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The Composition of Crude Corn Oil Recovered after Fermentation via Centrifugation from a Commercial Dry Grind Ethanol Process

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Abstract A study was conducted to examine the chemical composition of corn oil obtained after fermentation of corn to make fuel ethanol via centrifugation and compare its composition to that of corn germ oil (commercial corn oil) and experimental corn oils. The levels of free fatty acids in the post fermentation corn oil were high (11–16%), as previously reported. The levels of free phytosterols and hydroxycinnamate steryl esters (similar to oryzanol in rice bran oil) were higher than those of corn germ oil and were comparable to those of ethanol-extracted corn kernel oil. The levels of tocopherols were lower in post-fermentation oil than in either corn germ oil or ethanol extracted corn kernel oil. The levels of lutein and zeaxanthin in post-fermentation were much higher than those in corn germ oil and were comparable to those in ethanol-extracted corn kernel oil. Overall, exposure to all upstream processes of a fuel ethanol plant, including high-temperature liquefaction, saccharification and fermentation appeared to have the most notable effect on tocopherols, but it had little effect on the levels of free phytosterols, hydroxycinnamate steryl esters, lutein and zeaxanthin. It may be desirable to recover these valuable functional lipids prior to using the post-fermentation corn oil for industrial applications such as making biodiesel if a cost-effective recovery process can be developed.

Keywords Corn · *Zea mays* · Carotenoids · Fuel ethanol · Tocopherols · Tocotrienols · Vitamin E · Phytosterols · Plant sterols · Coproducts

Introduction

Most commercial corn oil is produced by either pressing corn germ (a fraction derived from the kernel, consisting mainly of embryos) or extraction with hexane or a combination of pre-pressing and hexane extraction, followed by refining, bleaching and deodorization (RBD) [1]. About 90% of the commercial corn oil is obtained from corn germ that is a byproduct (co-product) of corn wet milling, a process developed to efficiently remove the starch (~70%) from corn kernels [1].

Two other types of experimental corn oil have also been reported but neither has been produced on a commercial scale. Corn kernel oil has been obtained by the extraction of ground corn with ethanol [2–4]. Corn fiber oil has been obtained by the extraction of corn fiber with hexane and other solvents [5, 6]. The properties of these experimental oils were compared in a recent book chapter [7].

Like many other seed oils, linoleic acid is the predominant fatty acid in all three types of corn oil [7], with smaller proportions of oleic, palmitic, and linolenic acids. All three corn oils contain significant amounts of tocopherols which serve as natural antioxidants, but corn kernel oil and corn fiber oil also contain significant amounts of tocotrienols [7]. Among the common commodity seed oils, commercial corn oil contains the highest levels of

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phytosterols (approximately 0.5% free phytosterols and 0.5% phytosteryl fatty acyl esters) [7]. Corn fiber oil and corn kernel oil contain even higher levels of total phytosterols (~10 and 2%, respectively) [7]. Although commercial corn oil contains very low levels of carotenoids, we recently reported that corn kernel oil obtained by the ethanol extraction of ground corn kernels contains high levels of lutein and zeaxanthin (~100-fold higher than those in commercial corn oil) [7, 8].

In recent years the proportion of the US corn harvest used as a feedstock for fuel ethanol has increased dramatically and it reached about 33% in 2008 [9]. Because only the starch (~70% by weight) in corn kernels is fermentable, there has been great interest in trying to extract the corn oil from corn kernels (~4% by weight) and utilize it for edible or nonedible applications. One strategy has been to remove corn germ by wet or dry fractionation methods before fermentation and then extract the oil via hexane or via aqueous enzymatic methods [10, 11]. Another strategy (recently patented by Greenshift Co., New York, NY, USA) has been to remove the corn oil after the fermentation step in a dry grind ethanol process [12, 13]. The proprietary process basically involves recovering the oil from the thin stillage by a process that combines heating, condensation to remove water and centrifugation. In this study we determined the composition of corn oil recovered by this type of process, hereafter called "post fermentation corn oil" and compared it to the composition of corn kernel oil. Winkler et al. [14] reported the composition of oil extracted from corn Distillers Dried Grains and Solubles (DDGS), but we are not aware of any published information about the composition of corn oil recovered via centrifugation using the process described above.

