# The Composition of Corn Oil Obtained by the Alcohol Extraction of Ground Corn

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**ABSTRACT:** All commercial corn oil is obtained by the hexane extraction of corn germ. The chemical composition of commercial corn oil has been well characterized. This study was undertaken to quantitatively evaluate the lipid composition of corn oil obtained by the ethanol extraction of ground, whole corn kernels. When corn oil was obtained by extracting ground corn kernels (ground corn) with polar or nonpolar solvents, the resulting corn oil contained much higher levels of hydroxycinnamate steryl esters (~0.3%) than those found in commercial hexane-extracted corn (germ) oil (~0.02%). The levels of valuable tocopherols and tocotrienols were also significantly higher in kernel oil than in traditional corn germ oil. We previously reported that when corn oil was obtained by extracting corn kernels with polar solvents, the oil contained two polyamine conjugates, diferuloylputrescine and *p*-coumaroyl feruloylputrescine. In the current study, when ground corn was extracted with ethanol, the resulting corn oil contained about 0.5% diferuloylputrescine and about 0.2% *p*-coumaroyl feruloylputrescine. This is the first study to quantify these unique compounds in corn oil extracted by new techniques. This compositional information is important because this new oil is being considered for human food use.

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**KEY WORDS:** Corn, oil, phytosterols, polyamines, *Zea mays*.

We have previously shown that oil extracted from wet mill-derived corn fiber (corn fiber oil) and ground corn (corn kernel oil) contains significant levels of a unique class of ferulate esters called hydroxycinnamate steryl esters (HSE), the most abundant of which is sitostanol ferulate (Fig. 1A) (1,2). We also previously reported that corn kernels contain two polyamine conjugates, diferuloylputrescine (DFP) and *p*-coumaroyl feruloylputrescine (CFP) (Figs. 1B, 1C) (3). When extracted with polar solvents such as methylene chloride, ethanol, or isopropanol, corn fiber oil and corn bran oil (oil extracted from corn bran, a by-product of the corn dry-milling industry) were demonstrated to contain up to 10% polyamine conjugates. Corn germ oil (both crude and refined), has very low levels of these compounds (4). With the current research interest in using alcohols to extract vegetable oils (5,6), this study was undertaken to quantitatively evaluate the levels of hydroxycinnamic acid phytosterol esters, DFP, and CFP in corn oil obtained *via* alcohol extraction of ground whole corn kernels and corn germ.

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## MATERIALS AND METHODS

Corn bran (NC02080, Coarse, DIETFIBER, from a corn dry-milling plant) and dry-milled corn germ were kindly provided by Will Duensing, Lauhoff/Bunge Foods (Danville, IL). Wet-milled corn germ was kindly provided by a commercial corn wet mill. Yellow dent #2 corn kernels (Pioneer H3361) were grown at the University of Illinois (Urbana, IL). The corn bran was ground by the supplier to an average particle size of about 1 mm. Samples (20–30 g) of dry-milled corn germ and wet-milled corn germ were ground for 10 s with a coffee mill (Krups, Model 203B). Samples of corn kernels were milled to a 20-mesh particle size with a Wiley mill (Thomas Scientific, Philadelphia, PA).

*Extraction.* Extractions were performed with a Dionex accelerated solvent extractor, model ASE 200 (Dionex Inc., Sunnyvale,

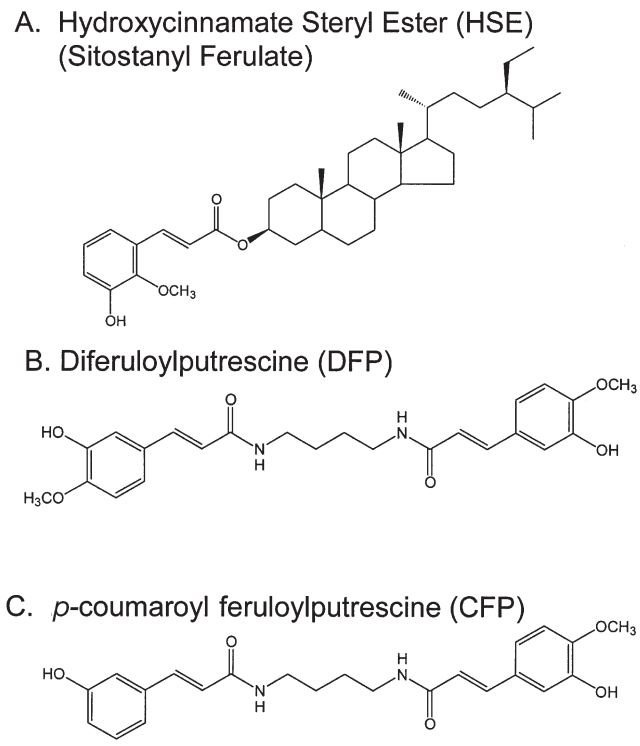


**FIG. 1.** Unusual lipid components detected in significant concentrations in alcohol-extracted corn oil. (A) The most common lipid component, hydroxycinnamate sterol ester (HSE); (B, C) the polyamine conjugates diferuloylputrescine (DFP) and *p*-coumaroyl feruloylputrescine (CFP).

