Oil Production from Catfish Viscera

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ABSTRACT: Oil was extracted from catfish viscera and purified (degummed, neutralized, bleached, and deodorized). The yield of catfish oil after purification was 65.7%. The major yield loss took place during the degumming process. The FA found in crude catfish visceral oil were 14:0, 16:0, 16:1, 18:0, 18:1, 18:2, 18:3, 20:0, 20:1, 20:2, 20:3, 20:4, and 22:6, the predominant FA being 18:1, 16:0, 18:2, and 18:0. The total unsaturated FA in the purified catfish oil amounted to 67.7%. The combined n-3 FA content of the purified catfish oil was 4.6 mg/g of oil. The purified catfish oil contained 1.21 mg/g DHA. FFA, water activity, and some mineral contents decreased during purification. Bleaching removed pigments, thus resulting in oil with greater lightness and less yellowness.

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There is a sizable and growing world market demand for high-quality fish oils, and commercial fish oil production can be quite profitable if suitable raw materials are available. The fish industry should carefully handle by-products from gutting, filleting, and other fish-processing operations because they are good raw materials for fish meal and oil production. The channel catfish (*Ictalurus punctatus*) is now the fourth-most popular fish product consumed in the United States (1). In the United States about 46.5 million lb (21.1 million kg) of catfish was processed in 1980, and by 2000 this number had increased to about 594 million lb (269.4 million kg) (2). The waste and by-products of catfish processing consist of heads, frames, skin, and viscera, which often end up in landfills or rendering plants. The average weight of viscera is about 265 g, which is about 10% by weight of a live whole catfish. The fat content of viscera is 33.6% (wet basis) (3), and the viscera can be used for recovering oil that could be converted into edible products. Producing edible oil from viscera may add value to catfish viscera, which is currently a processing waste. For the last two decades, interest in the dietary effects of marine n-3 FA has increased because they play a major role in human health (4). Natural fish oils may help maintain heart and vascular health in humans (5).

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Fish oil refining steps include extraction of crude oil, degumming, neutralizing, bleaching, and deodorizing. Both insoluble and soluble impurities are removed through a degumming step (6), and neutralization of crude oil with caustic soda removes FFA. Bleaching removes soap, trace metals, sulfurous compounds, and part of the more stable pigments and pigment-breakdown products, aldehydes, and ketones (6). The purpose of deodorization is to remove residual FFA, aldehydes, and ketones, which are responsible for an unacceptable oil odor and flavor (6–8).

Many species of marine fish have been studied for fish oil production, but little attention has been paid to the production of catfish oil from processing waste. A major question is whether it is feasible to produce edible oil from catfish viscera, a processing waste. Catfish oil is a new product and has not yet been produced on a pilot scale, so it is important to understand the FA composition and quality of the oil at different purification steps. Therefore, the objectives of this study were to produce edible oil from catfish viscera and to determine the effect of purification on the composition of FA and the quality of the catfish visceral oil.

EXPERIMENTAL PROCEDURES

Sample preparation. Catfish viscera were obtained in three separated batches from a local seafood store in Baton Rouge, Louisiana. The viscera were frozen at −20°C for 2 d. The thawed 1-kg portion of viscera was finely ground in a Hobart chopper bowl (Model 84181D; Hobart Corporation, Troy, OH) at 3,450 rpm for 10 min. Water was added (water/ground viscera, 5:1 vol/wt) and the mixture was heated at 70°C for 15 min. The solid particles were separated from the liquid phase by filtering through cheesecloth, and the particles were pressed to remove most of the liquid. The crude oil was separated from the water phase and visceral particles by centrifuging at 5,000 rpm (2,560 × *g*) for 30 min. The resulting crude oil was collected and stored at −20°C for 2 d. Three experimental crude oil extractions were conducted separately. Crude menhaden oil was supplied by Omega Protein Inc. (Reedville, VA). Both crude catfish and menhaden oils were refined as explained below.

