Development of a low commercial value fish-sausage from the fish trawling “mix” category

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Abstract
The decreasing stock of the main species consumed in Brazil has stimulated studies regarding the use and development of products using low commercial value species. The shrimp trawling modality produces a large volume of a “mix” category comprising low commercial value fish. However, it is necessary to develop products with greater practicality to meet the current demands of the Brazilian consumer. The aim of the present study was to develop a sausage using a low commercial value fish, the Brazilian flathead (*Percophis brasiliensis*), caught in the fish trawling “mix” category landed in Niterói/RJ, and perform physico-chemical, microbiological, toxicological and sensorial analyses. The centesimal composition of the analyzed fish were as follows: humidity 73.78%, proteins 16.17%, carbohydrates 3.75%, total lipids 4.41% and ash 1.78%. The microbiological and toxicological results were within the legislation standards. Twenty varieties of fatty acids (AG) were detected, with the most frequent being palmitic acid (22.81%). Among the omega-3 series, the most frequent were eicosapentaenoic acid (5.36%) and docosahexaenoic acid (15.95%). Regarding minerals, the most noteworthy were sodium, calcium and iron (1114.24 mg, 112.32 mg and 1.73 mg respectively). The sausage showed 87.30% acceptability. The results demonstrate that the low commercial value fish Brazilian flathead resulted in a viable product for use in fish technology, with excellent nutritional results.

Keywords: acceptability; bacteriological analyses; physico-chemical analyses; fish derivatives; accompanying fauna.

Practical Application: The developed sausage showed excellent quality.

1 Introduction

Due to the low selectivity and the small mesh sizes of the traditional nets, shrimp trawling has become responsible for the largest catches of accompanying fauna among all fishing modalities in the world. Part of the species caught as accompanying fauna are selected and marketed in the “mix” category, basically composed of low value species and some small, commercially important, species (Castro & Yamaguti, 2000). The volume of the “mix” category is significant, since it has become an alternative to improve the value of trawling fisheries, due to the depletion of marine fish stocks (Perez et al., 2010).

Fishery Brazilian production has, increasingly, presented smaller stocks each year, bringing uncertainty to processors and wholesalers. The raw material often does not appear in the desired quantity and quality (Kubitza, 2007). Canned sardines, for example, from 2002 to 2012 showed approximately a 94% increase in imports in Brazil due to the decreasing stocks of this resource (Martins et al., 2016). Therefore, the use of low economic value fish species has been the subject of research in Brazil. Pires et al. (2014), showed the potential in the use of fish of the accompanying fauna in the development of products such as mechanically separated meat (MSM) and Surimi.

Commercial, low-value, fish species have been used as raw material in the production of mechanically separated meat (MSM), surimi and sausages (Tenuta-Filho & Jesus, 2003). These technologies are employed for the use of solid fish waste mainly destined for animal feed, but which can also be used for the production of fertilizers or chemical products (Cavalcante et al., 2005). However, the nutritional value of these residues, rich in proteins and omega-3 and omega-6 fatty acids, encourages the full use of fish for the development of products destined for human consumption (Feltes et al., 2010).

Polyunsaturated fatty acids (PUFAs) with 18 to 22 carbons make up omega-3 (w-3 or n-3) and omega-6 (w-6 or n-6). The major n-6 fatty acids are 18:2 linoleic acid and 20:4 arachidonic acid, and the major n-3 fatty acids are linolenic acid 18:3, eicosapentaenoic acid (EPA) 20:5 and docosahexaenoic acid (DHA) 22:6. Linoleic acid and α-linolenic acid are essential fatty acids (EFAs) because the double bonds located on the third and sixth carbon atoms cannot be produced by the human body. Thus, essential fatty acids must be obtained from the diet (Martin et al., 2006).

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A-linolenic acid is converted to eicosapentaenoic acid-EPA and docosahexaenoic acid-DHA. However, the conversion rate is considered to be very low in humans, and, because this compound competes for the same enzyme system as linoleic acid, it tends to decrease as the amount of this acid increases. Therefore, it is of fundamental importance to obtain n-3 sources through food intake (Food and Agriculture Organization, 1994).

The presence of n-3 fatty acids, mainly EPA and DHA, in fish-containing diets and their by-products, has been related to beneficial health effects (Guiné & Henriques, 2011). Among the physiological effects in humans, are the prevention and treatment of various types of cancer, cardiovascular diseases, hypertension, inflammation in general, asthma, arthritis and psoriasis (Mayser et al., 1998). Therefore, regardless of the market value of the captured species and the technology used for the development of derived products, the presence of dietary fish is essential in increasing the intake of the essential fatty acids so important for human health.