CA) using hexane, isopropyl alcohol, or ethanol, as previously described (7). For some samples the extractor was programmed to deliver 70–100% ethanol, with the remainder being deionized water. The ground sample (1 g germ, or 4 g bran or ground corn kernels) was placed in an 11-cc stainless steel extraction vessel and the remaining volume was filled with sea sand. The extractor was programmed to extract at a pressure of 1000 psi (69 bar) and a temperature of 50°C, extracting each sample with a total of 22 mL of solvent, delivered in three 10-min extractions (3 × 7.3 mL). Three separate extractions were performed for each sample and duplicate HPLC injections were made from each extract.

*Nonpolar lipid HPLC analyses.* Nonpolar lipids [including phytosterol fatty acyl esters, retention time (rt) = 1.7 min; hydroxycinnamate phytosterol esters, rt = 26 min; TAG, rt = 3–5 min; and FFA, rt = 7–10 min] were quantitatively analyzed by an updated version of a normal-phase HPLC method with ELSD (1). These nonpolar lipid components were identified by comparison with the rt of commercial standards. Quantitative analysis of each component was achieved by injecting multiple samples of each standard (in the range of 1 to 50 μg per injection) and constructing a standard curve. These analyses were performed on a Hewlett-Packard model 1050 high-performance liquid chromatograph, with autosampler, and detection by both an HP model 1050 diode-array UV/vis detector (Agilent Technologies, Avondale, PA) and an Alltech-Varex MKII ELSD (Alltech Associates, Deerfield, IL), operated at 40°C and a nitrogen gas flow rate of 1.7 standard liters per minute. The column was a LiChrosorb 7-μm DIOL column (3 × 100 mm, packed by Chrompack, Raritan, NJ). The binary gradient had a constant flow rate of 0.5 mL/min, with solvent A as hexane/acetic acid (1000:1) and solvent B as hexane/isopropanol (100:1). The gradient timetable was as follows: 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.

*Polar lipid HPLC analyses.* Polar lipids (including acylated steryl glucosides, steryl glucosides, phospholipids, and polyamine conjugates) were quantitatively analyzed by a similar HPLC-ELSD method (7) using a Hewlett-Packard model 1100 high-performance liquid chromatograph, with autosampler, and detection by both an HP model 1100 diode-array UV/vis detector (Agilent Technologies) and a Sedex model 55 ELSD (Richard Scientific, Novato, CA), operated at 40°C and a nitrogen gas pressure of 2 bar. The polar lipid components were identified by comparison with the rt of commercial standards, and CFP and DFP were identified using standards prepared in our previous study (3). The diol column and flow rates were the same as above. The ternary gradient consisted of: solvent A, hexane/acetic acid (1000:1); solvent B, isopropanol; and solvent C, water. The gradient timetable was as follows: 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 was about 1 μg per injection. Mass vs. peak area calibration curves were constructed for the range of 1–20 μg per injection.

*Tocopherol and tocotrienol HPLC analyses.* Tocopherols and tocotrienols were quantified using a modified version of the previously published method (8). The HPLC was a Hewlett-Packard model 1100, with autosampler, and detection was by an HP model 1100 fluorescence detector (Agilent Technologies), with excitation at 294 nm and emission at 326 nm. The diol column and flow rates were the same as above. The binary gradient consisted of solvent A, hexane/THF (980:20), and solvent B, isopropanol. The gradient timetable was as follows: at 0 min, 100:0 (%A/%B); at 40 min, 100:0; at 45 min, 95:5, at 50 min, 95:5; at 51 min, 100:0; and at 60 min, 100:0. The minimum limits of quantitative detection of tocols was about 0.1 ng per injection, and a standard curve was constructed with α-tocopherol in the range of 1–200 ng per injection. This curve was used to quantify all tocopherols. 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) and tocotrienols (Tocopherol Complex, Solgar, Leonia, NJ) at a local vitamin store: α-tocopherol ($M + 1 = m/z$ 431.4), α-tocotrienol ($M + 1 = m/z$ 425.3), α-tocopherol ($M + 1 = m/z$ 416.3), γ-tocotrienol ($M + 1 = m/z$ 411.2), γ-tocopherol ($M + 1 = m/z$ 402.3), and δ-tocotrienol ($M + 1 = m/z$ 397.1) were confirmed by LC-MS, performed with an Agilent model 1100 mass selective detector 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 350ºC, vaporizer gas temperature at 325ºC, capillary voltage at 4000 V, corona current at 4.0 μA, and fragmentor at 80 V).

