Original article

Carp (*Cyprinus carpio*) oils obtained by fishmeal and ensilage processes: characteristics and lipid profiles

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Summary   Fish oil is an important source of long-chain ω-3 polyunsaturated fatty acid. The common carp (*Cyprinus carp*) is a major fish species in world aquaculture production. This study aimed towards obtaining carp viscera oil by ensilage and fishmeal processes. Characteristics of crude oils obtained were also compared with oil extracted by Bligh and Dyer methods. Crude oils obtained by the three processes resulted in significant difference ($P < 0.05$) for free fatty acids, peroxide, thiobarbituric acid and Lovibond colour values; however, iodine and saponification values were not significantly affected ($P > 0.05$). Recovery yield of crude oils was approximately 85% in relation to carp viscera oil. Carp crude oils obtained by the ensilage and fishmeal processes resulted in high unsaturated and polyunsaturated fatty acid contents (67.4%), and $\omega3/\omega6$ ratios around 1.15. These oils are applicable in fish diets; however, crude oils require refinement for human consumption.

Keywords   Colour, fatty acids, fish, lipids, oils.

Introduction

The common carp (*Cyprinus carpio*) is a major fish species in world aquaculture production, farmed exclusively in traditional extensive or semi-intensive ponds, where fish growth is highly dependent (50% or more) on natural food (plankton and benthos) (Vandeputte *et al.*, 2008). Among several alternative food sources, carp residues present elevated nutritional quality and high potential for animal production (Geron *et al.*, 2007).

Fish silage and fishmeal are sources of proteins and products of high biological value for animal feeding. They may be produced from fish species that are sub-utilised in the fish industry, or from marine fishing, commercial fish waste and industry residues. These are considered low-quality raw materials, and if unused may cause environmental, sanitary and economical problems (Geron *et al.*, 2007). Fish oil can be obtained from fish residues through the acid ensilage process (Reece, 1981; Zhou *et al.*, 1995) or the fishmeal process (Arvanitoy-anni & Kassaveti, 2008).

Dietary lipids play an important role as an energy source for fish growth, as well as carriers for fat-soluble vitamins. Fish oil contains high quantities of long-chain $\omega3$ ($\geq C_{20}$)-polyunsaturated fatty acids ($\omega3$-PUFA) nec-

essary for marine fish, similarly to the nutritional composition of their natural diet during the marine phase of their life cycle (Miller *et al.*, 2007; Peng *et al.*, 2008). Recently, studies have evaluated the replacement of fish oil in aquafeeds manufacturing with vegetable and animal oils, and oils obtained from cell organisms such as microalgae. However, diets containing replacement oils such as terrestrial plant and animal oils have reduced eicosapentanoic acid (EPA) and docosahexanoic acid (DHA) concentrations. Extracting oil from single-cell biomass also increases production costs of commercial aquafeeds (Miller *et al.*, 2007). Thus, fish oil is an economical alternative in relation to terrestrial plant and animal oils for use in aquaculture, because they represent a lipid source with elevated PUFA content.

There is also a growing world market for inclusion of high-quality fish oils in food and dietary supplements for human consumption. Presently, most available fish oils are produced by pressing/heating of headed and gutted pelagic fish, to release oil concentrated mainly in the fish's flesh (Zhong *et al.*, 2007).

Few reports have been published on carp crude oil characteristics and fatty acid composition (Rasoarahoma *et al.*, 2004; Druzian *et al.*, 2007; Guler *et al.*, 2008). The aim of this work was to obtain crude carp (*Cyprinus carpio*) viscera oil by acid ensilage and fishmeal processes, and subsequently compare oil characteristics and fatty acids profiles.

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Materials and methods

Raw material

The utilised raw material was carp (*Cyprinus carpio*) viscera from a commercial fish-processing plant; these residues were transported and immediately frozen in plastic containers at the Unit Operations Laboratory/FURG.

Acid ensilage process methodology

For elaboration of the ensiling, 35 kg of carp viscera were thawed overnight at ambient temperature, minced in a meat grinder, conditioned in 5 L plastic buckets and acidified with glacial acetic acid 10% (v/w); also, 250 mg kg^{-1} antioxidant tertiary butyl hydroxyquinone (BHT) were added for reducing lipid oxidation. During the initial liquefaction period, silage was agitated occasionally. Temperature during liquefaction ranged from 25 to 35 °C. After a 15-day period, silage was sifted in a tyler sieve no. 14 for removal of spines and scales, for subsequent centrifugation.

