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Fatty acid composition of three freshwater fishes under different storage and cooking processes

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Abstract

Fatty acid composition of common carp (*Cyprinus carpio*), Nile tilapia (*Oreochromis niloticus*) and tambacu, a hybrid of tambaqui (*Colossoma macropomum*) and pacu (*Piaractus mesopotamicus*), was evaluated by gas chromatography. Raw, roasted and steamed fishes with and without skin were analyzed fresh and after 15, 30 and 45 storage days at -20 °C. Total lipid content was 9.3 g/100 g in carp, 0.79 g/100 g in tilapia and 1.3 g/100 g in tambacu with skin, with reductions of about 63%, 39% and 71% in the fishes without skin, respectively. The carp showed a high content of monounsaturated fatty acids (about 50%). In tilapia, palmitic and oleic acids were present in larger proportion, 26.55% and 23.86%, respectively. In tambacu, the fatty acid profile was 37% saturated, 34% monounsaturated and 21% polyunsaturated. Fatty acid composition did not present wide variations due to storage time and preparation, indicating that the storage and cooking methods used did not interfere in fatty acid composition.

Keywords: Common carp; Nile tilapia; Tambacu; Fatty acids; Storage conditions; Processing

1. Introduction

Fish lipids have been intensely investigated since their protective effect on cardiovascular diseases was first studied. Fish oils are rich in long-chain polyunsaturated fatty acids (LC-PUFA), namely eicosapentaenoic (EPA) and docosahexaenoic (DHA), which reduce some risk factors associated with arteriosclerosis (Calder, 2004). Polyunsaturated fatty acids ω -3 (PUFA ω -3) play a role in preventing heart disease and have anti-inflammatory and anti-thrombosis effects (Connor, 2000). Also, ω -3 and ω -6 polyunsaturated fatty acids are considered essential but since they

cannot be synthesized in the human body, they must be obtained through diet (Mahan & Escott-Stump, 2005).

Despite the vast ichthyofauna diversity and extensive Brazilian hydrographic system, there is a lack of data on fish fatty acid composition (Maia, Rodriguez-Amaya, & Franco, 1994). Also, the composition of fatty acids in freshwater fishes is influenced by the environment and type of feed (Moreira, Visentainer, Souza, & Matsushita, 2001; Suzuki, Okazahi, Hayakama, Wada, & Tamura, 1986). Thus, the study of lipid composition of fish consumed in different regions of Brazil is of major importance to further research on the physiological effects of fish lipids on human health.

Freshwater Nile tilapia (*Oreochromis niloticus*), was chosen for this study for its good market acceptance and rusticity for handling. Tilapia is considered promising for aquaculture because of its rapid growth, late reproduction

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and high multiplication rate. It has a firm, consistent and tasty meat of great market acceptance (Kubitza, 2000). The common carp (*Cyprinus carpio*) is originated from Asia and reared almost worldwide (Padua, 2001). It has important nursery production quality, such as resistance to diseases and easy handling and reproduction. Besides, it adapts well to the most different feed types and develops quickly (Castagnolli, 1992). With the remarkable development of fish production in aquaculture in Brazil (Valle et al., 2000), the common carp seems to be a promising dietary choice. Tambacu, a hybrid derived from the cross of the species tambaqui (*Colossoma macropomum*) and pacu (*Piaractus mesopotamicus*), was also chosen for being well-accepted commercially and easy to handle.

Many studies analyze the profile of fatty acids in fresh raw fish; however, fish is usually consumed after frozen storage and/or after some type of culinary preparation. Freezing can cause alterations in fish, such as increase in the amount of free fatty acids and compounds derived from lipid oxidation (Sarma, Vidya Sagar Reddy, & Srikar, 2000). Few studies are available in the literature on the effect of freezing on the fatty acid profile of fish, with some investigating rancidity and change in flesh texture (Aubourg, 1999; Chevalier, Sequeira-Munoz, Le Bail, Simpson, & Ghoul, 2000).

According to Silva, Kuga, and Filho (1993), some factors, such as lipid contents, cooking temperature, species size and surface contact can affect lipid composition in fish after cooking. Additional data are certainly needed on this subject, since most fish are consumed cooked in many ways.

Since data related to the effects of storage and processing on the lipid composition of freshwater fish are scarce in the literature, the objective of this study was to evaluate the fatty acid profile of common carp, Nile tilapia and tambacu, with and without skin and the effects of the cooking methods (roasted and steamed) on both fresh and frozen fish stored for 15, 30 and 45 days.

2. Materials and methods

2.1. Raw material

Fish were cultivated under the semi-intensive system, at the Fish Culture Sector of the Department of Animal Biology of the Universidade Federal de Viçosa (UFV), Minas Gerais, Brazil. Fingerlings of carp (*C. carpio*) were placed in tanks previously fertilized with swine manure (200 g/ m^2) and were kept there until slaughtering. Their growing period was December 2001 through September 2002. For Nile tilapia (*O. niloticus*) and tambacu, the growing period was February 2001 through November 2002. After the adaptation period, the fish were fed extruded feeds containing 28% crude protein.

2.2. Experimental design

Raw fresh samples were frozen at -20 °C and kept under frozen conditions (-20 °C) for 15, 30 and 45 days

and then cooked using two methods: dry heat (baking) and humid heat (steaming). One fish was used for analysis in each preparation method (raw, roasted and steamed) at each sampling time. The analyses were performed in triplicate.

2.3. Sample treatment

The fish samples were transported to the Laboratory of Experimental Food Studies, at the Department of Nutrition and Health of the UFV, manually cleaned, descaled, eviscerated, individually packed into plastic bags and stored in a freezer. Thawing took place at cooling temperature for 8 h.

Fish were cooked whole and with skin. Steaming was performed under boiling water without salt in a steamer for 20 min. Roasting was performed in a conventional gas oven at 180 °C for 25 min. After the cooking process, fish were cut in the middle, longitudinally. One of the halves was used to analyze the skin-on material and the other half had the skin removed before the analyses.

2.4. Centesimal composition analysis

Moisture, total lipids and protein were determined according to Cunniff (1998), in an oven, at 105 °C for 24 h; extracted with a Soxhlet extractor and by the Kjeldhal method, respectively.

2.5. Fatty acid determination

About 4 g of each sample were homogenized for lipid extraction by the method of Folch, Lees, and Stanley (1957), followed by esterification of the lipid fraction by the method of Hartman and Lago (1973). All reagents were p.a. quality purchased from Merck (Brazil). The fatty acids methyl esters were analyzed by gas chromatography using a GC 17A (Shimadzu), equipped with a flame ionization detector, split injector, and fused silica capillary columns Supelco 2560 with 100 m and 0.25 mm internal diameter. The following operation parameters were used: detector and injector temperatures: 270 and 250 °C, respectively; column temperature programmed at 100 °C, increasing up to 180 °C at 10 °C min⁻¹ and from 180 °C to 240 °C at $1 \,^{\circ}\mathrm{C}\,\mathrm{min}^{-1}$, holding at this end temperature for 10 min; carrier gas: nitrogen at 0.6 mL/min and linear velocity of 14 cm/s; split ratio of 1:75 with a total flow of 52 mL/min and column pressure of 167 kPa. Retention time and peak areas were recorded with the help of a microcomputer using the software CLASS GC 10. Fatty acids were expressed in %.

2.6. Fatty acid identification

The fatty acids methyl esters were identified by comparing the retention time of the samples and appropriate fatty acids methyl esters standards, purchased from Sigma (St. Louis, MO, USA). The relative percentage of the area was obtained by using the following equation: Area % $FA_X = [A_X/A_R] \times 100$, where: $FA_X = fatty$ acid to be quantified, $A_X =$ area of the methyl esters X and $A_R =$ total area of the chromatogram. Peak areas lower than 0.1% of the total area were not considered.

2.7. Statistical analysis

Sigma Stat software, version 2.0, was used for the statistical analysis. The samples with and without skin were compared using the paired t test for each fatty acid and freezing time. Analysis of variance was applied to verify the existence of significant differences between the preparation methods. Statistically significant means were compared by the Tukey test (P < 0.05).

