (vol:vol) ratio was 1:5 for flax oil and 1:12 for hemp oil.
[c]Absorbance between 430 and 460 nm indicates the presence of carotenoids, and between 550 and 710 nm indicates the presence of chlorophylls.

TABLE 2
FA Composition (Area Percentage) of Nonstripped and Stripped Flax and Hemp Oils[a]

FA	NFO	SFO	NHO	SHO
C16:0	4.63 ± 0.02^{a,b}	4.72 ± 0.03^{a,b}	5.42 ± 0.03^{c}	6.31 ± 0.12^{d}
C18:0	4.16 ± 0.05^{c,d}	4.40 ± 0.09^{c,d}	2.58 ± 0.14^{a}	3.36 ± 0.09^{b}
C18:1n-9	20.3 ± 0.11^{c,d}	21.1 ± 0.17^{c,d}	9.19 ± 0.88^{a}	10.9 ± 0.24^{b}
C18:2n-6	15.3 ± 0.03^{a,b}	15.2 ± 0.03^{a,b}	52.1 ± 0.41^{c}	53.8 ± 0.24^{d}
C18:3n-6	—	—	3.37 ± 0.03^{a}	2.71 ± 0.07^{b}
C18:3n-3	54.13 ± 0.18^{d}	53.1 ± 0.29^{c}	23.3 ± 0.45^{b}	18.2 ± 0.42^{a}
Others	1.55	1.5	4.04	4.72
Total PUFA	69.4 ± 0.17$^{a\,b}$	68.3 ± 0.18^{ab}	78.8 ± 0.93^{d}	74.7 ± 0.72^{c}

[a]Values are means of three determinations ± SD. Values with different superscripts in each row are different ($P < 0.05$) from one another. For abbreviations see Table 1.

stripped hemp oil (SHO). Similarly, Khan and Shahidi (7) found that stripped borage and evening primrose oils had good oxidative status compared with their nonstripped counterparts.

The main tocopherols in NFO, as determined by HPLC, were α-tocopherol (40 ppm) and γ-tocopherol (800 ppm); no d-tocopherol was detected. However, NHO contained 900 ppm of γ-tocopherol and 70 ppm of δ-tocopherols as shown in Table 1. Thus, NHO had higher amounts ($P < 0.05$) of total tocopherols than NFO, which might contribute to its greater OS.

Pigments such as carotenoids, which absorb at 430–460 nm (15), were present in high amounts in NFO and NHO (Table 1). Meanwhile, chlorophylls, which absorb light at 550–710 nm (15), were present at a significantly ($P < 0.05$) higher quantity in NHO than NFO (Table 1). On the other hand, NFO and NHO contained more ($P < 0.05$) carotenoids and chlorophylls than their stripped counterparts, SFO and SHO. SFO and SHO retained traces of carotenoids (Table 1), which might interfere with their stability under light.

Hemp oil was stripped of its minor components more effectively than flax oil by using a modified multilayer column chromatographic technique developed by Lampi *et al.* (16). This procedure required only 2 h to strip 50 g of flax or hemp oil. Lower contents of CD and pigments were detected in SHO compared with NHO. However, more secondary oxidation products were retained or produced in SFO than NFO. This might be due to oxidation during the stripping process.

(ii) FA composition of NFO, SFO, NHO, and SHO, The FA composition of nonstripped and stripped flax and hemp oils is given in Table 2. The results in this table indicate that NHO and SHO contained higher amounts ($P < 0.05$) of PUFA than NFO and SFO. The main PUFA in NHO and SHO was LA, which was present at >50%, but this acid was present at only 15% in NFO and SFO. NHO also contained up to 3% GLA. NFO had up to 54% ALA, whereas hemp oil had only 23% ALA. The results of this work are similar to those of Moes *et al.* (17), who found that hempseed oil samples contained about 54–57.7% LA , 1.2–3.8% GLA, and 15.1–17.9% ALA.

OS of NFO, SFO, NHO, and SHO stored under Schaal oven conditions at 60°C. (i) Primary oxidation products. Based on CD values obtained during oxidation in the dark (Fig. 1a), NFO and NHO were more ($P < 0.05$) stable than the corresponding SFO and SHO. However, NFO and NHO were also highly