All experiments were performed at least twice with triplicate samples each time. The data presented are means ± SD.

## RESULTS AND DISCUSSION

All commercial corn oil in the United States is obtained by the hexane extraction of corn germ or the hexane extraction of corn germ that has been prepressed (4). In the first experiment, the composition of hexane-extracted corn germ (from a corn wet mill) was examined and, as expected, was found to contain high levels of TAG and low levels of DAG, FFA, and two phytosterol lipid classes (Table 1). Extracting ground corn kernels with ethanol at 50°C yielded similar levels of the typical types of nonpolar lipids found in corn oil from hexane-extracted corn germ (which was equivalent to an unrefined commercial corn oil), except that significant levels of three unique compounds, HSE (0.23%), DFP (0.66%), and CFP (0.18%) (Fig. 1; Table 1), were also detected. We previously reported high levels of HSE in hexane-extracted corn fiber oil (4–6%) and low levels in commercial hexane-extracted corn (germ) oil (<0.01%) (1). Additional studies were performed to evaluate the levels of these unusual lipids in detail.

The second experiment was designed to quantify the overall yields of oil and the levels of three sterol lipid classes (steryl esters, free sterols, and HSE) and polyamine conjugates (DFP and CFP) in extracts (oils) prepared by extracting ground corn kernels, ground corn bran, ground wet-milled corn germ, and

**TABLE 1**
**The Composition of Lipids in Corn Oil Obtained by Extracting Corn Germ with Hexane and by Extracting Ground Corn with 100% Ethanol at 50°C and 1000 psi[a]**

| Lipid class | Corn oil (wt%) | |
|---|---|---|
| | From hexane-extracted wet-milled corn germ | From ethanol-extracted ground corn kernels |
| TAG | 97.70 ± 0.78 | 95.78 ± 2.33 |
| DAG | 0.07 ± 0.00 | 0.05 ± 0.01 |
| FFA | 1.26 ± 0.03 | 0.68 ± 0.09 |
| Free phytosterols (FS) | 0.48 ± 0.01 | 0.76 ± 0.07 |
| Phytosterol fatty acyl esters (SE) | 0.47 ± 0.00 | 0.50 ± 0.04 |
| Hydroxycinnamate sterol esters (HSE) | 0.02 ± 0.00 | 0.23 ± 0.03 |
| Diferuloylputrescine (DFP) | 0 | 0.66 ± 0.05 |
| *p*-Coumaroyl feruloylputrecine (CFP) | 0 | 0.18 ± 0.01 |

[a]Both oil samples were unrefined.

ground dry-milled corn germ with three solvents (hexane, isopropanol, and ethanol) at either 50 or 100°C (Fig. 2, Table 2). Based on the results of a previous study in our laboratory (9), we estimate that the proportions of mass of ground corn kernels constitute about 5% germ, 5% bran (pericarp), and about 90% endosperm.

The extraction temperature of 50°C was chosen because this temperature was used in a previous paper (6) and patent (10) to extract corn oil from ground corn with ethanol. Extraction at 100°C was chosen for comparative purposes to evaluate whether higher concentrations of certain components in the oil may be achieved at higher temperatures. Yields of oil were significantly affected by solvent type and temperature. The yield of oil extracted from ground kernels with ethanol at 100°C, for instance, was 72% higher than yields using hexane at 50°C.

Examination of the three phytosterol lipid classes in ground corn extracts revealed that similar levels of the three were extracted by the three solvents. The data in Table 2 indicate that HSE were present at levels of 0.3% in the corn oil obtained from extracting ground corn with ethanol, and at similar levels in the corn oils obtained using the other solvents. Although numerous clinical studies have demonstrated the cholesterol-lowering efficacy of fatty acyl phytosterol esters and free phytosterols, the efficacy of HSE has not been investigated. We recently reported the first evidence that HSE are hydrolyzed by mammalian digestive enzymes (11), to release free phytosterols that are known to lower LDL cholesterol levels. More research is needed to evaluate their *in vivo* cholesterol-lowering efficacy and to evaluate their overall safety in humans.

