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Formulation of fish waste meal for human nutrition

Maria Luiza Rodrigues de Souza[1*], Grazyella Massako Yoshida[2], Daniel Abreu Vasconcelos Campelo[3], Lorena Batista de Moura[4], Tadeu Orlandi Xavier[4] and Elenice Souza dos Reis Goes[5]

*[1]Departamento de Zootecnia, Centro de Ciências Agrárias, Universidade Estadual de Maringá, Avenida Colombo, 5790, 87020-900, Maringá, Paraná, Brazil. [2]Programa de Pós-Graduação em Genética e Melhoramento Animal, Universidade Estadual Paulista, Jaboticabal, São Paulo, Brazil. [3]Instituto de Estudos Costeiros, Universidade Federal do Pará, Bragança, Pará, Brazil. [4]Programa de Pós-Graduação em Zootecnia, Universidade Estadual de Maringá, Maringá, Paraná, Brazil. [5]Faculdade de Ciências Agrárias, Universidade Federal da Grande Dourados, Dourados, Mato Grosso do Sul, Brazil. *Author for correspondence. E-mail: mlrsouza@uem.br*

ABSTRACT. This study aimed to elaborate and characterize meals containing waste from processing of tilapia, tuna, salmon and sardine for human consumption. Carcasses of tilapia and salmon, tuna torsos without fins and sardine tails were cooked, pressed, milled and dehydrated, resulting in waste meal. Greater protein (83.28%) and lower mineral matter (5.31%) were observed in tuna meal. Salmon meal presented greater content of lipids (18.81%) and sardine meal, lower content (3.98%). Tilapia meal presented greater mineral matter (37.66%), calcium (9.37%) and phosphorus (6.08%). Higher content of iron was observed in sardine and tuna meals. Higher amounts of fatty acids from n-3 series were found in salmon (53.71 g kg^{-1}), sardine (47.46 g kg^{-1}) and tuna (36.98 g kg^{-1}). Concerning amino acids, glutamic acid showed greater proportion in all meals, followed by lysine, leucine, glycine and aspartic acid. All meals presented high biological and nutritional values and are regarded as important sources of calcium, phosphorus and iron.

Keywords: waste reuse, Nile tilapia, salmon, tuna, sardine.

Elaboração de farinhas de resíduos do beneficiamento de peixes para alimentação humana

RESUMO. Este estudo objetivou elaborar e caracterizar farinhas de resíduos do beneficiamento da tilápia, atum, salmão e sardinha para consumo humano. Carcaças de tilápia e salmão, troncos de atum sem nadadeiras e caudas de sardinha foram cozidas, prensadas, moídas e desidratadas, resultando nas farinhas. Maior proteína (83,28%) e menor matéria mineral (5,31%) foram observados na farinha de atum. Farinha de salmão apresentou maior teor de lipídeos (18,81%) e a de sardinha, menor teor (3,98%). Farinha de tilápia apresentou maior matéria mineral (37,66%), cálcio (9,37%) e fósforo (6,08%). Observou-se maior teor de ferro nas farinhas de sardinha e atum (121,95 e 106,38 mg kg^{-1}). Maiores quantidades de ácidos graxos da série n-3 foram encontradas nas farinhas de salmão (53,71 g kg^{-1}), sardinha (47,46 g kg^{-1}) e atum (36,98 g kg^{-1}). No perfil de aminoácidos, o ácido glutâmico apareceu em maior proporção em todas as farinhas, seguido pela lisina, leucina, glicina e ácido aspártico. Todas as farinhas apresentaram alto valor biológico e nutricional, sendo boa fonte de cálcio, fósforo e ferro.

Palavras-chave: aproveitamento de resíduos, tilápia-do-Nilo, salmão, atum, sardinha.

Introduction

Aquaculture has taken over the responsibility of supporting the consumer market of fish, once extractive fishery tends to stagnation. In 2009, the Brazilian aquiculture production reached 337,353 tons, and tilapia, mainly *Oreochromis niloticus* species, represented almost 40% of total, with a production of 132,958.3 tons (Ministério da Pesca e Aquicultura [MPA], 2012). On the other side, the salmon (*Salmo salar*) consumed in Brazil come from importations, with Chile as the main supplier (MPA, 2012). Likewise, the majority of tuna (*Thunnus* sp.) consumed in Brazil come from other countries, as the Brazilian marine fishing of this species was 725 tons in 2010 (MPA, 2012). The main fishery resource in Brazil is the sardine (*Sardinella brasiliensis*), with production of 62,134 tons in 2010 (MPA, 2012).

