Inclusion of dehydrated mixture made of salmon and tilapia carcass in spinach cakes

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ABSTRACT. The present study deals with dehydrated mixture of salmon (20%) and tilapia (80%) at 0, 5, 10 and 15% inclusion levels in spinach cake, and evaluates its proximate composition, sensory and microbiological characteristics. It was included 0, 5, 10 and 15% of the dehydrated mixture in the dough of the cake. Moisture and lipid contents were not modified (p >0.05) when the mixture was included in the cake. There was a positive linear effect for crude protein (y=6.24 + 0.29x, R²=0.77) and mineral (y=0.60 + 0.19x, R²=0.85). Carbohydrate and calorie contents decreased linearly (p <0.01) according to increasing levels of the mixture in the cake. With regard to sensory analysis, there was a negative linear effect for taste (y=7.48 – 0.07x, R²=0.98), texture (y=7.48 – 0.047x, R²=0.93), acceptance (y=7.13 – 0.046x, R²=0.99) and purchase intention (y=3.82 – 0.039x, R²=0.93), demonstrating that increasing levels of inclusion of the dehydrated mixture caused a decrease in the acceptance of the cake. It can be concluded that the inclusion of up to 15% of salmon and tilapia dehydrated mixture in spinach cake increases the contents of crude protein and mineral matter, and reduces the carbohydrate content and calorie. However, based on the sensory results, it is recommended to include 10% of the dehydrated mixture of fish in the spinach cake.

Keywords: Salmo salar, Oreochromis niloticus, processing waste, proximate composition, organoleptic characteristics.

Bolo de espinafre com inclusão de mix desidratado de carcaça de salmão e tilápia: composição química e sensorial

RESUMO. O objetivo deste trabalho foi incluir em bolo de espinafre um mix desidratado de salmão (20%) e tilápia (80%) e avaliar suas características sensoriais, microbiológica e composição centesimal. Foram incluídos 0, 5, 10 e 15% do mix na massa do bolo. Os teores de umidade e lipídios não foram alterados (p >0,05) com a inclusão de mix na massa. Observou-se efeito linear positivo (p <0,01) para proteína bruta (y=6,24 + 0,29x, R²=0,77) e matéria mineral (y=0,60 + 0,19x R²=0,85). Os carboidratos e valor calórico apresentaram diminuição linear (p <0,01), conforme acréscimo do nível de inclusão de mix no bolo. Na análise sensorial, observou-se efeito linear negativo para sabor (y=7,48 – 0,07x, R²=0,98), textura (y=7,48 – 0,047x, R²=0,93), aceitação (y=7,13 – 0,046x, R²=0,99) e intenção de compra (y=3,82 – 0,039x, R²=0,93), demonstrando que com o acréscimo dos níveis de inclusão do mix, houve redução na aceitação do bolo. A inclusão de até 15% de mix desidratado de salmão e tilápia no bolo de espinafre é capaz de aumentar o teor de proteína bruta e matéria mineral, além de diminuir o teor de carboidratos e valor calórico. Porém, com base nos resultados sensoriais, recomenda-se a inclusão máxima de 10% de mix desidratado de salmão e tilápia em bolo de espinafre.

Palavras-chave: Salmo salar, Oreochromis niloticus, resíduos de beneficiamento, composição centesimal, características organolépticas.

Introduction

Fish have high quality protein and are rich in essential amino acids. They represent a source of B complex vitamins and several minerals, including phosphorus, magnesium, iron, zinc and iodine in marine fish (Ariño, Beltrán, Herrera, & Roncalés, 2013).

Fish processing industries produce more than 60% by-products as waste, such as, viscera, skin, head, fins and fish-bones, which are rich in protein, minerals and fatty acids. They are usually transformed into low value products and sold on the market as fishmeal for animal consumption and as fertilizers (Chalamaiah, Hemalatha, & Jyothirmayi,
Due to the nutritional value in fish waste, they may be processed and transformed into products for human consumption by means of fish protein concentrates. Fish protein concentrates are defined as dehydrated and ground products with variable protein content that may provide or not the flavor and aroma of fish, depending on the processing method (Ordóñez et al., 2005). The above mentioned concentrates are not directly consumed but are employed as raw material in the preparation of products with aggregated high value (Fontana, Centenaro, Palezi, & Prentice-Hernández, 2009) and with excellent amino acids composition.