Silage temperature was increased to 50 °C in a thermostatic bath (QUIMIS model Q-304-264; Scientific Equipments, Diadema, Brazil); afterwards, the material was centrifuged for 20 min at $7000 \times g$ (Centrifuge model SIGMA 6–15, D-37250; Sigma, Osterode, Germany). The resulting product included three fractions: a bottom solid cake, an aqueous middle layer and an upper oil layer. Oil fractions were separated and stored in an amber bottle at −20 °C for analysis.

Fishmeal process methodology

Raw viscera processing was carried out by extracting crude oil on a laboratory scale, under conditions similar to commercial fish-processing plants, through the following steps: grinding, cooking, screening and centrifugation.

For elaboration of the fishmeal, 35 kg of carp viscera were used, being thawed overnight at ambient temperature and minced in a meat grinder. Afterwards, the raw material was cooked at 95–100 °C during 30 min. It was then sifted in a tyler sieve no. 14 for removal of spines and scales. Separation of fractions through centrifugation and storage of the oil fractions was carried out under conditions similar to the ensilage process.

Crude oil yield

Yield was expressed as percentage of recovered crude oil in relation to carp viscera crude oil. Yield was calculated as presented in eqn 1:

$$\% \text{ Yield} = \frac{W_{RCO}}{W_{CCO}} \times 100 \qquad (1)$$

where W_{RCO} is the weight of recovered crude oil (kg) obtained by ensilage and W_{CCO} is the carp viscera crude oil (kg) extracted by the Bligh and Dyer method.

Analytical methodology

The following parameters were determined in duplicate for the raw material: Bligh and Dyer lipid (Bligh & Dyer, 1959), moisture (method 925.10) and protein (method 960.52) contents. Determination was carried out according to Association of Official Analytical Chemists (AOAC) (1995) methodologies.

For crude oils, the following parameters were determined in triplicate, according to American Oil Chemists' Society (AOCS) (1980) methodologies. Free fatty acids method (FFA, Ca 5a-40) was based on a sodium hydroxide solution titration (employing phenolphthalein as an indicator) of the oil, suitably diluted with an ethyl alcohol–ethyl ether mixture. Results are expressed in % oleic acid. Peroxide value method (PV, Cd 8–53) was based on a sodium thiosulphate solution titration of oil diluted with an acetic acid–chloroform mixture followed by treatment with potassium iodide. Results are expressed as meq kg^{-1} oil. Iodine value method (IV, Cd 1–25) was based on a sodium thiosulphate solution titration (employing amid as an indicator) of chlorophorm-diluted oil treated with Wij's solution and potassium iodide. Results are expressed as cg I$_2$ g^{-1}. Saponification value method (SV, Cd 36–76) was based on a hydrochloride acid titration (employing phenolphthalein as an indicator) of oil diluted with a potassium hydroxide solution. Results are expressed in mg KOH g^{-1}.

Thiobarbituric acid value (TBA) was determined according to methodologies proposed by Vyncke (1970), utilising the spectrophotometric method (Spectrophotometer model QUIMIS Q-108DRM; Scientific Equipments, Diadema, Brazil), calculated from a standard curve obtained by reacting known amounts of 1,1,3,3 tetramethoxypropane with TBA. Results are expressed as mg malonaldehyde kg^{-1} oil.

Oil colour was measured using the Lovibond method (Lovibond Colour Staler Tintometer; model F, Amesbury, UK), by fixing the colour yellow in 30 units and varying the colour red, as described by Windsor & Barlow (1984). Samples were placed in a cube and inserted into the provided space in the tintometer. The oil's reflected colour was determined by combining different red and yellow slides, until the reflected colour matched the slide colour combination. Yellow and red values of the slide colour were noted and repeated three times for each sample.

Fatty acid profiles were determined by preparation of methyl esters as described by Metcalfe & Schimitz (1966).