Fish and its derivatives are distinguished for high protein content of high biological value, with balanced amino acids, containing great proportions of methionine and cysteine, the majors limiting amino acids in vegetable proteins (Neves, Mira, & Marquez, 2004). In addition, they present high values of fat-soluble vitamins (A, D, E, K) and water-soluble from complex B, minerals (calcium, iron and phosphor) and lipids (Belda & Pourchet-Campos, 1991). Lipids found in marine fish are rich

in polyunsaturated fatty acids, mainly the ones from omega-6 series (n-6) and omega-3 (n-3), such as arachidonic acid (ARA or C20:4n-6) and the eicosapentaenoic acid (EPA or C20:5n-3) respectively, with EPA being able to originate docosahexaenoic (DHA or C22:6n-3) (Tapiero, Nguyen, Couvreur, & Tew, 2002, Tocher, 2003).

The usage of waste generated by fish industry in Brazil is very low (Godoy et al., 2010) and the quantity of produced residue is significant, varying from 8 to 16% when the final product is gutted fishery or 60 to 72% when the final product is skinless fillet (Kubitza & Campos, 2006). These residues present great quantity of material rich in protein, which are usually converted into products for animal nutrition, such as meals and fertilizers of low market value (Chalamaiah, Kumar, Hemalatha, & Jyothirmayi, 2012), fish oil, protein concentrates, surimi, pâtés, silage, among others (Morales-Ulloa & Oetterer, 1995, Bimbo, 2000).

According to Dekkers, Raghavan, Kristinsson, and Marshall (2011), only 40% of fishery products are addressed to human consumption. Considering nutritional quality of waste of fish industry, alternative options of transforming them into adequate products for human consumption have been sought, aiming to recover essential nutrients and bioactive compounds that might assist human health improvement, besides solving pollution issues (Chalamaiah et al., 2012).

Several studies were conducted in order to develop fish meals for human consumption (Franco et al., 2009, Godoy, Franco, Souza, Stevanato, & Visentainer, 2013), by adding it in products like cookies and biscuits (Franco et al., 2013), snacks (Justen et al., 2011), stocks and soups (Godoy et al., 2010), among others. This inclusion was considered viable to increase nutritional value of products; apart from the fact that developing products with addition of fish and its derivatives is a way of supporting this product consumption and incrementing fishery productive chain.

Thus, the objective of this work was to develop and characterize meals containing waste from processing of tilapia, tuna, salmon and sardine.

Material and methods

The meal production was carried out in the Laboratory of Fishery Technology of Iguatemi Experimental Farm, Universidade Estadual de Maringá (UEM), in November, 2012. For that, 4 kg tilapia finless carcasses (*Oreochromis niloticus*), 8.1 kg tuna whole torsos (*Thunnus* sp.) without fins (with bones, skin and muscles, considered out of standards of industry classification), 4 kg salmon finless carcasses (*Salmo salar*) and 4.73 kg sardine tails (*Sardinella brasiliensis*). Bones with adherent meat was considered carcass. All raw material came from waste processing of fishery industries in Rolândia, state of Paraná region (SmartFish) and Itajaí, state of Santa Catarina (GDC Alimentos SA).

The raw material was washed in chlorine water and weighed, then stored separately in industrial pressure cookers (20 L capacity) containing enough water to cover the raw material. In sequence, 0.5% BHT antioxidant was added and 0.5% Peroxitane. The residue was cooked in industrial stove for sixty minutes. After this period, the cooked feedstock was pressed in hydraulic press. The resulting cake was milled in an electric meat grinder, with the resulting mass subjected to dehydrator for drying at 60°C for 24 hours. After dehydrated, the mass was milled in a knife mill, resulting in fish meals used in the evaluations. Fish meals were conditioned in plastic bags and stored in freezer at -18±2ºC until analysis.

