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Production and Characterization of Emulsified Fish Mortadella From Nile Tilapia (*Oreochromus niloticus*)

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Abstract

This study produced fish mortadella from Mechanically Separated Meat (MSM) of Nile tilapia added with animal fat. Three formulations were developed: M1 (MSM - 89 % and 5 % pork fat), M2 (MSM - 84 % and 10 % pork fat) and M3 (MSM - 79 % and 15 % pork fat). The elaborated products were tested for technological, physical, physico-chemical, microbiological and sensory parameters. The results showed that the fish mortadella were microbiologically stable with a particular texture for an emulsified meat product, attractive colour and characteristic flavour. All formulations met the expected identity and quality requirements. They also achieved good acceptance by the judges, in which formulation M1 may be highlighted for presenting an emulsion stability of 97 %, higher protein content (18.09 %) and lower lipids (16.31 %). In addition, it also reached higher mean scores for texture attributes and purchase intent. Therefore, it is possible to prepare fish mortadellas from tilapia MSM using less animal fat.

Keywords: Fish waste; Fish mortadella; *Oreochromus niloticus*; Sensory analysis; Waste processing

1 Introduction

Fish are a highly beneficial food to human nutrition as they are a source of vitamins and minerals, and contain a high proportion of long chain polyunsaturated fatty acids and all the essential amino acids for humans (Fernandes et al., 2014; Sleder et al., 2015; Zotos & Vouzanidou, 2012). The Nile tilapia (*Oreochromis niloticus*) can be

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highlighted as being among the most cultivated species in the world. It has high productivity, good adaptation to diverse environments, good acceptance in the consumer market, reduced fat content and excellent fillet yield (FAO, 2017; Liu et al., 2015; Olopade, Taiwo, Lamidi & Awonaike, 2016). On the other hand, the mechanical filleting process generates approximately 70 % by-products (processing waste or discarded meat), resulting in large fish meat and oil losses. The head, carcass and guts constitute 54 % of the waste, while the skin represents 10 %. In general, these residues are destined to produce tilapia flour and oil used in manufacturing animal feed for fish, pigs and birds (Campagnoli de Oliveira Filho, Netto, Ramos, Trindade & Macedo Viegas, 2010).

Several studies have been carried out with tilapia and its by-products to promote and improve the sustainability of the fish industry, seeking to increase its consumption and maximize its use. One of its by-products is Mechanically Separated Meat (MSM), which has been used in developing new products such as emulsified meat products (sausages or mortadella, nuggets, surimi, fishburgers, etc.) It also stands out for having good unsaturated fat content which can reduce the addition of animal fat without causing sensory changes to the texture, flavour or consistency of the elaborated products (dos Santos Fogaca, Otani, De Gaspari Portella, Alves dos Santos-Filho & Sant' Ana, 2015; Ferreira Silva Bartolomeu et al., 2014).

In view of the recognized technological and market potential of emulsified meat products, the present study had the objective of producing an emulsified fish mortadela from Nile tilapia (*Oreochromus niloticus*) MSM added to different animal fat contents, aiming to obtain a differentiated product with high biological value and low fat content.

2 Materials and Methods

2.1 Tilapia slaughter and MSM production

The fish were obtained from an association of tilapia farmers from the city of Bananeiras/PB, Brazil. They were purged in clean water for 24 hours, captured in the early hours of the morning, then transported in plastic boxes where they were desensitized by thermal stunning. After respiratory activity ceased they were hung on hooks, where they were then eviscerated, and the fish were flaked, washed and filleted following a linear flow. Next, the carcasses were crushed using a pulping machine (Bresi®, 60 kg/h, 1/2, hp motor, stainless steel), thereby obtaining the MSM. The MSM was then vacuum packed into 2.0 kg polyethylene bags, labelled, and frozen (at -18 ^{o}C) until it was used in the production of fish mortadella.

2.2 Nile tilapia (*Oreochromis niloticus*) MSM quality assessment

Acidity, pH, moisture, dry matter, ash, protein (AOAC, 2012) and lipid (Floch, 1957) parameters were evaluated. The microbiological quality of the MSM was verified based on coliform counts at 45^{o}C MPN/g, coagulase-positive Staphylococcus CFU/g, sulphite-reducing *Clostridium sp.* CFU/g, *Salmonella sp.*/25 g and molds and yeasts (APHA, 2001).

2.3 Preparation of fish mortadella

The fish mortadella was prepared as described by Ferreira Silva Bartolomeu et al. (2014) and Campagnoli de Oliveira Filho, Netto et al. (2010). The present study used different concentrations of tilapia MSM and refined pork fat (Table 1). The other ingredients used in preparing the fish mortadella were added in the same concentrations to all treatments (Table 1).

A high-yielding tabletop cutter (METVISA®, CUT 2.5 L; 1/3 hp motor) was used for preparing the fish mortadella according to the flowchart and the steps described below (Figure 1).

Comminution began with the MSM, salt, curing agents and polyphosphate for thirty seconds. Ice water, pork fat, and other ingredients were added at thirty-second intervals until the formation of a paste (similar to a pâté), at a controlled temperature with a skewer thermometer (INCOTERM® Digital thermometer) with a

Table 1: Formulations of emulsified fish mortadella with different concentrations of tilapia MSM and pork fat

Ingredients	Concentrations (%) M1	M2	M3
MSM	74.00	69.00	64.00
Pork lard	5.00	10.00	15.00
ISP	5.00	5.00	5.00
Ice/cold water	8.00	8.00	8.00
Polyphosphate	0.30	0.30	0.30
Curing salts	0.20	0.20	0.20
Antioxidant	0.60	0.60	0.60
Starch	4.00	4.00	4.00
Mortadella seasoning	0.70	0.70	0.70
Flavour enhancement	0.15	0.15	0.15
Black pepper	0.05	0.05	0.05
Garlic powder	0.25	0.25	0.25
Smoke aroma	0.50	0.50	0.50
Salt	1.25	1.25	1.25
Total	1	1	1

MSM - Mechanically Separated Meat;
ISP - Isolated Soybean Protein;
M1 - Fish mortadella with 5% pork fat;
M2 - Fish mortadella with 10% pork fat;
M3 - Fish mortadella with 15% pork fat

maximum temperature of 12 ^{o}C at the end of comminution.
The obtained pâté was packed into artificial shrinkable casings using a manual stuffer (METVISA®, EL.10). The fish mortadella was cooked in moist heat until it reached an internal temperature of 72±1 ^{o}C, monitored with the aid of a thermometer equipped with a thermocouple, Digital Cooking Thermometer). After the cooking was finished, the mortadella was submitted to thermal shock by immersing it in ice water for 15 minutes until it reached temperatures between 15 to 20 ^{o}C, and then it was vacuum packed.