Fatty acid methyl esters (FAME) were identified by gas chromatography (GC) (chromatographer model VARIAN-3400 CX, Palo Alto, CA, USA) equipped with a DB-17 J&W Scientific (50% phenyl methylpolysiloxane) capillary column. Fatty acid esters analysis was carried out in duplicate by injecting 1.0 µL, split ratio 1:50, into the capillary column (30 m × 0.25 mm, 0.25 µm film in thickness). GC setting conditions were as follows: injection temperature 250°C and flame ionisation detector temperature 300 °C, helium gas carrier flow rate 1.0 mL min^{-1}, linear speed 24 cm s^{-1} and oven temperature held at 100 °C for 1 min, then increased to 160 °C at 6 °C min^{-1} and held at 230 °C at 6 °C min^{-1}. FAME were identified by direct comparison of the retention times with standards (SUPELCO^{TM} 37, Bellefonte, PA, USA), and were quantified as the percentage area of each FAME mixture.

Statistical analysis

Characterization values for the carp crude oils, as well as fatty acids profile, were compared using Tukey's HSD test of differences of means (Box *et al.*, 1978), with STATISTICA 6.0 (Statsoft, Tulsa, OK, USA). Values were considered significant at a $P < 0.05$ level.

Results and discussion

Raw material characteristics

Characterization results of the raw material (carp viscera) used in the fishmeal and ensilage processes were: moisture content of 75 ± 2%, protein content of 12 ± 1% and lipid content of 13 ± 1%.

Results presented for the raw material utilised in this work were compared with Druzian *et al.* (2007), who studied the proximate composition of carp; these authors reported moisture, protein and lipid content results of approximately 72.2%, 14.8% and 9.9% respectively. It could be verified that the carp viscera in this study presented lower moisture and protein contents and higher lipid content than those reported by these authors. These variations are closely related to the utilised raw material; Druzian *et al.* (2007) studied whole fish, while in this study only viscera were used. Also, natural factors such as food shortage, or physiological factors such as spawning or migrations, influence the chemical composition, occurring a higher variation of the lipid fraction (Borlongan & Benitez, 1992; De Silva *et al.*, 1997).

Crude oils obtained by the fishmeal and ensilage processes were recovered at 11.0 g per 100 g$_{viscera}$ and 11.1 g per 100 g$_{viscera}$, respectively, and yields obtained for crude oils (eqn 1) recovered by both processes were approximately 85%. Reece (1980) studied oil recovered from fish silage, encountering a maximum oil recovery level of approximately 11.4 g per 100 g$_{viscera}$, representing an oil content similar to the one found in this study.

Fish oil characteristics

Characteristics of carp crude oil extracted by the Bligh and Dyer method and oils obtained by the fishmeal and ensilage processes are presented in Table 1.

Significant differences ($P < 0.05$) for FFA, PV, TBA and Lovibond colour (LC) values for crude oil extracted by the Bligh and Dyer method and oils obtained by the two processes are shown in Table 1.

Regarding FFA content, a higher value could be observed in crude oil obtained by the ensilage process (Table 1). FFA content increase of oil from fish silage, recovered by centrifugation, has been shown to be associated mainly with FFA release from solid material during fish liquefaction (Reece, 1981). Much of the initial FFA is due to the action of endogenous enzymes present in viscera prior to acidification. Zhou *et al.* (1995) studied the FFA content of lipids during ensilage acidification of mince herring; in this study, the maximum FFA level was about 6%.

Crude oil extracted by the Bligh and Dyer method presented FFA content higher to crude oils obtained by fishmeal process (Table 1). The Bligh and Dyer

Table 1 Characterization of the carp crude oils obtained by Bligh and Dyer extraction and by two processes

Indices	Bligh dyer extraction*	Fishmeal process*	Ensilage process*
FFA (% oleic acid)	4.26 ± 0.02[a]	3.35 ± 0.02[b]	6.63 ± 0.01[c]
PV (meq peroxide kg^{-1})	3.50 ± 0.02[a]	3.38 ± 0.05[b]	3.68 ± 0.02[c]
TBA (mg malonaldehyde kg^{-1} oil)	1.22 ± 0.02[a]	6.69 ± 0.05[b]	1.17 ± 0.01[c]
IV (cg I$_2$ g^{-1})	115 ± 2[a]	115 ± 1[a]	114 ± 2[a]
SV (mg KOH g^{-1})	205 ± 1[a]	204 ± 2[a]	205 ± 3[a]
LC red (30 yellow)	10.5 ± 0.1[a]	5.0 ± 0.1[b]	16 ± 0.2[c]

Different superscript letters in the same line are significantly different ($P < 0.05$).
FFA, free fatty acids; PV, peroxide value; TBA, thiobarbituric acid; IV, iodine value; SV, saponification value; LC, lovibond colour.
*Mean value ± standard error (in triplicate).

extraction method does not use temperatures as high as the fishmeal process. Therefore, the small FFA content in the crude oil obtained by the fishmeal process can be attributed to the volatilization of low molecular weight FFA, due to the higher temperature used during this process. FFA content of fishmeal oil presented in Table 1 is similar to the one found by Chantachum *et al.* (2000). These authors studied the separation and quality of fish oil from precooked and non-precooked tuna heads, obtaining FFA of approximately 3% for oils separated from non-precooked heads at 95 °C during 30 min.