Contents of moisture, proteins, lipids and ash of fish meals were determined in different fish species. The quantifications were carried out in triplicate, according to AOAC methodology (Horwitz, 2005). Phosphorus content was determined in accordance with the methodology described by Silva and Queiroz (2002). Calcium and iron verifications in samples required digestion in acidic medium and the quantification was performed by flame atomic absorption spectrometry (Zhou, Cheng, Chan, & Wong, 1998). The quantification of fatty acids was conducted according to AOAC methodology (Horwitz, 2005). Concerning amino acids analysis of different fish meals, the methodology described by White, Hart, and Fry (1986) and Hagen, Frost and Augustin (1989) was employed.

Data obtained in proximate composition analysis and minerals, were subjected to analysis of variance (ANOVA) at 5% significance and, in the case of significant differences ($p < 0.05$) the Tukey test was applied; using software SAS. Data regarding fatty acids and amino acids had only descriptive character.

Results and discussion

The proximate composition indicates, in general, the nutritional value of the food, thus the meal produced with the carcass of different fish species used had varied values of moisture, protein, lipid and ash (Table 1). Salmon meal presented the highest lipid content, significantly differing from the others ($p < 0.05$), likewise sardine meal, which showed the lowest value. Tilapia and tuna meals did not differ regarding lipid content.

Table 1. Proximate composition of meals produced with waste from fish processing.

Parameters (%)	Meals from processing waste			
	Tilapia	Salmon	Tuna	Sardine
Moisture	1.78±0.11b	2.62±0.06b	4.57±0.03a	4.86±0.81a
Crude Protein	51.13±1.44c	44.63±0.37d	83.28±0.33a	61.88±0.91b
Lipid	5.82±1.16b	18.81±0.22a	5.60±0.25b	3.98±0.28c
Ash	37.66±0.45a	30.20±0.07b	5.31±0.39d	26.13±0.17c

Means in same row followed by distinct letters are significantly different by the Tukey's test ($p < 0.05$). Values expressed in Mean ± Standard deviation.

Concerning ash content, there was a significant difference ($p < 0.05$) for all studied species, the same was observed for meal protein content of different fish species. Moisture content was significantly higher in tuna and sardine meals (Table 1). The highest protein content and the lowest ash content were observed for tuna meal, which can be explained due to the use of fish torsos in the meal elaboration, once these torsos showed more meat and fewer bones, when compared to carcasses used in the other meals.

The moisture content recommended by the Regulation of Industrial and Sanitary Inspection of Animal Products (*Regulamento da Inspeção Industrial e Sanitária de Produtos de Origem Animal* [Riispoa], 1997) is 12%. Thus, the moisture values found for meals of all species were satisfactory. Low moisture value, as well as a package impermeable to liquids and gases, increases the product's shelf life (Haj-Isa & Carvalho, 2011).

Stevanato et al. (2007) found 6.0% moisture in tilapia-head meal, while for protein, lipids and ash, the contents were 38.4, 35.5, and 19.4%, respectively. Data from chemical composition of tilapia carcass meal, reported by Petenuci et al. (2010), were 14.2% moisture, 40.8% protein content, 18.3% ash, and for lipid content was 25.3%. In the present study, the tilapia carcass meal presented higher contents of ash and protein. Regarding the protein concentrate developed with meat mechanically separated from waste from tilapia filleting, Vidal, Rodrigues, Zapata, and Vieira (2011) obtained a product with protein content of 62.39 and 32.63% of fat.

All the evaluated meals showed great proximate composition, being considered satisfactory sources of proteins and minerals. High values of protein, lipids and minerals are directly related to low moisture content, since a reduction in meal moisture content causes an increase in the concentration of other compounds (Stevanato et al., 2007).

The contents of calcium, phosphorus and iron differed between the meals (Table 2). Tilapia meal showed the highest value ($p < 0.05$) of calcium and phosphorus, but also the lowest value ($p < 0.05$) of iron. Salmon and sardine meals presented similar values of calcium and phosphorus. As it contains lower percentages of ash, tuna meal exhibited the lowest content of calcium and phosphorus, however, the greatest value of iron, similar to the one found in sardine meal.

Table 2. Contents of calcium, iron and phosphorus in meals produced with waste from fish processing.