The mixture of protein concentrates from different fish species (dehydrated mixture) may combine into a single product the nutritional characteristics of marine species, such as the salmon (Salmo salar) and freshwater species, such as the Nile tilapia (Oreochromis niloticus). Lipids in sea water fish are rich in polyunsaturated fatty acids, especially Omega 3, known for its cardiovascular benefits (Psota, Gebauer, & Kris-Etherton, 2006). Protein from tilapia meat has high nutritional value with a balance of essential amino acids, especially rich in lysine (Pizato, Kraieski, Sarmento, & Prentice, 2012).

Several studies have developed fish protein concentrates from different fish species (Murueta, Toro, & Carreño, 2007; Vidal, Rodrigues, Zapata, & Vieira, 2011; Godoy, Franco, Souza, Stevanato, & Visentainer, 2013) and obtained crude protein between 60 and 85%, coupled to several lipids. Protein concentrates may be included in sweet and salty products, such as biscuits (Ibrahim, 2009), broths and soups (Godoy et al., 2010), extruded snacks (Justen et al., 2011), cookies and biscuits (Franco et al., 2013), and others. Due to an improvement in the product nutritional profile and good sensory acceptance, protein concentrates may be included in cakes appreciated by all, especially children. Chocolate and carrot cakes with inclusion of tilapia fillets were developed by Veit et al. (2013) achieving an improvement in nutritional composition of cakes without any fish addition, and with acceptability index over 88% among children aged 8 – 10 years.

The present study aimed at including a dehydrated mixture of salmon (20%) and tilapia (80%) in a spinach cake and assessing its proximate composition, sensory and microbiological characteristics.

Material and methods

The dehydrated mixture of fish and spinach cakes were prepared at the Fish Technology Laboratory of the Iguatemi Experimental Farm, Universidade Estadual de Maringá, Maringá, State of Paraná, Brazil.

Preparation of fish dehydrated mixture

Carcasses (spine and ribs), without head and fins, of Nile tilapia (O.niloticus), donated by SmartFish (Rolândia, Paraná State, Brazil) and of salmon (S. salar), donated by Tomita and Tomita Ltda (Maringá, Paraná State, Brazil), were washed in chlorine water and cooked for 60 min in an industrial pressure cooker. They were then ground, pressed under a 10t pressure and the mass was dehydrated in a forced air chamber for 24 hours at 60ºC. After dehydration, fishmeal was ground in a Wiley mill and vacuum-packed. This product was the protein concentrate of tilapia and salmon. The dehydrated mixture was prepared with 20 and 80% of the protein concentrate of salmon and tilapia, respectively. This mixture was prepared with the purpose of including salmon nutrients (especially polyunsaturated fatty acids) to the spinach cake; but to not affect the sensory characteristics, we used a higher proportion of tilapia meal, which has less pronounced flavor and odor compared to the salmon meal.

Preparation of the spinach cake

The spinach cake consisted of the dough and different levels of the dehydrated mixture (0, 5, 10 and 15%) according to the weight of the wheat flour in the cake. Table 1 provides details on ingredients. The different inclusion levels of the dehydrated mixture were the different treatments used, namely, T1=0, T2=5, T3=10 and T4=15%.

<table>
<thead>
<tr>
<th>Ingredients (%)</th>
<th>Inclusion levels of fish dehydrated mixture (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wheat flour</td>
<td>0%   5%  10% 15%</td>
</tr>
<tr>
<td>Dehydrated mix of salmon/tilapia</td>
<td>0 1 3 4</td>
</tr>
<tr>
<td>Sugar</td>
<td>20   20  20  20</td>
</tr>
<tr>
<td>Oil</td>
<td>9    9   9   9</td>
</tr>
<tr>
<td>Fresh spinach</td>
<td>10   10  10  10</td>
</tr>
<tr>
<td>Corn starch</td>
<td>3    3   3   3</td>
</tr>
<tr>
<td>Milk</td>
<td>10   10  10  10</td>
</tr>
<tr>
<td>Baking powder</td>
<td>1    1   1   1</td>
</tr>
<tr>
<td>Granulated chocolate</td>
<td>4 4 4 4</td>
</tr>
<tr>
<td>Egg yolk</td>
<td>12   12  12  12</td>
</tr>
<tr>
<td>Beaten egg white</td>
<td>5    5   5   5</td>
</tr>
<tr>
<td>Total</td>
<td>100  100 100 100</td>
</tr>
</tbody>
</table>

1Levels of the dehydrated mixture (0, 5, 10 and 15%) added according to the weight of the wheat flour in the cake.