Examination of the polar lipids in ground corn kernels extracted with one nonpolar (hexane) and two polar solvents (ethanol and isopropanol) revealed high levels (0.1 to 0.6%) of the two polyamine conjugates (DFP and CFP) in the oils obtained by both polar solvents and none in the oil obtained by hexane extraction (Table 2). Extraction of corn bran with the same solvents revealed that the levels of polyamine conjugates in bran extracts with the polar solvents were several fold higher than in the extracts from ground corn, similar to the high levels we reported previously in methylene chloride extracts of corn bran and corn fiber (3). Since very low levels of polyamine conjugates are found in corn germ and very high levels are found in

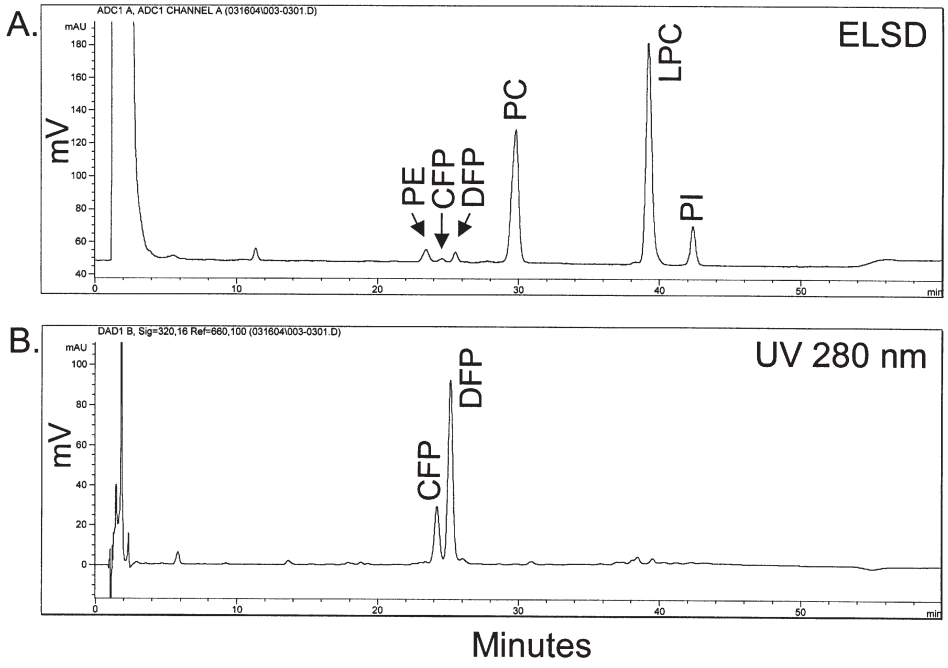


**FIG. 2.** HPLC chromatogram showing the peaks of CFP, DFP, and phospholipids in the unrefined oil obtained by extracting ground corn kernels with ethanol. Detection was with both (A) a UV detector (280 nm) and (B) an ELSD. LPC, lysophosphatidylcholine; for other abbreviations see Figure 1.

**TABLE 2**
**Sterols and Polyamine Conjugates (CFP and DFP) in Oil Extracted at 50 and 100°C from Ground Corn, Corn Bran, Wet-Milled (WM) Corn Germ, and Dry-Milled (DM) Corn Germ[a]**