3. Results and discussion

The content of moisture, proteins and lipids were 72.33%, 9.32% and 16.71%, respectively, in skin-on carp. After skin removal the content of lipids reduced to 3.45% and moisture raised to 77.90%. Skin-on tambacu and Nile tilapia presented 77.93% and 78.49% of moisture, 1.30% and 0.79% of lipids and 18.80% and 18.73% of proteins, respectively. The contents of lipids were 0.38% in skinless tambacu and 0.48% in skinless Nile tilapia.

Moisture values were within the range cited by other authors for freshwater fish, i.e., 70% and 85% (Yeannes & Almandos, 2003). According to Penfield and Campbell (1990), skin-on carp can be classified as a fat fish, because its lipid content was above 5%, while Nile tilapia and tambacu can be classified as lean fish. There was a reduction in the lipid percentage when the skin was removed from the three species. In the case of carp, skin removal modified its classification to low-fat, since its lipid content became lower than 5%. The subcutaneous layer contains a good part of an animal's fat storage (Undeland et al., 1998), but with skin removal, a considerable amount of fat is eliminated.

The total lipid contents found in this study for common carp are above the percentage reported by Andrade, Rubira, Matsushita, and Souza (1995) in a work developed in southern Brazil, where the lipid percentage in carps without skin was 1.19%. In contrast, the same study reported 2.86% of total lipid percentage in Nile tilapia without skin, which is above the value found in the present work. The lipid content of tambacu in the present study was much lower than that found in pacu (18.31%), according to Andrade et al. (1995).

In another study, the total lipid content found in tilapias was 1.1 g/100 g (Visentainer, Souza, Makoto, Hayashi, & Franco, 2005). Still in Brazil, Souza, Baccarin, Viegas, and Kronka (2004) reported values of 8.06% for total lipids in gutted tilapias. In Thailand, the total lipid, protein and moisture contents reported by Puwastien et al. (1999) in raw tilapias were 1.8, 19.8 and 78.1 g/100 g, respectively.

Tables 1–3 show the percentage of each fatty acid in raw, fresh carp, Nile tilapia and tambacu, with and without skin, and after 15, 30 and 45 days of frozen storage.

Sixteen fatty acids were identified in carp, with monounsaturated fatty acid accounting for 42.0–59.4%. Oleic acid (18:1) was the fatty acid present in the largest proportions in the raw carp samples, followed by linoleic (18:2) and palmitic acids (16:0), polyunsaturated and saturated, respectively. In raw samples of Nile tilapia, saturated fatty acids presented the largest percentage (43.34% mean with and without skin), predominantly palmitic (16:0) and stearic (18:0) acids that together accounted to, on average, 36.7% of the total lipid. Oleic acid was the main representative among the monounsaturated fatty acids (18:1). Linoleic (18:2), arachidonic (20:4) and DHA (22:6) acids were, in general, the polyunsaturated fatty acids found in the largest percentages. For tambacu with and without skin, the mean percentage distribution in the fatty acid groups was 37% of saturated, 34% monounsaturated and 21% polyunsaturated acids. Although the saturated fatty acids were found in higher proportions, oleic acid (18:1) had the highest percentages, individually, surpassed by palmitic acid after 30 days under frozen storage. A study carried out by Sant'Ana and Mancini-Filho (2000) showed that pacu contained 27.9% of saturated, 44.3% monounsaturated and 27.8% polyunsaturated fatty acids.

In general, there was little variation in the percentage of fatty acids after skin removal and even when this variation was significant, the observed difference was reduced. For carp, there was a decrease in the linoleic acid (18:2) percentage after skin removal, except for 30 days of frozen storage, with the inverse occurring. There was an increase in the percentage of arachidonic acid (20:4) after 45 days of freezing and of DHA (22:6) after 15 days of freezing, after the skin was removed, suggesting that these fatty acids were present in the subcutaneous layer. This is an important point, since fish skin is usually removed before consumption. In Nile tilapia, a significantly larger percentage of palmitic acid (16:0) was observed in the skinless samples, compared with the samples with skin, for fresh samples. Conversely, a significantly lower level of heneicosanoic acid (21:0) was found in skinless samples, after 45 days of frozen storage. Significant differences were also observed in the percentage of palmitoleic acid (16:1) that decreased in samples stored under freezing for 15 and 30 days, after skin removal. The same was observed in the linoleic acid percentage in fresh samples. In contrast, there was an increase in the percentage of cis-8,11,14 eicosatrienoic (20:3), arachidonic (20:4), EPA (20:5) and DHA (22:6) with skin removal, after 45 days of frozen storage.

Of the 22 fatty acids identified in tambacu, the following presented the highest percentage: oleic (18:1), palmitic (16:0), linoleic (18:2) and stearic acids (18:0), as shown in Table 3. A significant reduction occurred in myristic (14:0) and palmitic acids (16:0) in the fresh sample following skin removal. Conversely, heptadecaenoic (17:0) and stearic (18:0) acid percentages were significantly higher in

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Table 1 Fatty acid (%) of raw common carp, with and without skin, under different frozen storage times (mean \pm standard deviation)

Fatty acid		WS0 ^{a,c}	WOS0 ^b	WS15 ^d	WOS15	WS30 ^e	WOS30	WS45 ^f	WOS45
Saturated									
Miristic	14:0	0.6 (±–)	$0.6 (\pm -)$	1.2 (±0.1)	$1.1 (\pm 0.1)$	1.2 (±0.1)	$1.1 (\pm 0.1)$	0.9 (±0.1)	0.9 (±0.2)
Palmitic	16:0	13.9 (±0.5)	15.3 (±1.4)	18.2 (±0.4)	19.7 (±1.0)	18.3 (±0.5)	18.8 (±0.9)	16.5 (±0.2)	17.9 (±0.7)
Heptadecanoic	17:0	0.4 (±0.1)	0.7 (±0.3)	0.9 (±0.2)	1.5 (±0.4)	0.7 (±0.1)	$0.6 (\pm -)$	0.6 (±0.1)	0.8 (±0.1)
Stearic	18:0	3.5 (±0.2)	4.4 (±0.7)	3.4 (±0.5)	5.1 (±0.8)	4.1 (±0.3)	4.1 (±0.1)	4.3 (±0.1)	5.3 (±0.5)
Behenic	22:0	0.6 (±0.1)	0.7 (±0.1)	0.6 (±0.1)	0.7 (±0.1)	0.9 (±0.1)	0.7 (±0.1)	0.9 (±0.1)	1.1 (±0.1)
\sum		19.0	21.7	24.3	28.1	25.2	25.3	23.2	26.0
Monounsaturated									
Palmitoleic	16:1	4.9 (±0.1)	4.5 (±0.4)	9.2 (±0.4)	7.4^{*} (±0.7)	10.1 (±0.5)	9.8 (±0.2)	7.2 (±0.1)	6.8 (±0.4)
Oleic	18:1 9c	48.2 (±1.4)	44.2 (±4.1)	36.7 (±1.5)	$30.2^{*}(\pm 2.7)$	44.8 (±1.4)	44.0 (±0.7)	44.9 (±0.6)	42.2 (±1.4)
Vacenic	18:1 7c	4.0 (±0.2)	3.7 (±0.2)	4.5 (±0.3)	4.4 (±0.1)	4.5 (±0.1)	5.6^{*} (±0.3)	4.0 (±0.1)	4.4^{*} (±0.1)
\sum		57.1	52.4	50.4	42.0	59.4	59.4	56.1	53.4
Polyunsaturated									
Linoleic	18:2 ω-6	16.6 (±0.6)	$15.4^{*} (\pm 0.6)$	17.4 (±0.6)	15.7^{*} (±1.1)	8.8 (±0.4)	9.4^{*} (±0.4)	12.5 (±0.4)	12.1 (±0.5)
γ-Linolenic	18:3 ω-6	$0.4 (\pm -)$	$0.4 (\pm -)$	$0.4 (\pm -)$	$1.1 (\pm 1.0)$	$0.2 (\pm -)$	$0.2 (\pm -)$	0.3 (±0.1)	$0.0^{*}~(\pm -)$
α-Linolenic	18:3 ω-3	0.6 (±0.6)	0.7 (±0.2)	$0.0 (\pm -)$	0.7^{*} (±0.1)	0.6 (±0.1)	0.6 (±0.1)	0.7 (±0.1)	0.5 (±0.4)
cis-11,14 Eicosadienoic	20:2	$0.4 (\pm -)$	$0.4 (\pm -)$	0.9 (±0.2)	1.5 (±0.4)	0.7 (±0.1)	$0.6 (\pm -)$	0.6 (±0.1)	0.8 (±0.1)
<i>cis</i> -8,11,14	20:3	$0.5 (\pm 0.1)$	0.8 (±0.2)	$0.7 (\pm 0.1)$	0.9 (±0.2)	$0.4 \ (\pm 0.1)$	0.3 (±0.1)	0.5 (±0.1)	0.5 (±0.1)
Eicosatrienoic									*
Arachidonic	20:4 ω-6	2.2 (±0.6)	4.1 (±1.6)	3.3 (±1.0)	$7.0(\pm 2.0)$	2.1 (±0.5)	$1.8 (\pm 0.2)$	2.7 (±0.1)	$3.9^{*} (\pm 0.5)$
EPA	20:5 ω-3	$0.0 (\pm -)$	$0.0~(\pm -)$	$0.4 (\pm 0.1)$	$0.6(\pm 0.2)$	0.3 (±0.2)	0.3 (±0.1)	$0.0 (\pm -)$	$0.0 (\pm -)$
DHA	22:6 ω-3	$0.7 (\pm 0.3)$	$1.7 (\pm 0.8)$	$0.9 (\pm 0.9)$	3.5 [*] (±1.0)	$1.2 (\pm 0.3)$	$1.0 (\pm 0.1)$	$1.2 (\pm 0.2)$	$1.6 (\pm 0.3)$
\sum		21.4	23.5	24.0	31.0	14.3	14.6	18.5	19.4
$\sum NI^{g}$		2.6	2.6	1.8	0	1.7	1.4	2.4	1.9