The term "neutralized" oil refers to catfish oil that has been sequentially degummed and neutralized; "bleached" oil refers to oil that has been sequentially degummed, neutralized, and

bleached; "deodorized" oil refers to oil that has been sequentially degummed, neutralized, bleached, and deodorized.

Degumming. The method of Dijkstra and Opstal (9) was used with modifications for degumming the crude catfish and menhaden oils. A sample of crude oil (100 g) was removed from storage and placed in a 600-mL beaker that was then heated to 70°C in a water bath. Three milliliters of 3% aqueous citric acid solution was added to the oil, and the mixture was thoroughly mixed at 70°C for 1 min. The oil was then cooled to room temperature and was next centrifuged at 2,560 × g for 10 min to remove impurities.

Neutralization. The degummed oil was neutralized according to AOCS Official Method Ca 9b-52 (10). Sodium hydroxide (12.6 g of 9.5% NaOH solution) was added to 100 g of degummed oil, and the mixture was heated to 65°C for 30 min with constant stirring using a magnetic stirrer bar. The sample was then cooled to room temperature and kept undisturbed for 6 h. After centrifugation at 2,560 × *g* for 10 min, the oil was decanted from the precipitated soap. Fifty milliliters of demineralized water was added to the centrifuged sample to wash out any remaining soap. This operation was repeated three times. The remaining water and impurities were removed by centrifugation at 2,560 × *g* for 10 min.

Bleaching. The neutralized oil was bleached according to the method of Scott and Latshaw (11) with modifications. The neutralized oil sample was heated in a water bath and bleached with 4% (w/w) activated earth (AOCS CS Z1077) at 70°C for 10 min with constant stirring with a magnetic stirrer bar. The activated earth with absorbed impurities was removed from the oil by centrifugation at 2,560 × *g* for 30 min.

Deodorization. The bleached oil was deodorized according to the method of Bitner *et al.* (12) with modifications. The bleached oil was deodorized using a laboratory distillation unit. That consisted of a 500-mL round-bottomed boiling flask with three outlets. One outlet was connected to a vacuum pump, another outlet was connected to a glass distillation column, and the third outlet was sealed with a thermometer inserted. The flask was placed on a heating system. The bleached oil (100 mL) was added to the flask and heated to 100°C for 30 min under vacuum (5 mm Hg). The temperature was controlled manually. Volatile products were condensed in a cooling system installed on the vacuum line, and the distillate was collected from the column.

Esterification of FA. FAME were prepared according to AOAC procedure 969.33 (13). The crude oils and oils from each purification step were each placed into a 50-mL flat-bottomed boiling flask containing approximately 4 mL of methanolic sodium hydroxide (2 g of NaOH dissolved in 100 mL of methanol), and 10 boiling chips were then added to the flask. The condenser and reflux units were attached to the flask, and refluxing took place for 12 min immediately after the addition of 7 mL of boron trifluoride through the condenser. The esterified FA were extracted from the mixture by adding 5 mL of heptane and refluxing for 1 min. The esterified solution was allowed to cool to room temperature. A saturated solution of sodium chloride was added, and the flask was gently rotated. Saturated sodium chloride solution was added until the heptane solution containing FAME reached the neck of the flask. The heptane solution containing FAME was recovered, dehydrated with 1.5 g anhydrous sodium sulfate, and stored under nitrogen in Teflon-capped vials at –20°C until analyzed.

