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Effect of essential oils of copaiba and oregano in substitution of synthetic antioxidants in Tambaqui fish meat balls

Efeito dos óleos essenciais de copaíba e orégano em substituição de antioxidantes sintéticos em quibes de Tambaqui

Efecto de los aceites esenciales de copaiba y orégano en sustitución de antioxidantes sintéticos en kibbehs de tambaqui

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Abstract

The objective of the present experiment was to evaluate the effects of including the essential oil of copaiba and oregano as a possible replacement for synthetic butylated hydroxytoluene (BHT) additives in fish meat ball. PH, cooking loss, texture, color, sensory acceptability and chemical composition were evaluated. The addition of oregano essential oil had an effect on the color of the fish meat ball (P <0.05), showing better results for a * and Hue compared to BHT, and b * and Croma compared to the control (COM). In addition, the essential oils of copaiba and oregano showed the lowest water losses by cooking and the essential oil of oregano improved the aroma of the fish meat ball (P <0.05). The treatments had no effect (P> 0.05) on the chemical composition of the fish meat ball. The essential oils of copaiba and oregano are promising natural antioxidants to increase the useful life of fish by-products, in addition to being a viable solution to replace the synthetic antioxidant BHT, bringing benefits to its commercialization.

Keywords: Antioxidant; Fish; Processed; Meat quality.

Resumo

O objetivo do presente experimento foi avaliar os efeitos da inclusão do óleo essencial de copaíba e orégano como possível substituição de aditivos sintéticos butylated hydroxytoluene (BHT), em fish meat ball. Foram avaliadas o pH, perda por cocção, textura, cor, aceitabilidade sensorial e composição química. A adição de óleo essencial de orégano apresentou efeito sobre a cor dos fish meat ball (P< 0,05), apresentando melhores resultados de a* e Hue em relação ao BHT, e b* e Croma em comparação ao controle (COM). Além disso, os óleos essenciais de copaíba e orégano apresentaram as menores perdas de água por cocção e o óleo essencial de orégano melhorou o aroma das fish meat ball (P< 0,05). Os tratamentos não tiveram efeito (P> 0,05) na composição química dos fish meat ball. Os óleos essenciais de copaíba e orégano são antioxidantes naturais promissores para aumentar a vida útil de subprodutos do pescado, além de ser uma solução viável para substituir o antioxidante sintético BHT, trazendo beneficios para sua comercialização.

Palavras-chave: Antioxidante; Pescado; Processados; Qualidade de carne.

Resumen

El objetivo del presente experimento fue evaluar los efectos de incluir el aceite esencial de copaiba y orégano como posible reemplazo de los aditivos sintéticos de hidroxitolueno butilado (BHT) en la bola de carne de pescado. Se evaluó el PH, la pérdida de cocción, la textura, el color, la aceptabilidad sensorial y la composición química. La adición de aceite esencial de orégano tuvo un efecto sobre el color de la bola de carne de pescado (P <0.05), mostrando mejores resultados para a * y Hue en comparación con BHT, yb * y Croma en comparación con el control (COM). Además, los aceites esenciales de copaiba y orégano mostraron las menores pérdidas de agua por cocción y el aceite esencial de orégano mejoró el aroma de la bola de carne de pescado (P <0.05). Los tratamientos no tuvieron efecto (P> 0.05) sobre la composición química de la bola de carne de pescado. Los aceites esenciales de copaiba y orégano son antioxidantes naturales prometedores para aumentar la vida útil de los subproductos del pescado, además de ser una solución viable para reemplazar el antioxidante sintético BHT, aportando beneficios a su comercialización.

Palabras clave: Antioxidante; Pescado; Procesado; Calidad de la carne.

1. Introduction

Concerns about food quality have led consumers to seek healthier nutritionally foods, leading to an increase in the consumption of fish and its processed products, being sources of proteins, lipids and micronutrients, which are essential for a good diet (Angiolillo et al., 2018; Vieira et al., 2019). The consumption of fishery products is gradually increasing as consumers learn about the benefits of fish for health (Ben Atitallah et al., 2019; Thilsted et al., 2016).

