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Semi Preserved of Marine Fish, Physical-Chemical, Microbiological and Nutritional Characterization

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Abstract**—** *Fish is a food rich in nutrients and highly perishable. In this sense, finding suitable forms for its processing and conservation is essential to maintain its quality. Thus, the objective of this work is the preparation of semi-preserves based on marine fish and to analyze their physicochemical, microbiological and nutritional composition, optimizing the time for sterilization of the glasses, so that the product is free of microorganisms. Sea fish preserves have been prepared, having as cover sauces: olive oil (extra virgin), sauce with tomatoes and water with salt (in the natural). These preserves, after their elaboration, were stored at room temperature for 40 days. The microbiological results obtained, for all the samples, were considered within the standard established in the RDC number 12 of January 2, 2001, for this type of product. Where the limit values for coagulase positive Staphylococcus, Salmonella sp. and Coliforms at 45 ° C, showing that the protocols used in the present study were adequate for the commercialization of the products.*

Keywords**— Fish, preserves, processing, preservation, analysis, sterilization.**

I. INTRODUCTION

Fish are natural resources that, if well used, may serve to improve the nutritional quality of human food [1].

Fish is a food rich in nutrients but highly perishable precisely because of its nutritional quality and pH close to neutrality, which gives it greater perishability and sensitivity to chemical and microbiological reactions, compromising its shelf life, if not adequately preserved [2].

From the nutritional point of view, the fish has specific characteristics that make it a beneficial food. Among these characteristics, the following stand out: fish is rich in high quality and fast digestible proteins rich in lysine and essential amino acids and an important source of vitamins A and D. If their fats are ingested, they contain thiamine and riboflavin B1 and B2) and is a source of minerals like iron, phosphorus and calcium. Marine fish still contain iodine [3].

Fish has a high nutritional value, proteins, lipids, vitamins and minerals, which are necessary for growth.

In the last decades, there has been a significant evolution in the state of the art of processing and preserving food and, consequently, fish. Technologies such as refrigeration (dry ice and cold air), freezing, salting (wet and dry), smoking (cold and hot), canning (canning, glass and other packaging) with sterilization were cited by Cantu (1997) [4] appropriate for fish processing. Also used to preserve fresh processed foods, including fish, preserved in modified atmosphere packaging, without sterilization, Santos & Oliveira (2012) [5] reports. The evolution of drying and dehydration techniques, including fish, has been described by Wilhelm, Suter & Brusewitz (2004) [6] and more recently Jeantet et al. (2016) [7] that presented the latest techniques of refrigeration, freezing and also dehydration and drying, such as roller drying, spray drying and freeze drying. The same authors also addressed the recent techniques of food stabilization by chemical inhibition, such as the use of condoms as antifungal and antibacterial substances. They also addressed advances in preservation techniques by fermentation.

There is a growing demand for fish products that are easier to prepare. With the hectic life, many people are increasingly preferring quality products with simplicity and speed in preparation [8].

In this sense, finding suitable forms for its processing and conservation is essential to maintain its quality. The forms of fish processing and preservation have evolved

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and today there is room for the implementation of modifications that can guarantee products to lower costs, longer shelves of supermarkets and stores, without losing quality [9].

Therefore, it is important that the fish processing industry is aware of the new technologies being developed in this area, as the quality of the fish products will be influenced by each action during its processing and preparation. Complementing that, if fish processing techniques are applied in an innovative way, quality products with high nutritional value will be achieved [9].

The industry of canning of marine fish, in Brazil and also in the world, goes through many difficulties due to the scarcity of its main inputs, such as tuna and sardine [10].

Therefore, there is a need to develop technological innovations in the area of fish processing and industrialization that result in the development and elaboration of new products with other species of fish that are attractive and healthy for consumers [11].

Arraial do Cabo RJ, is a region of great fishing potential. This is due to a local phenomenon called resurgence. The phenomenon known as resurgence refers to the outcropping of deep cold water bodies due to the movement of the sea currents and their interaction with winds and coastal structures [12].

he nutrients brought by the cold waters promote the growth of the phytoplankton and zooplankton, resulting in an increase in the population of the marine organisms and favoring the intense proliferation of the local fish fauna and, consequently, the increase of the fish catch [13],[14]. Fishing in the municipality of Arraial do Cabo, Rio de Janeiro, is located on a stretch of the coast of Rio de Janeiro that is characterized by the presence of restingas, lagoons and lowlands, extending from Arraial do Cabo to Itacuruçá Island in Mangaratiba, where the capture of species of great commercial value is practiced [13].