FA analysis. FA analysis was done according to the method of Sathivel *et al.* (3). The FAME were quantified with a Hewlett-Packard (HP) 5890 Series II gas chromatograph equipped with a 7673A autosampler (Agilent Technologies, Palo Alto, CA) and interfaced to an HP 5970 mass selective detector. The GC was equipped with an EZ-Flash fast-temperature programmable column (Thermedics Detection, Inc., Chelmsford, MA). The column phase was RTX-2330 (90% bis-cyanopropyl/10% phenylcyanopropyl polysiloxane) with the following dimensions: 5 m long, 0.25 mm i.d. with 0.2-µm phase thickness. One microliter of FAME was injected in split mode. The head pressure was set at 2 psi, and the split vent flow was 7 mL/min. The injector temperature was 260°C, the column flow rate at 2 psi was 0.68 mL/min, and the split ratio was 10.4:1. The column temperature was held at 50°C for 6 s, ramped from 50 to 260°C at a 1°C/s, and held at 260°C for 84 s. Run time was 5 min. The transfer line temperature was 280°C. The mass selection detector was operated in the selected ion-monitoring mode. FA were identified with retention times obtained from commercial FAME standards (Sigma Company, St. Louis, MO). Concentrations of individual FA from each oil sample were calculated from the standard curves. The internal standard (IS) solution used for quantification of FA contained 1 mg nonadecanoic acid (19:0)/mL heptane. For the recovery studies, 1 mg nonadecanoic acid methyl ester/mL heptane was used as the IS. The calculated concentration of individual FA through the standard curves was quantified as mg FA/g dry-sample weight, taking into account the recovery of IS and sample weight. Three experimental replications (batches) were conducted for both catfish and menhaden oils, each with three extractions and three GC injections per extraction.

FFA analysis. The FFA content of the oils was determined in triplicate by the titration method according to AOAC official method 949.28 (13). FFA were expressed as mg oleic acid/g oil.

Water activity. The water activities of the oils were measured in triplicate using a water activity meter (AW Sprint, Novasina, Switzerland) at 25°C.

Mineral analysis. The mineral content of catfish oil from each purification step was determined in triplicate by the acid digestion method involving microwave technology (CEM microwave, MDS-2000; CEM Corporation, Matthews, NC). A 0.5-g oil sample was placed in a vessel and 6 mL of HNO_3 was added to the vessel. The sealed vessel was then heated. The heating program was run until the digestion process was completed. The sample was then cooled for 5 min, and the digested solution was transferred to a flask and filtered. An inductively coupled argon plasma machine (Model CIROS; SPECTRO Analytical Instruments, Kleve, Germany) was used to analyze Ca, Fe, Mg, and P contents.

TABLE 1
Quantity of Oil (g) Produced from Each Purification Step[a]

Fish oil	Degummed	Neutralized	Bleached	Deodorized
Catfish	810 ± 7.23	713 ± 6.24	683 ± 3.79	657 ± 6.65
Menhaden	890 ± 4.58	807 ± 4.16	783 ± 2.66	763 ± 5.1

[a]Based on an initial 1 kg of crude oil.

*CIE-L*a*b* color measurement*. The color of catfish oils from different processing steps was determined using a spectrophotometer (Model CM 3500d; Minolta Instrument Systems, Ramsey, NJ). The spectrophotometer was set to obtain color values based on a 10° standard observation and D65 illuminants. Results were expressed as L*, a*, and b* values. L* values measure lightness (0 = black and 100 = white); +a* values represent redness and −a* values represent greenness; +b* values represent yellow and −b* values represent blue. Psychrometric color terms involving hue angle (h) [$\tan^{-1}$ (b*/a*)] and chroma (C*) [$(b^{*2} + a^{*2})^{1/2}$] were calculated. The hue angle represents an actual color, and chroma evaluates purity or intensity of the color. An additional commercial refined menhaden oil purchased (from Omega Protein Inc., Reedville, VA) was used as a standard for comparing the total color difference (ΔE). Color difference (ΔE) was calculated by $[(\Delta L^{*2} + \Delta a^{*2} + \Delta b^{*2})]^{1/2}$ using the refined menhaden oil as a reference.

Statistical analysis. All data were analyzed using SAS software (14). ANOVA was performed to determine differences in the FA content attributable to the purification process. Tukey's Studentized range test was performed for *post hoc* multiple comparisons. All analyses were done at $\alpha = 0.05$.

RESULTS AND DISCUSSION

Material balance. The quantities of catfish and menhaden oils produced from each purification step are given in Table 1. Whole catfish viscera includes liver, digestive tract (intestine and stomach), gall bladder, and visceral storage fat. An average of 3.15 kg of whole catfish viscera was required to produce 815 g of crude oil. For 1 kg of crude oil, 657 g of the deodorized catfish oil was produced, compared to 763 g of the deodorized menhaden oil. The major weight loss of oil was observed during the degumming process, which resulted in a loss of 19 and 11% for catfish and menhaden oils, respectively.