^a WS: with skin.

^b WOS: without skin.

^c 0: zero frozen storage time (fresh sample).

^d 15: 15 days under frozen storage.

^e 30: 30 days under frozen storage.

f 45: 45 days under frozen storage.

^g NI: not identified.

* Significant difference by the paired t test ($P \le 0.05$). The test was applied for each fatty acid separately, under each frozen storage time. Comparisons refer to form of consumption (with and without skin).

fresh tambacu without skin. A significant reduction was observed in the percentage of palmitoleic acid (16:1) following skin removal, except for the samples analyzed after 15 days under frozen storage. Also worthy noting was the reduction of oleic acid percentage (18:1) and cis-10 heptadecaenoic (17:1) acid in fresh tambacu without skin and after 45 days of frozen storage, respectively. Conversely, an increase was observed in the percentage of arachidonic (20:4) and cis-11,14 eicosadienoic (20:2) acids, statistically significant at some storage times. Higher percentages of EPA (20:5) and DHA (22.6) were found at all frozen storage times in tambacu following skin removal. This percentage increase could be attributed to the fact that the muscle portions contain higher amounts of long-chain polyunsaturated fatty acids than the subcutaneous layer. The analysis of this profile shows that the percentage of monounsaturated fatty acids tends to decrease and that of polyunsaturated fatty acids tends to increase after skin removal. This is a positive fact, since fish are usually skinned before consumption, although the skin may be kept during preparation to maintain flesh firmness.

Generally, the fatty acid profile found in the present work agrees with the results reported by Andrade et al. (1995) and Geri et al. (1995), although the first found linolenic acid (18:3) in greater percentage and arachidonic acid (20:4) was not mentioned by those authors. Moreover, Geri, Poli, Gualtieri, Lupi, and Parisi (1995) reported a much higher amount of ω -3 fatty acids. Suzuki et al. (1986) studying carps cultivated in Japan also found a DHA percentage above the one reported in the present work.

The results for tilapia are in agreement with the values presented by Rasoarahona, Barnathan, Bianchini, and Gaydou (2005) for tilapias analyzed in the spring. However, in an analysis of tilapias purchased at a local market in Sao Paulo, Brazil, the presence of linoleic (18:2), α -linolenic (18:3) and arachidonic (20:4) acids was not reported in the studied samples, whereas the EPA (20:5) percentage was quite high (Luzia, Sampaio, Castellucci, & Torres, 2003).

The fact that tambacu is a hybrid made it difficult to compare the results with those in the literature. In some cases, the fatty acid profile of this fish was quite different from that of its precursor pacu. While studies indicated high percentages of oleic acid in tambacu, a study carried out in Brazil obtained opposite results in pacu (Andrade

Table 2			
Fatty acid (%) of raw Nile tilapia	, with and without skin	, under different frozen storag	e times (mean \pm standard deviation)