Due to the fact that it is located in a resurgence area near the city of Cabo Frio, fishing in Arraial do Cabo has a very interesting role in the feasibility studies of new fish products [13].

The preparation of semi-preserved fish uses techniques of elaboration and sterilization that make the shelf life can be extended, with a guarantee of quality of this product. When establishing an appropriate fish processing program or protocols, one can program the industrialization activities, making them more efficient. For each species of fish, a protocol can be developed for the elaboration of healthy products. In this way, the semi-preserves become a great alternative, because they generate conditions that increase the yield of the fish, through the development of protocols adapted to each type of species and their processing and conservation.

The objective of the present study was to study three species, *Carcarhinus spp*, *Sardinella brasiliensis* and anchovy, or shellfish *Pomatomus saltatrix* [13],[15],[16]. For the development and elaboration of semi preserved marine fish.

The values of moisture, proteins, fats and ashes were evaluated in the products developed and the total count of thermotolerant coliforms, total coagulase positive Staphylococcus counts and presumptive detection of Salmonella spp in the developed products.

The working hypothesis was that it is possible to keep the fish product sterile in hermetically sealed containers and retain their physical-chemical and nutritional qualities.

II. MATERIAL AND METHODS

2.1 Experimental Procedure

An important aspect in the development of a new product is the availability of raw material and, in case of scarcity, the possibility of establishing crops. In the case of this study, 3 species of fish were recorded, with the highest production volume in the catches, according to FIPERJ [13], of the last 10 (ten) years, dog fish (*Carcarhinus spp*), sardine (*Sardinella brasiliensis*) and anchovy (*Pomatomus saltatrix*).

The fish used were purchased in a store specialized in fish and crustaceans. Sardines were used in the sliced cut, fillets of anchovies and cation places.

The methodology was adapted from Silva (2011) [17]. The fish was thawed, washed in running water, diced and seasoned. For the seasoning, 1g of fish, 5g salt, 5g dehydrated herbs and 5g of dehydrated parsley were used for each seasoning (Tables 1, 2 ,3). After seasoning, the fish were allowed to stand for 1 hour, then the fish were drained and pre-steamed for 10 minutes, for the exudation of water and fats.

The precooked fish was pre-sterilized in pre-sterilized hexagonal glasses, in the following proportion: half fish and half cover sauce (Figures 1, 2, 3). Both the fish and the cover dressing were at a temperature of 80°C, which was monitored with a laser thermometer.

The cover sauces used were: extra virgin olive oil industrialized brand La Violetera, sauce with traditional tomatoes industrialized Heinz traditional brand and water and salt (300 ml water for 12g salt). 5g of dehydrated garlic and two bay leaves were added to each glass, before the fish and cover dressing were added. 0,5 cm of ridge was left between the cover dressing and the lid. The metal-capped glasses were capped and then placed in a pan-type

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pan in a water bath where the panes were covered with up to 3 cm of water above the cap.

After sterilization, 30 minutes, the glasses remained in the container until they cooled. Afterwards, they were removed and placed inverted, with the lid down, on a white cotton cloth.

Fig.1: Semi Natural Cider Preserve.

Fig.2: Sardine Semi Preserved in Olive Oil.

Fig.3: Semi canned anchovy in sauce with tomatoes.

The glasses remain so for 24 hours to evaluate possible leaks in the lids. After 24 hours, the glasses were untapped and left on a shelf for 10 (ten) days, so that the cover dressing and seasonings were incorporated into the fish.

After that, they were taken to the laboratory for physical-chemical and microbiological analyzes. The table below summarizes the formulations (recipes) of the products developed.

Table 1: Formulation of Semi Cage Preservation

Ingredients	Net Weight
Quotation booths	1,0 kg
Water	300 ml
Fine herbs	8 g
Bay leaves	2 in each glass
Salt	12,0 g
Crushed dehydrated parsley	8,0 g
Crushed dehydrated onion	8,0 g
Minced dehydrated garlic	8,0 g
Water and salt at 10%	1,5 L
Yield	5 glasses

Source: Author

Table 2: Formulation of Semi Preserved Anchovies

Ingredients	Net Weight
Anchovy fillets	1,0 kg
Water	300 ml
Fine herbs	8 g
Bay leaves	2 in each glass
Salt	12,0 g
Crushed dehydrated parsley	8,0 g
Crushed dehydrated onion	8,0 g
Minced dehydrated garlic	8,0 g
Sauce with tomatoes	1,5 L
Yield	5 glasses

Source: Author

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Table 3: Formulation of Semi Preserved Sardines

Ingredients	Net Weight
Steaks of sardines	1,0 kg
Water	300 ml
Fine herbs	8 g
Bay leaves	2 in each glass
Salt	12,0 g
Crushed dehydrated parsley	8,0 g
Crushed dehydrated onion	8,0 g
Minced dehydrated garlic	8,0 g
Olive oil (liquid cover)	1,5 L
Yield	5 glasses

Source: Author

For all formulations, the same experimental procedure was used, as described above.