Materials and Methods

Materials

A sample of corn kernels (commodity grade, yellow dent #2) was obtained from Western New York Energy, Medina, NY, USA and represented a mixture of several corn hybrids used by a typical dry grind ethanol plant. Corn kernels were ground to 20 mesh with a Wiley Mill. Corn kernel oil was obtained by extracting ground corn kernels with ethanol at 100 °C and 1,000 psi, in a Dionex Accelerated Solvent Extractor as previously described [15, 16]. Corn germ was obtained from a commercial corn wet mill. Corn germ oil (unrefined) was obtained by extracting ground corn germ (~1 mm particles) with hexane at 100 °C and 1,000 psi, in a Dionex Accelerated Solvent Extractor as previously described [15]. Two samples of post fermentation corn oil were obtained using the proprietary GreenShift oil extraction system from Western New York Energy, LLC, Medina, NY, USA (Sample A in September 2008 and Sample B in September 2009). A third sample (Sample C) of post-fermentation corn oil was obtained in September 2009 from another dry grind ethanol plant that uses the same GreenShift system. For some analyses, the oil was centrifuged at 25 °C in order to recover a white precipitate, which was subsequently removed and dissolved in hexane for HPLC analysis.

HPLC Analysis of Nonpolar Lipids

Nonpolar lipid classes were analyzed via a previously reported method [17]. The HPLC was an Agilent Model 1100 with autosampler. The detector was an ESA Corona™ Charged Aerosol Detector (ESA Biosciences, Chelmsford, MA, USA) operated with nitrogen as a nebulizing gas and at a range of 500 pA. A LiChrosorb 7-µm DIOL column (3 × 100 mm, packed by Chrompack, Raritan, NJ, USA) was used. The binary gradient had a constant flow rate of 0.5 ml/min, with Solvent A = 99.9% hexane/0.1% acetic acid (all solvent compositions are v/v), Solvent B = 99% hexane/1% isopropanol. Gradient timetable: at 0 min, 100/0 (%A/%B); at 8 min, 100/0; at 10 min, 75/25; at 40 min, 75/25; at 41 min, 100/0; and at 60 min, 100/0. These nonpolar lipid components were identified by comparison to the retention times of commercial standards.

HPLC Analysis of Polar Lipids

Polar lipids (including glycolipids and phospholipids) were quantitatively analyzed by a similar HPLC–ELSD method [16], using a Hewlett Packard Model 1100 HPLC, with autosampler, and detection by both an HP Model 1100 diode-array UV–visible detector (Agilent Technologies, Avondale, PA, USA) and a Sedex Model 55 Evaporative Light Scattering Detector (Richard Scientific, Novato, CA, USA), operated at 40 °C and a nitrogen gas pressure of 2 bar. The polar lipid components were identified by comparison to the retention times of commercial standards and *p*-coumaroyl feruloylputrescine (CFP) and diferuloylputrescine (DFP) were identified using standards prepared in our previous study [18]. The diol column and flow rates were the same as above. The ternary gradient consisted of: Solvent A = hexane/acetic acid, 1,000/1, Solvent B = isopropanol, and Solvent C = water. Gradient timetable: at 0 min, 90/10/0 (%A/%B/%C); at 30 min, 58/40/2; at 40 min, 45/50/5; at 50 min 45/50/5; at 51 min, 50/50/0; at 52 min, 90/10/0; and at 60 min 90/10/0. The minimum limits of quantitative detection with both HPLC methods were about 1 µg per injection. Mass versus peak area

calibration curves were constructed for the range of 1–20 μg per injection.