2.4 Evaluation of technological properties

Emulsion stability - ES

Emulsion stability was performed according to the method described by Parks and Carpenter (1987). Emulsion stability values were obtained by calculating the weight loss and its percentage using the following formula:

$$\%\ Emulsion\ stability = 100 - \%loss \quad (1)$$

Texture profile analysis

Hardness, springiness, cohesiveness, chewiness, fracturability, gumminess, adhesiveness and resilience parameters, were evaluated using the TA XT-2i texture analyser with Exponent® software (Stable Micro Systems, Godalming, UK) with a probe of 35 mm diameter (SMS P/36R) moving at a constant speed of 0.8 mm/s, according to Bourne (2002).
A Warner Braztler accessory (3 mm thick) was used to measure shear force at a test speed of 200 mm/min (Andres, Garcia, Zaritzky & Califano, 2006). Mean and standard deviation were calculated from 16 determinations (Lin & Chao, 2001).

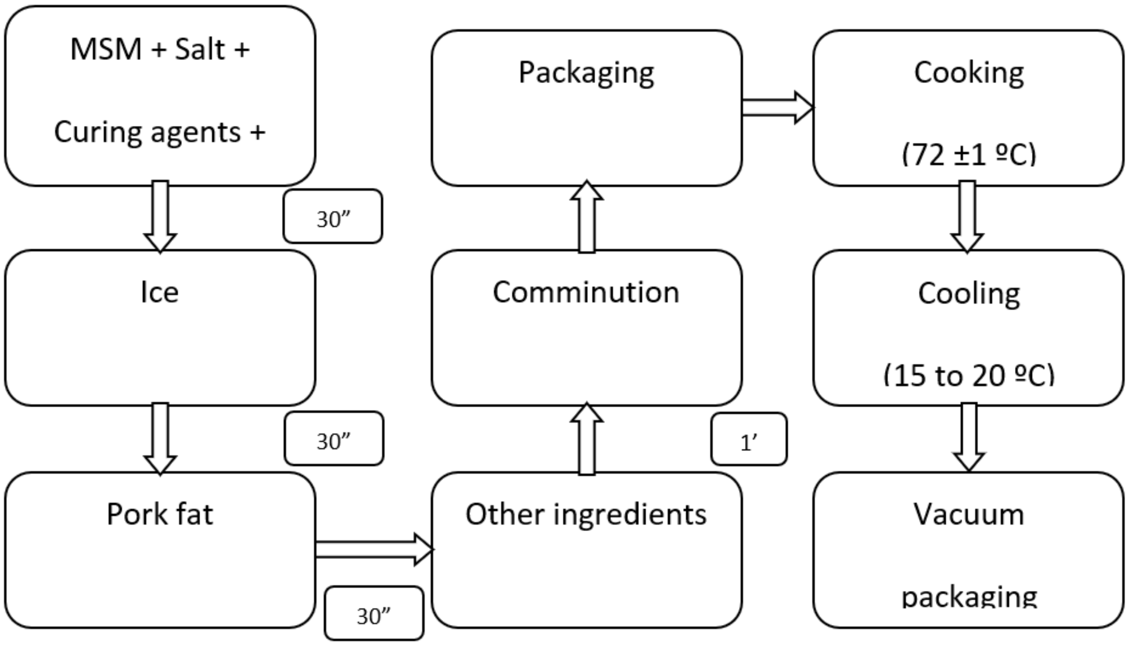

Figure 1: Flowchart of fish mortadella preparation

Physical and physical-chemical analyzes

The analyses carried out were: water activity (a_w), pH, normal acidity, moisture, dry matter, protein, ash (AOAC, 2012), chloride according to the Mohr Method, calcium content determined by EDTA volumetry (AOAC, 2012) and lipids (Floch, 1957). Water Retention Capacity (WRC) was evaluated according to the methodology by Grau (1953).
Lipid oxidation was evaluated by determining the peroxide index method (mEq/kg) based on the measurement of thiobarbituric acid reactive substances (TBARS) according to Ganhao, Estevez and Morcuende (2011) and based on Brasil (1999). The peroxide concentration was given by mEq/kg of the sample.
The instrumental colour of each fish mortadella was determined according to the methodology described by Abularach, Rocha and de Felício (1998), using a digital Minolta® colorimeter (Model CR-300, Minolta, Osaka, Japan). Luminosity parameters (L*), green (-)/red (+) (a*) and blue (-)/yellow (+) (b*) were determined according to the specifications of the *Commision Internationale de L'éclairage* (CIE, 1986).

Microbiological analyzes

Coliform counts at 45^{o}C MPN/g, coagulase-positive *Staphylococcus* CFU/g, sulphite-reducing *Clostridium* CFU/g, and checking for *Salmonella sp.*/25 g counts were carried out as recommended for fish mortadella and for counting molds and yeasts (APHA, 2001).