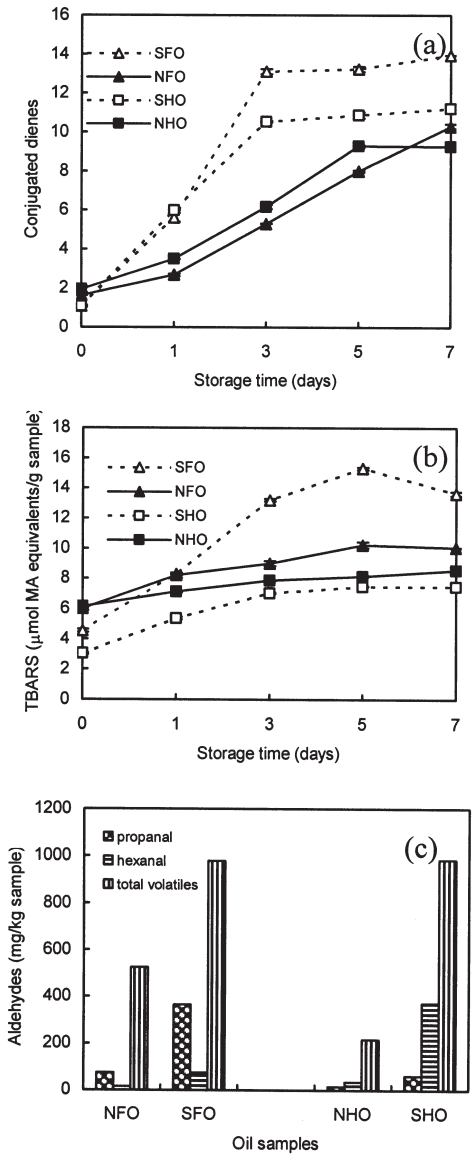

FIG 1. (a) Conjugated diene values; (b) TBARS; and (c) propanal, hexanal, and total volatiles of nonstripped and stripped flax and hemp oils stored under Schaal oven conditions at 60°C. SFO, stripped flax oil; NFO, nonstripped flax oil; SHP, stripped hemp oil; NHO, nonstripped hemp oil; MA, malonaldehyde.

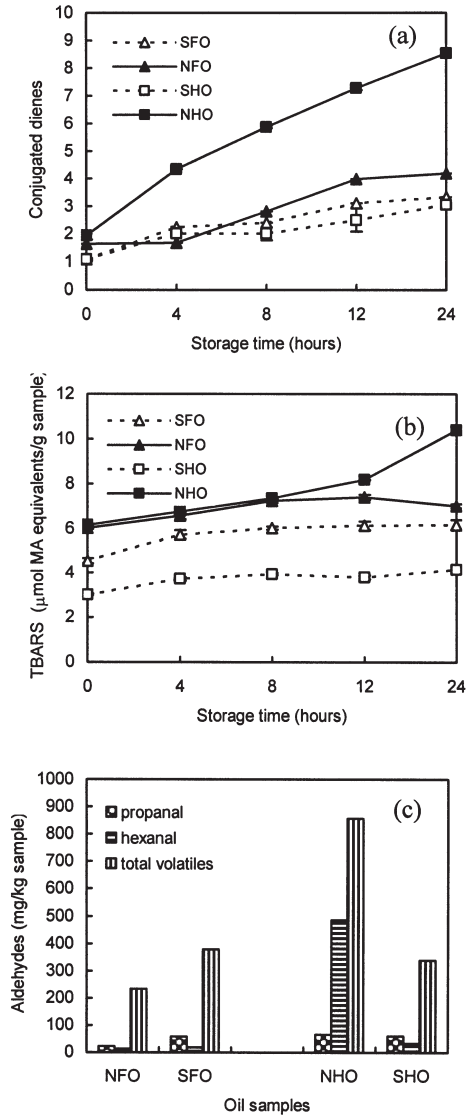

FIG. 2. (a) Conjugated dienes values; (b)TBARS; and (c) propanal, hexanal, and total volatiles of NFO, NFO, SFO, and NFO stored under fluorescent light at 27°C.