On the other hand, the higher consumption of processed foods is caused by the changes in the routine of consumers where the consumption of ready-made meals is more practical (Monteiro et al., 2010). For the preservation of these foods, it is necessary to use antioxidant substances, the addition of these chemical preservatives can minimize or prevent lipid peroxidation, delaying the formation of toxic compounds and increasing the shelf life.

The most widely used synthetic antioxidants are butylated hydroxyanisole (BHA) and butylated hydroxytoluene (BHT) (Kempinski et al., 2017), but there is a concern about the possible carcinogenic effects, respiratory and neurological disorders that these substances can cause, threatening health consumers (Ayoughi et al., 2011; Carocho et al., 2014; Wilson & Bahna, 2005).

Thus, to deal with these challenges, new food preservation methods have been developed, including essential oils from medicinal plants that have been studied as natural food preservatives attributed to their antioxidant and antimicrobial activities, with possible substitution to synthetic preservatives (Fachinello et al., 2018; Fernandes et al., 2016; Vital et al., 2016).

The Tambaqui (*Colossoma macropomum*) is a fish native to the Amazon River that is currently grown widely and intensively in the region, with the potential to be highly exported in the future, but one of the main challenges for its commercialization is to improve preservation by reducing lipid peroxidation of its fatty meat (Paulino et al., 2018; Vieira et al., 2019).

The essential oil of copaíba (CO) is widely distributed in northern South America, especially in the Brazilian Amazon rainforest and its main bioactive components are phenolic compounds, such as sesquiterpenes and diterpenes (Brochini & Lago, 2007). Oregano essential oil (EO), is rich in carvacrol and thymol, has antioxidant and antimicrobial activity (Bentayeb, et al., 2014; Kempinski et al., 2017) and can be successfully incorporated into food formulations.

Given the above, the objective was to evaluate the physical-chemical and sensory characteristics of fish meat ball from Tambaqui with the addition of copaiba and oregano essential oil, in comparison with the most commonly used preservative, BHT.

2. Material e Methods

This research is characterized as of a quantitative nature, in which the collection of numerical data is carried out to generate a set of data that will be later analyzed statistically (Pereira et al., 2018).

2.1. Preparation of fish meat balls

The fish meat ball were divided into four groups: Control - without antioxidants (CON), fish meat ball containing hydroxytoluenobutylated (BHT), fish meat ball with 0.125% of Copaiba essential oil (CEO), and fish meat ball with 0.125% of Oregano essential oil (OEO). These filets were minced in a grinder and the meat was mixed and added 10 g of NaCl / kg of meat and 50 mg / kg BHT according to (Fernandes et al., 2016) and the essential oils. The CEO and OEO was obtained from Ferquima® (Vargem Grande Paulista, São Paulo, Brazil) and the concentration used was based on the observed results of (Chaleshtori et al., 2014).

The fish meat balls with 20g of weight (n = 4 per treatment), were made with cuts of Tambaqui fillet with an average weight of the animals 1.5 kg. These cuts were chopped in a grinding machine and the meat was mixed with the other ingredients, shown in able 1.

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Table 1. Proportions (in grams) used in Tambaqui-based fish meat balls formulations.

Ingredients	**Grams**
Fish	1776
Kibe flour	125
Salt	51
Black-pepper	3.0
Mint "in natura"	36
Water	240
Garlic powder	6
Dehydrated Onions	120
Dehydrated Parsley	18

Source: Research Data (2019).

2.2. Instrumental meat color

The color was evaluated by using the CIELab system with a Minolta CR- 400 Chroma meter (Japan) (with a 10° view angle, D65 illuminant, 8 mm of aperture with a close cone). Six measurements at randomly selected points were recorded per sample, obtaining lightness (L*), redness (a*) and yellowness (b*). Chroma and hue values were calculated as follows:

Chroma = ($a^{*2}+ b^{*2}$) and hue angle (h^0) = (arctan (b^* a^*)).