Austreng & Gjerfsen (1981) studied the effect of FFA level as a quality criterion of fats and oils in salmon diets, utilising capelin oils with varying FFA contents (0.1–11.0%). The authors verified that there was no relationship between mortality and FFA level in the fish oil; protein and fat digestibility and gross energy in the diet did not seem to be influenced by the FFA content, however several FFA experiments in fish reported that diets with low FFA content produced fish with higher body fat content. The reported publications show a great divergence of results within and between species, making it impossible to generalise the use of FFA in feeds (Stickney & Andrews, 1972; Viola & Amidan, 1978). In this manner, the effect of FFA level as a quality criterion of fats and oils in fish diets should be evaluated for each species in this study.

In terms of PV, it can be observed in Table 1 that this value is higher in crude oils obtained by the Bligh and Dyer extraction and ensilage process. PV indicates lipid oxidation; such oxidation is important for defining the deterioration state, especially the presence of low molecular weight compounds proceeding from lipid degradation (Boran *et al.*, 2006; Manral *et al.*, 2008). The PV is in accordance with the use required for quality and acceptability of oils for human consumption, of 8 meq kg^{-1} oil (Boran *et al.*, 2006). PV for carp crude oil was below the results cited by Ganga *et al.* (1998), who encountered 4.02 meq kg^{-1} for sardine oil.

In this study, thiobarbituric acid (TBA) value of oil obtained by the fishmeal process was higher to oils obtained by Bligh and Dyer extraction and ensilage (Table 1). TBA value is widely used as an indicator of the lipid oxidation degree, with decomposition of hydroperoxides and formation of secondary products of oxidation such as aldehydes, ketones and alcohols (Boran *et al.*, 2006). The use of high temperatures during the fishmeal manufacturing process potentially causes higher oxidation. However, TBA value is in agreement with the use required for quality and acceptability of oils for human consumption of 7–8 mg malonaldehyde kg^{-1} oil (Boran *et al.*, 2006).

Lovibond colour in Bligh and Dyer extraction and ensilage oils was higher to the one obtained in the fishmeal process (Table 1). Oil pigmentation during ensiling is shown to be caused by the release of the acid hydrolysis product of haemoglobin, heme (Reece, 1981). Proteins are considered to be the prime target of FFA and become highly unextractable in their presence. It is generally believed that the interactions between proteins and FFA occur primarily through electrostatic, Van der Walls, hydrogen bonding and hydrophobic forces (Sequeira-Munoz *et al.*, 2006). In ensilage oil an increase in FFA content is verified, with the formation of lipid–protein complexes and a consequent colour increase. Bligh and Dyer extraction and ensilage are methods that do not use temperatures as high as the fishmeal process. In this manner, the low LC observed for crude oils obtained by the fishmeal process can be attributed to the larger removal of volatiles and destruction of pigments, which occurred due to the higher temperature employed during this process. Also, Bligh and Dyer extraction oils present FFA content higher than crude oils obtained by the fishmeal process, with the formation of lipid–protein complexes and a consequent increase in colour.

Iodine and saponification values were not significantly affected ($P > 0.05$), according to Table 1. These values are associated, respectively, to the unsaturated degree of fatty acids and the average weight of fatty acids esterified to glycerol. Therefore, oils presented similar compositions in terms of the unsaturated degree of fatty acids and saponified matter content. This can be attributed to the fact that these oils are originated from the same raw material. In this manner, the processes used for obtaining carp oil did not affect fatty acid composition. IV and SV for crude oils obtained by the Bligh and Dyer extraction and the fishmeal and ensilage processes were not within the range cited by Bernardini (1986) for different marine fish, of 120–190 cg I$_2$ g^{-1} and 160–190 mg KOH g^{-1} respectively. Such variations are due to water temperature and salinity (Borlongan & Benitez, 1992; De Silva *et al.*, 1997).

