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Quality Changes during Frozen Storage of Hot Smoked Fillets and Spreads Processed from Indian Mackerel (*Rastrelliger kanagurta*) and Pangasius (*Pangasius hypophthalmus*) Fish

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ABSTRACT

Indian mackerel and *Pangasius* fish fillets were both subjected to a hot smoking procedure, and their products were used in the spread preparation. Proximate analysis, physicochemical, microbial and sensory properties were monitored before and after frozen storage for three months. The fat content (12.1%) and nutritional value (215.3 Kcal/100g) of the smoked *Pangasius* fillets were more than those recorded for smoked Indian mackerel fillets (2.5% & 144.1 Kcal/100g, respectively). During frozen storage of smoked Indian mackerel and *Pangasius* fillets, moisture content was reduced to 61.25 and 51.8%, while an increment was noticed in salt content (2.73 & 2.45%), pH (6.7 & 6.82), TVB-N (24.94 & 18.55 mgN/100g), TBA (2.4 & 2.98 mgMDA/kg), TPC counts (2.5×10^3 & 5.3×10^3 CFU/g), and yeast and mold counts (0.97×10^2 & 1.3×10^2 CFU/g), respectively. Sensory scores for smoked Indian mackerel and *Pangasius* fillets ranged between very good and good, and the overall acceptability scores became 38 and 35, respectively, at the end of frozen storage period. By adding feta cheese and ketchup to produce Indian mackerel and *Pangasius* spreads, the protein content decreased to 21.5 and 19.8%, while carbohydrates (10.2 & 8.5%), fat (4.1 & 13.3%) and nutritional value (163.7 & 232.9 Kcal/100g) increased, respectively. At the end of frozen storage, moisture content decreased to 58.08 and 50.97%, while salt content (3.81 & 2.85%), pH (5.84 & 6.72), TVB-N (33.44 & 23.91 mg N/100g), TBA (2.03 & 2.67 mg MDA/kg), TV counts (7.9×10^3 & 9.2×10^3 CFU/g), and yeast and mold counts (3.1×10^2 & 3.8×10^2 CFU/g) increased, respectively. Sensory scores for the Indian mackerel and *Pangasius* spreads ranged between very good and good, and the overall acceptability scores became 39 and 37, respectively. The quality of hot smoked fillets and spreads coincides with Egyptian standards for smoked products.

INTRODUCTION

Fish and fishery products are a valuable source of proteins, lipids, essential micronutrients and minerals in many societies. Fish can be consumed fresh, cooked, grilled or in different product forms such as salted, smoked, cured, canned, or as a

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component of ready-to-eat products. Fresh fish accounts for 40% of all fish marketed for human consumption, while frozen, canned, and cured products account for 32, 16, and 12%, respectively (**Ababouch, 2006**; **Amagliani *et al.*, 2012**).

Egypt's overall fisheries and aquaculture production was estimated by the **FAO (2022)** to be 1.59 MT (million tons) on average in 2020 and is predicted to be 1.9 MT in 2030. Fisheries and aquaculture in Egypt play an important role in contributing to the national food supply. Fisheries are vital as a source of animal protein for the majority of poor families in a developing country such as Egypt, accounting for almost 20% of their animal protein intake (**Samy-Kamal, 2015**, **2021**). Fish production in Egypt comes mostly from aquaculture (about 80%), with declining wild fisheries representing only about 20%. Egypt has the largest aquaculture industry in Africa. The total market value of this industry was US$ 2.2 billion in 2015. The Egyptian aquaculture currently provides almost 79% of the country's fish needs, with almost all the output coming from small and medium-sized privately owned farms (**FAO, 2020**).

According to **Shehata and Eldali (2022)**, the average annual per capita consumption of fish grew from approximately 15.8kg in 2001 to almost 21kg in 2018. In Egypt, exploiting fish resources to produce fish products desired by the Egyptian consumer will enhance the chances of maximizing fish production, reducing the import of fish from abroad and achieving food security.

Rastrelliger kanagurta, an Indian mackerel species that live in the marine environment and have a delicious taste; belongs to the family *Scombridae* and has been a target of purse seine fishing in the Egyptian Red Sea, primarily in the Gulf of Suez, Hurghada, and Foul Bay. The Indian mackerel catch grew from 1.283 thousand tons in 2009 to 2.804 thousand tons in 2018 in the Red Sea, Egypt (**GAFRD, 2020**). From a socioeconomic standpoint, the Indian mackerel market prices are rather cheap for low-income households, offering an alternative seafood species to cultivated species (**Samy-Kamal, 2021**).

Freshwater fish from the Pangasiidae family called the *Pangasius* catfish (*Pangasius hypophthalmus*) is regarded as one of the most productive aquaculture species in several nations (**Rahman & Ali, 2012; Sokamte *et al.*, 2020**). In Egypt, this species is commonly known as "Basa" and is underutilized despite its firm texture, white flesh, easily digestible protein and numerous beneficial nutrients (**Økland *et al.*, 2005; Orban *et al.*, 2008**).

Smoking is an old process that includes exposing food to smoke produced by the combustion of wood (**Knockaert, 2002**). The reduction in the water activity of the smoked product and the antioxidant and antibacterial smoke components that permeate the items are what give this method its preservation effect (**Abdel-Naeem *et al.*, 2021**). This suggests the production of smoked fillets with high moisture content (60–65%) and a relatively low content of sodium chloride (2.5–3.5%) and total phenols, which is inadequate to keep the stability of the smoked product, with the result that its flavor characteristics are lost early during storage due to microbial growth and lipid oxidation (**Rørvik, 2000; Dauphin *et al.*, 2001; Neunlist *et al.*, 2005**). Lipid oxidation may have a detrimental effect on the smoked product's sensory and commercial aspects (poor odor) as well as its nutritional worth (loss of unsaturated fatty acids). Because the consumption of the final product frequently does not require further processing, microbial contamination can result in a product that is harmful to human consumption, as well as causing both physical and financial losses (**Ordóñez *et al.*, 1999; Basti *et al.*, 2006**).

Recently, there has been a lot of opportunity to improve fish-based goods and raise fish consumption. According to social and cultural developments, a variety of creative value-added ready-to-eat, "convenient" fish products are made, and their demand grows (**Rathod & Pagarkar, 2013**). By using various processing techniques like the smoking process, or adding specialized ingredients like fish spread products, one can add value to fish products by enhancing their organoleptic qualities and nutritional value in addition to their convenience and shelf life (**Rathod *et al.*, 2018**).

Fish consumption is beneficial to human health. It is a nutritionally valuable product with high protein content; it also contains valuable fatty acids (such as omega-3 fatty acids), minerals and vitamins (**Mohanty *et al.*, 2019**). Many factors such as fat contents, the temperature during processing, and processing methods can affect the quality of fish products (**Silva *et al.*, 1993**). The purpose of this study was to increase the value of two varieties of fish, the Indian mackerel and *Pangasius*, using hot smoking and employing the resultant smoked fillets in the processing of spread products. These products' nutritional values and quality indices were monitored for three months under proper storage circumstances.