Dried ingredients were weighed, sieved and mixed, whilst spinach, oil, egg yolk and milk were...
placed in a blender and homogenized. The other
ingredients were then added, except the baking
powder, which was mixed together with the
granulated chocolate and beaten egg white. The
dough was poured into an aluminum pan greased,
powdered with wheat flour, baked for 40 min at
180°C in a pre-heated electric oven. The products
were then cooled to room temperature and stored in
a fridge until analysis.

**Proximate composition and calorie**

Analyses of proximate composition were
performed at the Food Analysis Laboratory of the
Animal Science Department, Universidade Estadual
de Maringá. Moisture, protein, lipid and ash were
conducted in triplicate, according to AOAC
methodology (Associations of Official Analytical
Chemists [AOAC], 2005). Carbohydrate were
assessed by the difference of the other components
(Brasil, 2003) and total calories was obtained by the
sum of the multiplication of average rates of protein,
lipids and carbohydrates multiplied by 4, 9 and 4,
respectively (Souci, Fachman, & Kraut, 2000).

**Sensory analysis**

Sensory analysis was carried out by 50 non-
trained tasters (undergraduates and staff of the
Universidade Estadual de Maringá). A sample of
approximately 30 g of each treatment was provided
to each taster on disposable containers and identified
by three randomized numbers each. A sensory
analysis form was provided, along with a glass of
water to remove the residual flavor between the
samples. External aspect, aroma, flavor, texture and
general acceptance were evaluated by a 9-point
hedonic scale, from 1 = “dislike very much” to
9 = “like very much” (Dutcosky, 2007). The
Acceptability Index (AI) was calculated by the
equation 1(Dutcosky, 2007):

$$AI\% = \frac{A}{B} \times 100$$

where:

A = is the average score for the product;
B = maximum score of the scale.

The purchasing intention of the product was
calculated by a 5-point hedonic scale, between
1(definitely would not buy) and 5 (definitely would
buy) (Damásio & Silva, 1996).

**Microbiological analysis**

Microbiological analysis of the fish dehydrated
mixture and spinach cake was performed for each
treatment at the Food Microbiology and Microscopy
laboratory of the Department of Clinical Analyses,
Universidade Estadual de Maringá. Microbiological
analysis of samples was conducted for the Most
Probable Number (MPNg) of coliforms at 35 and
45°C, counts of coagulase-positive Staphylococcus
in a Colony-Forming Unit (CFUg) and research on
Salmonella sp. (American Public Health Association
[Apha], 2001).

**Statistical analysis**

Data of proximate composition and calorie was
tested by regression analysis at 5% using the
Statistical Analysis System (SAS Institute Inc., Cary,
NC, USA). Results of sensory analysis underwent
Proc genmod of SAS, with distribution of variables
as an inverse connection range. Effect of treatment
and tasters were taken into account testing scores
from the hedonic scale at the inclusion levels by the
analysis of regression at 5% significance.

**Results**

Moisture and lipid were not affected (p >0.05)
by the inclusion of the mixture in the dough
(Table 2). There was a positive linear effect
(p <0.01) for crude protein (γ=6.24 + 0.29x, R²=0.77)
and minerals (γ=0.60 + 0.19x, 
R²=0.85). The increase in inclusion levels caused an
increase in nutrients. Carbohydrates and calories
had a linear decrease (p <0.01), according to the
increase of inclusion levels of the mixture in the
cake.