FA composition. The compositions of catfish oil and menhaden oil from each purification step are shown in Tables 2 and 3, respectively. The 12 major FA found in catfish oil included 14:0, 16:0, 16:1, 18:0, 18:1, 18:2, 18:3, 20:0, 20:1, 20:2, 20:4, and 22:6. Based on our previous study (3), a low level of EPA was observed in catfish oil and was therefore not evaluated for this study. The saturated FA from crude, degummed, neutralized, bleached, and deodorized catfish oils accounted for 46.6, 42.1, 41.7, 40.0, and 33.6 mg/g of oil, respectively (Table 2). Stearic acid was the predominant FA in catfish oil, accounting for about 50% of all saturated FA. The total unsaturated FA content in catfish oil was almost twofold greater than that of the total saturated FA and amounted to 87.1, 81.1, 79.4, 75.8, and 68.6 mg/g of oil for crude, degummed, neutralized, bleached, and deodorized oils, respectively. Among unsaturated FA, oleic acid was the predominant FA, accounting for almost 30% of total unsaturated FA. The deodorized catfish oil contained 1.21 mg of DHA/g of oil (Table 2), whereas the deodorized menhaden oil contained 18.7 mg of DHA/g of oil (Table 3). The PUFA are considered to be of major importance in terms of human health. Combined n-3 FA (18:3 and 22:6) in the deodorized catfish oil accounted for 4.5% of total FA, whereas in the deodorized menhaden oil they constituted about 20.8% of total FA. On a quantitative basis, the deodorized menhaden oil had approximately five times more combined n-3 (18:3 and 22.6) than did the deodorized catfish oil. The amount of DHA in the deodorized menhaden oil was at least 15.5 times greater than that of the deodorized catfish oil; however, the amount of DHA present in our deodorized menhaden oil was greater than that

TABLE 2
FA Profile (w/w%) of Catfish Oil from Each Purification Step[a]

FA	Crude	Degummed	Neutralized	Bleached	Deodorized
14:0	4.87	3.95	3.78	4.10	2.71
16:0	13.92	12.85	14.83	13.00	16.41
16:1	13.06	11.34	10.55	11.40	9.93
18:0	11.97	13.40	13.93	12.25	16.51
18:1	19.82	24.61	23.60	23.12	16.36
18:2	16.45	18.28	17.75	20.07	24.39
18:3	4.51	3.88	3.75	3.99	2.49
20:0	1.21	0.82	0.94	0.92	0.94
20:1	8.82	6.45	6.76	6.91	7.06
20:2	2.98	2.46	2.52	2.60	2.72
20:4	1.77	1.41	1.20	1.22	0.42
22:6	0.62	0.51	0.39	0.42	0.06

[a]Only 12 FA were analyzed.

TABLE 3
FA Profile (w/w%) of Menhaden Oil from Each Purification Step[a]

FA	Crude	Degummed	Neutralized	Bleached	Deodorized
14:0	9.58	14.67	14.37	11.42	10.47
16:0	14.38	15.57	13.67	13.70	13.86
16:1	8.67	12.75	9.98	9.88	14.86
18:0	10.51	8.87	9.52	9.98	9.93
18:1	13.10	14.24	14.00	14.15	14.93
18:2	5.18	3.48	3.97	3.07	4.95
18:3	5.75	4.40	4.93	5.25	4.04
20:0	1.53	0.89	1.17	1.47	0.91
20:1	6.61	4.26	5.25	5.88	6.28
20:2	0.72	0.33	0.44	0.49	0.74
20:4	5.30	3.84	4.39	4.88	1.52
22:6	18.67	16.70	18.31	19.83	17.51

[a]Only 12 FA were analyzed. FA such as C18:4, C20:5, C22:1, and C22:5, which are readily found in menhaden oil, were not listed.

given by Young (15), who reported that DHA in menhaden oil was between 0.1 and 8.8%.