Fatty acid profiles

Table 2 presents the fatty acid profiles of fishmeal and ensilage oils. In this table, through Tukey's HSD tests of differences of means, it can be verified that crude oils obtained by both processes were not significantly different ($P < 0.05$).

In Table 2, the major fatty acids identified in the carp oils were C18:1ω9 (oleic), C16:0 (palmitic), C16:1 (palmitoleic), C18:2ω6 (linoleic), C18:3ω3 (linolenic), constituting approximately 67% of the total fatty acids of fishmeal and ensilage oils.

Marine fish require ω3-PUFA, in particular EPA and DHA but also arachidonic acid (AA), considering that the elongation and desaturation capacity of α-linolenic acid (ALA 18:3ω3) is insufficient to meet the needs of the fish (Miller *et al.*, 2007). In Table 2, it can be observed that DHA and AA contents in fishmeal and

Table 2 Fatty acid profiles of carp crude oils obtained by two processes

Fatty acids	Fishmeal process (%)*	Ensilage process (%)*
C11:0	0.08 ± 0.01^a	0.08 ± 0.01^a
C12:0	0.30 ± 0.01^a	0.31 ± 0.02^a
C13:0	0.24 ± 0.01^a	0.25 ± 0.01^a
C14:0	3.79 ± 0.02^a	3.82 ± 0.03^a
C15:0	1.20 ± 0.03^a	1.20 ± 0.01^a
C16:0	16.19 ± 0.03^a	16.14 ± 0.04^a
C17:0	1.11 ± 0.01^a	1.10 ± 0.02^a
C18:0	3.15 ± 0.01^a	3.17 ± 0.02^a
C20:0	0.25 ± 0.01^a	0.25 ± 0.01^a
C21:0	0.17 ± 0.02^a	0.18 ± 0.01^a
C22:0	0.18 ± 0.02^a	0.17 ± 0.02^a
C23:0	0.20 ± 0.01^a	0.20 ± 0.01^a
C14:1ω5	0.17 ± 0.01^a	0.18 ± 0.01^a
C15:1ω5	0.42 ± 0.01^a	0.43 ± 0.02^a
C16:1ω7	8.08 ± 0.04^a	8.04 ± 0.03^a
C17:1ω7	1.27 ± 0.03^a	1.30 ± 0.01^a
C18:1ω9	26.05 ± 0.03^a	26.01 ± 0.02^a
C20:1ω9	1.83 ± 0.05^a	1.85 ± 0.03^a
C22:1ω9	0.08 ± 0.01^a	0.08 ± 0.02^a
C 24:1 ω9	3.99 ± 0.02^a	3.99 ± 0.01^a
C18:2 ω6	9.48 ± 0.01^a	9.47 ± 0.01^a
C 18:3ω6	0.34 ± 0.02^a	0.32 ± 0.01^a
C18:3 ω3	7.16 ± 0.01^a	7.17 ± 0.01^a
C 20:2ω6	0.43 ± 0.02^a	0.42 ± 0.01^a
C20:3 ω6	0.42 ± 0.02^a	0.43 ± 0.02^a
C20:3ω3	1.43 ± 0.01^a	1.44 ± 0.02^a
C20:4ω6 (AA)	1.24 ± 0.02^a	1.22 ± 0.01^a
C20:5ω3 (EPA)	3.82 ± 0.01^a	3.81 ± 0.2^a
C22:2ω6	0.02 ± 0.01^a	0.02 ± 0.01^a
C22:6ω3 (DHA)	1.20 ± 0.02^a	1.20 ± 0.01^a
Σ u[†]	5.73 ± 0.03	5.71 ± 0.02

Different superscript letters in the same line are significantly different ($P < 0.05$).
AA, arachidonic acid; EPA, eicosapentanoic acid; DHA, docosahexanoic acid.
*Mean value ± standard error (n = 3 replicate).
[†]Sum of unidentified.