Fatty acid		WS0 ^{ac}	WOS0 ^b	WS15 ^d	WOS15	WS30 ^e	WOS30	WS45 ^f	WOS45
Saturated									
Lauric	12:0	0.3 (±0.1)	0.4 (±0.1)	$0.4 (\pm -)$	0.4 (±0.2)	0.4 (±0.1)	0.3 (±0.1)	0.4 (±0.1)	$0.4 (\pm -)$
Tridecanoic	13:0	$0.2 (\pm -)$	$0.2 (\pm -)$	$0.2 (\pm -)$	$0.2 (\pm -)$	$0.2(\pm 0.1)$	$0.2 (\pm 0.1)$	0.3 (±0.1)	$0.4 (\pm -)$
Miristic	14:0	4.4 (±0.9)	4.3 (±0.8)	4.7 (±0.3)	4.6 (±0.2)	4.2 (±0.8)	3.1 (±0.1)	4.4 (±0.6)	4.0 (±0.1)
Palmitic	16:0	25.9 (±0.1)	$27.5^{*}(\pm 0.6)$	26.8 (±0.7)	26.9 (±0.2)	27.1 (±2.0)	28.0 (±1.7)	25.3 (±0.9)	24.9 (±0.2)
Heptadecanoic	17:0	$1.4 (\pm -)$	1.4 (±0.2)	1.2 (±0.2)	$1.2 (\pm 0.1)$	1.3 (±0.2)	1.4 (±0.2)	1.5 (±0.2)	1.5 (±0.1)
Stearic	18:0	10.7 (±0.7)	11.6 (±1.0)	9.0 (±0.5)	9.1 (±0.3)	9.4 (±1.0)	11.0 (±0.8)	9.8 (±0.3)	10.2 (±0.2)
Heneicosanoic	21:0	$0.4 (\pm 0.2)$	0.5 (±0.1)	0.5 (±0.1)	$0.4 (\pm -)$	0.4 (±0.2)	$0.3 (\pm -)$	$0.4 (\pm -)$	0.0^{*} (±–)
Behenic	22:0	0.3 (±0.1)	0.3 (±0.1)	$0.2 (\pm -)$	0.2 (±0.1)	$0.0 (\pm -)$	0.1 (±0.1)	$0.0 (\pm -)$	$0.0 (\pm -)$
\sum		43.6	46.2	43.0	43.0	43.0	44.4	42.1	41.4
Monounsaturated									
Miristoleic	14:1	$2.1 (\pm 0.2)$	$1.7 (\pm 0.7)$	$1.7 (\pm 0.1)$	$1.7 (\pm -)$	$1.7 (\pm 0.3)$	1.9 (±0.4)	1.9 (±0.5)	2.2 (±0.1)
Palmitoleic	16:1	5.3 (±1.4)	5.3 (±1.3)	$6.8(\pm 0.1)$	$0.5^{*}(\pm 0.2)$	5.9 (±0.7)	$4.0^{*}(\pm 0.4)$	5.5 (±0.2)	4.7 (±0.2)
cis-10 Heptadecanoic	17:1	$0.7 (\pm 0.2)$	$0.8 (\pm 0.2)$	$0.8 (\pm 0.1)$	$0.7 (\pm -)$	0.8 (±0.2)	$0.5(\pm 0.2)$	$1.0(\pm 0.1)$	$1.0(\pm 0.1)$
Elaidic	18:1 9t	3.4 (±0.6)	$2.6(\pm 0.1)$	$2.7 (\pm 0.3)$	2.7 (±0.1)	2.6 (±0.8)	$2.2(\pm 0.3)$	2.7 (±0.2)	$2.5(\pm 0.1)$
Oleic	18:1 9c	22.5 (±3.1)	22.7 (±3.4)	27.0 (±2.0)	25.6 (±1.3)	26.5 (±3.3)	23.6 (±3.3)	23.9 (±3.0)	19.1 (±0.4)
Vacenic	18:1 7c	$0.2(\pm 0.1)$	$0.2(\pm 0.1)$	$0.1 (\pm -)$	$0.3^{*}(\pm 0.1)$	0.7 (±0.3)	0.3 (±0.1)	$0.0(\pm -)$	$0.0(\pm -)$
\sum		34.2	33.3	39.1	31.5	38.2	32.5	35.0	29.5
Polyunsaturated									
Linoleic	18:2 ω-6	4.7 (±0.1)	3.5^* (±0.6)	4.6 (±0.1)	4.8 (±0.1)	4.8 (±0.1)	4.6 (±0.3)	5.5 (±0.1)	5.5 (±0.1)
α-Linolenic	18:3 ω-3	$1.0 (\pm 0.3)$	1.4 (±0.2)	2.3 (±0.2)	2.1 (±0.1)	2.4 (±0.9)	1.5 (±0.2)	2.2 (±0.3)	$2.1 (\pm 0.1)$
cis-11,14 Eicosadienoic	20:2	$0.7 (\pm 0.1)$	$0.8 (\pm -)$	$1.1 (\pm 0.1)$	$1.1 (\pm -)$	1.4 (±0.2)	1.2 (±0.1)	$1.0 (\pm 0.1)$	$1.1 (\pm -)$
cis-8,11,14	20:3 ω-6	0.9 (±0.4)	$1.0 (\pm 0.4)$	0.5 (±0.1)	0.6 (±0.1)	0.6 (±0.1)	0.8 (±0.2)	0.8 (±0.1)	$1.0^{*} (\pm 0.1)$
Eicosatrienoic									
cis-11,14,17	20:3	$0.2 (\pm 0.1)$	0.1 (±0.1)	0.3 (±0.1)	0.3 (±0.1)	0.3 (±0.1)	0.3 (±0.1)	0.3 (±0.1)	$0.4 (\pm -)$
Eicotrienoic									
Arachidonic	20:4 ω-6	4.1 (±3.3)	4.9 (±3.0)	1.5 (±0.1)	2.2 (±0.4)	2.1 (±0.7)	4.0 (±1.0)	3.6 (±0.4)	$6.1^{*} (\pm 0.1)$
cis-13,16	22:2	$0.3 (\pm -)$	$0.3 (\pm -)$	0.6 (±0.1)	0.5 (±0.1)	0.5 (±0.1)	0.4 (±0.1)	0.4 (±0.1)	$0.4 (\pm -)$
Docosadienoic									
EPA	20:5 ω-3	0.6 (±0.4)	0.8 (±0.4)	$0.5 (\pm -)$	0.6 (±0.1)	0.5 (±0.2)	0.9 (±0.2)	0.6 (±0.1)	1.0^{*} (±–)
DHA	22:6 ω-3	3.9 (±3.3)	4.8 (±3.1)	2.2 (±0.3)	3.2 (±0.5)	2.6 (±2.3)	6.3 (±1.5)	3.8 (±0.5)	6.9 [*] (±0.1)
\sum		15.5	17.4	13.6	15.4	15.2	20.0	18.2	24.5
$\sum NI^{g}$		5.5	5.0	4.6	4.3	3.9	3.1	4.8	4.5

^a WS: with skin.

^c 0: zero frozen storage time (fresh sample).

^d 15: 15 days under frozen storage.

^e 30: 30 days under frozen storage.

^f 45: 45 days under frozen storage.

^g NI: non-identified.

* Significant difference by the paired t test ($P \le 0.05$). The test was applied for each fatty acid separately, under each frozen storage time. Comparisons refer to form of consumption (with and without skin).

et al., 1995). On the other hand, Sant'Ana and Mancini-Filho (2000) reported that the fatty acid composition of pacu fed a control diet was similar to the fatty acid profile of tambacu in the present study. A study on tambaqui, another tambacu precursor, also showed high contents of oleic, palmitic and stearic acids (40.0%, 28.8% and 9.8%, respectively) (Maia & Rodriguez-Amaya, 1992).

A number of factors can influence fish fatty acid composition, such as water temperature, time of capture, salinity and feed type. Therefore, these factors must be considered when analyzing differences among studies.

The high temperature of tropical environments is a conditioning factor of fatty acid composition in Brazilian fish, which accumulate 16 and 18 carbon acids, mainly 16:0, 16:1, 18:0 and 18:1. In some fish, saturated fatty acids are overcome by monounsaturated acids with the opposite occurring in others; however, there is no predominance of one or another group of fatty acid (Contreras-Guzmán, 1994).

A study developed in Madagascar showed a variation in the fatty acid composition of common carp throughout the year, with lipid accumulation and high PUFA levels during the coldest months (Rasoarahona, Barnathan, Bianchini, & Gaydou, 2004). In a study on the influence of salinity, Haliloglu, Bayýr, Sirkecioglu, Aras, and Atamanalp (2004) observed that the muscle tissue from saltwater trout contains less linoleic acid (18:2) than that of freshwater trout. Van Vliet and Katan (1990) confirmed that modern aquaculture, with grain-based feeds, produces fish with lower levels of ω -3 fatty acids than those growing naturally

^b WOS: without skin.

Table 3 Fatty acid (%) of raw tambacu, with and without skin, under different frozen storage times (mean \pm standard deviation)