FA comparisons. In Table 4, saturated, unsaturated, DHA, and n-3 FA of the deodorized oil from catfish viscera are compared with values reported by the USDA for selected fish oils (16). The amounts of total saturated FA present in sardine oil and menhaden oil were 31.3, and 33.3%, respectively, both of which were similar to that of our deodorized catfish oil (32.9%). However, the total saturated FA contents of herring (22.8%) and cod liver (24.6%) oils were lower than that of the deodorized catfish oil. The total unsaturated FA content of the deodorized catfish oil (67.7%) was somewhat similar to that reported for sardine (68.7%) and menhaden (66.7%) oils but lower than those for herring (77.2%) and cod liver (75.4%) oils.

The total n-3 FA (22:6 and 18:3) in the deodorized catfish oil accounted for 4.5% of the total FA content, whereas they accounted for 13.3, 13.0, 12.4, and 5.3% in sardine, cod liver, menhaden, and herring oils, respectively (Table 4). The predominance of 18:2n-6 in catfish oil compared to menhaden oil may be attributed to the fish feed, especially if it is made from soy products. Diet has a major effect on the FA composition of lipids (17). Fish can accumulate n-3 FA in lipids when the diet contains either linolenic acid (18:3n-3) or DHA (17). Marine plankton, a major food source for marine fish, contains a high quantity of PUFA. Ackman and Sipos (18) examined a number of fish oil FA contents and noted that the FA in marine fish were similarly found in phytoplankton. The FA composition of fish lipids was highly dependent on a number of factors, especially the diets of the fish (19,20).

FFA content. FFA were gradually removed throughout the oil purification process (Table 5). Crude oil contained the highest amount of FFA (4.53%), whereas the final deodorized oil contained the lowest level (3.25%). The neutralization step removed a minute amount of FFA compared to other purification steps. This may be due to the foam generated during neutralization, which may have lessened the efficiency of the process. The decreases in FFA during the deodorization process may have been due to the vaporizability of FFA. Considerable amounts of FFA were vaporized during distillation (8,21). An acceptable level of FFA in refined fish oil is between 1.8 and 3.5% (15). Under appropriate processing conditions, the FFA can be reduced up to 50% during deodorization (15).

Water activity. The water activity gradually decreased from crude oil to deodorized oil. The highest water activity was found in crude oil (0.838), and this was reduced to 0.555 after deodorization (Table 5).

Minerals. Table 5 shows the mineral content of catfish oil at different purification steps. Mg, Ca, Fe, and P were major minerals found in crude catfish oil; other minerals were not reported since they were present only in trace amounts. The purification steps reduced the mineral content of catfish oil. Crude catfish oil contained a high amount of phosphorus

TABLE 4
Comparisons of FA (%) of Catfish Oil with Major Marine Fish Oils

FA	Sardine[a]	Menhaden[a]	Herring[a]	Cod liver[a]	Purified catfish[b]
Saturated	31.3	33.3	22.8	24.6	32.9
Unsaturated	68.7	66.7	77.2	75.4	67.7
DHA	11.1	9.8	4.5	11.9	1.1
n-3 (22:6 + 18:3)	13.3	12.4	5.3	13.0	4.5

[a]Reference 16.
[b]Our results.

TABLE 5
FFA, Water Activity, and Selected Minerals in Catfish Oils from Different Purification Steps

Properties	Crude	Degummed	Neutralized	Bleached	Deodorized
FFA (%)	4.53 ± 0.25	4.28 ± 0.18	4.25 ± 0.41	3.80 ± 0.06	3.25 ± 0.1
Water activity	0.838 ± 0.01	0.756 ± 0.02	0.696 ± 0.03	0.651 ± 0.02	0.555 ± 0.02
Magnesium (ppm)	4.99 ± 0.15	4.64 ± 0.1	—[a]	—	—
Calcium (ppm)	10.7 ± 0.21	—	—	—	—
Iron (ppm)	0.64 ± 0.02	—	—	—	—
Phosphorus (ppm)	107.6 ± 2.1	99.2 ± 0.02	—	—	—

[a]Indicates a trace amount.