Table 3 Lipid classes in carp crude oils obtained by two processes

Fatty acids	Fishmeal process*	Ensilage process*
Σ SFA (%)	26.86 ± 0.19	26.87 ± 0.20
Σ MUFA (%)	41.89 ± 0.20	41.88 ± 0.15
Σ PUFA (%)	25.54 ± 0.15	25.50 ± 0.13
ω3 (%)	13.61	13.62
ω6 (%)	11.93	11.88
ω3/ω6	1.14	1.15

Σ SFA, sum of unsaturated; Σ MUFA, sum of monounsaturated; Σ PUFA, sum of polyunsaturated.
*Mean value ± standard error (n = 3 replicate).

silage oils were lower than values cited by Rasoarahoma *et al.* (2004) for carp oil, 6.7% and 5.9% respectively; and were higher than those cited by Druzian *et al.* (2007), of 1.02% and 1.16% respectively. However, EPA content was higher than those cited by these authors, who presented contents of 3.4% and 2.4% respectively. The cause for variation in results observed for PUFAs, EPA, DHA and AA is likely to be the feeding habits of fish. PUFA percentages, such as EPA and DHA, in fish muscle, depend on their diet. Variations in fatty acid composition might be related to changes in fish nutritional habits (Borlongan & Benitez, 1992; De Silva *et al.*, 1997; Arts *et al.*, 2001).

Palmitic acid was the most abundant saturated fatty acid (SFA) (Table 2). In general, fish are relatively low in SFA (< 30%), with the exception of certain species. It can be verified in Table 2 that SFA content was around 28.9%. Oleic acid was identified as the most abundant monounsaturated fatty acid (MUFA) in carp fishmeal and silage oils. When researching the effect of climate on fatty acids composition, Guler *et al.* (2008) and Kolakowska *et al.* (2000) found similar results for palmitic acid and oleic acid in carp oil. These fatty acids are an important source of energy. Palmitoleic acid was the second most abundant MUFA for fishmeal and silage oils. High levels of oleic, palmitoleic and arachidonic acids have been reported as a characteristic property of freshwater fish oils (Andrade *et al.*, 1995). MUFA contents in carp oil (41.9%) were higher than SFA in fishmeal and silage (approximately 26.9%). The reported publications of Guler *et al.* (2008) and Druzian *et al.* (2007) found similar results for carp oil in the winter, of 41.1% and 43.12% respectively.

Table 3 presents lipid classes in carp crude oils obtained by two processes. In this study, the PUFAs content in fishmeal and silage oils (approximately 25.5%) were higher to the ones found by Druzian *et al.* (2007), of 23%. PUFAs content has been reported to vary over a wide range: 11.6–15.7% (Bieniarz *et al.*, 2000) and 32.3–34.5% (Geri *et al.*, 1995) of total fatty acids. MUFAs and PUFAs contents of fishmeal and silage oils represent around 67.4% of the total fatty acids, making it a rich source of unsaturated and polyunsaturated fatty acids.

Differences in dietary concentrations of AA, EPA and ω3/ω6 ratio may influence physiology and health status via the complex effects of synthesis of various eicosanoics from AA and EPA (Miller *et al.*, 2007). Fatty acid composition, essentially PUFAs, and the ω3/ω6 series ratio in different tissues, are affected by salinity. This ratio is more elevated in marine fish than in freshwater fish. The ω3-PUFAs/ω6-PUFAs ratio in total lipids of freshwater fish varies mainly between 0.5 and 3.8, whereas for marine fish variation may range from 4.7 to 14.4. The ω3/ω6 ratio has been suggested to be a useful indicator of fish oil relative nutritional values (Guler *et al.*, 2008). In this study, the ω3 and ω6 contents in fishmeal and silage oils were 13.61%,

13.62% and 11.93%, 11.88% respectively. The $\omega 3/\omega 6$ ratios in fishmeal and silage oils were approximately 1.14 and 1.15 respectively, superior to the ratio cited by Guler *et al.* (2008) for carp oils, of approximately 1.06.

Conclusions

Characteristics of carp crude oil obtained from visceral carp (*Cyprinus carpio*) by the fishmeal and ensilage processes and extracted by the Bligh and Dyer method presented respectively: FFA values of 3.35%, 6.63% and 4.26%; PV of 3.38, 3.68 and 3.50 meq peroxide kg^{-1}; TBA values of 6.69, 1.17 and 1.22 mg malonaldehyde kg^{-1} oil; and LC at 5 red and 30 yellow, 16 red and 30 yellow and 10.5 red and 30 yellow. Crude oils by the three processes presented significant difference ($P < 0.05$) for FFA, PV, TBA and LC. IV and SV presented no significant difference ($P < 0.05$) between the two processes and the Bligh and Dyer extraction. Recovery of carp crude oils obtained by fishmeal and ensilage processes was 11 g per 100 g viscera, and yield was approximately 85% in relation carp viscera oil.