Sensorial analysis

After approval of the project by the Research Ethics Committee under number 821.481, the fish mortadella was submitted for sensory acceptance, intention to purchase and order preference (Faria & Yotsuyanagi, 2002; Meilgaard, Carr & Civille, 1991; Stone & Sidel, 1993).
One hundred twenty-six (126) untrained consumers aged between 20 and 50 years of age who liked to consume fish and emulsified fish mortadella were recruited. These tasters evaluated

the samples in individual booths with white artificial lighting, away from noise and odours, and at previously established times.
The fish mortadella was offered sliced, refrigerated and codified, accompanied by salt crackers and a glass of water. Attributes of appearance, colour, aroma, flavour (specific mortadella flavour), flavour (specific fish flavour), texture and overall assessment were evaluated using a mixed nine-point structured hedonic scale (1 = I greatly disliked it; 9 = I liked it a lot). Next, the tasters were asked to indicate their intention to purchase, also using a mixed five-point hedonic scale (1 = I would never buy it; 5 = I would buy it). The samples were considered accepted when they obtained a mean $\geq$ 5.0 (equivalent to the hedonic term "I neither liked it nor disliked it"). An Acceptability Index (AI) was also calculated according to the equation:

$$AI(\%) = A \times 100/B \qquad (2)$$

In which, A = average score obtained for the product and B = maximum score given to the product. An AI with good repercussion was considered $\geq$ 70 % (Teixeira, Meinert & Barbetta, 1987).
The overall preference for the different fish mortadella was also evaluated through the preference order test in decreasing order (from the most preferred to the least preferred).

2.5 Statistical analysis

Data were submitted to analysis of variance (ANOVA) and Tukey test at 5 % probability ($p<0.05$) for comparison of means using the STATISTICA 7.0 program (Statistica, 2005). Principal component analysis (PCA) was performed for the different groups of variables and based on the correlation matrix of these variables in order to provide graphical representation of the significant sensory attributes.
The results of sensory preference ordering tests were analyzed according to the Friedman test using the Newell-MacFarlane table (Faria & Yotsuyanagi, 2002).

3 Results and Discussion

3.1 Nile tilapia (*Oreochromis niloticus*) MSM quality assessment

The percentage values found for the MSM in the quality assessment are presented in the table 2. Regarding the pH, similar values in analyzing MSM from fish were reported by Cavenaghi-Altemio, Alcade and Fonseca (2013) of 7.10, and Calil Angelini et al. (2013) of 6.9. There is no legal parameter for the pH limit of fish MSM; however, for fresh fish a maximum value of 6.8 is imposed for this parameter (Brasil, 1952). The results of this study are justified by the fact that fish have a close to neutral pH. In addition, the processing for obtaining MSM disintegrates tissue, which exposes muscle fibres to degrading substances and may facilitate an increase in pH values, according to Pereda et al. (2005).
Moisture, ash and protein values were close to those found by Dallabona et al. (2013) (71.0 %, 1.22 % and 15.37 %), by Campagnoli de Oliveira Filho, Favaro-Trindade, Trindade, de Carvalho Balieiro and Macedo Viegas (2010) (74.45 %, 1.14 % and 12.76 %), and by dos Santos Fogaca et al. (2015) (73.87 % and 15.87 %) for moisture and protein, respectively.
The values for lipids were different from those found by Cavenaghi-Altemio et al. (2013) of 2.53 %, dos Santos Fogaca et al. (2015) of 7.62 %, and by Kirschnik and Macedo-Viegas (2009) of 2.91 %, due to the fact that in the present study, the MSM had the presence of the ventral musculature which contains the highest fat content, around 15 %.
The results complied with the parameters determined by the relevant legislation Brasil (2000), which recommends a minimum protein content of 12 % and a maximum fat content of 30 %, confirming that tilapia MSM is a good protein source, and thus demonstrating its viability for use in protein products of good biological value Rebouças, Rodrigues, de Castro and Vieira (2012).

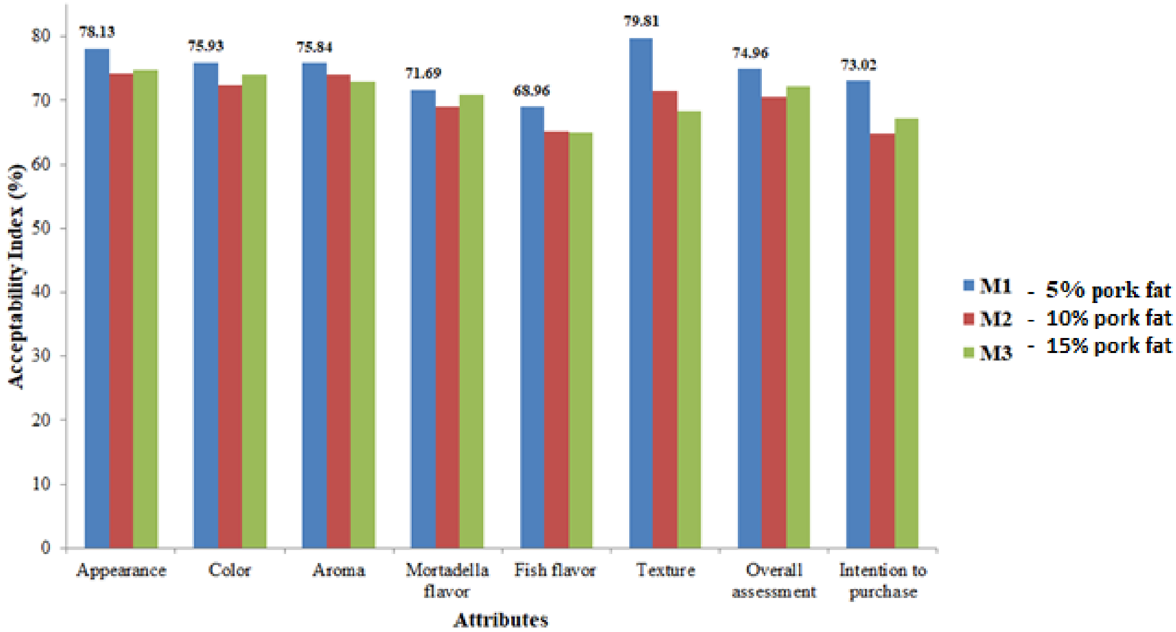

Figure 2: Acceptability index of the sensory attributes of fish mortadella with different concentrations of pork fat

Table 2: Mean values obtained in the physical and physicochemical analyses of fish mortadella with different concentrations of tilapia MSM and pork fat.