Despite proven nutritional quality, fish consumption in Brazil is still considered low (Brasil, 2011). This is explained by problems in distribution and marketing, and also by the absence of consumption habits, caused by the lack of practicality in fish preparation (Bonacina & Queiroz, 2007). There is also a deficiency of products on the market to meet consumer demands resulting from an urban lifestyle, and it is increasingly necessary to offer ready or semi-ready products that are easy and quick to prepare (Battistella, 2008). However, in order to produce fish-based products, quality control of fish health and the use of technologies that ensure product safety are required (Gonçalves, 2011).

Thus, the aim of this study was to develop Brazilian flathead (Percophis brasilensis) sausages, a species caught in the trawling “mix” category, and perform physico-chemical, microbiological, toxicological and sensorial analyses of the final product, in order to evaluate its adequacy for human consumption.

2 Materials and methods

Brazilian flathead samples were collected totaling 30 kg. The fish were randomly collected, already slaughtered, at the moment of landing on the quayside in the municipality of São Gonçalo (RJ) and transported in isothermal boxes containing ice to the laboratory of the Fundação Instituto de Pesca do Estado do Rio de Janeiro (FIPERJ). At the FIPERJ laboratory, the fish were washed under running water, ventrally cut to remove the viscera and the heads and fillets were then removed. The fillets and the filleting residue were packed separately in plastic bags and stored in a freezer at –18 °C.

The fillets and filleting residues were transported in isothermal boxes to the Laboratório de Processamento de Pescado da Embrapa Agroindústria de Alimentos (CTAA/EMBRAPA). Processing was performed according to the flowchart displayed in Figure 1. Subsequently, sausage samples were evaluated regarding physicochemical, microbiological, toxicological and sensorial analyses.

For the MSM, the filleting residue (bone skeleton and skin) was processed in a fish MSM machine (Mec Fish Brazil). Subsequently, the MSM was weighed and the samples were stored until the analysis (Fogaça, 2009).

Figure 1. Flow diagram of the fish sausage process.
Surimi was obtained by a wash cycle with water at 5 °C at a 3:1 (water: meat) ratio. The water and MSM mixture was stirred for five minutes and then allowed to stand for two minutes. The excess water was removed with a 100 micrometer screen followed by a vacuum pump for five minutes. At the end of processing, starch (10%) and cryoprotectants (2% sodium chloride and 1% sucrose) were added to the mixture. The surimi was then weighed, and the samples collected and frozen (-18 °C) (Fogaça, 2009).

One sausage formulation (50% surimi + 35% fillet + 15% ingredients) was prepared. The surimi, fillet and ingredients were mixed in a cutter mixer (Geiger, model 50-, São Paulo, Brazil) for five minutes until an emulsion was obtained. After the emulsification process, the mixture was removed from the cutter mixer and brought to a manual filler using collagen casings. They were then tied at every 10 cm in length using cotton string.

The physical-chemical analyses performed on the samples were the following: pre-dried matter at 105 °C (humidity); ashes; crude protein (MicroKjedahl) and ethereal extract (Soxlet method) (Brasil, 1981) for the determination of the centesimal composition. All samples were evaluated in triplicate at the Centro Estadual de Controle de Pesquisa em Qualidade de Alimentos (CEPQA), located in the Empresa de Pesquisa Agropecuária do Estado do Rio de Janeiro (PESAGRO-RJ). Carbohydrates (non-nitrogen extracted) were calculated by NIFEXT ("Nitrogen Free Extract"), by the difference between 100% of the other fractions of the centesimal composition (Oliveira et al., 1999). The total energetic value (TEV) of the foods was calculated based on the carbohydrate, protein and lipid values, knowing that carbohydrates and proteins provide 4 kcal/g of energy and lipids, 9 kcal/g (Food and Agriculture Organization, 2005).

Fatty acid analyses were performed at the Laboratório de Óleos Graxos da Embrapa Agroindústria de Alimentos, according to the official Association of Official Analytical Chemists (2005) International method. Saturated and unsaturated fat content and weight/weight of each fatty acid were performed on the raw materials according to the official Association of Official Analytical Chemists (2005) International method. All samples were evaluated in triplicate.

The mineral profile analyses were performed at the Embrapa Food Technology, according to the methodology described by Association of Official Analytical Chemists (2005) International. All samples were evaluated in triplicate.