The major fatty acids identified in the carp crude oils were C18:1$\omega 9$ (oleic), C16:0 (palmitic), C16:1 (palmitoleic), C18:2$\omega 6$ (linoleic), C18:3$\omega 3$ (olinolenic), constituting approximately 67% of the total fatty acids of crude oils obtained by the ensilage and fishmeal processes, as well as unsaturated and polyunsaturated fatty acids (MUFA + PUFA), with an approximately 67.4% content of the total fatty acids. The carp crude oils presented quality for use in aquafeeds; however, they require purification in order to generate a product acceptable for human consumption.

Acknowledgments

References

American Oil Chemists' Society (AOCS) (1980). *Official and Tentative Methods of American Oil Chemists' Society*, 3rd edn. Champaig, IL, USA: AOCS.

Andrade, A.D., Rubira, A.F., Matsushita, M. & Souza, N.E. (1995). Omega-3 fatty acids in freshwater from South Brazil. *Journal of the American Oil Chemists' Society*, **72**, 1207–1210.

Arts, M.T., Ackman, R.G. & Holub, B.J. (2001). 'Essential fatty acids' in aquatic ecosystems: a crucial link between diet and human health and evolution. *Canadian Journal of Fisheries and Aquatic Sciences*, **58**, 122–137.

Arvanitoyanni, I.S. & Kassaveti, A. (2008). Fish industry waste: treatments, environmental impacts, current and potential uses. *International Journal of Food Science & Technology*, **43**, 726–745.

Association of Official Analytical Chemists (AOAC) (1995). *Official Methods of Analysis*, 16th edn. Arlington, VA: AOAC.

Austreng, E. & Gjerfsen, T. (1981). Fish oils with different contents of free fatty acids in diets for rainbow trout fingerlings and salmon parr. *Aquaculture*, **25**, 173–183.

Bernardini, E. (1986). *Tecnologia de aceites y grasas*. Pp. 234–261. Madrid, Span: Alhambra S.A.

Bieniarz, K., Koldras, M., Kaminski, J. & Mejza, T. (2000). Fatty acids and cholestorol in some freshwater fish species in Poland. *Folia Universitas Agriculture Stetinensis*, **27**, 21–44.

Bligh, E.G. & Dyer, W.J. (1959). A rapid method of total lipid extraction and purification. *Canadian Journal of Biochemistry and Physiology*, **27**, 911–917.

Boran, G., Karaçam, H. & Boran, M. (2006). Changes in the quality of fish oils due to storage temperature and time. *Food Chemistry*, **98**, 693–698.

Borlongan, I.G. & Benitez, L.V. (1992). Lipids and fatty acid composition of milkfish (*Chanos chanos* Forsskal) grown in freshwater and seawater. *Aquaculture*, **104**, 79–89.

Box, G.E.P., Hunter, W.G. & Hunter, J.S. (1978). *Statistics for Experiments: An Introduction to Design, Data Analysis and Model Building*. New York: John Wiley & Sons.

Chantachum, S., Benjakul, S. & Sriwirat, N. (2000). Separation and quality of fish oil precooked and non-precooked tuna heads. *Food Chemistry*, **69**, 2089–2294.

De Silva, S.S., Gunasekera, R.M. & Austin, C.M. (1997). Changes in fatty acid profile of hybrid red tilapia *Oreochromis mossambicus* X *O. niloticus*, subjected to short-term starvation and comparison with changes in seawater raised fish. *Aquaculture*, **153**, 273–290.

Druzian, J.I., Marchesi, C.M. & Scamparini, A.R.P. (2007). Fatty acid profile and proximate composition of carp (*Cyprinus carpio*) feed artificial food and pig manure. *Ciência Rural*, **37**, 539–544.

Ganga, A., Nieto, S., Sanhuez, J., Romo, C., Speisky, H. & Valenauela, A. (1998). Concentration and stabilization of n-3 polyunsaturated fatty acids sardine oil. *Journal of the American Oil Chemists' Society*, **75**, 733–736.

Geri, G., Poli, B.M., Gualtieri, M., Lupi, P. & Parisi, G. (1995). Body traits and chemical composition of muscle in the common carp (*Cyprinus carpio*) as influenced by age and rearing environment. *Aquaculture*, **129**, 329–333.