HPLC Analysis of Tocopherols and Tocotrienol

Tocopherols and Tocotrienols were analyzed via a previously reported method [19]. The HPLC was a Hewlett Packard Model 1100 with autosampler, and detection by an HP Model 1100 Fluorescence Detector (Agilent Technologies, Avondale, PA, USA), with excitation at 294 nm and emission at 326 nm. Free phytosterols were analyzed simultaneously using the same HPLC method using a Sedex 55 Evaporative Light Scattering Detector. The diol column and flow rates were the same as above. The isocratic method employed a mobile phase consisting of hexane/THF, 970/30, v/v. The minimum limits of quantitative detection of tocols (tocopherols and tocotrienols) was about 0.1 ng per injection and a standard curve was constructed with the four tocopherol isomers (obtained from Calbiochem) in the range of 1–200 ng per injection and this curve was used to quantify all tocopherols. The masses of the corresponding tocotrienols were calculated using the standard curve for the analogous tocopherol. The structures and retention times of the tocopherols and tocotrienols were confirmed by purchasing gelcap supplements of tocopherols (Bio E Gamma Plex, Soloray Inc. Park City, UT, USA) and tocotrienols (Tocopherol Complex, Solgar, Leonia, NJ, USA) at a local vitamin store: α-tocopherol (αT) ($M + 1 = m/z$ 431.4), α-tocotrienol (αT3) ($M + 1 = m/z$ 425.3), β-tocopherol (βT) and γ-tocopherol (γT) ($M + 1 = m/z$ 416.3), β-tocotrienol (βT3) and γ-tocotrienol (γT3) ($M + 1 = m/z$ 411.2), δ-tocopherol (δT) ($M + 1 = m/z$ 402.3) and δ-tocotrienol (δT3) ($M + 1 = m/z$ 397.1), were confirmed by LC–MS, performed with an Agilent 1100 MSD equipped with an Atmospheric Pressure Chemical Ionization interface operated in the positive mode (drying gas at 6.0 L/min, nebulizer pressure at 60 psi, drying gas temperature at 350 °C, vaporizer gas temperature at 325 °C, Capillary voltage at 4,000 V, and corona Current at 4.0 μA, and fragmentor at 80 v).

HPLC Analysis of Carotenoids

Carotenoids were analyzed via a previously reported method [8], employing a LiChrosorb 7 Diol column, 3 × 100 mm (Varian Scientific, Lake City, CA, USA) with an isocratic mobile phase consisting of hexane/isopropanol/acetic acid, 96.9/3.0/0.1, v/v/v, at a flow rate of 0.5 ml/min. HPLC analyses were conducted with an Agilent Model 1100 HPLC equipped with an auto sampler and an Agilent Model 1100 Diode Array Detector to measure the visible absorbance of the carotenoids at 450 nm. Carotenoid standards (1–50 ng) were used to construct a standard curve to calculate mass values from peak area values. Carotenoid standards were purchased from Indofine Chemical Co., Hillsborough, NJ, USA (β-cryptoxanthin, lutein, and zeaxanthin) and Sigma Chemical Co., St. Louis, MO, USA (β-carotene).

Triplicate samples (1 mg/ml in hexane with 0.01% BHT) were prepared for each of the samples of post fermentation corn oils and precipitates. Each sample was injected one time in each of the four HPLC systems. Triplicate extractions of ground corn were conducted to prepare control samples of ethanol extracted corn kernel oil and each extract as evaporated and dissolved (1 mg/ml in hexane with 0.01% BHT) and injected one time in each of the four HPLC systems. The results presented are the means ± standard deviations.

Results and Discussion

Precipitate Enrichment

The three post fermentation corn oil samples were turbid ("milky") in appearance and upon storage at 4 °C they developed a white precipitate. A fourth sample that was enriched in this precipitate was also prepared by centrifuging corn oil sample A and the polar and nonpolar lipids in the precipitate-enriched sample were also analyzed.

Analysis of the Polar and Nonpolar Lipid Classes

The common polar and nonpolar lipids were analyzed in the six types of corn oil samples (all were unrefined): corn germ oil (RBD, refined, bleached and deodorized), corn kernel oil which was prepared by extracting ground corn with ethanol as previously described [15, 16], post fermentation corn oil (3 samples), and a centrifuged precipitate from the post fermentation corn oil. In all samples triacylglycerols were the predominant lipid class (Table 1). The levels of total free fatty acids were high in the two post fermentation oils and in the isolated precipitates (11–16%), but were low in the control corn kernel oil (~1%). Three phytosterol classes were also quantified. The levels of free (unesterified) phytosterols were similar in all samples (ranging from 0.5 to 0.93 wt%). The levels of hydroxycinnamate steryl esters, which are unique to corn, rice and a few other cereals [7], were absent in the corn germ oil and present the three other corn oil samples (ranging from 0.31 to 0. 41%). In contrast, a large variation was observed in the levels of the steryl ester/wax ester fraction (0.69–8.62%). Unfortunately, using our nonpolar HPLC method, this peak elutes very early and although it contains both steryl esters and wax esters, it also may contain

Table 1 Polar and nonpolar lipids in corn kernel oil and post fermentation corn oil