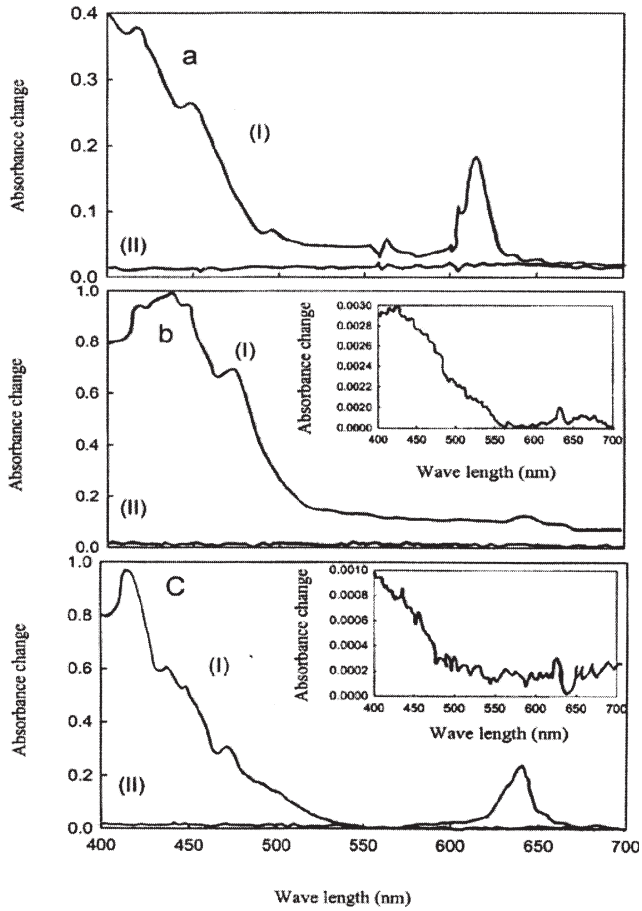

FIG. 3. Visible spectra in oil/hexane of pigments of nonstripped (I) and stripped (II) olive (a), flax (b), and hemp (c) oils. Conditions for oil/hexane: olive oil (1:1, vol/vol); flax oil (1:5, vol/vol); and hemp oil (1:12, vol/vol). Insets in panels (b) and (c) represent magnified spectra of stripped oils.

prone to oxidation. Flax oil is traditionally known as a drying oil owing to its high content of ALA. Hemp oil is also known to be highly unstable—despite the presence of different minor components that play a significant role in OS—because of its content of ALA and GLA, which are well known to be readily oxidized during storage and heating (18).

(ii) Secondary oxidation products. Secondary oxidation products of NFO, SFO, NHO, and SHO were determined by examining TBARS and headspace volatiles, mainly hexanal and propanal. Determination of TBARS is based on color intensity of the reaction between TBA and secondary oxidation products of PUFA, including MA. The TBARS values, expressed as mmol MA equiv per g of NFO and NHO, were lower than those of their corresponding stripped counterparts (Fig. 1b), in part owing to the presence of minor components such as tocopherols in the original oils. A sharp increase in TBARS values was noticed for SFO for the first 5 d, followed by a decrease. This might be due to the volatilization of secondary oxidation products or their further breakdown. Meanwhile, the difference between day seven and day zero of oxidation in the TBARS values for NHO was less than that for SHO, hence the oxidation of SHO was higher than that of NHO. TBARS values of SHO increased during the first 3 d, and then remained constant up to 7 d. The increasing trend in oxidation of SFO and SHO, as reflected in TBARS values, compared with NFO and NHO, was similar to that obtained for primary oxidation products as reflected in CD values (Fig. 1a). However, NHO was relatively more stable than NFO, as shown in Figure 1b. This might be due to a higher ($P < 0.05$) total amount of tocopherols (Table 1) in NHO than that found in NFO, in addition to possible effects due to FA composition of the oil. NFO contained almost double the amount of ALA, which is highly susceptible to oxidation.

The major volatile observed during autoxidation of NFO and SFO (Fig. 1c) was propanal, which is an oxidation product of ALA. Meanwhile, hexanal (Fig. 1c) was the major volatile observed during the oxidation of NHO and SHO. Hexanal is the major volatile produced from the oxidation of n-6 PUFA

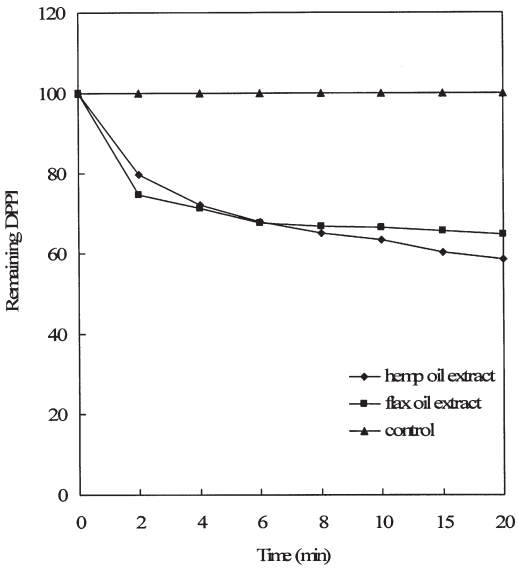

FIG. 4. Comparison of 1,1-diphenyl-2-picrylhydrazyl radical-scavenging activity of flax and hemp oil extracts after 20 min.

such as LA (11). NFO and NHO were more stable ($P < 0.05$) than their corresponding stripped counterparts. This can be explained, in part, by the presence of tocopherols in both oils.