MATERIALS AND METHODS

Materials

Fresh marine Indian mackerel (*Rastrelliger kangurta*) fish, which are locally called cascombry, were selected with an average length of 26cm and a weight of 150g for the present study. Fish specimens were bought in November 2020 from El-Ansary Fish Local Market of the Suez Governorate in Egypt. In the raceway unit of the college of fish resources at Suez University, freshwater *Pangasius hypophthalmus*, with an average length of 30cm and weight ranging from 0.950 to 1.200kg were used. To the laboratory of the fish processing unit belonging to the college of fish resources at Suez University, 25 kilograms of each variety of fish were brought in polystyrene boxes containing crushed ice.

Other ingredients including feta cheese (Abour Land Company), ketchup (28% TSS, Heinz Company), refined salt (El-Nasr Salines Company), sawdust of white hardwood were used to generate smoke in addition to polyethylene packing bags and glass jars, with capacity of 50 g and airtight lids were all bought from the Suez commercial market in Egypt.

Technological process

Hot smoking process

As shown in Fig. (1), the hot smoked fillets were prepared. With the use of a sharp knife, the fresh fish (Indian mackerel and Pangasius) were hand filleted. The fillets were brined in a 15% brine solution for 10 minutes, and then they were let to drain for 10 minutes over the trolley racks in the electric smoking kiln. Three stages composed the directed smoking regime inside the kiln; the first was a drying stage at 30°C for 60min with a mild amount of smoke, and the second was a cooking stage at 55°C for 60min with a medium amount of smoke. Condensed smoking was the last step, which took place at 85°C for 60min, while producing a lot of smoke. The hot smoked fillets were then allowed to cool at room temperature for 20 minutes before being frozen in an air blast freezer for about an hour at -25°C. The vacuum-sealed frozen smoked fillets were kept at a temperature of -20°C until analysis was done.

Spread products

To produce spread products, smoked fish fillets (84.7%) were minced twice, and then feta cheese (8.5%) and ketchup (6.8%) were added (Fig.1). The hand blender was then run at high speed for 5 minutes. The spread products were tightly sealed in little glass jars and kept in the freezer at -20°C until examination.

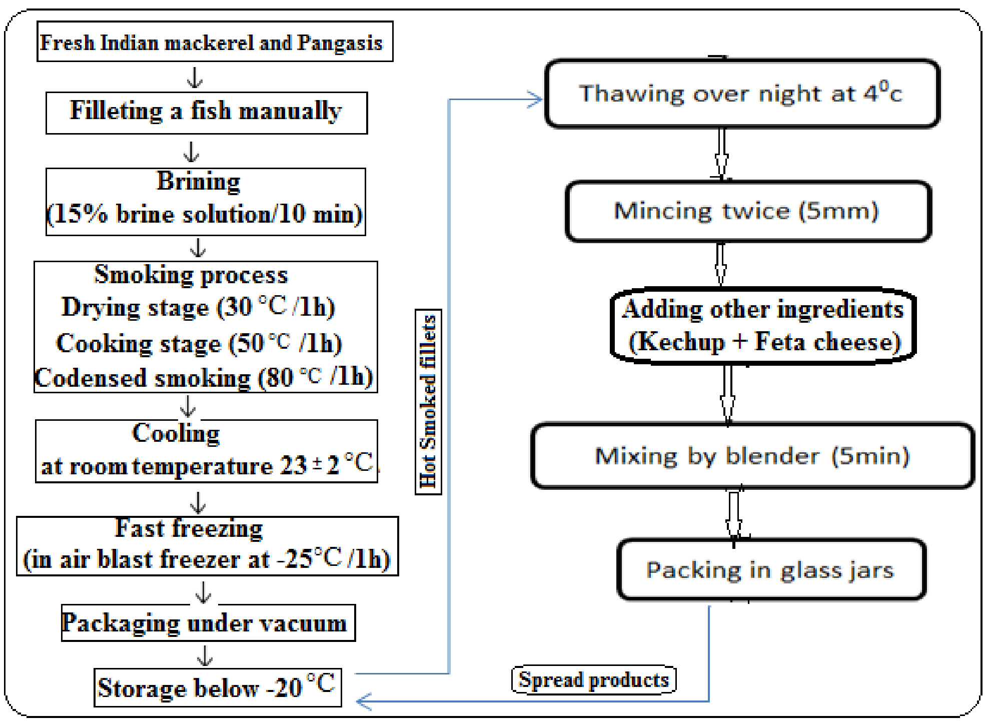

Fig.1. Layout of hot smoking fillets and spread products process

Analytical methods

Proximate composition

According to **AOAC (2000)**, the weight lost during the drying process of 5g of the minced sample at 105°C for 4h was used to estimate the moisture content. The Khjeldahl method was applied to determine the protein content (**ISO 5983-2: 2009**). Following the method of **Bligh and Dyer (1959)**, total lipids (TL) were isolated from samples (25g) using a solvent composed of methanol/chloroform/0.88% KCl (at 1/1/0.5, v/v/v). The ash and sodium chloride contents (NaCl) were determined according to **AOAC (2000)**. The difference was used to compute carbohydrates.

Physicochemical properties analysis

Thiobarbituric acid (TBA) value. TBA value was assessed as noted by **Pearson (1976)**. The process involved 35 minutes of simmering water bath boiling for the extracted sample (10g) and blank with TBA reagent. The optical density at 538nm was detected using the T60 UV-Visible Spectrophotometer after cooling, and the TBA number was calculated as follows:

TBA number (as mg malonaldehyde/ kg sample) = 7.8 × optical density

Total volatile basic nitrogen (TVB-N). The TVB-N content was determined according to **Antonacopoulos and Vyncke (1989)** distillation procedure, with some modifications. In the presence of tashiro indicator, the produced distillate from 10g sample was titrated with 0.1 N HCl till the neutral point. The titration value was converted to mg of TVBN/100 g of muscle using the following equation:

TVBN (mg)/100 g = ml 0.1 N HCl × 1.4 × 100 / weight of sample

pH value. pH values were examined using a calibrated pH meter (OHAUS STARTER 2100 Bench pH meter, OHAUS Instruments, USA) after homogenizing 10g of macerated sample with 90ml of distilled water.

Nutritional value. Based on the outcomes of their proximate analyses, the caloric values of smoked fish fillets and spread products of both Indian mackerel and Pangasius fish were estimated using the following equation as specified by **Falch *et al.* (2010)**.

Nutritional value (kcal/100g) = [(%protein× 4) + (%carbohydrate× 4) + (%lipid× 9)]

Microbiological examination

Samples preparation. Using a sterile electronic mixer, the samples of both raw and prepared fish products were aseptically ground. Serial dilutions were made using a representative amount of each product (5g), mixed with 45 ml of peptone water (0.1%).