Mineral	Meals from processing waste			
	Tilapia	Salmon	Tuna	Sardine
Calcium (%)	9.37±0.53a	8.88±0.75ab	0.90±0.002c	6.60±0.28b
Phosphorus (%)	6.08±0.26a	4.79±0.19b	0.91±0.06c	4.03±0.31b
Iron (mg kg^{-1})	35.18±2.46c	73.29±1.37b	106.38±3.04ab	121.95±15.24a

Means in the same row followed by distinct letters are significantly different by the Tukey's test ($p < 0.05$). Values expressed in Mean ± Standard deviation.

Flavored meals of tilapia carcasses elaborated by Godoy et al. (2013) presented 1.78% calcium, 5.47% phosphorus and 2.36 mg 100 g^{-1} iron; distinct values from the ones found in tilapia meal produced in this study. In sardine muscle, (*Opisthonema oglinum*), Andrade, Bispo, and Druzian (2009) found 25.36 mg 100 g^{-1} calcium and 0.71 mg 100 g^{-1} iron, superior values compared to findings of mullet muscles (*Mugil* spp) and lane snapper (*Lutjanus synagris*), according to the authors above. Thus, it is noticed that fish meal is able to aggregate nutrients, such as proteins and lipids, as well as minerals naturally found in fish.

The results of minerals in different fish meals prove that fish meat is source of mainly calcium and phosphorus but also contains iron, copper and selenium (Sartori & Amancio, 2012). The iron content in all meals produced in this study was higher than in other meats (Sarcinelli, Venturini, & Silva, 2007).

In the meal lipid fractions of different fish species, 29 fatty acids were found (Table 3). Sardine and tuna meals presented predominance of cis-docosahexaenoic acid (22:6n-3), with proportion of 36.49 and 30.91 g kg^{-1}, respectively. Regarding salmon meal, the most abundant fatty acid (54.52 g kg^{-1}), was oleic acid (18:1n-9 c), the same was predominant in the fatty acid profile of tilapias, in proportion of 25.66 g kg^{-1} (Table 3).

The eicosapentaenoic acid (EPA, 20:5n-3) and the docosahexaenoic acid (DHA, 22:6n-3) were found in inferior proportion in tilapia meal. The same was observed for the sum of polyunsaturated fatty acids (Pufa) and for the sum of n-3 series fatty acids (Table 3).

Petenuci et al. (2010) found a total of 24 fatty acids in tilapia carcass meal, and likewise this study, the predominant fatty acid was oleic acid (18:1n-9), with 35.15%, followed by palmitic (16:0) and linoleic (18:2n-6) acids, with contents of 27.4 and

11.82%, respectively. These fatty acids were also predominant in the lipid fraction of tilapia-head meals (Stevanato et al., 2007). The oleic acid (18:1n-9) is the precursor of arachidonic acid (ARA, C20:4n-6), which can be incorporated into phospholipids of cell membranes and has fundamental role in the eicosanoids production (prostaglandins, prostacyclins, tromboxanes and leukotrienes). The eicosanoids, in turn, perform regulatory functions in different tissues (Suárez-Mahecha, Francisco, & Beirão, 2002).

Table 3. Fatty acids profile (g kg^{-1}) of meals produced with waste from fish processing.