Fatty acid		WS0 ^{ac}	WOS0 ^b	WS15 ^d	WOS15	WS30 ^e	WOS30	WS45 ^f	WOS45
Saturated									
Miristic	14:0	1.3 (±0.1)	0.9^{*} (±0.1)	1.2 (±0.2)	1.2 (±0.1)	3.9 (±0.8)	3.0 (±0.1)	4.2 (±0.5)	3.8 (±0.1)
Palmitic	16:0	23.6 (±0.6)	21.0 [*] (±0.5)	24.4 (±1.0)	23.6 (±0.4)	25.5 (±2.1)	27.0 (±2.0)	24.2 (±0.9)	23.6 (±0.3)
Heptadecanoic	17:0	0.6 (±–)	$1.0^{*} (\pm 0.1)$	0.5 (±0.1)	0.7 (±0.4)	1.2 (±0.2)	1.3 (±0.2)	1.5 (±0.2)	1.5 (±0.1)
Stearic	18:0	9.2 (±0.5)	$11.5^{*} (\pm 0.2)$	8.6 (±0.5)	8.2 (±2.1)	9.0 (±1.0)	10.6 (±0.7)	9.3 (±0.3)	9.7 (±0.3)
Archachidic	20:0	$0.0 (\pm -)$	$0.0 (\pm -)$	$0.0 (\pm -)$	$0.0 (\pm -)$	0.2 (±0.2)	$0.3 (\pm -)$	0.4 (±0.1)	$0.3 (\pm -)$
Heneic	21:0	$0.0 (\pm -)$	$0.0 (\pm -)$	$0.0 (\pm -)$	$0.0 (\pm -)$	0.4 (±0.2)	$0.3 (\pm -)$	$0.4 (\pm -)$	0.2 (±0.2)
\sum		34.7	34.4	34.5	33.7	40.2	42.5	40.0	39.1
Monounsaturated									
Palmitoleic	16:1	5.0 (±0.4)	2.8^{*} (±0.4)	5.4 (±0.6)	4.3 (±0.8)	5.5 (±0.8)	3.9^{*} (±0.4)	5.3 (±0.2)	4.5 [*] (±0.2)
cis-10 Heptadecanoic	17:1	$0.0 (\pm -)$	$0.0 (\pm -)$	0.5 (±0.1)	0.7 (±0.4)	0.7 (±0.2)	$0.5(\pm 0.2)$	0.9 (±0.1)	0.0^{*} (±–)
Elaidic	18:1 9t	$0.1 (\pm 0.1)$	$0.0(\pm -)$	$0.0(\pm -)$	$0.0(\pm -)$	2.5 (±0.8)	2.1 (±0.3)	2.5 (±0.2)	2.4 (±0.1)
Oleic	18:1 9c	36.0 (±0.9)	26.3* (±2.1)	36.5 (±0.6)	33.7 (±5.5)	25.4 (±2.9)	22.9 (±3.4)	22.8 (±3.0)	18.2 (±0.4)
Vacenic	18:1 7c	$0.0 (\pm -)$	$0.0 (\pm -)$	$0.0 (\pm -)$	$0.0 (\pm -)$	0.6 (±0.3)	$0.3(\pm 0.1)$	$0.0 (\pm -)$	$0.0 (\pm -)$
\sum		41.1	29.1	42.4	38.7	34.7	29.7	31.5	25.1
Polyunsaturated									
Linoleic	18:2 ω-6	12.6 (±0.1)	11.8 (±0.4)	11.7 (±0.4)	13.0 (±3.3)	6.9 (±0.6)	6.3 (±1.3)	7.9 (±0.5)	8.1 (±0.2)
γ-Linolenic	18:3 ω-6	$0.0 (\pm -)$	$0.0 (\pm -)$	$0.0 (\pm -)$	$0.0 (\pm -)$	0.6 (±0.2)	$0.4 (\pm 0.1)$	0.8 (±0.1)	$0.6 (\pm 0.1)$
α-Linolenic	18:3 ω-3	0.7 (±0.1)	0.8 (±0.1)	0.6 (±0.1)	0.7 (±0.1)	2.3 (±0.9)	$1.5 (\pm 0.2)$	2.1 (±0.3)	2.0 (±0.1)
cis-11,14 Eicosadienoic	20:2	0.5 (±0.1)	0.8 (±0.1)	0.6 (±0.1)	0.4 (±0.2)	1.4 (±0.3)	$1.1 (\pm 0.1)$	$0.0 (\pm -)$	1.0^{*} (±–)
cis-11,14,17 Eicotrienoic	20:3	$0.0 (\pm -)$	$0.0 (\pm -)$	$0.0 (\pm -)$	$0.0 (\pm -)$	0.3 (±0.1)	0.3 (±0.1)	$0.3 (\pm -)$	0.4^{*} (±–)
Arachidonic	20:4 ω-6	1.2 (±0.6)	5.2^{*} (±0.6)	1.4 (±0.6)	2.7 (±2.0)	2.0 (±0.6)	3.9 (±0.9)	3.4 (±0.4)	5.7 [*] (±0.2)
cis-13,16 Docosadienoic	22:2	$0.0 (\pm -)$	$0.0 (\pm -)$	$0.0 (\pm -)$	$0.0 (\pm -)$	0.5 (±0.1)	$0.4 (\pm 0.1)$	0.3 (±0.2)	$0.4 (\pm -)$
EPA	20:5 ω-3	0.3 (±0.1)	1.2^{*} (±0.2)	$0.0 (\pm -)$	0.5 (±0.4)	0.5 (±0.2)	0.9 (±0.2)	0.6 (±0.1)	1.0^{*} (±–)
DHA	22:6 ω-3	1.5 (±0.8)	8.5 [*] (±1.5)	1.8 (±1.0)	3.9 (±3.3)	2.5 (±2.2)	6.1 (±1.4)	3.6 (±0.5)	6.5^{*} (±0.2)
Σ		16.8	28.3	16.1	21.2	17.0	20.9	19.0	25.7
$\frac{\sum_{i=1}^{n} NI^{g}}{(i)}$		5.9	4.0	5.7	4.3	6.5	6.0	7.2	6.7

^a WS: with skin.

^b WOS: without skin.

^c 0: zero frozen storage time (fresh sample).

^d 15: 15 days under frozen storage.

^e 30: 30 days under frozen storage.

^f 45: 45 days under frozen storage.

^g NI: non-identified.

* Significant difference by the paired t test ($P \le 0.05$). The test was applied for each fatty acid separately, under each frozen storage time. Comparisons refer to modes of consumption (with and without skin).

in rivers and lakes. Another study observed that the percentage of linoleic acid (18:2) in cultivated carps was greater than in wild carps (Suzuki et al., 1986).

Carps analyzed in this study were fed swine manure, while Nile tilapia and tambacu were raised in tanks and fed extruded ration (28% crude protein) what might have influenced fatty acid composition. Although the percentage of oleic (18:1) and linoleic (18:2) acids here reported for carp are usually higher than those found in the literature, most studies on the common carp (Andrade et al., 1995; Rasoarahona et al., 2004; Suzuki et al., 1986) reported the presence of three main fatty acids (oleic, linoleic and palmitic), as in this work.

The period of storage did not have a strong influence on the fatty acid profile. By increasing the period of storage for raw carp, with and without skin, a rise in the content of saturated and monounsaturated fatty acids and a decrease in polyunsaturated fatty acids were observed. Raw Nile tilapia with skin presented increased contents of monounsaturated and polyunsaturated fatty acids during storage and the same content of saturated fatty acids. However, when the skin was removed the content of monounsaturated fatty acids decreased. It was observed that the content of saturated fatty acids increased and monounsaturated fatty acids decreased in raw tambacu with and without skin as storage time progressed. But polyunsaturated fatty acid contents increased in the samples with skin and decreased in skinless samples after 45 days of storage. Shorter frozen storage periods seem to provide a better preservation of monounsaturated fatty acids, with saturated fatty acids increasing with increased storage time in tambacu.

Even though the cooked fish samples were analyzed at 0, 15, 30 and 45 days of frozen storage, only the results of samples cooked after 30 days of storage were presented in this study, since this is the most common way of purchasing and consuming fish in this region in Brazil. Tables 4–6 show the percent of each fatty acid in relation to the total fatty acid content present in the carp, Nile tilapia and tambacu samples kept under frozen storage for 30 days

Table 4
Fatty acid profile (%) of raw, roasted and steamed carp, after 30 days under frozen storage; with and without skin