2.2 Physical and Microbiological Physical Analyzes

2.2.1 Preparation of Samples

For each species of fish, 50g aliquots were separated, with 4 samples of each one, both fresh fish and developed (ready) products.

The physico-chemical and microbiological analyzes were performed in Food Analysis Laboratory, in triplicate, using the same standards used by Ribeiro et al. (2009) [18].

The physical and chemical analyzes for fresh fish and semi-preserved fish were carried out following the methodology of the Adolfo Lutz Institute. The moisture, ash, protein and lipid contents were determined [19].

Microbiological analyzes followed the following methodologies: M6-AFNOR-3M 01 / 2-09 / 89C, for Total Count of Thermotolerant Coliform at 45 ° C. M12A-ISO 6888-1: 1999, for Total Positive Coagulase Staphylococcus Count at 35 ° C + -1 ° C. M26- AOAC - 2011.03, for the presumptive detection of Salmonella spp, following what is recommended by the legislation of ANVISA and MAPA, according to DRC number 12 of January of 2001 [20].

III. RESULTS

The results of the centesimal composition for the in natura fish species (anchovy, dog fish and sardine) showed that the average of the results for the moisture content was around 77,95%, while the crude protein contents were on average 19,19%, for the lipid contents were on average 2,58% and for the ash content was 6,23%.

Table 4: Centesimal Characterization of Fish Type: Anchovy, Dog fish and Sardine in natura (wet weight or natural matter).

Variables (%)	Product		
	Anchovy	Dog fish	Sardine
Humidity	75,89±25,34	80,09±21,55	77,89±23,20
Crude protein	18,56±2,17	20,67±1,06	18,35±2,70
Lipids	3,15±5,40	0,24±21,45	3,35±6,04
Ashes	6,68±4,83	4,45±7,68	7,57±4,77

* * Results of Mean (%) and Standard Deviation.

The results observed, for moisture content in the semi-preserved anchovy and dog fish samples, were equivalent (Table 4). Although the pre-cooking exuded fat and water from the fish meat, after it was placed in the cover dressing and left on the shelf in quarantine, before the analysis was carried out, fish meat incorporated water from the cover dressing, reestablishing the content of moisture. For the sardine the same pre-baking processes were done for the exudation of fats and water, but during the quarantine in the olive oil it incorporated olive oil and not water, therefore the result of its moisture content was lower than the semi canned anchovies and cacao.

The results of the crude protein levels varied from 19,67% to 13,59%, not having a very significant distance between them.

As for lipid contents, the results were quite different: 9,11% for anchovy, 1,01% for dog fish and 65,02% for sardine samples.

For the results of the ash content, the anchovy obtained 27,82% and the dog fish 28,58%, while the results for the sardine were around 10,12% (Table 5).

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Table 5: Centesimal Characterization of Semi-preserved Fish Type: Anchovy (in tomato sauce), Dog Fish (in salt and water) and Sardine (in olive oil) (wet weight or natural matter).

Variables (%)	Product		
	Anchovy	Dog fish	Sardine
Humidity	83,68±17,50	85,52±15,64	36,06±12,88
Crude Protein	19,67±5,08	13,59±7,35	16,90±5,91
Lipids	9,11±2,54	1,01±2,77	46,97±6,20
Ashes	4,87±1,67	4,47±14,68	7,29±26,57

** Results of Mean (%) and Standard Deviation.

The results of the microbiological analyzes (Table 6) were satisfactory and within the standards recommended by ANVISA (National Agency of Sanitary Surveillance), confirming that it is possible to efficiently process and preserve these products based on the protocols of processing and elaboration of the products, as they were elaborated in this study [20].

If the samples analyzed presented satisfactory results, the food being in compliance with ANVISA standards, the fish meets the national standards indicated for microbiological quality, maintaining the commercial sterility of the product [18].

Table 6: Microbiological Analysis of Canned Fish Type: Anchovy, Cacao and Sardine.