Total plate count (TPC). The mesophilic bacteria were counted using Plate Count Agar (MPN, UK) media in accordance with the spread plate method of **Buck and Cleverdon (1960)**. For the bacterial total count, the incubation period and temperature was 35 ± 2°C for 24 hours (**APHA, 2001**). Colony forming units (CFU/g) were calculated using plates with between 30 and 300 colonies.

Yeasts and molds. On Potato Dextrose Agar media (lab M, UK), yeasts and molds were counted. To analyze the samples, 3-5 days of incubation at 25 ± 2°C were used and the results expressed as CFU/g of the plates with 10-150 colonies.

Sensory attributes

The appearance, odor, flavor, texture, color and taste of smoked fish fillets and spread products of both Indian mackerel and *Pangasius* were assessed by fifteen trained panelists from Suez University's Faculty of Fish Resources' Fish Processing and Technology Department. A nine-point hedonic scale was used for the evaluation, with excellent equaling nine and bad or unsatisfactory equaling one (**Muzaddadi, 2013**). The mean value was used to express each sensory attribute's score. The sum of the mean values for appearance, odor, flavor, texture, color and taste was taken to represent overall acceptability.

Statistical analysis

Using IBM SPSS Statistics version 20, a statistical analysis of all data was performed. All data were expressed as means ± standard deviation (SD). The analysis of variance (ANOVA) and Duncan's multiple range test were used to detect significant differences between samples. Significant differences were defined at $P< 0.05$.

RESULTS AND DISCUSSION

Proximate composition and nutritional values

Table (1) displays the chemical-nutritional content of raw, smoked fillets and spread products of both Indian mackerel as lean fish and *Pangasius* as fatty fish.

Fresh and hot smoked fish

The moisture, protein, fat, ash, carbohydrates and nutritional values before and after hot smoking of both fresh raw Indian mackerel and *Pangasius* fish were 74.4 and 63.2; 17.8 and 25.1; 3.0 and 2.5; 1.26 and 3.9; 2.54 and 5.3 g/ 100g; 108.36 and 144.1 K.Cal/100g for Indian mackerel raw and smoked fillets, respectively, and 69.4 and 57.2; 15.3 and 22.1; 12.9 and 12.1; 1.2 and 4.1; 1.2 and 4.5 g/ 100g; 182.1 and 215.3 K.Cal/100g for *Pangasius* raw and smoked fillets, respectively. There was a significant decrease in moisture and a slight decrease in fat, while protein, ash and carbohydrate contents showed a significant increase in both smoked fish which led to an increase in its nutritional value, especially with *Pangasius*. The short time of brining and cooking loss due to the rising temperature up to 80°C during the hot smoking process may be responsible for these changes. These findings resemble those of **Sokamte *et al.* (2020)**, who examined smoked *Pangasius hypophthalmus* fillets and opposed to those of **Hanumanthappa and Chandrasekhar (1987)** who reported that, the moisture, protein, fat and ash contents of smoked Indian mackerel were 44.88, 35.28, 13.72, and 7.0%, respectively, due to dry salting for 3h and smoking at 70 ± 5°C for 5h.

Spread products

Spreads with smoked fish are delicious, thus value- added products were developed from smoked fish fillets of Indian mackerel and *Pangasius*. The proximal composition of spread products of Indian mackerel and *Pangasius* were moisture (63.5 and 54.1), protein (21.5 and 19.8), fat (4.1 and 13.3), ash (4.0 and 4.3), carbohydrates (10.2 and 8.5 g/ 100g) and nutritional value (163.7 and 232.9 K.Cal/100g) as presented in

Table (1). Smoked *Pangasius* fillets as oily fish were simpler to form an excellent spread product than smoked Indian mackerel fillets since the latter has less fat. Adding feta cheese and ketchup as ingredients in fish spread products caused increasing the carbohydrates and fat while decreasing the protein content, which led to an increase in the nutritional values more than smoked fillets. **Khater and Farag (2016)** found that, the contents of moisture, protein, lipid, and ash of Salmon, Herring, and anchovy pastes were 29.1 to 31.35%; 47.99 to 52.59%; 3.76 to 4.34% and 2.9 to 3.05%; respectively. These results are completely different from our findings due to the difference in the added ingredients and the processing methods.

Table 1. Proximate composition (g/ 100g wet bases) and nutritional values (K.Cal/ 100g) of raw, smoked fillets and spread products of Indian mackerel and *Pangasius* fish

sample		Moisture	Protein	Fat	Ash	Carbohydrates	Nutritional value
Indian mackerel	Raw	74.40±0.38[a]	17.80±0.29[c]	3.00±0.23[b]	1.26±0.41[b]	2.54±0.25[c]	108.36
	Smoked fillets	63.20±0.27[b]	25.10±0.42[a]	2.50±0.22[b]	3.90±0.32[a]	5.30±0.23[b]	144.10
	Spread	63.50±0.1[c]	21.50±0.22[b]	4.10±0.41[a]	4.00±0.24[a]	10.20±0.32[a]	163.70
Pangasius	Raw	69.40±0.25[a]	15.30±0.23[c]	12.90±0.19[a]	1.20±0.37[b]	1.20±0.43[c]	182.10
	Smoked fillets	57.20±0.19[b]	22.10±0.25[a]	12.10±0.32[b]	4.10±0.43[a]	4.50±0.52[b]	215.30
	Spread	54.10±0.41[c]	19.80±0.46[b]	13.30±0.26[a]	4.30±0.31[a]	8.50±0.36[a]	232.90

a, b, c values in each column (for each fish) with different super- scripts are significantly different ($P< 0.05$); Data are Mean ± SD

Changes in physicochemical properties of Indian mackerel and *Pangasius* smoked fillets and spread products during frozen storage

To assess the quality of smoked fillets and spread products of both Indian mackerel and *Pangasius* fish during three months of frozen storage at -20°C, several physicochemical parameters, such as moisture, salt contents, pH value, TVB-N content, and TBA value were monthly examined (Figs. 2- 11).

Moisture content

Numerous biochemical processes and physiological changes that may impact the stability and quality of fish depend on the moisture content of the fish. The hot smoking process caused a decrease in moisture content from 74.4 in fresh Indian mackerel to 63.21g/ 100g and from 69.4 in fresh *Pangasius* to 57.25g/ 100g. After three months of storage under -20°C, a slight decrease of 3.1% in moisture content was observed in smoked Indian mackerel, and its value became 61.25 g/ 100g, while a moderate decrease of 9.52% was showed in smoked *Pangasius*, reaching a value of 51.8g/ 100g (Fig 2).

Sokamte *et al.* (2020) observed that, the hot smoking process induced a reduction (20.1%) in moisture content of *Pangasius hypophthalmus* from 77.32 % before smoking to 61.79 %.

In addition, the moisture contents of spread Indian mackerel and *Pangasius* slightly decreased to 60.23 and 54.13g/ 100g, respectively, after adding feta cheese and ketchup to smoked fish, reaching 58.08 and 50.97g/ 100g after frozen storage for three months (Fig. 3).