Fatty acids	Meals from processing waste			
	Tilapia	Salmon	Tuna	Sardine
12:00	0.08	0.13	0.08	0.13
14:00	2.87	5.42	3.13	4.46
15:00	0.13	0.34	0.66	0.72
16:00	21.05	28.83	18.86	16.97
17:00	0.24	0.52	0.78	0.69
18:00	5.59	8.16	6.52	3.67
20:00	0.1	0.4	0.19	0.18
22:00	0.08	0.21	0.16	0.12
24 00	1.63	0.38	0.39	0.24
23:00	0.03	0.06	0.06	ND
21:00	0.05	ND	ND	ND
14:1n-5	0.19	0.04	0.41	0.02
16:1n-7	5.2	7.35	3.32	3.45
20:1n-9	1.41	2.9	1.25	2.22
24:1n-9	0.07	0.68	0.52	0.47
17:01	ND	0.07	ND	ND
18:1n9 t	0.4	0.12	0.11	ND
22:1n-9	0.05	0.4	0.11	0.2
18:1n-9 c	25.66	54.52	11.28	5.16
20:2n-6	0.46	1.92	0.14	0.18
18:3n-3	0.84	7.54	0.61	0.8
20:3n-3	0.1	0.57	0.1	0.1
20:5n-3	0.15	15.79	5.36	10.07
22:6n-3	3.2	29.81	30.91	36.49
18:2n-6	11.24	28.18	1.41	1.25
18:3n-6	1.19	0.32	0.07	0.1
20:3n-6	0.98	0.84	0.08	0.1
20:4n-6	2.97	1.49	2.08	1.42
22:2n-6	0.04	0.2	ND	ND
SFA	31.85	44.45	30.82	27.17
Mufa	32.99	66.08	17	11.52
Pufa	21.16	86.67	40.76	50.51
Pufa/SFA	0.66	1.95	1.32	1.86
n-3	4.28	53.71	36.98	47.46
n-6	12.26	29.23	1.49	1.35
n-6/n-3	2.86	0.54	0.04	0.03

Abbreviations: ND = non-detected; SFA = sum of saturated fatty acids; Mufa = sum of monounsaturated fatty acids; Pufa = sum of polyunsaturated fatty acids; n-6 and n-3 = sum of fatty acids of n-6 and n-3 series, respectively; Pufa/SFA = ratio between the sum of polyunsaturated and saturated acids; and n-6/n-3 = ratio between the sum of n-6 and n-3 series acids.

In lipids of saltwater fish, such as sardine, salmon and tuna, the main fatty acids found are the polyunsaturated ones, like the eicosapentaenoic (EPA, 20:5n-3) and the docosahexaenoic (DHA, 22:6n-3) (Gunstone, Harwood, & Padley, 1994, Lands, 2005, Turon, Rwabwogo, Baréa, Pina, & Graille, 2005). These fatty acids have an important role in maintaining mechanical and osmotic stability in plasmatic membranes of cells (Steffens, 1997); they are also found in fish yolk sacs, considered important for the visual and neural development of larvae (Sargent, Mcevoy, & Bell, 1997, Glencross, 2009). Benefits for human beings are also attributed to these n-3 series fatty acids, combating and preventing cardiovascular diseases, brain disorders and cancer (Lands, 2005).

The proportions of EPA found in sardine, salmon and tuna meals were 10.07, 15.79 and 5.36 g kg^{-1}, and were much greater than in tilapia meals, 0.15 g kg^{-1}. The DHA was observed in proportions of 36.49, 29.81 and 30.91 g kg^{-1}, for sardine, salmon and tuna meals, respectively; these proportions were higher than in tilapia meals, which had 3.20 g kg^{-1}. The DHA is originated from EPA, which is synthesized from linolenic acid (18:3n-3) (Tapiero et al., 2002, Tocher, 2003).

Although presenting greater sum of polyunsaturated fatty acids in sardine, salmon and tuna meals, 50.51, 86.67, 40.76 g kg^{-1}, respectively, against 21.16 for tilapia meal, saltwater fish have lower capability of synthesizing these fatty acids compared to freshwater fish. This because of an inferior enzymatic activity of Δ6 and Δ5, responsible for lengthening and desaturating linoleic acids (18:2n-6) and linolenic acids (18:3n-3), which are precursors of the other (Olsen, 1998). The natural environment of saltwater fish explains this contradiction, as the saltwater phytoplankton and zooplankton are rich in polyunsaturated fatty acids, essentially the ones from n-3 series, thereby; saltwater fish naturally acquire these acids from their diet (Zenebe, Ahlgren, Gustafsson, & Boberg, 1998).

This preeminent distribution of polyunsaturated fatty acids reflects on the ratio, polyunsaturated/saturated fatty acids (Pufa/SFA), the meals derived from saltwater fish presented the highest values of this ratio, sardine meal demonstrated 1.86, salmon meal 1.95 and 1.32. Even the lowest value, 0.66, presented by tilapia meal, is not inferior to 0.40, the value that indicates that the product is unhealthy (Wood et al., 2003). The value of Pufa/SFA ratio found for tilapia-head meal was 0.47 (Stevanato et al., 2007) while Petenuci et al. (2010) found 0.41 in tilapia-carcass meal.