Analyze	Samples			Legislation (ANVISA, 2001)
	Dog fish	Sardine	Anchovy	
*Salmonella (*Ausência) *	Absent	Absent	Absent	Absence
Staphylococcus Positive Coagulase(UFC/g)**	$<1,0x10^1$	$<1,0x10^1$	$<1,0x10^1$	Max 10^3g^{-1}
Coliforms at 45°C (NMP/g)***	$<1,0x10^1$	$<1,0x10^1$	$<1,0x10^1$	Max 10^3g^{-1}

Results: * Absence; ** UFC - Colony Forming Unit; *** NMP - Most likely number.

The Port Authority Order No. 37, dated February 14, 2011, describes the quality and identification of canned fish, as well as the norms and requirements required for its processing [21].

In the results of the nutritional value table (Table 7), we can observe that the values of the protein contents remained very close. The results for carbohydrates were higher for dog fish and anchovy around 30% and 10% for sardines. For lipid contents, the results for the cacao were around 1%, due to the exudation of water and fats in their pre-baking and because their coating liquid was made with water and salt. The same occurred for the anchovy, which had as a cover liquid sauce with tomatoes.

As for the sardine samples, although the pre-cooking had exuded water and lipids, as its cover sauce was olive oil and the ready-made samples of the preserved half-lives awaited the 40 days of quarantine to be elaborated the analyzes, with that time the pre-cooked sardine meat incorporated well its sauce cover. Therefore, its result was around 65,02%, slightly higher than the other canned samples observed. This was due to the difference in the composition of the cover sauces for each type of fish evaluated.

Table 7: Nutritional value of semi-preserved fish type: Anchovy (in tomato sauce), Cação (in water and salt) and Sardine (in olive oil).

Variables	Nutritional value per serving (40g)		
	Dog fish	Sardine	Anchovy
Protein	13,59	16,90	15,32
Carbohydrate	30,90	10,22	32,11
Lipid	1,01	65,02	10,54
Minerals	28,58	10,12	32,54
Kcal	40,83	461,38	67,10
Humidity	84,36	27,75	77,60

Source: Author

IV. DISCUSSION

The present work obtained results for fresh fish samples, values of moisture contents, proteins, lipids and ashes, similar to those of the finished product, showing a good recovery of these parameters after the treatments. The results found in this study were equivalent to the results for the dog fish and sardines, according to the Table TACO (Brazilian Food Composition Table), which is 81,4% for the cacao and 76,6% for the sardine [22]. For anchovy, the value was higher than that found by Gonçalves & Prentice-Hernández (1998) [23], which was 69.38%.

In the study conducted by Cozer et al. (2014) [24], for canned jundiá, in tomato sauce it obtained values of 83,07% of humidity, 60,15% of crude protein. And in the results of microbiological analyzes, the results have remained within the standards required by current legislation. In this way, the results of Cozer et al. (2014)[24] were similar to those found in the present study.

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When we compare the fish in natura with the semi-preserved (ready-to-eat) product, it is observed that the protein content decreases drastically, which can be explained by the decrease of the proteins in the elaborated preserves, due to the pre-baking, where a exudation of water and fats, thus losing part of the original proteins of the flesh in natura.

The fish musculature has about 60 to 85% moisture [25]. Showing that the results for moisture in this study are within that average observed by these authors above. The muscle of fish of low commercial value such as acari (*Liposarcus pardalis*) and tamuatá (*Callichthys callichthys*), possesses about 70% of proteins and has excellent acceptability [26]. The results of the centesimal composition of the work of Silva Junior et al. (2017) [27] for fish protein concentrate resulted in the following mean values, for moisture 13,61%, for proteins 66,7%, for lipids 9,58% and for 10,2% ashes. The results showed that the results were much lower than those described in the present study. As regards protein results, the lipid results were the same and the ash contents were close.

V. CONCLUSION

The results of the microbiological analyzes were satisfactory and within the standards recommended by ANVISA. The results for the analysis of centesimal composition were also equivalent to the results observed by other authors. Confirming that it is possible to process and preserve these products efficiently, based on the protocols of processing and elaboration of the products, as they were done in this study, which proved the initial hypothesis of work.

We can observe that the development of semi-preserved marine fish can serve to increase the value added of fish to natural. Contribute to an increase in fishing, by using several species of fish, only making changes in seasoning, depending on the type of fish meat, due to the type of fibers and how that meat will absorb the seasoning. Therefore, future studies are recommended. In addition to this technological knowledge, it is necessary to improve fishing management, making it more sustainable, so that a constant supply of raw material (fish) is available at competitive prices. In this way, the diversification of a line of fish products can be made effective, without risk of lack of products due to overfishing.

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