TABLE 6
Color Characteristics of Catfish Oils from Different Purification Steps[a]

Oil	L*	a*	b*	C*	h	ΔE[b]
Crude	3.09 ± 0.01	–0.809 ± 0.02	1.79 ± 0.2	1.96 ± 0.01	114.37 ± 1.3	6.20 ± 0.02
Degummed	2.99 ± 0.02	–0.84 ± 1.2	2.04 ± 0.03	2.2 ± 0.15	112.34 ± 0.6	6.41 ± 0.03
Neutralized	1.29 ± 0.01	–0.48 ± 0.04	1.32 ± 0.02	1.40 ± 0.01	109.88 ± 0.02	7.65 ± 0.01
Bleached	5.15 ± 0.02	–1.21± 0.05	0.12 ± 0.01	1.22 ± 0.03	174.28 ± 0.03	3.59 ± 0.01
Deodorized	2.99 ± 0.02	–1.25 ± 0.07	1.29 ± 0.05	1.79 ± 0.01	134.18 ± 0.01	6.04 ± 0.01
Menhaden oil[a]	8.45 ± 0.01	–1.20 ± 0.02	–1.32 ± 0.01	1.78 ± 0.02	227.34 ± 1.00	0

[a]L* values mean lightness; a* values measure redness/greenness; b* values measure yellowness/blueness; h represents hue angle (in deg); and C* indicates chroma.
[b]Refined menhaden oil was used as a standard for color difference (ΔE) calculation.

(107.6 ppm), and it was reduced to 99.2 ppm after the degumming process. Ca and Fe were removed after the degumming process, whereas Mg and P were removed after the neutralization process. Crude fish oils are expected to contain a certain amount of minerals since phospholipids are reported to carry minerals into oil (22). Minerals that are complexed by phospholipids would presumably be removed by degumming and alkali refining since these steps remove phospholipids (23). Degumming and neutralization would reduce the phosphorous, iron, magnesium, and calcium in the oil to trace levels (9,24).

Color. The color characteristics of catfish oil at different processing steps compared with those of the refined menhaden oil are presented in Table 6. The refined menhaden oil and bleached catfish oil were lighter (higher L*) than crude, degummed, neutralized, and deodorized oils. No specific pattern was observed for color lightness changes during purification. Bleaching did increase the color lightness of catfish oil. All catfish oils had a negative a* value, indicating a slight greenish color, and a positive b* (yellowish) value. The lowest b* value was observed for the bleached catfish oil. The total color difference (ΔE) values of all catfish oils were greater than 1.0. Therefore, they may be perceptibly different from the refined menhaden oil from the consumers' point of view. Hue angle values of all catfish oils were higher than 90°. Oils with a hue angle value between 90 and 180° were more greenish-yellow in color. Commercially refined menhaden oil had the highest hue angle (227.34°); its color was observed to be a very light greenish-blue.

The color lightness (L*) of catfish oil decreased slightly after degumming and neutralization. When the citric acid solution was added during the degumming process, and when caustic soda was added during the neutralization process, the oil was observed to turn into a cloudy mixture and its color became dull. During the bleaching operation, the bleaching earth adsorbed pigments, water, minerals, and the remaining soap. As a result, bleached oil had a higher lightness (L*) value. However, the L* value of deodorized oil was lower than that of bleached oil. This difference may be imparted by the thermolytic products of heat decomposition from a more susceptible unsaturated FA.

This research has demonstrated a processing procedure that can be used to extract and purify oil recovered from catfish viscera. The results will give useful information for optimizing unit operations for catfish oil extraction and purification should the oil industry choose to produce edible catfish oil from a processing waste. Further studies on storage stability, quality changes during rendering, safety, and sensory evaluation are necessary to determine the potential of this oil for human consumption.

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