Fatty acid		With skin			Without skin			
		Raw	Roasted	Steamed	Raw	Roasted	Steamed	
Saturated								
Miristic	14:0	1.2b (±0.1)	1.3a (±0.1)	1.3ab (±–)	1.1b (±0.1)	1.3a (±0.1)	1.1b (±0.1)	
Palmitic	16:0	18.3b (±0.5)	21.0a (±0.3)	18.7b (±0.4)	18.8b (±0.9)	21.3a (±0.5)	19.0b (±0.2)	
Heptadecanoic	17:0	0.7a (±0.1)	0.6a (±0.1)	0.7a (±0.1)	$0.6c (\pm -)$	0.9b (±0.2)	1.1a (±0.1)	
Stearic	18:0	4.1b(±0.3)	3.5c (±0.1)	4.8a (±0.2)	4.1b (±0.1)	4.1b (±0.2)	5.7a (±0.1)	
Behenic	22:0	0.9a (±0.1)	0.3b (±0.3)	$1.0a~(\pm 0.1)$	0.7b (±0.1)	0.7b (±0.1)	$1.3a~(\pm 0.1)$	
\sum		25.2b	26.7a	26.5ab	25.3b	28.3a	28.2a	
Monounsaturated								
Palmitoleic	16:1	10.1b (±0.5)	11.1a (±0.2)	8.6c (±0.1)	9.8a (±0.2)	10.0a (±0.5)	7.4b (±0.2)	
Oleic	18:1 9c	44.8a (±1.4)	41.2b (±0.3)	43.6ab (±1.3)	44.0a (±0.7)	38.7b (±0.9)	39.2b (±0.6)	
Vacenic	18:1 7c	4.5b (±0.1)	5.4a (±0.3)	4.5b (±0.1)	5.6a (±0.3)	4.9b (±0.2)	4.6b (±0.2)	
\sum		59.4a	57.7a	56.7a	59.4a	53.6b	51.2c	
Polyunsaturated								
Linoleic	18:2 ω-6	8.8b (±0.4)	10.1a (±0.2)	8.2b (±0.3)	9.4b (±0.4)	10.3a (±0.2)	8.3b (±0.1)	
γ-Linolenic	18:3 ω-6	0.2a (±–)	0.2a (±–)	0.2b (±0.1)	0.2a (±–)	0.2a (±–)	0.0b (±–)	
α-Linolenic	18:3 ω-3	0.6a (±0.1)	0.6a (±–)	0.5b (±–)	0.6a (±0.1)	0.6a (±0.1)	0.5b (±0.1)	
cis-11,14 Eicosadienoic	20:2	0.7a (±0.1)	0.6a (±0.1)	0.7a (±0.1)	0.6b (±–)	0.9a (±0.2)	1.1a (±0.1)	
cis-8,11,14 Eicosatrienoic	20:3	0.4b (±0.1)	0.4b (±0.1)	0.5a (±0.1)	0.3c (±0.1)	0.6b (±0.1)	0.7a (±0.1)	
Arachidonic	20:4 ω-6	2.1ab (±0.5)	1.3b (±0.1)	3.0a (±0.5)	1.8c (±0.2)	2.7b (±0.4)	5.3a (±0.2)	
EPA	20:5 ω-3	0.3a (±0.2)	0.2a (±–)	0.2a (±0.1)	0.3b (±0.1)	0.3b (±0.1)	0.4a (±0.1)	
DHA	22:6 ω-3	1.2a (±0.3)	0.4b (±0.1)	1.3a (±0.3)	$1.0b~(\pm 0.1)$	1.1b (±0.2)	2.6a (±0.1)	
\sum	14.3a	13.8a	14.6a	14.2c	16.7b	18.9a		
$\sum NI^{a}$		1.1a	1.8a	2.2a	1.1a	1.4a	1.7a	
$\overline{\sum}$ UFA ^b		73.7	71.5	71.3	73.6	70.3	70.1	
$\sum \omega - 6^{c}$		11.5a	12.0a	11.9a	11.8b	13.8a	14.3a	
$\sum \omega - 3^d$		2.1a	1.2b	2.0a	1.9b	2.0b	3.5a	
U/S ^e		2.9a	2.7b	2.7b	2.9a	2.5a	2.5b	
P/S ^f		0.6a	0.5a	0.6a	0.6b	0.6b	0.7a	
ω-6/ω-3 ^g		5.5b	10.0a	6.0b	6.2a	6.9a	4.1b	

Variance analysis for dependent samples: when statistically significant, it was complemented with the Tukey's Procedure of Multiple Comparisons. For each fatty acid, and separately for fish with and without skin, the differences among raw, roasted and steamed fish are presented horizontally: numbers followed by the same letter were not statistically significant (P > 0.05).

^a NI: non-identified.

 $^{\rm b}$ $\sum UFA:$ sum of unsaturated fatty acids.

^c $\sum \omega$ -6: sum of ω -6 fatty acids (linoleic + *cis*-8,11,14 eicosatrienoic + arachidonic).

^d $\Sigma \omega$ -3: sum of ω -3 fatty acid (linolenic + EPA + DHA).

^e U/S: unsaturated/saturated fatty acid ratio.

^f P/S: polyunsaturated/saturated fatty acid ratio.

^g ω -6/ ω -3: ω -6/ ω -3 fatty acid ratio.

and analysed raw and cooked, using the two different methods.

After cooking using both methods, the carp showed a fatty acid profile similar to that of the raw carp, with the predominance of monounsaturated fatty acids, mainly oleic acid (18:1), followed by polyunsaturated and saturated fatty acids. Despite this, a decrease in monounsaturated fatty acids and an increase in saturated an d polyunsaturated fatty acids were observed in the samples without skin. In all the preparation methods, palmitic acid was found in a larger percentage in tilapias. Saturated fatty acids were prevalent, followed by monounsaturated and polyunsaturated fatty acids. The fatty acid profile of steamed and roasted tambacu was similar to that of raw tambacu, with higher proportions of saturated and mono-unsaturated fatty acids, particularly oleic (18:1) and palmitic (16:0).

A marked rise in the levels of arachidonic acid (20:4) and total PUFA occurs in carp after cooking, which may be explained by the loss of juices during thawing and water loss during the cooking process. A negative effect of dry heat cooking (baking) was also observed on the contents of ω -3 fatty acids due to the reduction of DHA (22:6) in carp with skin.

Although there was a significant statistical variation in the percentage of tridecanoic, miristic, heneicosanoic, behenic, *cis*-10 heptadecanoic, elaidic, vacenic, *cis*-11,14 eicosadienoic and *cis*-13,16 docosadienoic fatty acids in tilapias according to the preparation method, such variation was very small, but more accentuated for stearic, palmitoleic, oleic, α -linolenic, arachidonic and DHA acids. There was a reduction in the percentage of stearic, arachidonic and DHA acid in steamed skinless tilapia in comparison with raw and roasted fish. On the other hand, there

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Table 5					
Fatty acid profile (%) of raw	roasted and steamed	Nile tilapia, afte	er 30 days under	frozen storage:	with and without skin