2.3. pH measurements

The pH was measured using a pH meter Text, equipped with a penetration pH electrode. The pH Meter was calibrated at 20 °C using standard pH 4.0 and 7.0 buffers

2.4. Cooking losses

The fish meat ball were weighed and wrapped in aluminum foil. Each sample was cooked in a pre-heated grill (Grill Philco Jumbo Inox, Philco SA, Brazil) at 200 °C until an internal temperature of 72 °C was reached, which was monitored using an internal thermocouple (Incoterm, 145 mm, Incoterm LTDA, Brazil). The sample was then removed from the heat and left at ambient temperature to cool. Once the fish meat ball reached 25 °C, each fish meat ball was weighed. The cooking losses were calculated ach day as a percentage relative to the initial weight according to the following equation:

$$WL\ (\%) = \frac{Wi - Wf}{Wi * 100}$$

where, Wi = initial weight (at the Day of production) and Wf = final weight

2.5. Texture measurement

The fish meat ball was removed from the heat and left at ambient temperature to cool 25°C. Was cut into rectangular pieces of 1 cm^2 cross-section (eight pieces per treatment). The texture was analyzed using a texture analyzer with a Warner-Bratzler blade and set with a 50 kg load cell and a speed of 2 mm/s. The maximum shear force (Kgf) was recorded.

2.6. Characterization of hamburgers

The moisture, protein, lipid and ash were measured in raw fish meat ball (production day) according Association of Official Analytical Chemists (AOAC, 1990). Crude protein was obtained by the semi-micro Kjeldahl method following three distinct steps: digestion, distillation and titration (Silva & Queiroz, 2002).

2.7. Consumer acceptability

The acceptability of cooked fish meat ball attributes, with one day of storage, was evaluated: odor, tenderness, flavor, and overall acceptability; with a structured hedonic nine-point scale (1 = dislike extremely; 9 = like extremely), without the middle level (Font-i-Furnols & Guerrero, 2014).

Fish meat ball was cooked until the internal temperature reached 70°C on a pre-heated grill at 200°C. The fish meat ball were cut into four triangles and kept warm (50°C) until consumer evaluation (around 10 min after cooking).

Analyses were conducted with 71 untrained tasters at the Federal University of Roraima (Brazil) in a room adequately to perform the sensory test, according to the Brazilian national profile within quotas of gender and age. The consumers were randomly selected between people who attend the university (students, employees and visitors). Each one evaluated four samples (CON, BHT, CEO and OEO) codified with a random three-digit code.

2.8. Statistical analysis

Statistical analyzes were performed using SPSS (version 23.0) (IBM SPSS Statistics, SPSS Inc., Chicago, IL, USA) for Windows. The mean and standard deviation were calculated for each variable. The treatment was the only fixed effect evaluated, with triplicates per treatment, for each analysis. When the differences were statistically significant, the Tukey test was performed, with statistical significance set at $p <0.05$.

3. Results and Discussion

The color values (L*, a*, b*, C*, and h°) associated with treatments are given in Table 2.

Color is one of the most important factor in the consumer's perception of meat quality, as it is a characteristic that influences both the initial choice of the product by the consumer and acceptance at the time of consumption (Fletcher, 1999) Lightness (L*) not was significantly different between treatments ($P < 0.05$).

There was a notable difference in the a* values between the fish meat ball ($P < 0.05$), with redness intensity greatest for OEO, not differing from the CEO and CON and the BHT exhibited the lowest redness intensity. This difference may have occurred due to the high antioxidant power of the essential oil of oregano that decreased oxidation of the heme group within the iron atom and a lower formation of MetMb (Faustman et al., 2010).

Regarding the yellow color index (b), the OEO presented higher b * values (yellowness; $P <0.05$) relative to CEO and BHT, and, particularly, CON, which presented the lowest value. This difference occurred because the essential oil of oregano has a lemon yellow color (Quer et al., 1988), so the treatment with oregano showed higher levels of this color. The maintenance of color (redness and yellowing) can be attributed to the action of antioxidants added to treatments (BHT, CEO and OEO), which contributes to the stabilization of color by delaying discoloration (Kempinski et al., 2017; Mancini et al., 2015).

C* was affected ($P <0.05$) between treatments, with higher values for OEO, intermediary for BHT and CEO and, CON, which presented the lowest value. The h ° (shade angle) showed a difference (p <0.05) between treatments. BHT was with higher values, while OEO was the least. CON and CEO had intermediate values.

The meat product with low chroma values is considered pale (Cardoso et al., 2016). In this study, the OEO treatment had the highest chroma value compared to the CON. (Atitallah et al., 2019) also observed differences between treatments when evaluating microalgae-fortified canned fish burgers with decreased chroma values with the addition of microalgae.