| Sample, solvent | °C | Oil extracted (%) | wt% of oil | | | | |
|---|---|---|---|---|---|---|---|
| | | | SE | FS | HSE | CFP | DFP |
| Corn kernels | | | | | | | |
| Hexane | 50 | 2.70 ± 0.06 | 1.03 ± 0.02 | 0.74 ± 9.92 | 0.38 ± 0.03 | 0 | 0 |
| | 100 | 3.28 ± 0.04 | 1.01 ± 0.02 | 0.76 ± 0.04 | 0.30 ± 0.00 | 0 | 0 |
| Isopropanol | 50 | 3.36 ± 0.06 | 1.33 ± 0.29 | 0.59 ± 0.07 | 0.33 ± 0.02 | 0.081 ± 0.007 | 0.290 ± 0.023 |
| | 100 | 5.03 ± 0.24 | 0.86 ± 0.14 | 0.52 ± 0.03 | 0.27 ± 0.02 | 0.126 ± 0.003 | 0.459 ± 0.056 |
| Ethanol | 50 | 3.38 ± 0.04 | 0.69 ± 0.03 | 0.81 ± 0.04 | 0.31 ± 0.02 | 0.194 ± 0.024 | 0.568 ± 0.083 |
| | 100 | 5.53 ± 0.20 | 1.04 ± 0.25 | 0.52 ± 0.02 | 0.25 ± 0.01 | 0.120 ± 0.002 | 0.463 ± 0.022 |
| Corn bran | | | | | | | |
| Hexane | 50 | 1.92 ± 0.07 | 5.70 ± 0.25 | 1.49 ± 0.08 | 0.44 ± 0.01 | 0 | 0 |
| | 100 | 2.11 ± 0.00 | 5.01 ± 0.05 | 1.65 ± 0.05 | 0.42 ± 0.01 | 0 | 0 |
| Isopropanol | 50 | 2.46 ± 0.01 | 3.82 ± 0.08 | 0.93 ± 0.01 | 0.23 ± 0.04 | 0.99 ± 0.15 | 2.58 ± 0.32 |
| | 100 | 6.41 ± 0.28 | 2.80 ± 0.06 | 1.10 ± 0.09 | 0.28 ± 0.01 | 1.18 ± 0.11 | 4.36 ± 0.38 |
| Ethanol | 50 | 3.47 ± 0.61 | 3.03 ± 0.14 | 0.80 ± 0.04 | 0.23 ± 0.02 | 1.12 ± 0.07 | 3.24 ± 0.25 |
| | 100 | 5.08 ± 0.43 | 2.33 ± 0.08 | 2.20 ± 0.03 | 0.25 ± 0.01 | 0.88 ± 0.04 | 3.34 ± 0.21 |
| WM germ | | | | | | | |
| Hexane | 50 | 32.18 ± 0.22 | 0.51 ± 0.15 | 0.25 ± 0.04 | 0.03 ± 0.01 | 0 | 0 |
| | 100 | 34.21 ± 0.59 | 0.63 ± 0.01 | 0.34 ± 0.01 | 0.03 ± 0.01 | 0 | 0 |
| Isopropanol | 50 | 27.91 ± 0.29 | 0.49 ± 0.02 | 0.27 ± 0.02 | 0.02 ± 0.00 | 0 | 0 |
| | 100 | 33.43 ± 0.02 | 0.63 ± 0.01 | 0.37 ± 0.02 | 0.02 ± 0.00 | 0.02 ± 0.00 | 0.04 ± 0.00 |
| Ethanol | 50 | 27.18 ± 0.71 | 0.45 ± 0.06 | 0.26 ± 0.00 | 0.02 ± 0.00 | 0 | 0 |
| | 100 | 41.57 ± 0.03 | 0.59 ± 0.04 | 0.35 ± 0.02 | 0.03 ± 0.01 | 0.02 ± 0.00 | 0.06 ± 0.01 |
| DM germ | | | | | | | |
| Hexane | 50 | 14.31 ± 0.15 | 0.53 ± 0.06 | 0.48 ± 0.07 | 0.04 ± 0.00 | 0 | 0 |
| | 100 | 17.42 ± 0.30 | 0.44 ± 0.05 | 0.45 ± 0.05 | 0.04 ± 0.01 | 0 | 0 |
| Isopropanol | 50 | 16.26 ± 0.19 | 0.48 ± 0.04 | 0.31 ± 0.03 | 0.04 ± 0.01 | 0.07 ± 0.00 | 0.27 ± 0.00 |
| | 100 | 22.27 ± 0.50 | 0.97 ± 0.14 | 0.31 ± 0.03 | 0.03 ± 0.00 | 0.10 ± 0.01 | 0.31 ± 0.02 |
| Ethanol | 50 | 17.15 ± 0.59 | 0.72 ± 0.11 | 0.47 ± 0.04 | 0.05 ± 0.01 | 0.09 ± 0.01 | 0.33 ± 0.01 |
| | 100 | 23.57 ± 1.10 | 1.42 ± 0.55 | 0.47 ± 0.06 | 0.05 ± 0.01 | 0.11 ± 0.01 | 0.31 ± 0.03 |

[a]For other abbreviations see Table 1.

corn bran (pericarp), the probable source of polyamine conjugates in ground corn is the pericarp. Although distinct trends of polyamine conjugate localization were observed (high concentrations in bran, low in germ), care needs to be exercised in interpreting these data, since the particular hybrid (or combination of hybrids) used by commercial corn wet and dry mills is not specified and hybrid differences could account for some of the observed trends. Extraction of both germ samples (dry-milled germ and wet-milled germ) with hexane revealed very low levels of polyamine conjugates in the extracts. Interestingly, when extracting the ground corn with isopropanol, higher concentrations of DFP and CFP were achieved at 100°C than at 50°C. In contrast, when extracting ground corn with ethanol, higher concentrations were achieved at 50°C than at 100°C.