Fatty acid		With skin			Without skin		
		Raw	Roasted	Steamed	Raw	Roasted	Steamed
Saturated							
Lauric	12:0	0.4a (±0.1)	0.3a (±–)	0.4a (±0.1)	0.3a (±0.1)	0.3a (±0.2)	0.4a (±0.1)
Tridecanoic	13:0	0.2a (±0.1)	0.3a (±–)	0.2a (±0.2)	0.2b (±0.1)	0.3ab (±0.1)	0.4a (±0.1)
Miristic	14:0	4.2a (±0.8)	4.8a (±0.1)	4.2a (±0.8)	3.1b (±0.1)	4.5ab (±0.5)	5.7b (±0.9)
Palmitic	16:0	27.1a (±2.0)	27.6a (±0.5)	26.9a (±2.2)	28.0a (±1.7)	27.8a (±0.2)	29.2a (±0.7)
Heptadecanoic	17:0	1.3b (±0.2)	1.8a (±0.1)	1.3b (±0.2)	1.4 a (±0.2)	1.7a (±0.1)	1.7a (±0.1)
Stearic	18:0	9.4a (±1.0)	10.1a (±0.2)	9.3a (±0.9)	11.0a (±0.8)	10.4a (±0.1)	8.7b (±0.2)
Heneicosanoic	21:0	0.4a (±0.2)	0.5a (±–)	0.4a (±0.1)	0.3b (±–)	0.5a (±–)	0.5a (±0.1)
Behenic	22:0	0.0b (±–)	0.3a (±–)	0.1ab (±0.1)	0.1a (±0.1)	0.3a (±0.1)	0.3a (±0.1)
\sum		43.0b	45.7a	42.8b	44.4a	45.8a	46.9a
Monounsaturated							
Miristoleic	14:1	1.7a (±0.3)	2.3a (±0.1)	1.7a (±0.3)	1.9a (±0.4)	2.1a (±0.1)	2.2a (±–)
Palmitoleic	16:1	5.9a (±0.7)	6.6a (±0.1)	5.8a (±0.6)	4.0c (±0.4)	6.0b (±0.1)	7.8a (±0.3)
cis-10 Heptadecanoic	17:1	0.8a (±0.2)	1.0a (±0.1)	0.8a (±0.2)	0.5b (±0.2)	0.9a (±0.1)	1.0a (±–)
Elaidic	18:1 9t	2.6a (±0.8)	3.2a (±0.1)	2.6a (±0.8)	2.2b (±0.3)	3.2a (±0.3)	2.6ab (±0.1)
Oleic	18:1 9c	26.5a (±3.3)	21.2a (±0.9)	26.5a (±3.5)	23.6a (±3.3)	20.6ab (±1.6)	17.6b (±0.8)
Vacenic	18:1 7c	0.7a (±0.3)	0.1b (±–)	0.7a (±0.3)	0.3a (±0.1)	0.1b (±0.1)	0.1b (±–)
\sum		38.2a	34.4a	38.1a	32.5a	32.9a	31.3a
Polyunsaturated							
Linoleic	18:3 ω-6	4.8a (±0.1)	5.0a (±0.1)	4.8a (±0.2)	4.6a (±0.3)	5.0a (±0.1)	4.9a (±0.2)
Linolenic	18:3 ω-3	2.4a (±0.9)	2.5a (±0.1)	2.3a (±0.8)	1.5c (±0.2)	2.3b (±0.1)	3.9a (±0.2)
cis-11,14 Eicosadienoic	20:2	1.4a (±0.2)	1.3a (±0.1)	1.4a (±0.2)	1.2b (±0.1)	1.2b (±–)	1.5a (±0.1)
cis-8,11,14 Eicosatrienoic	20:3 ω-6	0.6a (±0.1)	0.8a (±0.1)	0.6a (±0.1)	0.8a (±0.2)	0.7a (±0.1)	0.6a (±0.1)
cis-11,14,17 Eicotrienoic	20:3	0.3a (±0.1)	0.4a (±–)	0.2a (±0.2)	0.3a (±0.1)	0.3a (±0.1)	0.3a (±0.3)
Arachidonic	20:4 ω-6	2.1a (±0.7)	2.5a (±0.3)	2.1a (±0.7)	4.0a (±1.0)	2.9ab (±0.2)	2.0b (±0.3)
cis-13,16 Docosadienoic	22:2	0.5a (±0.1)	0.6a (±0.1)	0.5a (±0.1)	0.4b (±0.1)	0.5b (±–)	0.7a (±–)
EPA	20:5 ω-3	0.5a (±0.2)	0.5a (±0.1)	0.5a (±0.2)	0.9a (±0.2)	0.6a (±–)	0.7a (±0.1)
DHA	22:6 ω-3	2.6a (±2.3)	2.9a (±0.4)	3.2a (±1.3)	6.3a (±1.5)	3.4b (±0.2)	3.2b (±0.5)
\sum		15.2a	16.5a	15.6a	20.0a	16.9a	17.8a
$\sum NI^{a}$		3.6a	3.3a	2.9a	3.1a	4.3a	2.6a
$\sum UFA^{b}$		53.4	50.9	53.7	52.5	49.8	49.1
$\sum \omega - 6^{c}$		7.5a	8.3a	7.5a	9.4a	8.6a	7.5a
$\sum \omega - 3^d$		5.5a	5.9a	6.0a	8.7a	6.3a	7.8a
Ī/S ^e		1.24a	1.11b	1.26a	1.18a	1.09b	1.05b
P/S ^f		0.35a	0.36a	0.37a	0.45a	0.37b	0.38b
ω-6/ω-3 ^g		1.36a	1.41a	1.25a	1.08b	1.37a	0.96b

Variance analysis for dependent samples: when statistically significant, it was complemented with the Tukey's Procedure of Multiple Comparisons. For each fatty acid, separately for fish with and without skin, the differences among raw, roasted and steamed fish are shown horizontally: numbers followed by the same letter were not statistically significant (P > 0.05).

^a NI: non-identified.

^b \sum UFA: sum of unsaturated fatty acids.

^c $\sum \omega$ -6: sum of ω -6 fatty acids (linoleic + *cis*-8,11,14 eicosatrienoic + arachidonic).

^d $\sum \omega$ -3: sum of ω -3 fatty acid (linolenic + EPA + DHA).

 e U/S: ratio unsaturated/saturated fatty acids.

^f P/S: ratio polyunsaturated/saturated fatty acids.

^g ω -6/ ω -3: ω -6/ ω -3 fatty acid ratio.

was an increase in the percentage of the palmitoleic, oleic and linolenic acids in the skinless steamed fish compared to raw and roasted fish.

In tambacu, a significant variation was observed in some fatty acids when submitted to cooking. For instance, the content of vacenic acid (18:1 7c) was reduced to about 83%, but this reduction had little practical significance, since the percentage of this fatty acid in the raw samples was lower than 1%.

It should be emphasized that the elevated percentages of myristic (14:0), palmitoleic (16:1) and linolenic (18:3) acids

in tambacu following cooking, particularly steaming, were found in both samples, with and without skin. However, such increase cannot be attributed to the cooking method, since different samples were used for the analyses before and after cooking; thus, it is possible that the initial content of these acids was higher in the samples submitted to cooking than in the raw ones.

Table 6 shows increased percentages of cis-10 heptadecaenoic (17:1) acid in the skinned tambacu samples, after applying the two cooking techniques. Conversely, the percentages of arachidonic (20:4) and DHA (22:6) acids were

Table 6
Fatty acid profile (%) of raw, roasted and steamed tambacu, after 30 days under frozen storage, with and without skin

Fatty acid		With skin			Without skin			
		Raw	Roasted	Steamed	Raw	Roasted	Steamed	
Saturated								
Miristic	14:0	3.9b (±0.8)	4.6ab (±−)	5.3a (±0.3)	2.3b (±0.1)	4.2ab (±0.5)	5.5a (±0.9)	
Palmitic	16:0	25.5a (±2.2)	26.0a (±0.4)	27.3a (±0.5)	27.0a (±2.0)	26.4a (±0.3)	28.1a (±1.2)	
Heptadecanoic	17:0	1.2b (±0.2)	1.7a (±0.1)	1.8a (±0.2)	1.33a (±0.23)	1.6a (±–)	$1.6a (\pm -)$	
Stearic	18:0	9.0ab(±1.1)	9.5a (±0.2)	7.7b (±0.5)	10.6a (±0.7)	9.9a (±0.2)	8.3b (±0.3)	
Arachidic	20:0	0.2a (±0.2)	0.4a (±0.1)	0.4a (±0.1)	0.3b (±–)	0.4a (±0.1)	$0.0c(\pm -)$	
Heneic	21:0	0.4a (±0.2)	$0.5a(\pm -)$	0.5a (±0.1)	$0.3a(\pm -)$	$0.4a(\pm -)$	$0.3a(\pm 0.2)$	
\sum		40.2b	42.7a	43.0a	42.5a	43.0a	43.9a	
Monounsaturated								
Palmitoleic	16:1	5.5b (±0.8)	6.2b (±0.1)	7.8a (±0.2)	3.9c (±0.4)	5.7b (±0.2)	7.5a (±0.4)	
cis-10 Heptadecanoic	17:1	0.7a (±0.2)	$1.0a(\pm 0.1)$	$1.0a(\pm 0.1)$	0.5b (±0.2)	$0.9a(\pm -)$	$1.0a(\pm -)$	
Elaidic	18:1 9c	2.5a (±0.8)	3.1a (±0.1)	2.4a (±0.2)	2.1b (±0.3)	3.0a (±0.4)	2.5ab (±0.1)	
Oleic	18:1 9c	25.4a (±3.0)	19.9b (±0.9)	$17.0b(\pm 1.0)$	22.9a (±3.4)	19.5ab (±1.3)	16.9b (±0.5)	
Vacenic	18:1 7c	$0.6a (\pm 0.3)$	0.1b (±–)	$0.6a (\pm 0.1)$	0.3a (±0.1)	$0.3a (\pm 0.1)$	0.6a (±0.1)	
\sum		34.7a	30.3b	28.8b	29.7a	29.4a	27.9a	
Polyunsaturated								
Linoleic	18:2 ω-6	6.9a (±0.6)	6.7a (±0.2)	6.1a (±0.1)	6.3a (±1.3)	6.9a (±0.6)	6.2b (±0.3)	
γ-Linolenic	18:3 ω-6	0.6b (±0.2)	0.9a (±0.6)	0.9a (±0.6)	0.4b (±0.1)	$0.8a~(\pm 0.1)$	0.8a (±0.2)	
Linolenic	18:3 ω-3	2.3b (±0.9)	2.4b (±0.1)	$4.0a~(\pm 0.1)$	$1.5c (\pm 0.2)$	2.2b (±0.1)	3.7a (±0.1)	
cis-11,14 Eicosadienoic	20:2	1.4a (±0.3)	$1.3a (\pm 0.1)$	1.5a (±0.1)	1.1a (±0.1)	1.1a (±–)	1.1a (±–)	
cis-11,14,17 Eicotrienoic	20:3 ω-3	0.3a (±0.1)	0.4a (±0.1)	0.4a (±–)	0.3a (±0.1)	0.3a (±–)	0.3a (±0.2)	
Arachidonic	20:4 ω-6	2.0a (±0.6)	2.3a (±0.3)	1.5a (±0.1)	3.9a (±0.9)	2.7ab (±0.2)	1.9b (±0.2)	
cis-13,16 Docosadienoic	22:2	0.5b (±0.1)	0.6ab (±0.1)	0.7a (±–)	0.4b (±0.1)	0.5b (±–)	0.7a (±–)	
EPA	20:5 ω-3	0.5a (±0.3)	0.5a (±0.1)	0.6a (±–)	0.9a (±0.2)	0.6b (±0.1)	0.7ab (±0.1)	
DHA	22:6 ω-3	2.5a (±2.2)	2.7a (±0.4)	2.3a (±0.1)	6.1a (±1.4)	3.2b (±0.2)	3.1b (±0.4)	
$\sum_{i=1}^{n}$		16.9a	17.7a	18.1a	20.9c	18.2b	18.8a	
$\overline{\sum} NI^a$		1.1a	1.3a	1.4a	1.0a	1.3a	1.4a	
$\sum UFA^{b}$		25.8a	24.0a	23.5a	25.3a	23.8a	23.3a	
$\sum \omega - 6^{c}$		9.5a	10.0a	10.0a	10.6a	10.4a	8.9a	
$\sum_{i=1}^{n} \omega - 3^{d}$		5.6 ^a	6.0a	7.4a	8.7a	6.2a	7.8a	
U/S ^e		1.3 ^a	1.1b	1.1b	1.2a	1.1a	1.1a	
P/S ^f		0.4^{a}	0.4a	0.4a	0.5a	0.4a	0.5a	
ω-6/ω-3 ^g		1.9 ^a	1.7a	1.2a	1.2b	1.7a	1.1b	