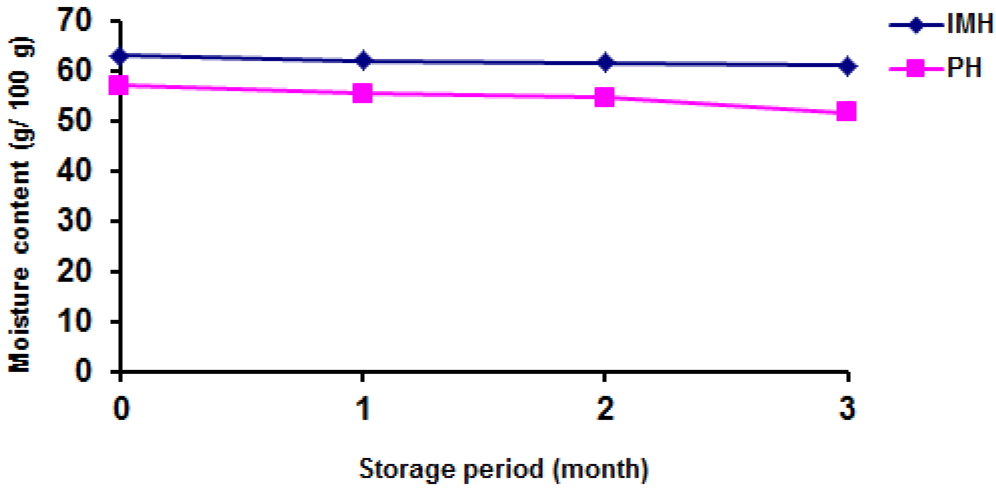

Fig. 2. Changes in moisture content (g/ 100g) of both hot smoked Indian mackerel (IMH) and *Pangasius* (PH) fillets during 3 months of frozen storage (-20 ºC). (Results are mean ± standard deviation).

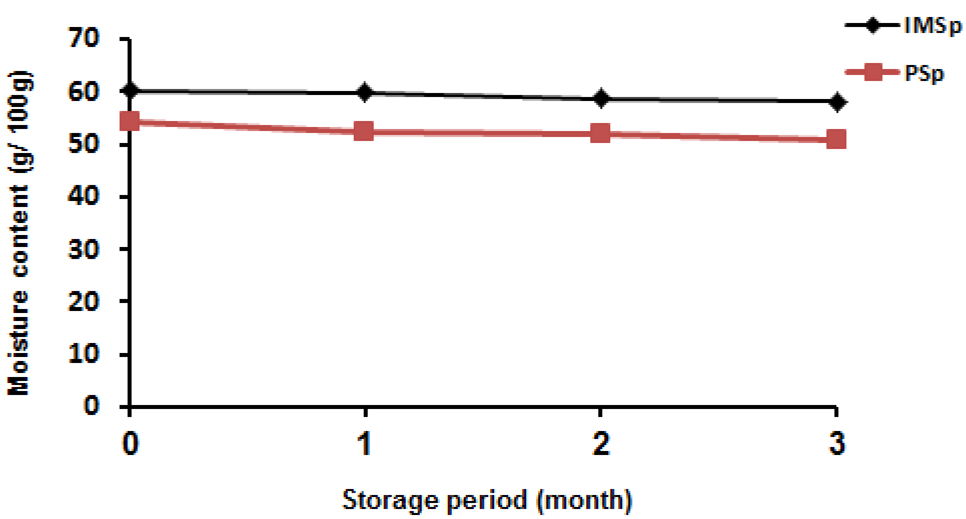

Fig. 3. Changes in moisture content (g/ 100g) in spread products of both Indian mackerel (IMSp) and *Pangasius* (PSp) during 3 months of frozen storage (-20 ºC). Results are mean ± standard deviation.

Salt content

As shown in Fig. (4), the salt contents of hot smoked Indian mackerel and *Pangasius* fish fillets increased from 0.28 to 2.45g/ 100g and from 0.19 to 2.01g/ 100g in raw and smoked fillets, respectively, due to the brining step before smoking process and the reduction in moisture contents after the hot smoking process. The findings of **Sokamte *et al.* (2020)** showed that, salt concentrations in raw and hot smoked *Pangasius* were 0.32 and 3.09%, respectively, which were consistent with this investigation. Additionally, due to a drop in moisture levels, the minor increase in NaCl content persisted throughout the period of three months of frozen storage and reached 2.73g/ 100g in hot smoked Indian mackerel fillets and 2.27 g/ 100g in hot smoked *Pangasius* fillets.

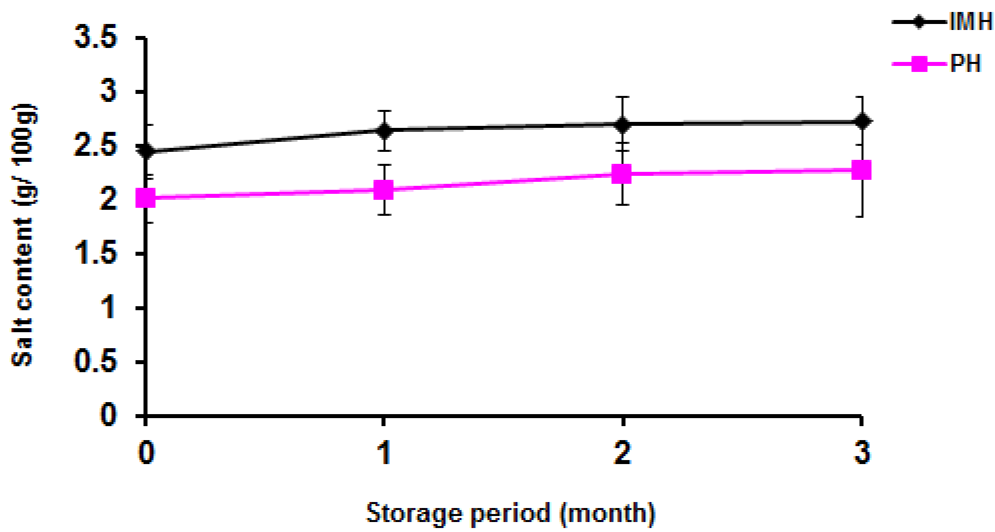

Fig. 4. Changes in salt content (g/ 100g) of both hot smoked Indian mackerel (IMH) and *Pangasius* (PH) fillets during 3 months of frozen storage (-20 ºC). Results are mean ± standard deviation

Regarding the spread Indian mackerel and *Pangasius* products, they contained the highest salt contents at zero time and after three months of frozen storage, which recorded 3.54; 3.81 and 2.79; 2.85 g/ 100g, respectively (Fig 5). **Khater and Farag (2016)** found that salmon, herring and anchovy paste samples had NaCl content ranging from 1.47 to 1.69 %.