	Mortadella		
Variable	**M1**	**M2**	**M3**
a_w	0.96 ±0.00^a	0.96 ±0.00^a	0.96 ±0.00^a
pH	7.11 ±0.00^a	7.13 ±0.01^a	7.16 ±0.05^a
WRC* (%)	57.93 ±2.21^a	58.51 ±3.11^a	58.15 ±2.79^a
Normal acidity (%)	4.12 ±0.14^a	3.80 ±0.01^a	3.68 ±0.13^a
Moisture (%)	63.63 ±0.01^a	59.58 ±0.05^b	56.73 ±0.05^c
Dry matter (%)	36.37 ±0.01^c	40.42 ±0.05^b	43.27 ±0.05^a
Ashes (%)	3.90 ±0.20^a	4.31 ±0.15^a	3.85 ±0.46^a
Proteins (%)	18.09 ±0.01^a	17.46 ±0.02^b	16.54 ±0.03^c
Lipids (%)	16.31 ±0.00^c	19.84 ±0.05^b	24.38 ±0.02^a
Sodium chloride (%)	1.88 ±0.03^c	2.35 ±0.02^a	2.17 ±0.00^b
Calcium (%)	0.32 ±0.00^a	0.28 ±0.00^b	0.21 ±0.00^c

Means ± standard deviation with different letters on the same line differed by Tukey's test ($p<0.05$). M1 - Fish mortadella with 5% pork fat; M2 - Fish mortadella with 10% pork fat; M3 - Fish mortadella with 15% pork fat. *WRC - Water Retention Capacity.

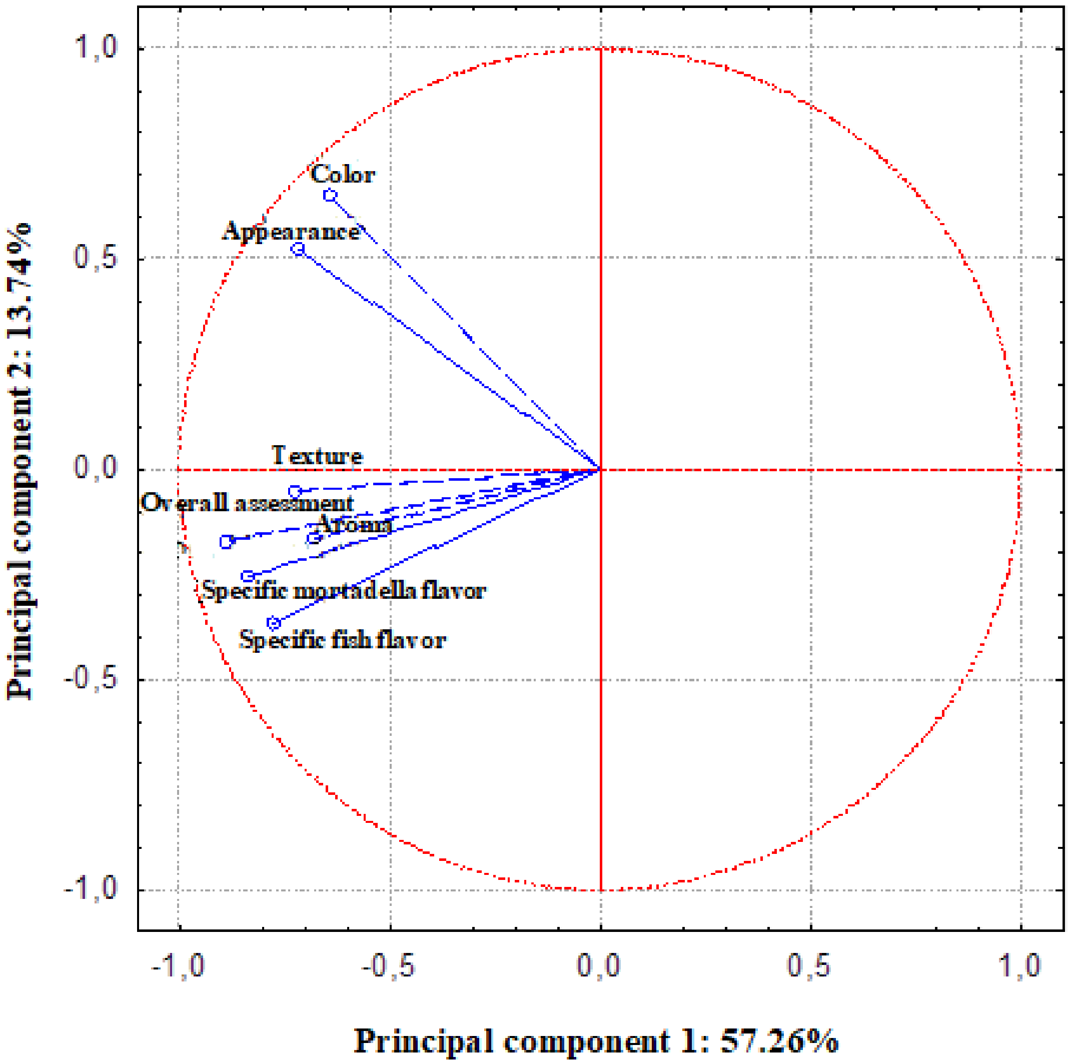

Figure 3: Projection of the PCA of the attributes in the sensory analysis of samples of fish mortadella with different concentrations of tilapia MSM and pork fat

3.2 Evaluation of the technological properties of the fish mortadella

The values obtained from the emulsion stability, shear force and texture profile evaluations are shown in Table 3.

Emulsion stability

In this study, the results for emulsion stability (ES) (97.03; 93.30 and 91.74 %) in the different fish mortadella treatments, M1, M2 and M3, respectively, were influenced by the fat content, so that the higher the pork fat concentration added, the lower the ES ($p < 0.05$). It is also important to point out that the higher amount of protein provided better emulsion stability results (Table 3) agreeing with the literature data. In evaluating sausages made from MSM and fish surimi, Yousefi and Moosavi-Nasab (2014) concluded that crude protein values of around 19% resulted in products with greater hardness and better gel consistency.
The findings of this study were attributed to the balance achieved for the protein concentrations (Table 3), which contributed to the formation of a good emulsion.