	Hexane-extracted corn germ oil (RBD) (wt%)	Ethanol-extracted corn kernel oil (unrefined) (wt%)	Post fermentation corn oil Sample A (wt%)	Post fermentation corn oil Sample B (wt%)	Post fermentation corn oil Sample C (wt%)	Post fermentation corn oil—precipitate (wt%)
Triacylglycerols	~96	~95	73.8 ± 1.4[a]	80.2 ± 2.3	79.3 ± 3.0	78.2 ± 2.7
Diacylglycerols	1.87 ± 0.07	3.22 ± 1.15	4.27 ± 0.03	3.99 ± 0.05	4.27 ± 0.04	3.99 ± 0.04
Free fatty acids						
Palmitic	0	0.22	5.01 ± 0.31	3.63 ± 0.03	3.86 ± 0.07	4.18 ± 0.42
Oleic	0	0.11	2.43 ± 0.70	1.74 ± 0.04	1.83 ± 0.01	2.04 ± 0.12
Linoleic	0	0.66	8.47 ± 0.50	5.66 ± 0.08	5.72 ± 0.10	7.25 ± 0.41
Linolenic	0	0.12	0.51 ± 0.14	0.18 ± 0.01	0.19 ± 0.01	0.47 ± 0.06
Total	0	1.11	16.42	11.21	11.60	13.94
Free sterols (FS)	0.52 ± 0.1	0.81 ± 0.04	0.93 ± 0.07	0.60 ± 0.01	0.74 ± 0.03	0.83 ± 0.01
Hydroxycinnamate steryl esters	ND	0.31 ± 0.02	0.38 ± 0.00	0.36 ± 0.02	0.41 ± 0.02	0.32 ± 0.01
Steryl esters and wax esters[b]	1.64 ± 0.16	0.69 ± 0.03	4.25 ± 0.07	3.64 ± 0.03	3.66 ± 0.06	8.62 ± 0.53
Glycolipids	ND	0.002	ND	ND	ND	ND
Phospholipids	ND	0.170	ND	ND	ND	ND
Polyamine conjugates	ND	0.84	ND	ND	ND	ND

RBD is refined, bleached and deodorized; *ND* not detected

[a] Mean ± standard deviation ($n = 3$)

[b] This chromatographic peak reported as this fraction may contain other nonpolar lipids such as various hydrocarbons

hydrocarbons and other very nonpolar compounds. Further studies will require steps to remove the triacylglycerols (perhaps by hexane washing) and confirm that these are waxes and to characterize their chemical composition.

Glycolipids and phospholipids were not detected in the hexane-extracted corn kernel oil. As expected, the low levels of glycolipids and phospholipids that might have been extracted from the germ with hexane were probably removed during RBD, which usually includes a degumming step. Previously we demonstrated that when corn germ or ground corn is extracted with either hexane or ethanol, the unrefined (crude) oil contains low levels of glycolipids and phospholipids [15, 16]. In contrast, glycolipids and phospholipids were not detectable in the post fermentation corn oil/precipitate samples (Table 1). A probable explanation for the absence of glycolipids and phospholipids is that the centrifugation process, used to recover the corn oil, also causes the oil to float through the aqueous phase. In effect, this step is similar to the "degumming" step in conventional oil refining where most of the glycolipids and phospholipids are removed from the crude oil by washing it with water or often with dilute citric acid. It is also possible that low levels of organic acids in the post fermentation broth might enhance the partitioning of glycolipids and phospholipids into the water phase.

We also previously reported that extraction of ground corn with a polar solvent such as ethanol or methylene chloride produced an oil that contained ~1% polyamine conjugates (mainly diferuloylputrescine and *p*-coumaroyl feruloylputrescine) [18]. However, no polyamine conjugates were detected in the post-fermentation corn oil (Table 1). It is possible that the polyamine conjugates also may have partitioned into the water phase, as suggested above for phospholipids and glycolipids.

Analysis of the Tocopherols and Tocotrienols

Previous studies reported that corn oil contains three tocopherol vitamers (α, γ, and δ) and three tocotrienol vitamers (α, γ, and δ) [19]. Using a sensitive normal phase HPLC method with fluorescence detection, five tocol vitamers were detected in the current samples (δ-tocotrienol was detected in corn oils in our previous study [19] but was not detected in any oil samples in the current study) (Fig. 1). This chromatogram (Fig. 1) is presented mainly to demonstrate the good chromatographic separation of all of the three tocopherols and two tocotrienols. The response factors are highest for alpha T/T3, lower for beta and gamma T/T3, and lowest for delta T/T3, so it is difficult to try to visualize the quantitative trends from the chromatogram. The levels of the five tocols were lower in the post-fermentation corn oil than in the control corn kernel oil (Table 2). Because tocol levels vary among various corn hybrids and vary in response to growing and storage