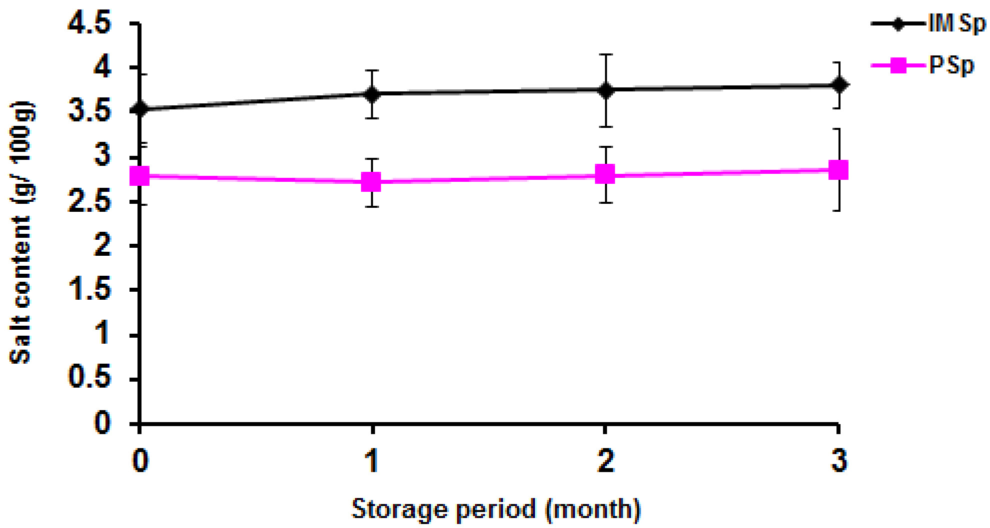

Fig. 5. Changes in salt content (g/ 100g) in spread products of both Indian mackerel (IMSp) and *Pangasius* (PSp) during 3 months of frozen storage (-20 ºC). Results are mean ± standard deviation.

pH value

The rate of food deterioration and microbial growth are both significantly influenced by the pH value of the food. The pH values of both Indian mackerel and *Pangasius* before and after hot smoking were approaching neutral pH ranging from 6.47 to 6.53 and 6.59 to 6.82, respectively (Fig. 6). The pH values somewhat decreased during the first two months of frozen storage due to acids, viz. carboxylic acids, acetic acid and others which are formed on the surface of fillets during wood burning **(Lingbeck *et al.*, 2014)**. By the end of the 3rd month of frozen storage at -20°C, the pH values reverted to a level approaching neutral due to an increase in volatile amines. **Leksono *et al.* (2014)** found that the pH of fresh *Pangasius hypophthalmus* was close to 7, and after hot smoking, it decreased to range between 6.62- 6.66.

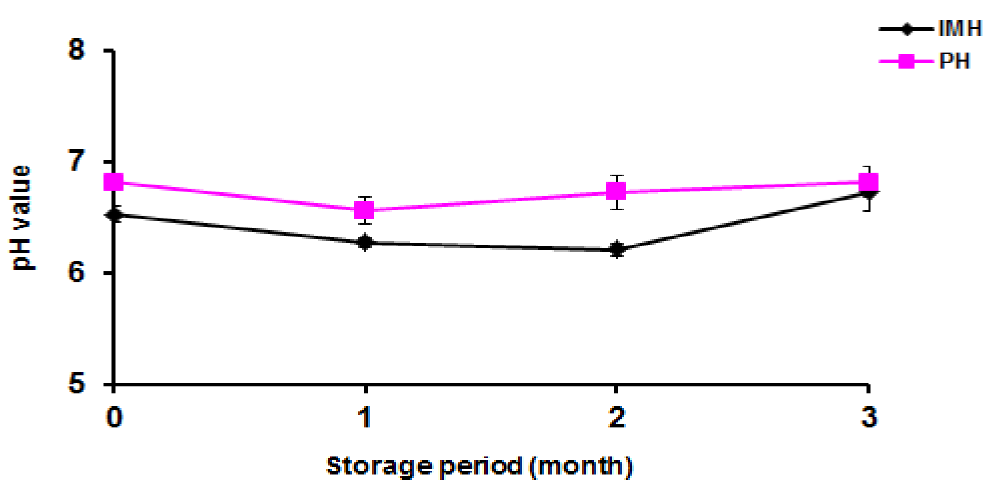

Fig. 6. Changes in pH value of both hot smoked Indian mackerel (IMH) and *Pangasius* (PH) fillets during 3 months of frozen storage (-20 ºC). Results are mean ± standard deviation.

The pH values of spread Indian mackerel and *Pangasius* fish products decreased to slight acid values (6.15 and 6.21, respectively) due to ketchup adding (Fig 7). The pH values of spread Indian mackerel continued to decrease during frozen storage and reached a value of 5.84. The results match with those of **Khater and Farag (2016)** who reported that, salmon, herring, and anchovy pastes had respective mean pH values of 6.09, 5.78, and 5.64, respectively. On the other side, the pH values of spread *Pangasius* returned to increase after the second month and became 6.72 by the end of the third month of frozen storage. These findings are close to those noted in anchovy cake (6.93) in the study of **Inanli *et al.* (2011)**.

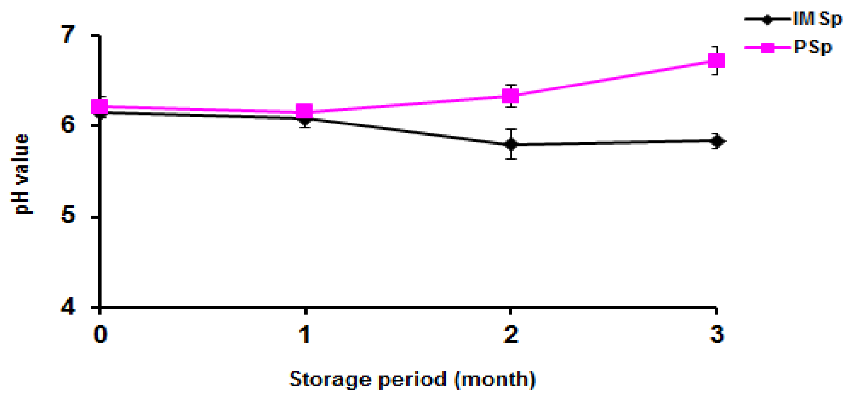

Fig. 7. Changes in pH value in spread products of both Indian mackerel (IMSp) and *Pangasius* (PSp) during 3 months of frozen storage (-20 ºC). Results are mean ± standard deviation

TVB-N content

The level of putrefaction, decomposition, and proteinase constituent breakdown in fish and fish products are assessed using total volatile basic nitrogen (TVB-N). The TVB-N contents before and after hot smoking and in spread products from either the Indian mackerel or *Pangasius* fish were determined periodically during frozen storage for three months (Figs. 8, 9). The TVB-N values in hot smoked Indian mackerel fillets increased significantly more than seven times after smoking due to the effect of high temperature (from 2.71 to 19.61mg/ 100g) (Fig 8). A gradual increase continued during the frozen storage and reached 24.94mg/ 100g, which indicates that it is within the limits of good quality according to **EOS (2005)**. These results agree with those of **Amitha *et al.* (2019)** for fresh Indian mackerel (4.57 mg/100g). Moreover, the current result concurs with that of **Hanumanthappa and Chandrasekhar (1987)** for hot smoked Indian mackerel (19.6mg/ 100g). On the contrary, the TVB-N values in hot smoked *Pangasius* fillets slightly increased from 12.97 to 16.42mg/ 100g, and this may be attributed to the fat contents which decrease the effect of heat on proteins besides the difference in the nature of proteins. During frozen storage of hot smoked *Pangasius* fillets, a progressive increase persisted and eventually reached 18.55mg/ 100g, which indicates that it is within EOS's guidelines for good quality **(EOS, 2005)**.