Geron, L.J.V., Zeoula, L.M., Vidotti, R.M. *et al.* (2007). Chemical characterization, dry matter and crude protein ruminal degradability *in vitro* intestinal digestion of acid and fermented silage from tilapia filleting residue. *Animal Feed Science and Technology*, **136**, 226–239.

Guler, G.O., Kiztanir, B., Aktumsek, A., Citil, O.B. & Ozparlak, H. (2008). Determination of the seasonal changes on total fatty acid composition and $\omega 3/\omega 6$ ratios of carp (*Cyprinus carpio* L.) muscle lipids in Beysehir Lake (Turkey). *Food Chemistry*, **108**, 689–694.

Kolakowska, A., Szcygielski, M., Bienkiewicz, G. & Zienkowicz, L. (2000). Some of fish species as a source of n-3 polyunsaturated fatty acids. *Acta Ichthyologica Psicatoria*, **30**, 59–70.

Manral, M., Pandey, M.C., Jayathilakan, K., Radhakrishna, K. & Bawa, A.S. (2008). Effect of fish (*Catla catla*) fring on the quality characteristics of sunflower oil. *Food Chemistry*, **106**, 634–639.

Metcalfe, L.D.A.A. & Schimitz, J.R. (1966). Rapid preparation of fatty acid esters from lipids for gas liquid chromatography. *Analytical Chemistry*, **38**, 510.

Miller, M.R., Nichols, P.D. & Cater, C.G. (2007). Replacement of fish oil with thraustochytrid *Schizochytrium* sp. L oil in Atlantic salmon parr (*Salmo salar* L) diets. *Comparative Biochemistry and Physiology*, **148**, 382–392.

Peng, S., Chen, L., Qin, J.G. *et al.* (2008). Effects of replacement of dietary fish oil by soybean oil on growth performance and liver biochemical composition in juvenile black seabream, *Acanthopagrus schlegeli*. *Aquaculture*, **276**, 154–161.

Rasoarahoma, J.R., Barnathan, G., Bianchini, J.P. & Gaydou, E.M. (2004). Annual evolution of fatty acid profile from muscle lipids of

the common carp (*Cyprinus carpio*) in Madagascar inland waters. *Journal Agriculture Food Chemistry*, **52**, 7339–7344.

Reece, P. (1980). Control and reduction of free fatty acid concentration in oil recovered 449 from fish silage prepared from sprat. *Journal Science Food Agriculture*, **31**, 147–155.

Reece, P. (1981). Recovery of high quality oil from mackerel and sprat by silage process. *Journal Science Food Agriculture*, **32**, 531–538.

Sequeira-Munoz, A., Chevalier, D., LeBail, A., Ramaswamy, H.S. & Simpson, B.K. (2006). Physicochemal changes induced in carp (*Cyprinus carpio*) fillets by high pressure processing at low temperature. *Innovative Food Science Emerging Technologies*, **7**, 13–18.

Stickney, R.R. & Andrews, J.W. (1972). Effects of dietary lipids on growth, food conversion, lipid and fatty acid composition of channel catfish. *Journal of Nutrition*, **102**, 249–258.

Vandeputte, M., Kocour, M., Mauger, S., Rodina, M., Launay, A., Gela, D., Dupont-Nivet, M., Hulak, M. & Linhart, O. (2008). Genetic variation for growth at one and two summers of age in the common carp (*Cyprinus carpio L.*): heritability estimates and response to selection. *Aquaculture*, **277**, 7–13.

Viola, S. & Amidan, G. (1978). The effects of different dietary oil supplements on the composition of carp's body fat. *Bamidgeh*, **30**, 104–109.

Vyncke, W. (1970). Direct determination of the thiobarbituric acid value in trichloroacetic acid extracts of fish as a measure of oxidative rancidity. *Fette-Seifen Anstrichmittel*, **12**, 1084–1087.

Windsor, M. & Barlow, S.. (1984). *Introduction to the Fishing Products*. Pp. 99–109, Zaragosa, Spain: Acribia Publishing House.

Zhong, Y., Madhujith, T., Mahfouz, N. & Shahidi, F. (2007). Compositional characteristics of muscle and visceral oil from steelhead trout and their oxidative stability. *Food Chemistry*, **104**, 602–608.

Zhou, S., Paulson, A.T. & Ackman, R.G. (1995). Release of free fatty acids from lipids during ensilation of herring mince: the possible role of ethoxyquin. *Journal Food Lipids*, **3**, 121–134.