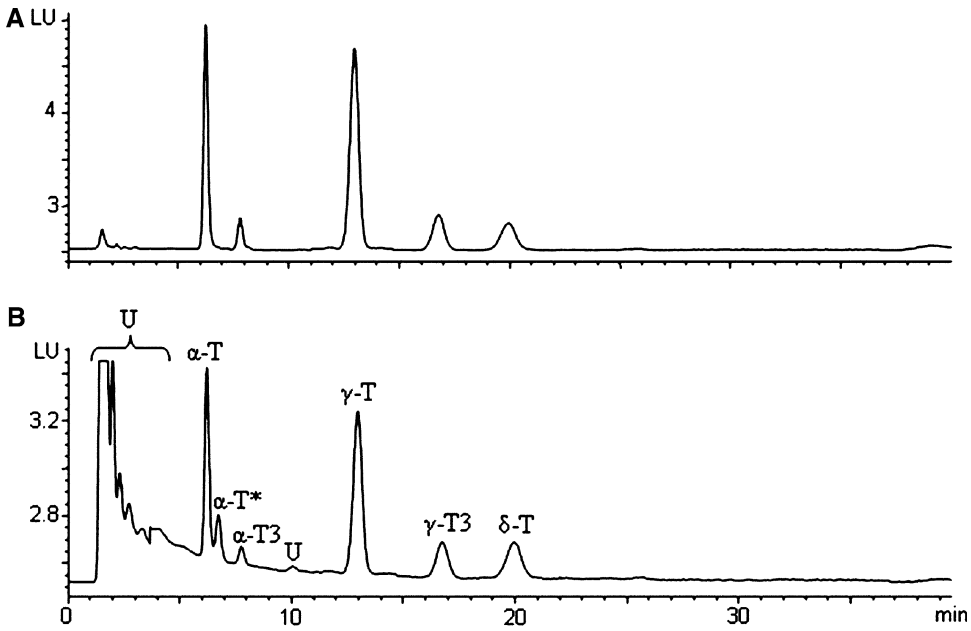

Fig. 1 An HPLC chromatogram of the tocopherols and tocotrienols in: **a** oil from ground corn extracted with ethanol and **b** post-fermentation corn oil

Table 2 Analysis of tocopherols and tocotrienols in corn oils (mg tocol/100 g oil)

	α-T	α-T*	γ-T	δ-T	α-T3	γ-T3	δ-T3	Total T	Total T3
Hexane-extracted corn germ oil (unrefined)[a]	21.8 ± 4.5[b]	0	275.6 ± 9.6	21.5 ± 0.2	1.5 ± 0.3	11.1 ± 0.3	0	318.9	12.6
Ethanol-extracted corn kernel oil (unrefined)[a]	42.6 ± 1.0	0	103.5 ± 0.5	11.3 ± 0.1	17.4 ± 0.0	32.4 ± 0.1	5.0 ± 0.3	157.4	54.8
Post-fermentation corn oil, Sample A	1.6 ± 0.7	14.2 ± 2.2	49.5 ± 0.4	14.1 ± 0.2	4.9 ± 0.2	14.5 ± 0.2	0	105.2 ± 1.4	19.4 ± 0.3
Post fermentation corn oil, Sample B	2.8 ± 0.2	55.6 ± 0.1	73.0 ± 1.7	14.4 ± 0.9	8.8 ± 0.5	19.9 ± 0.2	0	90.2 ± 1.9	28.7 ± 1.0
Post fermentation corn oil, Sample C	2.3 ± 0.1	262.9 ± 0.7	71.1 ± 0.7	34.3 ± 1.4	9.4 ± 0.2	22.0 ± 0.3	0	107.7 ± 1.6	31.4 ± 0.2

* Unknown peak, not considered in calculation of either total T or total T3

[a] Data from Ref. [19]

[b] Mean ± standard deviation ($n = 3$)

conditions we compared the composition of corn kernel oil in the current study to that determined in a previous study [19]. While considerable differences in levels of tocols existed in these control samples (including the presence of δ-tocotrienol in the previous study and absence in the current study), they each contained considerably more total tocols than the post fermentation corn oil samples. Thus, it appears that some of the tocols are destroyed during the fuel ethanol process, which can include extreme temperatures and pressures during pre-treatment, liquefaction, fermentation, and alcohol stripping processes. Previously, we reported that high temperature treatment can destroy tocols in wet milled corn germ [19]. When the germ was heated to 105 °C for 1 h and then extracted with hexane, the levels of tocopherols were reduced by 56% and the levels of tocotrienols were reduced by 64% [19].