The microbiological analyses included *Salmonella* sp., coliform counts at 35 °C, thermotolerant coliforms and coagulase positive *Staphylococcus*. All analyses were performed according to methodology reported by the Ministério da Agricultura (Brasil, 2003a). All samples were evaluated in triplicate.

For the toxicological analyses, the presence of hydrogen sulphide gas (H₂S), was determined according to the technique described by Laboratório nacional de Referência Animal (LANARA) (Brasil, 1981), while histamine content was determined according to the spectrophotometric methodology described by Glória & Soares (1993). All samples were evaluated in triplicate.

The sensorial test was performed with 87 testers, who evaluated the product using a seven-point hedonic scale (1 = very disagreeable, 2 = moderately disliked, 3 = mildly disliked, 4 = didn’t like or dislike, 5 = slightly liked, 6 = moderately liked; 7 = I liked it a lot) for odor, color, texture and flavor attributes and, finally, the overall aspect of the product. Observations could be written down by the tasters in the file itself. Finally, tasters were asked about their intention to buy the product divided into a five-point scale: I would definitely buy the product; probably buy; maybe yes/maybe not; I would probably not buy; I would decidedly not buy.

Regarding the results obtained by the hedonic scale method, the samples were accepted if 70% of the tasters assigned a score ≥ 4.

### 3 Results

Table 1 expresses the results of the centesimal composition and the mineral profile of the Brazilian flathead product.

The main results of the lipid profile of the Brazilian flathead sausage are displayed in Table 2.

Table 3 expresses the microbiological results of the Brazilian flathead (Percophis brasiliensis) product according to the Brazilian legislation.

Table 1. Mean results of the centesimal composition and mineral profile of the Brazilian flathead (*Percophis brasiliensis*) sausage.

<table>
<thead>
<tr>
<th>Components</th>
<th>Values</th>
</tr>
</thead>
<tbody>
<tr>
<td>Humidity (%)</td>
<td>73.78</td>
</tr>
<tr>
<td>Protein (%)</td>
<td>16.17</td>
</tr>
<tr>
<td>Lipids (%)</td>
<td>4.41</td>
</tr>
<tr>
<td>Carbohydrates (%)</td>
<td>3.75</td>
</tr>
<tr>
<td>Ashes (%)</td>
<td>1.89</td>
</tr>
<tr>
<td>Energetic value kcal&lt;sup&gt;1&lt;/sup&gt; 100 g&lt;sup&gt;-1&lt;/sup&gt;</td>
<td>119.37</td>
</tr>
<tr>
<td>Na (mg/100g)</td>
<td>1114.24</td>
</tr>
<tr>
<td>Ca (mg/100g)</td>
<td>112.32</td>
</tr>
<tr>
<td>Fe (mg/100g)</td>
<td>1.73</td>
</tr>
</tbody>
</table>

<sup>1</sup>% = percentage; grams per hundred grams of food; kcal = kilocalorie; mg/100g = Milligrams per hundred grams of food.
Sulfur-based compounds and histamine contents were evaluated for all sausage samples. None of these compounds were detected by the employed methodologies.

Regarding the sensory analysis, from the total of 87 tasters, 63.2% were female and 36.8% male, aged between 20 and 65 years old. The product was accepted at 87.3% for the overall product appearance, averaging 5.43 (±1.39). The acceptability results per attribute are listed in Table 4.

In the current study, regarding intention to buy the final product, the highest percentage of respondents attributed that they would probably buy it (34.5%), followed by perhaps yes/maybe (27.5%) and would decidedly buy the product (21.8%), while 8.0% would probably not buy or would definitely not buy.

### 4 Discussion

The Brazilian legislation states that common sausages made with beef, pork or poultry must have the following chemical composition values: maximum humidity of 65%, minimum protein content of 12% and maximum lipid content of 30% (Brasil, 2000). Therefore, according to the legislation, the produced Brazilian flathead sausage presented higher humidity values (73.78%), but met protein requirements (16.17%). The humidity value above the recommended level can be explained by the absence of fats from other sources and the inherent characteristics of fish meat, resulting in a product with high humidity content, but lipid content well below the maximum limit recommended by the legislation. These results confirm the nutritional quality and the association of fish products with healthy diets.