Texture profile analysis - TPA

Shear force and instrumental texture profile may vary according to the amount of water, proteins and fats found in the sample (Cengiz & Gokoglu, 2007; Cofrades, Guerra, Carballo, Fernandez-Martin & Colmenero, 2000). It was observed (Table 3) that as the MSM percentage was reduced and that of pork fat increased along with a consequent reduction in protein content, a significant decrease ($p < 0.05$) in the other parameters was also observed, with the exception of adhesiveness which increased ($p < 0.05$) with the increase in fat concentration.
Campagnoli de Oliveira Filho, Favaro-Trindade et al. (2010) observed an increase in the softness of the sausages made with increasing amounts of Nile tilapia MSM - 0 to 100 %. Similarly, it was observed in the present study that the softness improved (as decreases in the hardness and shear force) with the increase in fat concentration instead of MSM. These data were expected, considering that higher amounts of fat are responsible for a reduction in texture parameters, especially when fat increases are followed by a decrease in protein and water contents in the formulations, as observed in the present study (Dincer & Cakli, 2010; Hashemi & Jafarpour, 2016; Kin et al., 2013). The results in the present study agreed with Sleder et al. (2015) who also found that a reduction in the amount of protein and an increase in the amount of fat significantly decrease the shear force from 8.4 (N) to 5.92 (N) in sausages without added fat and with 95.5 % of tambaqui meat (*Colossoma macropomum*); and with 9 % fat and 86.5 % tambaqui meat, respectively. In evaluating sausages prepared with fish MSM of fish surimi, Yousefi and Moosavi-Nasab (2014) found that those made with MSM presented greater shear force than those prepared with surimi - 1.75 N cm^2 and 1.08 N cm^2, respectively, and concluded that it is the greater amount of protein present in the MSM rather than its quality that determined the best texture profile for MSM fish sausages.
According to Hedrick, Aberle, Forrest, Judge and Merkel (1994), meat protein, represented mainly by myofibrillar proteins (actin and myosin), is the main factor responsible for the hardness in emulsified products. Colmenero, Barreto, Mota and Carballo (1995) also confirmed this relationship in their experiments with sausages, verifying that the higher the protein content, the greater the firmness of the final product; a similar characteristic was observed with the fish mortadella in this research.
Adhesiveness is high in traditional emulsified meat products (Campos, Gonçalves, Mori & Gasparetto, 1989). However, the partial or total replacement of meaty matter by MSM and the inclusion of fats contribute to forming products with a softer consistency, as in this study (Table 3). As the fat content increased in the formulations, the products had a softer consistency, tending to disintegrate during slicing.

Instrumental colour analysis

The parameters L* (luminosity), a* (red/green colour intensity) and b* (intensity of yellow/blue

Table 3: Mean values obtained in the emulsion stability, shear force and texture profile (TPA) analyses of fish mortadella with different concentrations of tilapia MSM and pork fat.

Technological parameters	Mortadella M1	M2	M3
Emulsion stability (%)	97.03 ±0.00^a	93.30 ±0.00^b	91.74 ±0.00^c
Shear force (N)	3.73 ±0.01^a	2.47 ±0.02^b	1.25 ±0.02^c
Texture profile (TPA)			
Hardness (kg)	1.87 ±0.06^a	1.20 ±0.04^b	0.82 ±0.01^c
Springiness (mm)	0.80 ±0.03^a	0.67 ±0.04^b	0.49 ±0.08^c
Cohesiveness (kg)	0.53 ±0.01^a	0.47 ±0.01^b	0.35 ±0.02^c
Chewiness (g.mm)	0.81 ±0.08^a	0.41 ±0.03^b	0.22 ±0.02^c
Fracturability (kg)	2.03 ±0.02^a	0.94 ±0.03^b	0.61 ±0.04^c
Gumminess (kg)	1.01 ±0.06^a	0.58 ±0.08^b	0.37 ±0.06^c
Resilience (kg)	0.35 ±0.07^a	0.21 ±0.01^b	0.11 ±0.05^c
Adhesiveness (kg.s)	-0.78 ±0.06^c	-0.34 ±0.07^b	-0.12 ±0.08^a

Means ± standard deviation with different letters on the same line differed by Tukey's test (p<0.05). M1 - Fish mortadella with 5% pork fat; M2 - Fish mortadella with 10% pork fat; M3 - Fish mortadella with 15% pork fat.

colour) are shown in Table 4.

Colour is correlated with the acceptance of food products (Dincer & Cakli, 2010; Ferreira Silva Bartolomeu et al., 2014). It was observed that by decreasing the MSM content and increasing the fat content, the luminosity value (L*) was increased between samples M1 and M2, as well as between M1 and M3 ($p < 0.05$), confirming the theory that moisture and lipid contents have a considerable influence on L* values. The L* results were below those determined by Moreira, Visentainer, de Souza and Matsushita (2001), who found an L* value of 72.28 in a study with sausage formulated with tilapia filets, and close to those found by do Amaral et al. (2015), who also verified that the fat content positively affected luminosity indices in pork sausages as the fat content increased with values between 55 and 63.

Dantas Guerra et al. (2012) also found that the higher the fat content, the higher the L* and b* values, with values between 52.0 and 59.4 (L*) and 8.6 to 10.5 (b*) for sheepmeat mortadella prepared with different levels of pork fat ranging from 10 to 30 %, respectively. Similarly, differences were also found ($p < 0.05$) in the present study for the b* parameter when the fish mortadella with 5 % pork fat was compared with the other formulations; and the fish mortadella with 10 and 15 % fat in the formulation presented higher values for yellow colour intensity due to the higher fat percentage added to them.