The data in Table 2 indicate that polyamine conjugates (DFP and CFP) were present at levels of 0.1–0.6% in the corn oil obtained from extracting ground corn with ethanol or isopropanol. Very little is known about the physiological role of polyamine conjugates in plants or about their effects on humans and livestock when they are ingested. Some monoamine and polyamine conjugates have been shown to have antimicrobial activities (12–14). We recently found that DFP and CFP inhibit aflatoxin biosynthesis in *Aspergillus flavus* (15), and others reported that they inhibit α-glucosidase (16). There is a need to conduct further research to understand whether polyamine conjugates have beneficial or harmful biological activities if they are present in an edible oil.

The levels of tocopherols and tocotrienols in the various solvent extracts were also quantitatively analyzed (Table 3). These analyses confirmed previous reports (17,18) that corn kernels contain the isomers of α- and γ-tocopherol and α- and γ-tocotrienol. Low levels of δ-tocopherol (50–150 mg/kg) were observed in the extracts of all samples with all solvents at all temperatures, confirming one previous report of this isomer (19). Low levels of δ-tocotrienol (35–50 mg/kg) were observed in the extracts of ground corn with all three solvents, but only at a temperature of 100°C (Table 3). High levels of tocopherols and tocotrienols were observed in the corn oil obtained by extracting ground corn and wet-milled corn germ. Lower levels (about 50% lower) of both were observed in the corn bran oil and the lowest levels (about 25% of the levels in the corn kernel oils or in wet-milled germ oils) in the dry-milled corn germ oil.

The very high levels of tocopherols and tocotrienols in corn oil obtained by extraction of ground corn kernels with all three solvents may indicate that this type of corn oil has superior health-promoting properties when compared with commercial corn germ oil and could perhaps be marketed as a value-added nutraceutical oil. Several studies have indicated that tocotrienols benefit cardiovascular health by lowering the levels

**TABLE 3**
**Individual Tocopherols (T) and Tocotrienols (T3) in Oil Extracted at 50 and 100°C from Ground Corn, Corn Bran, WM Corn Germ, and DM Corn Germ[a]**