According to Lee et al. (2005), the increase in the value of H * indicates a process of discoloration of the meat product, making the product paler, which was observed in this test for BHT treatment in comparison to OEO.

Table 2. Evolution of color CIE parameters (L*, a*, b*, C* and H*) in fish meat ball treated with BHT and of copaiba essential oil and Oregano.

Variables	CON[1]	BHT[2]	CEO[3]	OEO[4]	SEM[5]	P value
L	54.05	50.50	48.89	47.96	0.933	0.073
a	1.92ab	1.69b	2.47ab	2.87a	0.174	0.036
b	12.11b	14.45ab	14.25ab	16.46a	0.606	0.049
Chroma	12.26b	14.55ab	14.47ab	16.71a	0.618	0.050
Hue	81.01ab	83.35a	80.21ab	80.08b	0.498	0.038

Different lower case letters in the same line are significantly different. [1]CON: fish meat ball, without antioxidants; [2]BHT: fish meat ball containing hydroxytolueneobutylated; [3]CEO: fish meat ball with 0.125% of Copaiba essential oil; [4]OEO: fish meat ball with 0.125% of Orégano essential oil. [5]SEM: Standard error of means. Source: Research Data (2019).

Table 3. Effect of copaiba essential oil and oregano and BHT inclusion on parameters pH, cooking loss e Shear force in fish meat ball.

Variables	CON[1]	BHT[2]	CEO[3]	OEO[4]	SEM[5]	P value
pH	6.05	6.50	6.35	6.40	0.106	0.585
PPC	11.05b	12.49a	8.39bc	6.68c	0.880	0.006
Texture (Newton)	3.54	3.29	3.79	3.76	0.227	0.913

Different lower case letters in the same line are significantly different. [1]CON: fish meat ball, without antioxidants; [2]BHT: fish meat ball containing hydroxytolueneobutylated; [3]CEO: fish meat ball with 0.125% of Copaiba essential oil; [4]OEO: fish meat ball with 0.125% of Oregano essential oil. [5]SEM: Standard error of means. Source: Research Data (2019).

For pH values and texture the addition of natural antioxidant or BHT did not imply differences (P > 0.05) between treatments (Table 3).

In most fish species, the pH post mortem is between 6.0 and 6.8, and the differences between these values may be due to species, diet, stress level during capture, season and type of muscle (Khalafalla et al., 2018). The pH value of this study is within the limits of the ideal value (6.32).

Atitallah et al. (2019) evaluating microalgae-fortified canned fish burgers, also found no effect between treatments for pH values, with mean values of (7.0) between treatments. According to the author, this result is necessary for a better stability of fishery products.

Higher pH values (6.56) were observed by (Albertos et al., 2019) in the elaboration of functional bio-based seaweed edible films for extending the shelf life of fresh fish burgers compared to this study (6.32). The treatments had affected ($P < 0.05$) the cooking loss (Table 3). Cooking loss was largest for BHT treatment and lowest for OEO, while CON had intermediate losses. CEO did not differ from CON and OEO.

The loss by cooking of the food is an important quality characteristic, associated with the meat yield at the moment of consumption, which can be influenced by the water retention capacity in the meat structures (Pardi et al., 1993; Silva et al., 2005). The lower value of cooking loss for the OEO treatment in comparison to the CON attributes better acceptability characteristics for the fish meat ball, and lower water losses during the preparation of a food provides better texture and juiciness of the food (Caldara et al., 2012). In contrast to this finding, (Vital et al., 2018) observed that fish fillet with alginate-based coating containing essential oil of ginger and oregano did not show a significant weight loss.

The EOs incorporation had no effect on this textural parameter of fish meat ball (Table 3). This behaviour could be due to the fact that the preparation of the burgers included mincing the fish; consequently, the muscle structure was destroyed, the same observed by Dolea et al., 2018, when evaluating the effect of thyme and oregano essential oils of salmon and seaweed burgers.

Textural modification of foods is a criterion commonly used to define the sensory quality of foods, with instrumental texture being strongly correlated with organoleptic characteristics providing more data than sensory evaluation (Atitallah et al., 2019; Steele et al., 2015).