The mean concentrations of TVB-N in spreads of Indian mackerel and *Pangasius* were 32.08 and 21.67mg/ 100g, respectively (Fig 9). The increase in TVB-N values may be related to the feta cheese and ketchup added to formulate the spread products. During three months of frozen storage, slight increases in TVB-N values were observed in both spread products. The degradation of some amino acids can be linked to an increase in TVB-N concentration with storage (**Lou *et al*., 2021**). **Khater and Farag (2016)** found that, TVB-N values were 13.80, 13.92 and 14.36 mg/100g in Herring, Salmon and Anchovy pastes, respectively. Although the current results showed high TVB-N values, they did not surpass the acceptable limit (35mg/ 100g of fish flesh) for fish as defined by the recommendations of **EOS (2005)**.

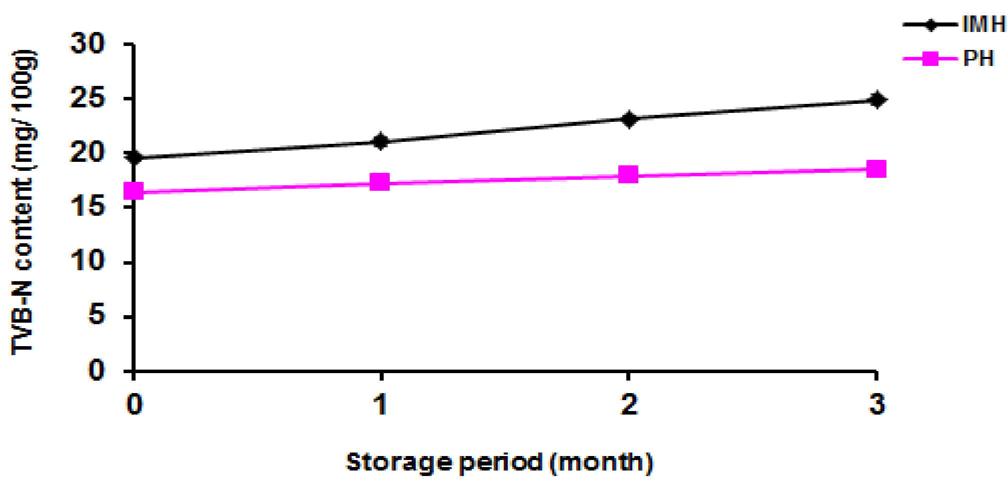

Fig. 8. Changes in TVB-N content (mg/ 100g) in both hot smoked Indian mackerel (IMH) and *Pangasius* (PH) fillets during 3 months of frozen storage (-20 ºC). Results are mean ± standard deviation.

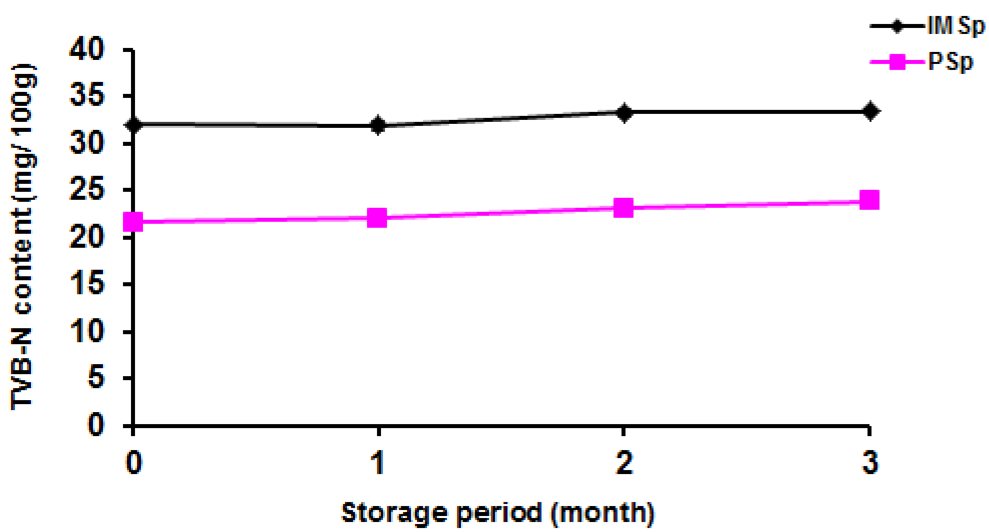

Fig. 9. Changes in TVB-N content (mg/ 100g) in spread products of both Indian mackerel (IMSp) and *Pangasius* (PSp) during 3 months of frozen storage (-20 ºC). Results are mean ± standard deviation

TBA value

The TBA test is often used to determine the degree of lipid oxidation in food products. The accumulation of secondary oxidation products, quantified as mg malonaldehyde/ kg of flesh was used to express the TBA **(Pegg, 2004)**. Chemicals causing secondary oxidation are a valuable tool for assessing the chemical changes caused by the storage process **(Ortiz *et al.*, 2013)**. As illustrated in Fig (10), TBA values before and after hot smoking of Indian mackerel (lean fish) and *Pangasius* (fatty fish) were 0.13; 1.43mg MDA/kg and 0.23; 2.01 mg MDA/kg, respectively. Exposure to heat and oxygen during hot smoking may quicken the oxidation of fish lipids leading to an increase in the TBA value. Slight increases were continued in both hot smoked fish fillets during three months of frozen storage.