| | | | mg/kg | | | | | |
|---|---|---|---|---|---|---|---|---|
| Sample, solvent | °C | % oil extracted | α-T | γ-T | δ-T | α-T3 | γ-T3 | δ-T3 |
| Corn kernels | | | | | | | | |
| Hexane | 50 | 2.70 ± 0.06 | 386.7 ± 2.3 | 1066.7 ± 26.7 | 72.2 ± 3.7 | 138.0 ± 3.5 | 214.8 ± 2.8 | 0 |
| | 100 | 3.28 ± 0.04 | 425.6 ± 11.0 | 1034.7 ± 5.2 | 112.4 ± 0.8 | 173.6 ± 0.4 | 324.2 ± 0.5 | 49.5 ± 2.8 |
| Isopropanol | 50 | 3.36 ± 0.06 | 330.9 ± 26.7 | 911.5 ± 61.5 | 60.7 ± 05.4 | 133.0 ± 12.7 | 239.1 ± 19.4 | 0 |
| | 100 | 5.03 ± 0.24 | 228.7 ± 9.7 | 616.0 ± 7.8 | 74.8 ± 01.1 | 123.2 ± 0.2 | 277.4 ± 9.8 | 39.4 ± 5.5 |
| Ethanol | 50 | 3.38 ± 0.04 | 284.2 ± 30.2 | 865.8 ± 148.0 | 61.1 ± 10.3 | 132.2 ± 14.6 | 257.3 ± 38.9 | 0 |
| | 100 | 5.53 ± 0.20 | 201.2 ± 5.4 | 548.0 ± 5.7 | 101.8 ± 0.4 | 113.2 ± 0.5 | 256.8 ± 5.3 | 35.6 ± 3.2 |
| Corn bran | | | | | | | | |
| Hexane | 50 | 1.92 ± 0.07 | 207.5 ± 9.2 | 473.4 ± 19.2 | 77.0 ± 4.2 | 62.9 ± 1.3 | 108.3 ± 8.8 | 0 |
| | 100 | 2.11 ± 0.00 | 174.9 ± 6.5 | 578.9 ± 15.4 | 113.0 ± 2.8 | 58.0 ± 2.8 | 168.5 ± 2.1 | 0 |
| Isopropanol | 50 | 2.46 ± 0.01 | 93.4 ± 27.4 | 426.8 ± 0.7 | 57.3 ± 2.1 | 0 | 110.0 ± 6.4 | 0 |
| | 100 | 6.41 ± 0.28 | 131.2 ± 3.7 | 385.7 ± 5.9 | 68.5 ± 1.4 | 47.6 ± 1.8 | 126.8 ± 1.1 | 0 |
| Ethanol | 50 | 3.47 ± 0.61 | 62.4 ± 12.2 | 335.3 ± 6.4 | 49.4 ± 2.7 | 0 | 90.3 ± 0.4 | 0 |
| | 100 | 5.08 ± 0.43 | 113.0 ± 3.5 | 330.4 ± 11.9 | 59.8 ± 1.1 | 40.4 ± 0.1 | 106.3 ± 3.2 | 0 |
| WM germ | | | | | | | | |
| Hexane | 50 | 32.18 ± 0.22 | 181.3 ± 8.5 | 827.1 ± 39.2 | 91.7 ± 8.7 | 0 | 71.1 ± 37.8 | 0 |
| | 100 | 34.21 ± 0.59 | 166.0 ± 1.4 | 993.3 ± 12.7 | 133.9 ± 3.7 | 0 | 67.7 ± 0.5 | 0 |
| Isopropanol | 50 | 27.91 ± 0.29 | 191.9 ± 10.7 | 880.3 ± 62.6 | 94.4 ± 7.2 | 0 | 48.7 ± 2.6 | 0 |
| | 100 | 33.43 ± 0.02 | 216.4 ± 11.9 | 1116.9 ± 51.5 | 138.9 ± 2.0 | 0 | 72.4 ± 3.4 | 0 |
| Ethanol | 50 | 27.18 ± 0.71 | 157.9 ± 6.5 | 889.7 ± 3.3 | 90.4 ± 5.5 | 0 | 52.5 ± 4.9 | 0 |
| | 100 | 41.57 ± 0.03 | 194.2 ± 1.9 | 1045.7 ± 25.7 | 134.4 ± 4.1 | 0 | 75.7 ± 1.2 | 0 |
| DM germ | | | | | | | | |
| Hexane | 50 | 14.31 ± 0.15 | 0 | 268.0 ± 3.5 | 57.8 ± 12.4 | 0 | 0 | 0 |
| | 100 | 17.42 ± 0.30 | 0 | 201.4 ± 3.0 | 90.6 ± 1.1 | 0 | 0 | 0 |
| Isopropanol | 50 | 16.26 ± 0.19 | 0 | 323.7 ± 10.4 | 68.7 ± 2.3 | 0 | 0 | 0 |
| | 100 | 22.27 ± 0.50 | 31.9 ± 0.8 | 348.5 ± 4.2 | 93.4 ± 5.5 | 0 | 38.0 ± 1.4 | 0 |
| Ethanol | 50 | 17.15 ± 0.59 | 0 | 291.2 ± 0.2 | 68.3 ± 2.8 | 0 | 0 | 0 |
| | 100 | 23.57 ± 1.10 | 31.1 ± 0.4 | 309.0 ± 3.5 | 84.3 ± 2.5 | 0 | 33.8 ± 3.9 | 0 |

[a]β-T and β-T3 were not detected in any of the samples. For other abbreviations see Table 2.

of serum cholesterol (20). Some have presented evidence that they lower serum cholesterol by inhibiting its synthesis *via* 3-hydroxy-3-methylglutaryl-CoA reductase (19). The total levels of tocotrienols in ethanol-extracted corn kernel oil (353 mg/kg in oil extracted at 50°C and 548 mg/kg in oil extracted at 100°C, Table 4) are comparable to those that have been reported in two other oils that are being marketed as "high in tocotrienols," i.e., palm oil (530 mg/kg) and rice bran oil (770 mg/kg) (21).

A final experiment was conducted to examine the effect of various concentrations of ethanol (70–100%, with the remainder being water) on the extraction of polyamine conjugates from ground corn (Table 5). Whereas Kwiatkowski and Cheryan (6) reported that primarily corn oil was extracted at 100% ethanol and that 70% ethanol was the optimal solvent mixture for the extraction of zein protein, we observed that similar levels of DFP (approximately 0.6%) and CFP (approximately 0.2%) were extracted at all four ethanol concentrations (Table 4). It is unusual for a natural product to be extracted in such constant yields over such a broad range of solvent polarities.