We also observed that in the chromatogram of the post-fermentation corn oil there was a large peak of unknown fluorescent material (labeled U in the chromatogram) that eluted at1-2 min with some minor peaks and "tailing" in the 3–5 min region (Fig. 1). It is possible that these unknown peaks are generated during the fuel ethanol production process, because the chromatogram of the control corn kernel oil contained a much smaller peak at 1–2 min and almost no peaks in the 3–5 min region.

We also detected an additional peak in the post-fermentation corn oil which was absent in the control corn oil. The peak area of this unknown was about one-third the size of the two largest peaks (α and γ tocopherols). This unknown peak was labeled α-T* because it eluted after α-T and before α-T3. It is possible that this unknown peak may

Table 3 The levels of carotenoids in ethanol extracted corn kernel oil and in oil obtained after fermentation via centrifugation from a dry grind ethanol process

	β-Carotene (μg/g) oil	β-Cryptoxanthin (μg/g) oil	Lutein (μg/g) oil	Zeaxanthin (μg/g) oil	Total carotenoids (μg/g) oil
Hexane-extracted corn germ oil (unrefined)	0	0	1.4 ± 0.2[a]	0.9 ± 0.1	2.3 ± 0.2
Hexane-extracted corn kernel oil (unrefined)	14.2 ± 0.8	6.5 ± 0.3	42.8 ± 4.0	21.5 ± 1.3	80.7 ± 8.1
Ethanol-extracted corn kernel oil (unrefined)	25.7 ± 0.6	21.9 ± 0.8	101.3 ± 21.9	71.8 ± 3.0	220.7 ± 25.8
Post fermentation corn oil, Sample A	35.3 ± 0.5	106.2 ± 1.1	85.8 ± 1.0	69.5 ± 3.3	296.8 ± 4.9
Post fermentation corn oil, Sample B	50.8 ± 1.1	102.6 ± 2.0	92.8 ± 3.6	55.6 ± 2.2	295.5 ± 7.3
Post fermentation corn oil, Sample C	56.5 ± 0.8	169.8 ± 0.3	90.7 ± 0.3	88.3 ± 1.1	405.3 ± 1.3

[a] Mean ± standard deviation ($n = 3$)

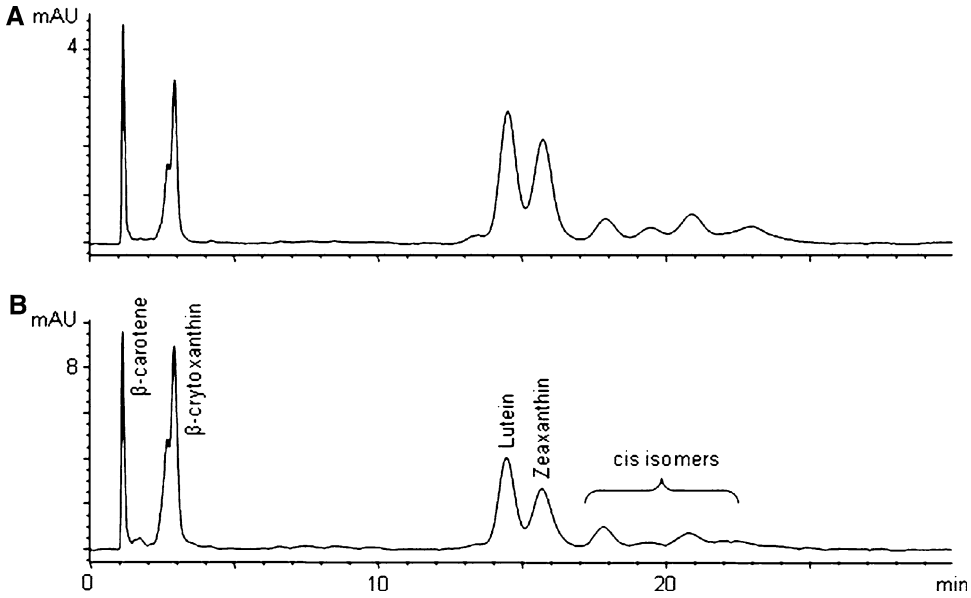

Fig. 2 An HPLC chromatogram of the carotenoids in: **a** oil from ground corn extracted with ethanol and **b** post-fermentation corn oil

be a "tocomonoenol", which has been reported in some other species [20, 21], but further work will be required to confirm its identity.