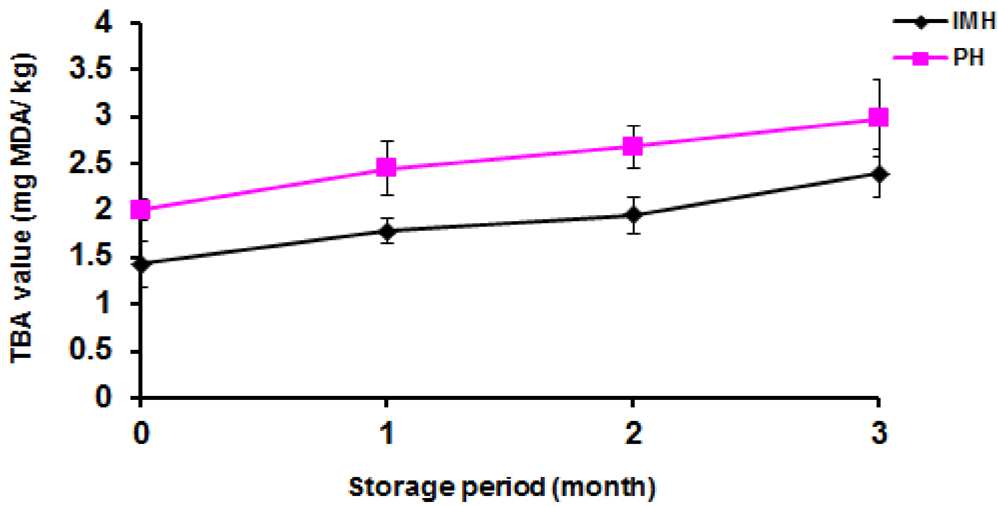

Fig. 10. Changes in TBA value (mg MDA/ kg) in both hot smoked Indian mackerel (IMH) and *Pangasius* (PH) fillets during 3 months of frozen storage (-20 ºC). Results are mean ± standard deviation

Adding feta cheese and ketchup to prepare spread products from hot smoked fish caused decreases in TBA values to reach 1.15mg MDA/kg in Indian mackerel spread and 1.49mg MDA/ kg in *Pangasius* spread (Fig 11). By the end of frozen storage time, the TBA values of spread products increased to a value of 2.03mg MDA/kg in Indian mackerel spread, reaching 2.67 mg MDA/kg in *Pangasius* spread. These results match with those of **Khater and Farag (2016)** who found that, anchovy paste contained 1.73 mg MDA/ kg of TBA; salmon paste contained 2.26mg of MDA/ kg, and herring paste contained 1.30mg of MDA/ kg. The known permissible threshold for TBA is 7.8mg of MDA/ kg. Researchers claim that the TBA number in a good material shouldn't be ≥5 and should be ≤3 in extremely good materials (**Sinnhuber, 1958; Varlik *et al.*, 1993**). Although the TBA values continued increasing throughout the whole period of frozen storage, results in this investigation indicate that all fish products fell into the category of extremely good products.

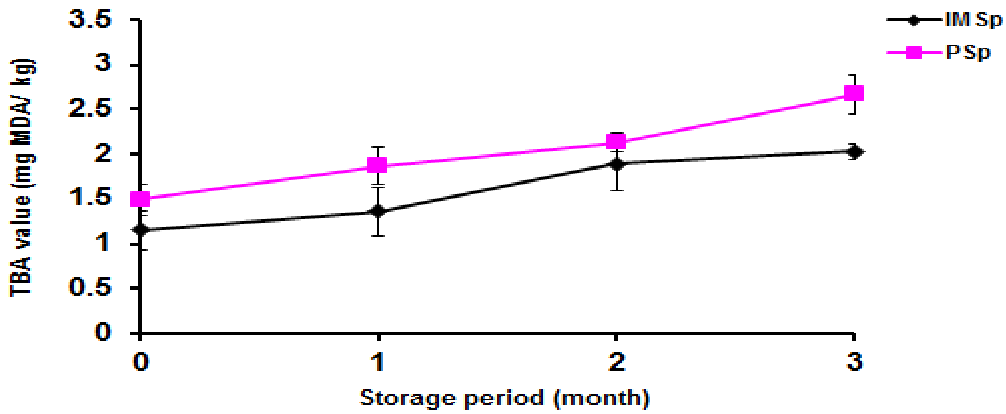

Fig. 11. Changes in TBA value (mg MDA/ kg) in spread products of both Indian mackerel (IMSp) and *Pangasius* (PSp) during 3 months of frozen storage (-20 ºC). Results are mean ± standard deviation.

3.3. Microbiological status of Indian mackerel and *Pangasius* smoked fillets and spread products during frozen storage

As represented in Table (2), despite the fact that the catch conditions for both species were different, the microbial loads in both varieties of raw Indian mackerel and *Pangasius* fish were comparable. The raw and hot smoked Indian mackerel and *Pangasius* fish fillets had total plate bacterial counts (TPC) of $3.8x10^4$ and $2.5x10^3$ CFU/g and $6.5x10^4$ and $5.3x10^3$CFU/g, respectively, while yeast and mold counts were $6.2x10^2$ and $0.97x10^2$ CFU/g and $5.2x10^3$ and $1.3x10^2$ CFU/g, respectively (Table 2). This good microbiological quality was the consequence of hot smoking techniques combined with antibacterial smoke components, which decreased moisture contents and raised salt contents (**Cyprian *et al.*, 2015**). A slight increase in microbial counts was noticed during frozen storage at -20°C, and after three months their counts reached $4.2x10^3$ and $2.3x10^2$ CFU/g and $7.3x10^3$ and $3.0x10^2$ CFU/g, respectively in both fillets of hot smoked Indian mackerel and *Pangasius*. A generation of psychrotrophic bacteria or the presence of oxygen may be the cause of microbial growth during frozen storage. These results of aerobic bacterial counts were less than the Egyptian standard (**ESO, 2005**) acceptable limit of smoked fish (10^5 CFU/g). **Hanumanthappa and Chandrasekhar (1987)** found that, the total plate count and yeast and mold of smoked Indian mackerel were 1.4 $x10^2$ CFU/g and 0.3 $x10^2$ CFU/g, respectively.

Table 2. Microbial counts (CFU/ g) of hot smoked fillets and spread products of Indian mackerel and *Pangasius* fish during three months of frozen storage (-20°C)

Treatments		TPC				Yeast and mold			
		0	1	2	3	0	1	2	3
Indian mackerel	Raw	$3.8x10^4±0.3$	ND			$6.2x10^2±0.4$	ND		
	smoked	$2.5x10^3±0.3^d$	$3.3x10^3±0.2^c$	$3.9x10^3±0.3^b$	$4.2x10^3±0.5^a$	$0.97x10^2±0.2^b$	$1.4x10^2±0.2^a$	$1.8x10^2±0.1^a$	$2.3x10^2±0.3^a$
	spread	$7.3x10^3±0.2^d$	$7.5x10^3±0.4^c$	$7.9x10^3±0.6^b$	$8.3x10^3±0.3^a$	$1.8x10^2±0.1^d$	$2.2x10^2±0.1^c$	$2.8x10^2±0.3^b$	$3.1x10^2±0.1^a$
Pangasius	Raw	$6.5x10^4±0.2$	ND			$5.2x10^3±0.3$	ND		
	smoked	$5.3x10^3±0.4^d$	$6.4x10^3±0.1^c$	$6.9x10^3±0.3^b$	$7.3x10^3±0.7^a$	$1.3x10^2±0.2^d$	$1.9x10^2±0.2^c$	$2.4x10^2±0.1^b$	$3.0x10^2±0.5^a$
	spread	$8.1x10^3±0.6^d$	$8.8x10^3±0.3^c$	$9.2x10^3±0.3^b$	$9.7x10^3±0.7^a$	$2.1x10^2±0.3^d$	$2.7x10^2±0.2^c$	$3.2x10^2±0.4^b$	$3.8x10^2±0.6^a$

a, b, c, d values in each raw with different super- scripts are significantly different ($p < 0.05$); Data are Mean ± SD. ND: not determined.