Variance analysis for dependent samples: when statistically significant, it was complemented with the Tukey's Procedure of Multiple Comparisons. For each fatty acid, separately for fish with and without skin, the differences among raw, roasted and steamed fish are shown horizontally: numbers followed by the same letter were not statistically significant (P > 0.05).

^a NI: non-identified.

 $^{\rm b}$ $\sum {\rm UFA:}$ sum of unsaturated fatty acids.

^c $\sum \omega$ -6: sum of ω -6 fatty acids (linoleic + *cis*-8,11,14 eicosatrienoic + arachidonic)

^d $\sum \omega$ -3: sum of ω -3 fatty acids (linolenic + EPA + DHA).

^e U/S: unsaturated/saturated fatty acids ratio.

^f P/S: polyunsaturated/saturated fatty acids ratio.

 $^{\rm g}$ $\omega\text{-}6/\omega\text{-}3$: $\omega\text{-}6/\omega\text{-}3$ fatty acid ratio.

lower in cooked tambacu without skin. The fatty acid DHA (22:6) has a high number of double bonds, thus being highly susceptible to oxidation (Silva et al., 1993), what may have led to its reduced percentage.

Lipids can undergo several reactions during the cooking process, such as hydrolysis and oxidation, which may affect flavour, scent, colour and texture, and its nutritional value (Silva et al., 1993). Alterations in lipid level after steaming and roasting are related with lipid content of each species, temperature, species size and exposed surface (Gall, Otwell, Koburger, & Appledorf, 1983; Silva et al., 1993). In general, even when a significant statistical variation was found between the cooking methods, the difference in the percentages of fatty acids was small, which demonstrates that baking and steaming had little influence on fatty acid composition of the species analyzed. Such differences may be rather attributed to the water and lipids leached out during thawing and cooking than to lipid reactions (such as oxidation) during heat treatment.

Several studies show that in general, baking, steaming or grilling saltwater fish have little influence on their lipid content and fatty acid profile, while frying increases the linoleic acid content (18:2), which is related to the cooking oil used for frying. Gladyshev, Sushchik, Gubanenko, Demirchieva, and Kalachova (2006) reported that heat treatment (boiling and roasting) did not decrease content of EPA and DHA in humpback salmon, except a modest reduction after frying. Applying roasting and grilling techniques, Gall et al. (1983) did not observe any significant changes in the composition of fatty acids of four species of sea water fish, differently from what occurred when vegetable oil frving was used. Heat treatment (cooking and frying) did not generally decrease the contents of EPA and DHA in sea trout, herring, rock sole and cod (Gladyshev, Sushchik, Gubanenko, Demirchieva, & Kalachova, 2007). Microwave cooking did not alter significantly the lipid total content and percentage of fatty acids in sea water fish, according to studies carried out by Hearn, Sgoutas, Sgoutas, and Hearn (1987). However, Silva et al. (1993), in a study carried out in Brazil, verified that conventional oven-baking and steaming did not significantly alter saltwater fish fatty acid stability, whereas microwave oven cooking reduced PUFA concentration.

The literature is scarce on the effect of cooking on freshwater fish. When analyzing three freshwater fish species, Mai, Shimp, Weihrauch, and Kinsella (1978) reported little influence of baking on fatty acid profile and an increase in linoleic acid content (18:2) after frying, except for trout.

During steaming or baking, it is common to keep fish skin to protect meat structure, as it is very tender, removing it before consumption. It was observed that skin removal did not reduce the contents of ω -3 and ω -6 fatty acid, which are considered beneficial, especially ω -3.

4. Conclusions

Skin removal reduced total lipid content in the three species of fish. However, fatty acid composition was little affected by the procedure, except in tambacu. This fish showed a tendency to increasing the total polyunsaturated fatty acid content and reducing the monounsaturated fatty acid content following skin removal. This suggests that the muscle portion has higher amounts of polyunsaturated fatty acids than the subcutaneous layer, which was removed with the skin.

Sixteen fatty acids were found in carp, with greater percentage (on average 53.78%) of monounsaturated fatty acids. The oleic (18:1), linoleic (18:2) and palmitic (16:0) acids were present in larger amounts, considering all the frozen storage times and preparation methods. Nile tilapia presented high levels of saturated fatty acids and lower amounts of polyunsaturated fatty acids when compared to other freshwater fish species. In spite of that, tilapia is considered a lean fish (average of 0.64 g of lipid/100 g sample). Palmitic (16:0) and oleic (18:1) acids were present in higher levels in tilapias, considering all the frozen storage times and preparation methods. The saturated and monounsaturated fatty acids were found in higher proportions in tambacu, particularly oleic acid (18:1) which amounted to 27.7% of the total percentage of fatty acids, on average, and palmitic acid (16:0), which amounted to 24%.

In general, the composition of fatty acids did not present great variations due to frozen storage time and preparation methods, indicating that these methods had no important interference in the fatty acid composition of common carp, Nile tilapia and tambacu. Exception was observed for raw tambacu, in which frozen storage increased the content of saturated fatty acids and reduced the content of monounsaturated acids. The analyses of the roasted and steamed fish following thawing showed that shorter frozen storage periods provided a better preservation of the monounsaturated fatty acids, with saturated fatty acids increasing with increased storage time. A small decrease in monounsaturated fatty acids and an increase in saturated and polyunsaturated fatty acids were also observed in samples of carp without skin after heat treatment.

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