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Table 4. Chemical composition analysis in fish meat ball treated with BHT and of copaiba essential oil and Oregano.

Variables	CON[1]	BHT[2]	CEO[3]	OEO[4]	SEM[5]	P value
Humidity	62.87	62.56	62.58	63.77	0.378	0.747
Ashes	0.96	0.96	0.91	0.99	0.013	0.145
Protein	20.59	18.35	21.10	18.82	0.492	0.080
Lipids	2.04	1.91	2.00	1.91	0.024	0.122

Different lower case letters in the same line are significantly different. [1]CON: fish meat ball, without antioxidants; [2]BHT: fish meat ball containing hydroxytolueneobutylated; [3]CEO: fish meat ball with 0.125% of Copaiba essential oil; [4]OEO: fish meat ball with 0.125% of Orégano essential oil. [5]SEM: Standard error of means. Source: Research Data (2019).

Regarding the compositional analysis (Table 4), there were no significant differences ($p > 0.05$) between the treatments. The average values of the main components were: humidity 63%, ashes 0.95%, protein 19.71%, and lipids 1.96%.

The values observed in this study for moisture, ash, protein and lipids are close to those found by Kakaei & Shahbazi (2016) for chopped trout fillet with chitosan-gelatin film incorporated with ethanolic red grape seed extract and *Ziziphora clinopodioides* essential oil. Protein and lipid values are consistent with desired levels for such products according to Tokur et al. (2004) when evaluating fish burger produced from Tilapia during frozen storage, crude protein (17.82%) and lipid values (5.29%).

Table 5. Consumer acceptability of fish meat ball (n = 71 consumers)

Variáveis	CON[1]	BHT[2]	CEO[3]	OEO[4]	SEM[5]	P value
Aroma	7.04b	7.19b	7.11b	8.07a	0.102	0.001
Flavor	7.19	7.42	7.11	7.28	0.101	0.733
Texture	7.47	7.50	7.42	7.59	0.097	0.944
General acceptance	7.35	7.42	7.16	7.47	0.099	0.709

Different lower case letters in the same line are significantly different. [1]CON: fish meat ball, without antioxidants; [2]BHT: fish meat ball containing hydroxytolueneobutylated; [3]CEO: fish meat ball with 0.125% of Copaiba essential oil; [4]OEO: fish meat ball with 0.125% of Orégano essential oil. [5]SEM: Standard error of means. Source: Research Data (2019).

The consumers were unable to distinguish ($P > 0.05$) differences in the taste, texture, and overall acceptability between the fish meat ball despite the different natural antioxidante. The effects of treatment on consumer sensory scores are presented in Table 5. Only the attribute odor showed significant differences ($p < 0.05$) between treatments, with oregano treatment (OEO) achieving the highest scores. The same observed by Vital et al. (2018) when evaluating the sensory acceptability of fish fillet with alginate-based coating containing ginger and oregano essential oil. Consumers are always in contact with a mixture of flavors, and some foods are more associated / related to a type of seasoning, such as oregano that is present in different foods, favoring its acceptability (Du et al., 2012; Vital et al ., 2018). Different results were found by Messias et al. (2016) in his study of fish burger sensory acceptability using oregano, basil and rosemary, where no significant differences were observed for the aroma, as well as the texture and flavor variables.

Ojagh et al. (2010) and Kakaei & Shahbazi (2016) also found no significant differences for the use of cinnamon essential oil and chitosan-gelatin film incorporated with *Ziziphora clinopodioides* essential oil on the quality of refrigerated rainbow trout and sensory properties of minced trout fillet.

4. Conclusion

The use of essential oil of oregano in fish meat ball of Tambaqui improved the color attributes and provided a higher score for the aroma in the sensory attributes.

The essential oils of copaiba and oregano caused less water losses by cooking the fish meat ball.

Nutritional characteristics were not altered with the addition of copaiba and oregano essential oil.

Thus, this study demonstrated that the essential oils of copaiba and oregano can be used in food products as natural additives to replace the synthetic additive BHT in the food industry, bringing benefits and advantages to the commercialization of fish by-products such as the fish meat ball of Tambaqui.

The use of these essential oils in food, opens up numerous possibilities for future work with processed animal products for commercialization, improving quality attributes.

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