As saltwater fish have a diet rich in n-3 series fatty acids, freshwater fish show a higher n-6/n-3 ratio (Olsen, 1998, Zenebe et al., 1998). This ratio was 2.86 for tilapia meal, in contrast; sardine, salmon and tuna meals showed values of 0.03, 0.54 and 0.04, respectively. A few authors assert that the ideal n-6/n-3 ratio for human is from 1.00 to 2.00 (Granados, Quiles, Gil, & Ramírez-Tortosa, 2006). In order to prevent cardiovascular disease, a proportion of 4:1 for n-6:n3 is recommended (Candela, López, & Kohen, 2011) however, in

human nutrition this ratio can reach values of 10.00 to 25.00, causing fatty acids imbalance (Stevanato, Souza, Matsushita, & Visentainer, 2007). Therefore, the use of meals with low n-6/n-3 ratio as part of the diet conveniently avoids this imbalance.

All the meals evaluated in the present study contained all the essential amino acids (EAA). The glutamic acid was the most abundant in all meals, followed by lysine, leucine, glycine and aspartic acid (Table 4).

Table 4. Amino acids profile (mg g^{-1} of protein) of meals produced with waste from fish processing.

Amino acid	Meals from processing waste				Standard WHO[1]	
	Tilapia	Salmon	Tuna	Sardine	Children	Adults
Alanine	68.44	65.21	59.57	64.47		
Arginine	68.88	65.89	60.33	65.67		
Aspartic acid	75.37	92.80	100.13	96.20		
Glycine	76.46	76.38	44.68	62.43		
Glutamic acid	133.20	83.22	137.92	140.54		
Proline	57.18	49.48	34.67	45.03		
Serine	44.62	45.37	40.93	43.83		
Taurine	1.30	ND	ND	ND		
Histidine	22.74	29.18	49.06	25.24	16.00	15.00
Isoleucine	44.62	42.86	47.43	43.49	31.00	30.00
Leucine	77.11	72.96	83.10	77.95	61.00	59.00
Lysine	86.85	131.33	95.37	89.03	48.00	45.00
Methionine + Cysteine	42.02	43.55	42.05	43.66	24.00	22.00
Phenylalanine + Tyrosine	87.72	86.64	87.23	88.01	41.00	38.00
Threonine	50.25	49.25	49.94	49.29	25.00	23.00
Valine	63.46	65.44	67.21	64.98	40.00	39.00

[1]World Health Organization (WHO, 2007). Children from 3 to 10 years old and adults above 18 years old.

Fish present 10 essential amino acids: arginine, histidine, threonine, tyrosine, valine, methionine, isoleucine, leucine, phenylalanine and lysine (Kubitza, 2000). All different evaluated meals presented all essential amino acids (EAA). Regarding fish muscle tissue, Furuya, Botaro, Neves, Silva, and Hayashi (2004) stated that lysine is the most abundant EAA, however, as the meals were produced from tilapia and salmon carcasses, sardine tails and whole torsos of tuna, such fact was not observed in the present study.

In fish protein hydrolysates, the amino acids found in greater quantities are aspartic acid and glutamic acid (Klompong et al., 2009, Yin et al., 2010, Ghassem et al., 2014, Hou, Li, & Zhao, 2011). Lysine and leucine were the most abundant EAA in acid, biological and enzymatic silages, prepared with tilapia residues, presenting respective values of 3.33, 2.41 and 3.22% for lysine and of 3.50, 2.41 and 3.31% for leucine (Borghesi, Arruda, & Oetterer, 2007).

Comparing the amino acid profile of all meals to the FAO standard (Food and Agriculture Organization) (WHO, 2007) in Table 4, it is verified that the produced meals showed higher values than the required, which highlights the quality of proteins found in tilapia, salmon, sardine and tuna meals.

Conclusion

The developed meals present high biological and nutritional values, regarded as excellent sources of calcium, phosphorus and iron, in which tilapia meal showed the highest ash content. Protein content was high in all meals, especially in the tuna meal, and the amino acid profile of all meals was superior to FAO standard. Salmon, tuna and sardine meals showed high concentration of omega-3 fatty acids, with salmon presenting the greatest lipid content compared to the others.

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