### Table 2. Proximate composition and calorie of the dehydrated mixture and spinach cake with the inclusion of the dehydrated mixture of salmon and tilapia carcass.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Dehydrated mixture of fish</th>
<th>Inclusion levels of fish dehydrated mixture (%)</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Moisture (%)</td>
<td>6.00±0.15</td>
<td>2.96±0.04</td>
<td>2.76±0.11</td>
</tr>
<tr>
<td>Crude protein (%)</td>
<td>48.57±0.05</td>
<td>6.33±0.03</td>
<td>7.13±0.49</td>
</tr>
<tr>
<td>Lipid (%)</td>
<td>12.95±0.05</td>
<td>16.37±0.79</td>
<td>15.30±0.95</td>
</tr>
<tr>
<td>Mineral matter (%)</td>
<td>30.89±0.05</td>
<td>0.67±0.37</td>
<td>1.27±0.62</td>
</tr>
<tr>
<td>Carbohydrate (%)</td>
<td>1.59±0.25</td>
<td>73.68±0.55</td>
<td>73.54±0.44</td>
</tr>
<tr>
<td>Calorie (kcal100g⁻¹)</td>
<td>317.21±0.34</td>
<td>467.35±3.27</td>
<td>460.41±3.32</td>
</tr>
</tbody>
</table>

Linear regression : $\gamma=6.2438 +0.2896x$, $R^2=0.77$; $\gamma=0.5962 + 0.1872x$, $R^2=0.85$; $\gamma=73.9787 - 0.2782x$, $R^2=0.77$; $\gamma=466.5328 - 0.0353x$, $R^2=0.85$. Values expressed asMean±Standard deviation.
There was a concentration of <1x10^2 CFU/g of positive coagulase Staphylococcus in the microbiological profile of the spinach cake containing different levels of the dehydrated mixture. All samples showed that coliforms at containing different levels of the dehydrated mixture. All samples showed that coliforms at 35 and 45°C were less than 3MPNg⁻¹, whereas research on Salmonella sp. indicate its absence in 25 g of each sample examined.

In the case of sensory analysis, there was a negative linear effect for flavor (y = 7.48 – 0.07x, R²=0.98), texture (y = 7.48 – 0.047x, R²=0.93), acceptance (y = 7.13 – 0.046x, R²=0.99) and purchase intention (y = 3.82 – 0.039x, R²=0.93). These results evidence increasing levels of the dehydrated mixture lead to a reduced acceptance of the spinach cake (Table 3). Aroma and color were not affected by different inclusion levels (p >0.05), averaging between 6.54 and 6.98 for aroma and between 6.56 and 6.78 for color.

### Table 3. Sensory analysis of spinach cake with different levels of inclusions of the dehydrated mixture made up of salmon and tilapia carcass.

<table>
<thead>
<tr>
<th>Inclusion levels of fish dehydrated mixture (%)</th>
<th>0</th>
<th>5</th>
<th>10</th>
<th>15</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aroma³</td>
<td>6.98±1.57</td>
<td>6.86±1.59</td>
<td>6.66±1.89</td>
<td>6.54±1.74</td>
<td>0.158</td>
</tr>
<tr>
<td>Flavor¹</td>
<td>7.36±1.64</td>
<td>7.26±1.45</td>
<td>6.84±1.82</td>
<td>6.28±1.97</td>
<td>0.001¹</td>
</tr>
<tr>
<td>Texture¹</td>
<td>7.44±1.47</td>
<td>7.34±1.35</td>
<td>6.92±1.61</td>
<td>6.80±1.65</td>
<td>0.016¹</td>
</tr>
<tr>
<td>Color¹</td>
<td>6.68±1.57</td>
<td>6.78±1.56</td>
<td>6.56±1.58</td>
<td>6.68±1.72</td>
<td>0.829</td>
</tr>
<tr>
<td>General acceptance¹</td>
<td>7.08±1.63</td>
<td>6.98±1.58</td>
<td>6.72±1.74</td>
<td>6.40±1.86</td>
<td>0.035¹</td>
</tr>
<tr>
<td>Purchase intention²</td>
<td>3.84±0.93</td>
<td>3.56±1.15</td>
<td>3.52±1.15</td>
<td>3.20±1.23</td>
<td>0.047¹</td>
</tr>
</tbody>
</table>

n=30; ¹Hedonic scale between 1 (definitely would not buy) and 5 (definitely would buy). Linear regression: y=7.48 – 0.07x, R²=0.98; y=7.48 – 0.047x, R²=0.93; y=7.13 – 0.046x, R²=0.99; y=3.82 – 0.039x, R²=0.93. Values expressed as Mean±Standard deviation.