Table (2) shows that, the microbial counts of spread products increased slightly compared to the smoked fillets due to the ingredients added during spread forming. The TVC and yeasts and molds in spread Indian mackerel ranged from $7.3x10^3$ to $8.3x10^3$CFU/ g and from $1.8x10^2$ to $3.1x10^2$ CFU/g, respectively, during three months of frozen storage. Additionally, the TPC and yeasts and molds counts in spread *Pangasius* ranged from $8.1x10^3$ to $9.7x10^3$ CFU/g and from $2.1x10^2$ to $3.8x10^2$ CFU/g, respectively, during three months of frozen storage. The results confirmed the microbial keeping quality for spreads products, which were processed by mixing minced hot smoked fillets of either Indian mackerel or *Pangasius* with feta cheese and ketchup. **Khater and Farag (2016)** reported that, Salmon, Herring, and Anchovy paste samples' respective mean total aerobic counts were 5.34, 5.35, and 5.53 log CFU/g, respectively. Total aerobic counts are regarded as a quality marker, a measure of the shelf life of food samples and the potential for growth of the harmful microorganisms present, even though there is no direct association between these two factors (**Arvanitoyannis *et al.*, 2005**).

3.4. Organoleptic acceptability of Indian mackerel and Pangasius smoked fillets and spread products during frozen storage:

Table 3 displays the mean sensory evaluation scores for hot-smoked fillets and spreads produced from Indian mackerel and Pangasis fish. The panellists gave smoked fillets favourable ratings for appearance, colour, flavour, odour, and texture. For Indian mackerel, the ratings ranged from excellent (9) to very good (8), while for Pangasius, they were very good (8) to good (7). Therefore, smoked Indian mackerel fillet had a higher overall acceptance score (40) than Pangasius (38). Sensorial scores were slightly decreased during frozen storage, particularly in the colour and texture of smoked Indian mackerel, which changed to good (7) after three months of storage with an overall acceptability score of 38. After the first month of storage, the majority of the attributes of smoked Pangasius changed, and at the end of the storage period, the total acceptability score was 35. The distinctive organoleptic characteristics, higher moisture content, and lower salt concentrations in the flesh make modern smoked foods stand out today. According to **Praveen Kumar *et al.* (2022)** a decrease in sensory scores of smoked Pangasius fillets was observed with increase in refrigerated storage period, but these smoked fillets were acceptable till the end of storage (63 days). **Goulas and Kontominas (2005)** observed that after 30 days of storage at 2°C, the sensory scores of smoked chub mackerel declined.

Table 3: Sensory properties of smoked fillets and spread products of Indian mackerel and Pangasius fish during three months of frozen storage (-20 °C)

Sensory characters	Storage time	Smoked fillets		Spread	
		Indian mackerel	Pangasius	Indian mackerel	Pangasius
Appearance	0	8±0.5[a]	8±0.5[a]	8±0.7	8±0.6[a]
	1	8±0.5[a]	7±0.4[b]	8±0.6	7±0.5[b]
	2	8±0.2[b]	7±0.7[b]	8±0.5	7±0.4[b]
	3	8±0.3[b]	7±0.5[b]	8±0.4	7±0.3[b]
Color	0	8±0.5[a]	7±0.7	8±0.4	8±0.3[a]
	1	8±0.4[a]	7±0.6	8±0.3	8±0.5[a]
	2	8±0.3[a]	7±0.3	8±0.2	8±0.2[a]
	3	7±0.2[b]	7±0.2	8±0.1	7±0.1[b]
Taste	0	8±0.4	8±0.3	8±0.3	8±0.5[a]
	1	8±0.3	7±0.5	8±0.2	8±0.4[a]
	2	8±0.2	7±0.2	8±0.2	7±0.3[b]
	3	8±0.1	7±0.4	7±0.4	7±0.2[b]
Odour	0	8±0.5[a]	8±0.5[a]	8±0.2	8±0.4
	1	8±0.4[b]	8±0.5[a]	8±0.3	8±0.4
	2	8±0.3[b]	8±0.3[a]	8±0.4	8±0.2
	3	8±0.2[b]	7±0.5[b]	8±0.5	8±0.3
Texture	0	8±0.4	8±0.3[a]	8±0.6	8±0.5
	1	8±0.3	8±0.2[a]	8±0.5	8±0.4
	2	8±0.3	8±0.5[a]	8±0.4	8±0.3
	3	7±0.1	7±0.7[b]	8±0.1	8±0.1
Overall acceptability	0	40	38	40	40
	1	40	36	40	39
	2	40	36	40	38
	3	38	35	39	37

a, b, c, d values in each column with different super- scripts are significantly different ($p < 0.05$); Data are Mean ± SD.

As a result of the necessity to make a variety of products and the fact that the quality of hot smoked fillets changed during frozen storage, it was necessary to use these ingredients to produce substitute products, such spreads, to increase their shelf life and sensory qualities. The panelist′s sensorial scores of spread Indian mackerel and Pangasius varied from very good (8) to good (7). The overall acceptability of both spreads recorded

40 at the beginning of the storage period. Spread Indian mackerel was more stable than spread Pangasius during frozen storage as the overall acceptability for both spread products became 39 and 37, respectively by the end of the storage period. **Freitas *et al.* (2012)** developed two spread formulations of the Tilapia-based spread. The overall impression, appearance, and flavour of the spread produced with common salt were considerably (P≤0.05) poorer when taking into account the acceptability of all consumers. However, the spread prepared with the seasoned salt was chosen as the best, but both products were well received.

4. CONCLUSION

As a sustainable alternative to imported herring fish, which is consumed in Egypt, local Indian mackerel and Pangasius, both marine and freshwater species, could be utilized for the smoking process. Short-term hot smoking is a promising technique for achieving distinctive organoleptic qualities with low salt content and minimal moisture loss. In comparison to hot smoked Pangasius fillets, hot smoked Indian mackerel fillets exhibited higher salt, moisture, TVB-N values, and overall acceptability scores but lower nutritional value, TBA, TVC, and yeast and mold counts. After three months of frozen storage, smoked Indian mackerel and Pangasius fillets showed a slight deterioration in their qualitative characteristics.

The need to reintroduce smoked fillets of Indian mackerel and Pangasius in substitute products like spreads is highlighted by a decrease in organoleptic qualities during frozen storage, particularly in colour and texture. When mixed to make spreads, feta cheese and ketchup led to increase of TVB-N, salt content, TVC, and yeast and mold counts. According to ESO (2005), the variations in some physicochemical and microbiological counts were within permissible limits. Particularly when Pangasius was spread out and stored frozen for three months, an improvement in the sensory characteristics was observed.

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