There was no observed decrease for the a* values when the amount of MSM also decreased. This occurred due to the fact that we chose to use isolated soybean protein to improve the acceptance of the fish mortadella, which probably contributed to an increase in the red colour between the M1 fish mortadella and the other formulations ($p < 0.05$).

3.3 Evaluation of physical and physical-chemical characteristics of fish mortadella

The physical and physico-chemical parameters of the fish mortadella are shown in Table 2.

The results of water activity (a_w) obtained for the fish mortadella were equal ($p > 0.05$), corresponding to 0.96 and above 0.85, which is the

Table 4: Mean values obtained in the instrumental color analyse of fish mortadella with different concentrations of tilapia MSM and pork fat.

Technological parameter	Mortadella M1	Mortadella M2	Mortadella M3
Instrumental Colour			
L*	62.44 ±0.26^{b}	65.21 ±0.13^{a}	65.31 ±0.09^{a}
a*	9.57 ±0.25^{b}	10.23 ±0.17^{a}	10.76 ±0.31^{a}
b*	10.76 ±0.06^{b}	11.21 ±0.16^{a}	11.35 ±0.05^{a}

Means ± standard deviation with different letters on the same line differed by Tukey's test (p<0.05). M1 - Fish mortadella with 5% pork fat; M2 - Fish mortadella with 10% pork fat; M3 - Fish mortadella with 15% pork fat.

limit for considering them as foods with high water activity. This characteristic favours the proliferation of microorganisms, indicating the need to keep these products under refrigeration during storage, as well as the adoption of other measures during processing to inhibit microbial proliferation. These results are similar to those established by Fellows (2009) for a cooked cured product (0.95); by Ferreira Silva Bartolomeu et al. (2014) who determined a_w of 0.98 for a smoked tilapia mortadella; and to the values reported by Campagnoli de Oliveira Filho, Favaro-Trindade et al. (2010) of 0.98 for tilapia MSM sausage; and also those by Dallabona et al. (2013) corresponding to 0.97-0.98 for tilapia MSM smoked sausages under different heat treatment conditions and packaging.

The pH values were similar to the results obtained for MSM. Other researchers such as Sleder et al. (2015) reported values lower than those found in this study, with mean pH values of 6.6 in tambaqui sausages (*Colossoma macropomum*); Cavenaghi-Altemio et al. (2013) determined mean values of 6.76 in analysing sausages produced from protein concentrates of tilapia; and (Mélo et al., 2011) found mean values of 6.39 in tilapia MSM mortadella.

In the present study, the pH values can be justified since the material used was raw, which already has higher values of pH, as well as sodium polyphosphate in the formulation (Mélo et al., 2011) which has a pH between 9.5 and 10.5.

There were no significant differences in the WRC values between the different fish mortadella formulatios (Table 2). However, these results were lower than those reported by Sleder et al. (2015). These authors identified WRC values ranging between 71.83 and 74.71 % in tambaqui mortadella with different fat percentages. Despite the low WRC values in this study, there was no damage to the structure of the formed emulsion, such as fat coalescence or syneresis, and it did not affect the sensory evaluations.

Regarding to moisture content, similar results have been reported for different types of emulsified meat products with MSM. For example, Sleder et al. (2015) observed a decrease in moisture values (72.73 %, 71.05 % and 68.82 %) as the fat content in the tambaqui sausage formulation increased (0 %, 4.5 % and 9 %, respectively). The results found in this study were similar to those of Yousefi and Moosavi-Nasab (2014), who studied MSM sausages or tilapia suprimi and found moisture values of 65.46 and 67.45 %, respectively; and those of Mélo et al. (2011), who analysed the moisture of tilapia MSM mortadella and determined decreasing values between 75.90 and 59.38 % as the fat percentage varied between 0 to 20 %, a similar behaviour to that found in the present investigation.

Despite the variations observed according to the added pork fat content, all formulations prepared in this study remained within the limits of moisture recommended by legislation Brasil (2000), which is 65 %. This differed from other studies with other types of emulsified meat products in which the average moisture of their products was above those quantified here, such as Campagnoli de Oliveira Filho, Favaro-Trindade et al. (2010), who produced tilapia MSM sausages with a mean moisture content of 70.75 %; and Cavenaghi-Altemio et al. (2013), who processed emulsified meat products with concentrated tilapia protein and found average moisture values of 75.83 %.

Increasing pork fat content caused the dry matter content to increase significantly ($p < 0.05$) in all formulations (Table 2). In addition to the increasing pork fat concentration in the formulations, the values were in line with the composition of the MSM extracted from the abdominal muscle, which contained considerable adiposity (Bordignon et al., 2010; de Oliveira, Henriques Lourenco, Sousa, Peixoto Joele & do Amaral Ribeiro, 2015). It is worth mentioning that fish fat is a source of omega-3, a fatty acid beneficial to consumer health (Menegazzo, Petenucci & Fonseca, 2016; Moreira et al., 2001; Nestel, 2000).

The values for ash represented the overall mineral content of the formulations, which did not differ significantly between them ($p > 0.05$) despite the fat percentage and MSM variations. Similar results were found by Sleder et al. (2015), who found quantities between 3.86 and 4.20 % in tambaqui sausages with different fat contents; by Dallabona et al. (2013), who found values between 3.44% in pasteurized sausage and 3.97 % in smoked sausage; and by Campagnoli de Oliveira Filho, Netto et al. (2010), who obtained similar results (3.40 %) in cooked emulsified meat products. The high levels of ash occurred due to the addition of ingredients such as salt and additives, as well as the incorporation of bone fragments to the mass.

The protein and fat content of the fish mortadella varied significantly ($p < 0.05$). Consequently, a concomitant decrease in protein content and increase in lipid content was observed for M1, M2 and M3. The importance of these nutrients and their equilibrium in forming a good emulsion and maintenance of their shelf life stability is known, in addition to the importance of the amount of proteins for greater firmness of the formed gel (Yousefi & Moosavi-Nasab, 2014). Thus, having a balanced percentage in the amount of proteins and fats is of fundamental importance to ensure obtaining a product with all the desirable characteristics present.