Regarding protein levels, the levels of the present study were higher than those found by Gonçalves et al. (2009) in “piramutaba” (Brachyplatystoma vaillantii) sausage (12.6%). Ribeiro et al. (2017), in a study in which kibbe and meatballs were baked from fish pulp obtained from the trawling “mix” category, found lower protein levels (kibbe: 15.10%, meatball: 15.10%) compared to the present study (16.17%). When comparing the lipid results of this same study (kibbe: 4.50%, meatball: 4.40%), the Brazilian flathead sausage was similar (4.41%), although higher than lipid content reported by Silva et al. (2012) in a study on the development of croaker sausage (0.57%). On the other hand, the present study observed lower ash (1.89%) contents when compared to Gonçalves et al. (2009), of 3.51%. Such differences in results can be explained by the intrinsic characteristics of each species.

Regarding the nutritional requirements recommended by the Brazilian National Health Surveillance Agency (Brasil, 2003b), a 100 g portion of Brazilian flathead sausage would serve 5.9% of the recommended daily energy value and 21.56% of the daily requirement (75g/day) of crude protein, proving to be an alternative source of protein containing low calories, ideal for 2.000 kcal/day diets. The 1,114.24 mg of sodium in each 100 g portion would attend 46.43% of the daily necessities (2.400mg/day) of an adult individual, although more studies are necessary regarding the sausage formulation in order to create adjustments concerning this mineral. However, in a study in which the sodium content in hot dog meat sausages was evaluated, the average sodium content was of 551 mg of sodium per 50 g portion (Brasil, 2010), in the same range as the results found herein.

### Table 2. Lipid profile and fatty acid quality of the sausage Brazilian flathead (Percophis brasiliensis).

<table>
<thead>
<tr>
<th>Fatty acid</th>
<th>Sample Sausage (g/100g of fat)</th>
</tr>
</thead>
<tbody>
<tr>
<td>ΣSFAs¹</td>
<td>33.97</td>
</tr>
<tr>
<td>ΣMUFA²</td>
<td>33.05</td>
</tr>
<tr>
<td>ΣPUFA³</td>
<td>28.11</td>
</tr>
<tr>
<td>ΣAG ω3⁴</td>
<td>23.40</td>
</tr>
<tr>
<td>ΣAG ω6⁵</td>
<td>1.97</td>
</tr>
<tr>
<td>ΣAG ω6/ω3⁶</td>
<td>0.08</td>
</tr>
<tr>
<td>ΣPUFA/ΣSFAs⁷</td>
<td>0.82</td>
</tr>
</tbody>
</table>

¹ΣSFAs = sum of saturated fatty acids; ΣMUFA = sum of monounsaturated fatty acids; ΣPUFA = sum of polyunsaturated fatty acids; ΣAG ω3 = sum of omega-3 fatty acids; ΣAG ω6 = sum of omega-6 fatty acids; ΣAG ω6/ω3 = ratio of the sum of omega-6 fatty acids by the sum of omega-3 fatty acids; ΣPUFA/ΣSFAs = ratio of the sum of polyunsaturated fatty acids by the sum of saturated fatty acids.

### Table 3. Microbiological results of the Brazilian flathead (Percophis brasiliensis) product according to the Brazilian legislation (Brasil, 2003a).

<table>
<thead>
<tr>
<th>Product</th>
<th>Salmonella sp.</th>
<th>Coliforms at 35°C</th>
<th>Thermotolerant coliform</th>
<th>Positive coagulase Staphylococci</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sausage</td>
<td>absent/25g⁸</td>
<td>&lt;10 CFU/g⁹</td>
<td>&lt;10 CFU/g⁹</td>
<td>&lt;10⁹ CFU/g⁹</td>
</tr>
</tbody>
</table>

⁸Absent/25g = absent in twenty-five grams of food; CFU/g = colony forming unit per gram of food.

### Table 4. Mean values of the scores of each attribute in the sensorial analysis.

<table>
<thead>
<tr>
<th>Attribute</th>
<th>Taste</th>
<th>Odor</th>
<th>Texture</th>
<th>Color</th>
</tr>
</thead>
<tbody>
<tr>
<td>Means</td>
<td>5.6 (±1.43)⁹</td>
<td>5.5 (±1.16)⁹</td>
<td>5.4 (±1.6)⁹</td>
<td>4.8 (±1.76)⁹</td>
</tr>
</tbody>
</table>