These experiments demonstrate that the composition of crude corn oil obtained by extracting ground corn with ethanol is very different from the composition of commercial corn oil (obtained by extracting corn germ with hexane). The ethanol-extracted corn kernel oil contains significantly higher levels of LDL cholesterol-lowering free phytosterols and phytosterol fatty acyl esters as well as tocopherols and tocotrienols, which are known to have antioxidant and/or vitamin E activity. Significantly increased yields of oil are also obtained when ethanol, rather than hexane, is used to extract ground corn kernels (especially at high temperatures). The extra yield is not just due to more of the same type of oil, but also to the presence of unique components that are extracted with the more polar solvent under these conditions.

The most abundant unique chemical components in oil obtained by the ethanol extraction of ground corn are hydroxycinnamic acid phytosterol esters and polyamine conjugates. Before the commercialization of ethanol-extracted corn kernel oil as a edible product can be considered, it will be necessary to understand the fate of these compounds during the processing of corn oil (refining, bleaching, and deodorization) and to better understand their physiological properties and health effects (benefits or toxicity).

**TABLE 4**
**A Summary of Total Levels of T and T3 in Oil Extracted at 50 and 100°C from Ground Corn, Corn Bran, WM Corn Germ, and DM Corn Germ[a]**

| Sample, solvent | °C | Total T (mg/kg) | Total T3 (mg/kg) | Total T + T3 (mg/kg) | %T3 (T3/total T + T3) |
|---|---|---|---|---|---|
| Corn kernels | | | | | |
| Hexane | 50 | 1525.6 | 352.8 | 1878.4 | 18.8 |
| | 100 | 1572.7 | 547.3 | 2120.0 | 25.8 |
| Isopropanol | 50 | 1303.1 | 372.1 | 1675.2 | 22.2 |
| | 100 | 919.5 | 440.0 | 1359.5 | 32.4 |
| Ethanol | 50 | 1211.1 | 389.5 | 1600.6 | 24.3 |
| | 100 | 851.0 | 405.6 | 1256.6 | 32.3 |
| Corn bran | | | | | |
| Hexane | 50 | 757.9 | 171.2 | 929.1 | 18.4 |
| | 100 | 866.8 | 226.5 | 1093.3 | 20.7 |
| Isopropanol | 50 | 577.5 | 110.0 | 687.5 | 16.0 |
| | 100 | 585.4 | 174.4 | 759.8 | 23.0 |
| Ethanol | 50 | 447.1 | 90.3 | 537.4 | 16.8 |
| | 100 | 503.2 | 146.7 | 649.9 | 22.6 |
| WM germ | | | | | |
| Hexane | 50 | 1100.1 | 71.1 | 1171.2 | 6.1 |
| | 100 | 1293.2 | 67.7 | 1360.9 | 5.0 |
| Isopropanol | 50 | 1166.6 | 48.7 | 1215.0 | 4.0 |
| | 100 | 1472.2 | 72.4 | 1544.6 | 4.7 |
| Ethanol | 50 | 1138.0 | 52.5 | 1190.5 | 4.4 |
| | 100 | 1374.3 | 75.7 | 1450.0 | 5.2 |
| DM germ | | | | | |
| Hexane | 50 | 325.8 | 0 | 325.8 | 0 |
| | 100 | 292.0 | 0 | 292.0 | 0 |
| Isopropanol | 50 | 392.4 | 0 | 392.4 | 0 |
| | 100 | 473.8 | 38.8 | 511.8 | 7.4 |
| Ethanol | 50 | 359.5 | 0 | 359.5 | 0 |
| | 100 | 424.4 | 33.8 | 458.2 | 7.4 |

[a]For abbreviations see Tables 2 and 3.

**TABLE 5**
**Effect of Ethanol Concentration (70 to 100%) on the Concentration of Polyamine Conjugates[a] in Oil Extracted from Ground Corn at 50°C**

| Ethanol (%) | Oil extracted (%) | CFP (%) | DFP (%) |
|---|---|---|---|
| 100 | 3.56 ± 0.04 | 0.209 ± 0.037 | 0.588 ± 0.051 |
| 90 | 3.87 ± 0.09 | 0.183 ± 0.013 | 0.688 ± 0.012 |
| 80 | 3.87 ± 0.33 | 0.222 ± 0.048 | 0.746 ± 0.028 |
| 70 | 4.07 ± 0.03 | 0.160 ± 0.003 | 0.643 ± 0.012 |

[a]For abbreviations see Table 1.

## DISCLAIMER

Mention of trade names or commercial products in this publication is solely for the purpose of providing specific information and does not imply recommendation or endorsement by the U.S. Department of Agriculture.

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