It should be noted that most of the oil samples in this study were "crude," meaning that they were not refined, bleached, or deodorized (RBD). Hexane-extracted corn germ oil (RBD, commercial corn oil) was used as the control in Table 1, mostly to confirm the absence of free fatty acids, but crude corn germ oil was used as a control in Tables 2 and 3 because higher levels of tocols and carotenoids would be expected in the crude germ on than in the RBD germ oil. Almost all edible oils are RBD to remove free fatty acids (which are very susceptible to oxidation), to remove off-flavors and odors, and to extend the shelf life of the oils. Although the purpose of RBD is to remove undesirable components, the processes also remove some (~5–50%) of tocopherols, tocotrienols and phytosterols [22]. This information should be taken into consideration when comparing the levels of tocopherols, tocotrienols, and phytosterols in crude oils (such as those used in this study) and in commercial RBD oils.

Analysis of the Carotenoids

Using our normal phase HPLC method, we were able to separate four peaks of carotenoids in the corn oil samples. β-Carotene eluted at 1–2 min and the three xanthophylls (oxygen-containing carotenoids) β-cryptoxanthin, lutein, and zeaxanthin, eluted at 3, 15, and 16 min, respectively (Fig. 2). Lutein and zeaxanthin have been reported to be present in high levels in the macula portion of the retina, and ingesting at least 6 mg of lutein + zeaxanthin per day has been reported to be an effective dosage to delay the onset of age-related macular degeneration [8]. The levels of lutein and zeaxanthin were very low in the corn germ oil as we reported previously [8]. The levels of lutein and

zeaxanthin in the post-fermentation corn oil were 85–93 and 56–88 μg/gram of oil, respectively (Table 3). These levels were similar to those in the control corn oil sample prepared freshly for this study (by extracting ground corn with ethanol), but they were slightly lower than the levels that we previously reported in a similar corn oil preparation [8]. As with the sterols and tocols, it should be noted that differences in their levels vary with individual yellow corn hybrids, so it is difficult to conclude whether lutein and zeaxanthin levels are affected by the dry grind ethanol processing conditions and by removal of the post-fermentation oil via centrifugation. The levels of β-carotene are about 35–57 μg/g of oil, in post-fermentation corn oil (Table 3), which is lower than in the fresh corn oil control but higher than in our previous publication. Finally, the levels of β-cryptoxanthin in the post-fermentation corn oil were about 103–170 μg/g of oil. This value is much higher than in the ethanol-extracted corn kernel oil. Further studies will be needed to confirm whether more β-cryptoxanthin is actually being extracted by this process or whether another compound may be created and co-elute with β-cryptoxanthin in this HPLC system. Structurally, β-cryptoxanthin contains one oxygen and lutein and zeaxanthin each contain two oxygens. Although lutein and zeaxanthin have been reported to be valuable for eye health, we are not aware of reports on the health-promoting properties of β-cryptoxanthin, although we expect that its antioxidant properties would be similar to those of other carotenoids.

In summary, in this study we measured the levels of three valuable classes of lipids in crude corn oil recovered by centrifugation, post fermentation from a commercial fuel ethanol plant, and compared their levels to those in corn oil prepared by ethanol extraction of whole corn kernels. We found that there was no significant difference in the levels of free sterols and hydroxycinnamate steryl esters in these two oils. The levels of carotenoids (especially lutein and zeaxanthin) were somewhat similar, but the levels of tocopherols and tocotrienols were considerably lower in the post-fermentation corn oil than in the corn kernel oil control. If the post-fermentation corn oil is used for non-edible applications such as making biodiesel, it may be desirable to remove/fractionate some or all of these valuable "functional lipids" before converting the triacylglycerols and fatty acids into biodiesel if an economically feasible process can be developed to accomplish this.

Immediately after this manuscript was accepted for publication, the authors learned that another paper on the topic of post-fermentation corn oil and precipitate composition had just been published [23]. The paper by the other authors focused mainly on the chemical analysis of the precipitates, whereas our current study focused on the analysis of the oil.

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