⁹Equal letters between different attributes indicate that the scores do not differ from each other.
England (Department of Health and Social Security, 1984); ΣPUFAs/ΣSFAs should not be less than 0.45, which translates into an unhealthy diet, mainly in relation to cardiovascular diseases. The Brazilian flathead sausage presented ΣPUFAs/ΣSFAs of 0.82, above dietary recommendations. Menezes et al. (2009), in a study that evaluated the fatty acid profile of two marine fish species from the Brazilian state of Alagoas, reported ΣPUFAs/ΣSFAs for striped mojarra (Eugenes plumieri) and mackerel (Scomberomorus cavalla) of 0.82 and 1.34 respectively, above the values found in the present study. However, the sum of EPA + DHA was of 15.32% for mackerel and 6.02% for the striped mojarra, below what was observed herein, of 21.31%. Scherr et al. (2015), in a study that analyzed the n-6/n-3 ratio in grilled fish comprising the main fish species consumed in Brazil, found lower results for all species (Sea bass (Dicentrarchus labrax), 0.34, Grouper (Epinephelus lanceolatus): 2.50, “Pirarucu” (Brachyplatystomai flattened): 3.73, “Namorado Sandperch” (Pseudopercis numida): 0.63, “Pescadinha” (Cynoscion acoupa): 5.98, “Piracuru” (Arapaima gigas): 1:21, Common snook (Centropomus undecimalis): 0.34, Sardine (Sardinella brasiliensis): 0.60; Trout (Onchorhynchus mykiss): 4.03; Salmon (Salmo salar): 2.41) when compared to the results found in the present study.

The Agência Nacional de Vigilância Sanitária - ANVISA (Brasil, 2001) establishes a maximum permitted tolerance of fecal coliforms of 10^6 CFU/g in refrigerated fish, the absence of Salmonella spp.and maximum values of <5 x 10^6 CFU/g for coagulase positive Staphylococcus. In the Brazilian flathead product, values lower than the limit determined by current legislation were found. Thus, the product was in accordance to established standards. Similarly to the results found in the present study, Galvão (2009), in a study conducted on “piramutaba” (Brachyplatystoma vaillanti) sausage, found results in accordance to the legislation regarding coagulase positive Staphylococcus.

The Brazilian legislation considers that fish belonging to the Scombridae, Scombresocidae, Clupeidae, Coryyphaenidae families (Brasil, 1997) with a positive reaction for sulfuric gas (Brasil, 2017) and with histamine levels above the maximum level of 100 ppm in muscle are deteriorated. Studies to detect these quality and freshness indicators in fish have been developed, preferably, in fresh or frozen products. In the current study, no histamine and sulfur compounds levels higher than those recommended by the legislation were detected. However, Souza et al. (2017), when evaluating the freshness of frozen marine fish, detected sulfide gas (H,S) in 70.8% of the analyzed samples, indicating an advanced stage of decomposition. Soares et al. (1998), in a study analyzing frozen fish fillets, found histamine concentrations above the maximum permissible levels in 37% of the samples.

Reports of lesser acceptability of the final product than the current study (87.3%) are available in the literature. Oliveira (2009), testing different tilapia sausage formulations, obtained an overall acceptability of 78.1%, similar to Gonçalves et al. (2009), who obtained 75.6% overall acceptance regarding “piramutaba” sausage containing 30% shrimp flavor.

In a study on sausage elaboration with Nile tilapia mechanically separated meat (MSM), Lago (2015) evaluated odor, color, flavor and texture attributes obtaining 75.0%, 53.12% and 75.0% and 78.1% for each, respectively, all below the results found observed herein (94%;72%;85% and 88%). Similarly to the results found in the present study, the color attribute was the least accepted and statistically different from the other attributes, possibly explained by the clear and unusual appearance of the sausages. For color, some opinions were registered in the current study that indicate the cause of low acceptance values in the present study, namely “very pale”, “very clear”, “pale” and “very opaque”.

The purchase intention results of the present study were lower (34.5% “probably buy it; 27.5% “perhaps yes/maybe”) than in the study conducted by Lago (2015), who obtained a higher percentage in intention to buy on the second scale, “would probably buy”, followed by the first scale, “would certainly buy”, in a study on Nile tilapia sausage. In the present study, the highest percentages were attributed to the “would likely buy” option, followed by “maybe yes/no”, but only 16.0% of the testers “decidedly” and “would probably not buy the product”, indicating that the product would have excellent market acceptance.

5 Conclusions

The developed product, Brazilian flathead sausage, was shown to display satisfactory hygienic-sanitary conditions, good nutritional quality, with emphasis on polyunsaturated fatty acid content, and a high index of sensorial acceptance emphasizing odor and flavor, with excellent purchase intention. Thus, fish that could be underutilized or even discarded proved to be a viable alternative for the development of a safe, versatile and nutritious consumer product.

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