OS of NFO, SFO, NHO, and SHO under fluorescent light at 27°C. (i) Primary oxidation products. The CD values obtained during the photooxidation of NFO, SFO, NHO, and SHO are presented in Figure 2a. Photooxidation of NHO, in the initial stages, progressed rapidly compared with NFO. Thereafter, the oxidation, as demonstrated by CD values, progressed gradually for NFO. Meanwhile, the CD values for SFO and SHO were lower ($P < 0.05$) than those of NFP and NHO. Therefore, NFO and NHO are more ($P < 0.05$) photo-oxidizable than their corresponding SFO and SHO. This may be explained by considering the presence of pigments, mainly chlorophylls, in NHO and carotenoids in NFO. The visible spectra of pigments in nonstripped and stripped olive oil (19) and NFO, NHO, SFO, and SHO are presented in Figure 3. The characteristic visible absorption peaks occurring below 500 nm correspond to carotenoids, whereas absorptions at 605, 647, and 667 nm correspond to different types of chlorophylls. These peaks qualitatively confirm the presence of chlorophylls in NHO, and lower levels of chlorophyll in NFO. Edible oils containing natural pigments such as chlorophylls and pheophytin reportedly are highly susceptible to light-induced oxidation or photo-oxidation (13,20). The role of photosensitizers in light-induced oxidation has also been observed in nonstripped borage and evening primrose oils (19). Meanwhile, the CD values of NHO were higher ($P < 0.05$) than those of NFO. This may be due to the presence of higher levels of cholorophylls in NHO compared with those in NFO. Chlorophylls and carotenoids were effectively removed from SHO (Fig. 3) and therefore, the oxidation was not affected by fluorescent light, as in SFO (Fig. 3). Traces of carotenoids might be retained in SFO and act as prooxidants.

(ii) Secondary oxidation products. Figure 2b displays TBARS values of NFO, NHO, SFO, and SHO stored under fluorescent light at 27°C for 24 h. The TBARS of NFO and NHO were higher ($P < 0.05$) than those of their stripped counterparts. However, TBARS values of NHO were higher ($P < 0.05$) than those of NFO. This might be due to higher contents of photosensitizers or other unknown factors in NHO as compared with NFO.

The primary hydroperoxides generated in photo-oxidation of unsaturated FA are decomposed to volatiles, which in turn have detrimental effects on flavor stability of edible oils (21). The major volatile detected in photo-oxidized SFO and NFO (Fig. 2c) was propanal, and that obtained from the oxidation of hemp oil under fluorescent light was hexanal (Fig. 2c). Hexanal may be formed *via* photooxidation of n-6 FA, namely LA and GLA. Meanwhile, propanal may be formed *via* photo-oxidation of ALA. SFO had a higher ($P < 0.05$) content of propanal, as it was more susceptible to oxidation than NFO (Fig. 2c). Meanwhile, the hexanal content of NHO was higher ($P < 0.05$) than that of its counterpart, SHO (Fig. 2c).

DPPH radical-scavenging activity of flax and hemp oil methanolic extracts. DPPH radical was used to evaluate free radical-scavenging properties of flax and hemp oil extracts, mainly their minor components (Fig. 4). Hemp oil extract, after 20 min of reaction with DPPH radical, exhibited a greater ($P < 0.05$) DPPH radical-scavenging activity than that observed for flax oil extract. This was indicated by the least amount of DPPH radical remaining after 20 min. The more DPPH remaining after reaction of free radical with antioxidant extracts, the lower the antioxidant capacity was. Similar kinetics were detected in antioxidant-DPPH radical reaction for the two oil extracts examined (Fig. 4). Meanwhile, depending on the standard curve prepared to calculate the tocopherol equivalents, hemp oil extract had a higher ($P < 0.05$) capacity, expressed as α-tocopherol equivalents (mM), than flax oil extract.

Determination of total phenolic contents of oil samples. Phenolic compounds are well known to contribute to the overall antioxidant capacity of oils. Thus, phenolics have a great effect on the stability, sensory, and nutritional characteristics of oil samples and might prevent their deterioration through quenching of radical reactions responsible for lipid oxidation (22). It has been reported that oil stability is correlated not only with the total amount of phenolics but also with the type of phenolics present (22).

Total phenolics of flax and hemp oils, obtained by methanol extraction, were evaluated as GAE. The levels of total phenolics determined in this way are not absolute measurements of the amounts of phenolic materials but are in fact based on the reducing capacity of gallic acid. Data demonstrated that extracts of flax and hemp oils had nearly the same ability to reduce Folin-Ciocalteu's reagent (data not shown). The presence of these phenols might contribute to better stability of NFO and NHO under Schaal oven conditions as compared with their stripped counterparts.

Thus, OS of hemp and flax oils was influenced to a large degree by their minor constituents, both antioxidative and prooxidative. Therefore, overprocessing of oils should either be

avoided or desirable minor components and antioxidant constituents should be added to the processed oils in order to ensure their adequate stability. Storage of oils in appropriate containers is also recommended in order to control photo-oxidative deterioration of hemp and flax oils.

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