The fish mortadella formulated in the present study are products of high biological value and with reduced fat content, in addition to being a source of unsaturated fat. This is especially true for the M1 formulation, which presented a percentage of 18.09 % for proteins and 16.31 % for fats (almost half of that recommended by the legislation, which is at most 30 %). These results reinforced the relevance of this new product being proposed for the food industry.

In relation to sodium chloride, a statistical difference ($p < 0.05$) between the treatments was observed (Table 2), probably due to the fact that the processing is semi-industrial and carried out in batches of 500 g each, which may have caused differences in the homogenization pattern of the ingredients.

Calcium levels are well below that recommended by the legislation of a maximum of 0.90 % (Brasil, 2000). As expected, it was also observed that calcium values decreased ($p < 0.05$) as MSM was replaced by animal fat in the formulations. This fact is due to the reduction of the tilapia filleting residues, which contain bone fragments in their composition (calcium source).

Although there are no legal standards limiting the levels of lipid oxidation expressed as peroxide value, values above 1.51 mg of mEq/kg and 3.0 mEq/kg in fish and fish products, respectively, are classified as unacceptable (Campagnoli de Oliveira Filho, Favaro-Trindade et al., 2010). Thus, it is essential to verify the presence of oxidation in emulsified meat products through an evaluation of the peroxide index (mEq/kg), considering that it is possible to have important alterations in the food depending on its presence in the product due to lipid oxidation.

No changes to this parameter were found in this study with results of 0.0 mEq/kg, which showed that all the processing was in accordance with what is recommended by Good Manufacturing Practice (Brasil, 1997), and unlike other studies such as Ferreira Silva Bartolomeu et al. (2014), who found levels of 0.99 mEq/kg in tilapia mortadella; Calil Angelini et al. (2013), who quantified a mean of 0.22 mEq/kg in tilapia quenelles; and Campagnoli de Oliveira Filho, Favaro-Trindade et al. (2010), who found levels between 0.67 mEq/kg and 1.24 mEq/kg in sausages with percentages of tilapia MSM ranging from 0 to 100 %.

3.4 Microbiological evaluation of MSM and fish mortadella

All the formulations were in accordance with RDC No. 12 of January 2001 - National Health Surveillance Agency (Brasil. Agência Nacional de Vigilância Sanitária. Collegiate Board Resolution No. 12 of 2001. Approves the technical regulation on microbiological standards for food. Diário Oficial da República Federativa do Brasil, Poder Executivo, Brasília, DF, Brazil (in Portuguese)., 2001), and no viable cells were found for any of the microorganisms that were tested for. These results agree with those found by Ferreira Silva Bartolomeu et al. (2014), who formulated smoked tilapia MSM mortadella; and those of Campagnoli de Oliveira Filho, Netto et al. (2010) and Dallabona et al. (2013), who also elaborated tilapia MSM mortadella and tested its microbiological quality. These findings affirmed the feasibility of producing this type of emulsified meat product.

3.5 Sensory evaluation

Only the attributes texture and intention to purchase differed significantly among the treatments ($p < 0.05$) (Table 5). This occurred due to the correlation of the data for the instrumental texture and sensory texture. The increase in the fat content in the formulations corresponded to a reduction in the instrumental texture, which probably contributed to the reduction in the scores attributed to sensory texture ($p < 0.05$).
Nonetheless, all formulations presented mean scores equal to or above 6.0 (Table 5), demonstrating that the fish mortadella was sensorially well-accepted for this parameter; a behaviour also similar to that found by Rahman, Al-Waili, Guizani and Kasapis (2007), who observed a correlation between instrumental and sensory texture in fish sausages.
A positive correlation between instrumental hardness and fracturability with the sensory texture was observed, although they were higher for the M1 formulation (Table 3). There was good sensory acceptance for the texture attribute, presenting a higher mean score of 7.3 ($p < 0.05$) when compared to the other formulations. These findings were similar to the data found by Rahman et al. (2007), which showed that instrumental hardness is highly correlated with the sensory hardness of fish sausage.
The sensory scores for colour, odour, flavour and texture were higher than from studies with other emulsified products such as by Campagnoli de Oliveira Filho, Netto et al. (2010) and Cavenaghi-Altemio et al. (2013). In the present study, scores were similar to the results of Ferreira Silva Bartolomeu et al. (2014), who evaluated smoked tilapia MSM mortadella and found scores ranging from 7 (I moderately liked it) to 8 (I liked it a lot).
In addition, despite being a new product to the consumers, it was found that their overall acceptance varied between the hedonic terms “I slightly liked it” and “I liked it a lot”, with a mean score ranging from 6.8 ($\pm$1.5) to 7.1 ($\pm$1.4). Campagnoli de Oliveira Filho, Favaro-Trindade et al. (2010) found lower results than these for the overall assessment of sausages prepared with increasing percentages of MSM (0 to 100 %), with scores ranging from 6.1 to 5.0.
In an emulsion, the lack of balance between the amounts of fat and protein can harm its sensory evaluation (Yousefi & Moosavi-Nasab, 2014), which was not observed in this study considering that all formulations generally obtained average scores for the sensory attributes between 6 and 7. The M1 formulation stood out with scores for the appearance attributes, mortadella flavour, texture and overall assessment between 7 and 8, whose hedonic terms correspond to “I moderately liked it” and “I liked it a lot”.
These scores were reinforced by the comments made by the judges of which 105 out of the 126 tasters made some kind of comment with an emphasis on taste and texture attributes, with about 32.38 % and 10.47 % reporting that the samples presented “pleasant taste” and “pleasant texture”, respectively. The other comments were basically summarized as: “strong and/or smoky flavour” (13.33 %), “fishy taste” (7.61 %), “pleasant smell” (5.7 %), among others.
Other important data relates to the purchase intention, whose attributed scores ranged from the hedonic terms of 3 (“I would perhaps buy

Table 5: Mean scores of the sensory acceptance and purchase intention tests performed with of fish mortadella with different concentrations of tilapia MSM and pork fat.