### Discussion

The inclusion of fish dehydrated mixture increased the crude protein, as expected due to the high concentration of such nutrient in the dehydrated mixture, which presented 48.57% crude protein. The highest inclusion level (15%) had the greatest protein percentage, corroborated by Stevanato et al. (2007), who added fishmeal from tilapia head in soups offered in school meals, and by Franco et al. (2013), who included fishmeal from tilapia carcasses in cookies and biscuits. An increase in the content of crude protein in the products containing protein concentrates was reported. It should be highlighted that protein in fish contains high biological value, with a balanced composition of aminoacids especially those restricted to protein of vegetal origin, such as methionine and cysteine (Neves, Mira, & Marquez, 2004).

Minerals and crude protein also increased as the levels of inclusion of dehydrated fish mixture increased, may be because of the spine in tilapia and salmon carcasses used as a raw material for the preparation of protein concentrates. The above evidences the mineral enrichment in spinach cakes, since fish are composed of a great variety of minerals, which include phosphorus, magnesium, iron, zinc and iodine in marine fish (Ariño et al., 2013).

Carbohydrates and calorie decreased linearly (p <0.01) in proportion to the increase in the inclusion level of the mixture in the cake. This may have occurred due to the low carbohydrate content in the fish dehydrated mixture (1.59%).

The microbiological profile of dehydrated mixture and of the spinach cake with different inclusion levels showed that the products complied with standards required by the National Health Surveillance Agency (Anvisa), with the Resolution 12 of January 2nd, 2001 (Brasil, 2001), or rather, mixture and cakes were prepared under adequate hygiene conditions and thus proper for human consumption.

In the case of sensory analysis, increasing level of inclusion of fish dehydrated mixture caused a decrease in the acceptance of spinach cakes with regard to flavor, texture and general acceptance. Our results show that in spinach cake, the inclusion of dehydrated mixture of fish should not exceed 10%, possibly due to the protein concentrate of the sea water salmon with its more pronounced flavor and odor, even with the inclusion of only 20% together with the tilapia concentrate (80%). However, scores varied between 6.28 and 7.34 and revealed that tasters liked the product slightly (score 6) to moderately (score 7).

Veit et al. (2013) developed and characterized chocolate and carrot cakes with tilapia fillets. The acceptance of the cakes showed that the students liked the chocolate and carrot cake with fillets. Mean values were very close to the highest scores of the scale. Justen et al. (2011) observed that the inclusion of up to 12% of aromatized tilapia fishmeal failed to affect the sensory parameters (aroma, flavor, physical aspect, texture, color and general acceptance) of maize extruded snacks, with scores between 6.92 and 8.15 for all attributes.

In addition, there was a reduction in purchase intention, scores from 3.84 to 3.20, due to the increase of inclusion levels of fish dehydrated mixture. However, 32% of tasters of the spinach cake with an inclusion of up to 10% of dehydrated mixture affirmed they would possibly buy the spinach cake and 22% said they would certainly buy.

Our findings demonstrate that fish dehydrated mixture may be included in the preparation of candies, such as cakes, cookies, bread, and others, that may make part of school meals. The spinach...
Spinach cakes with dehydrated mixture of fish

cake may be a good alternative to be included in school meals since it has excellent and healthy nutritional characteristics. Once fish is an excellent quality food, its inclusion in school meals in products highly appreciated by children is a promising alternative to consuming fish. Besides, an alternative manner for using filleting waste has been found.

Conclusion

The inclusion of up to 15% of the dehydrated mixture (20% of protein concentrate of salmon and 80% of tilapia) in spinach cake increases the contents of crude protein and mineral matter, besides reducing the carbohydrate content. However, based on the sensory results, we recommend a maximum inclusion level of 10% of the dehydrated mixture of fish in the spinach cake.

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