Attributes	**Mortadella**		
	M1	**M2**	**M3**
Appearance	7.0 ±1.5[a]	6.7 ±1.6[a]	6.7 ±1.6[a]
Colour	6.8 ±1.6[a]	6.5 ±1.7[a]	6.7 ±1.8[a]
Aroma	6.8 ±1.8[a]	6.7 ±1.7[a]	6.6 ±1.7[a]
Specific mortadella flavour	7.0 ±1.5[a]	6.8 ±1.5[a]	6.9 ±1.5[a]
Specific fish flavour	6.8 ±1.4[a]	6.7 ±1.4[a]	6.5 ±1.5[a]
Texture	7.3 ±1.4[a]	6.6 ±1.7[b]	6.0 ±1.1[c]
Overall assessment	7.1 ±1.4[a]	6.8 ±1.5[a]	6.9 ±1.5[a]
Intention to purchase	3.9 ±1.1[a]	3.2 ±1.3[b]	3.0 ±1.0[c]

Means ± standard deviation with different letters on the same line differed by Tukey's test ($p<0.05$). M1 - Fish mortadella with 5% pork fat; M2 - Fish mortadella with 10% pork fat; M3 - Fish mortadella with 15% pork fat.

Table 6: Contribution to the formation of the principal component of the attributes evaluated in the sensorial analysis of mortadella samples.

Analyzed Attributes	**Principal Components**	
	1	**2**
Appearance	-0.716	0.526
Colour	-0.646	0.653
Aroma	-0.681	-0.167
Specific mortadella flavour	-0.836	-0.256
Specific fish flavour	-0.776	-0.367
Texture	-0.723	-0.052
Overall assessment	-0.889	-0.168

Table 7: Distribution of grades according to the ordination of general preference by the judges (n = 126) in the sensorial analysis of fish mortadella with different concentrations of tilapia MSM and pork fat.

Mortadella	**Number of Judges per Order***			**Sum of orders****
	1	2	3	
M1	34	33	59	277[a]
M2	55	44	27	224[b]
M3	57	44	25	220[b]

* 1 = least preferred, 3 = most preferred.
** Sum of the orders of each sample = (1 x number of judges) + (2 x number of judges + (3 x n^o judges). a, b - lower case letters indicate the significant differences between of the mortadellas ($p<0.05$) by the Friedman test.
M1 - Fish mortadella with 5% pork fat; M2 - Fish mortadella with 10% pork fat; M3 - Fish mortadella with 15% pork fat.

it/maybe not buy it") and 5 ("I would buy it"), with emphasis on the sample prepared with 5 % pork fat in its formulation which had a higher mean score ($p < 0.05$) when compared to the other formulations.

Figure 2 shows the acceptability indexes (AI) for the fish mortadella with different concentrations of tilapia MSM and pork fat. According to Teixeira et al. (1987), it is necessary that a product reaches an AI of at least 70 % in order for it to be considered as accepted in terms of sensory properties. Thus, it was observed that the AI for most of the attributes evaluated in the 3 fish mortadella formulations was above the minimum, with emphasis on the M1 fish mortadella.

The findings of this study agreed with Sleder et al. (2015) who verified that the best acceptance rates achieved in a study with tambaqui meat sausages were for those with lower pork fat content (9 %), with mean acceptance rates of 80 % for all the tested parameters. Mélo et al. (2011) also found expressive acceptance levels (78.43 %) for the overall assessment attribute in fish mortadella prepared with corn oil and wheat fibre.

The intent to purchase test indicated a tendency of purchasing fish mortadella with 5 % pork fat by a large proportion of the judges who participated in the sensory evaluations (73.02 %), as shown in Figure 2. Results similar to this were observed by Mélo et al. (2011), who reported an intent to purchase of 78.43 % for mortadella prepared with MSM, corn oil and wheat fibre. The data obtained in this study indicate that there is room in the market for the product when it is available, increasing the supply of nutritious and healthier products.

The principal component analysis of the attributes in the sensory acceptance analysis of the fish mortadella was important to verify which attributes most contributed to differentiating the three formulations, which is shown in Figure 3.

In the PCA, the first main component contributed to 57.26 % of the total variance and the second with 13.74 %, representing the first two factorial axes (71 %) in the total variance. If the first two or three components in a PCA accumulate a relatively high percentage of the total variance (generally above 70 %), they will satisfactorily explain the variability manifested among the samples (Mardia, 1979), and corroborate the data found in the present study.

For the Principal Component 2, colour and appearance were the attributes that most contributed to the group separation, as can be seen in Table 6. Thus, for the target audience in question who analyzed the fish mortadella formulations under study, the most important quality to differentiate them from one another were these sensory attributes, with particularly little influence by the fish taste and mortadella flavour attributes.

The preference order test allowed us to determine which product was most preferred by a particular target audience through indications of the judges, ordering samples from the "most preferred" to "least preferred". The results for the preference order are shown in Table 7.

According to the statistical analysis, a significant difference ($p < 0.05$) was observed between the M1 samples and the other formulations, in which the sample with 5 % pork fat in its constitution was more preferred, most probably due to the texture attribute since it also presented a higher score ($p < 0.05$) on the sensory acceptance test, also justifying its higher purchase intent.

When judges were asked which sensory characteristics they enjoyed the most in their preferred sample (M1), the majority of the answers were the flavour, the texture and the aroma. These attributes were similarly mentioned as characteristics that were not appreciated in the less preferred samples (M2 and M3).

4 Conclusion

In general, it can be affirmed that the different fish mortadella formulations had a good acceptability, with emphasis on the fish mortadella with 5 % of pork fat which, in addition to exceeding the minimum limits predicted to be considered approved by the consumer public, also presented excellent results for technological, physical parameters and nutritional value. The development of this study proved that technological processing of an emulsified meat product from fish capture to its consumption is feasible from the logistic, sanitary, and technological points of view, among others. The use of MSM did not depreciate the quality of the final product. We suggest that the production of this mortadella can represent a viable technological alternative for the cluse of tilapia processing waste.

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