

Changes in Biochemical Criteria of Tilapia Fish Samples during Frozen Storage at -18°C for 180 Days and their Fried Products

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Abstract

Changes in values of biochemical criteria of Tilapia samples during frozen storage for 180 days at -18°C and their fried products were studied. Quality criteria of raw samples were 6.74, 6.2 pH; 10.64, 12.04 mg/100 gm TVB-N and 0.21, 0.23 mg/kg TBA for farms A and B samples, respectively. After frying values of TBA, TVB-N and pH increased in all fried samples prepared from frozen samples throughout storage periods. TVB-N and TBA values increased with the storage periods until the end the storage, while pH value of frozen samples decreased to 6.61 in farm A frozen samples, on other side pH values were increased to 6.53 in samples of farm A after 60 days of freezing. After that the values of pH were increased until the end of storage.

Keywords: Tilapia fish; Biochemical criteria; Frying; Frozen storage

Introduction

Seafood has high nutritional value being rich in unsaturated fatty acids (omega 3), essential amino acids, minerals and vitamins and low cholesterol and saturated fatty acids [1].

In addition, fish is an important source of high-quality proteins, vitamins especially vitamin A, E and D and minerals such as potassium, sodium, calcium, magnesium, iron, copper, zinc and manganese [2,3]. However, availability of these necessary nutrients depends on the storage methods [4], such as salting, roasting, drying and freezing.

Food freezing considered preservation method was used through thousands of years because it maintains the quality of products. Freezing is a perfect method to preservation the commercial fishes because it stops chemical and microbiological degradation, and is an excellent method of preservation the sensory attributes of fish flesh during frozen storage [5].

Concern, thermal processing techniques (smoking, frying, etc.), are widely used for improving the eating and safety of food products and used to prolong shelf life of food products. There are different ways of seafood cooking to enhance the flavour and taste and to improve its safety and hygienic quality by inactivation of pathogenic microorganisms. Physical and chemical reactions take place during cooking which either improve or impair the nutritional properties of food, but fat-soluble vitamins, the contents of thermo labile compounds, or polyunsaturated fatty acids are reduced [6].

This work is aimed to determine the quality of frozen tilapia fish, and effect of frying on its quality criteria.

Materials and Methods

Fish samples

Tilapia (*O. niloticus*) fish samples (20 kg) were collected from two fish farms (A and B) located in Fayoum governorate, Egypt, during June 2016. The main resources of irrigation water were El batts drain for (A) and El-Wadi drain for (B). The averages of weight and length were 303 ± 1.5 gm. and 25.9 ± 0.22 cm for fresh samples from farm A, while they were 327 ± 93.8 g. and 26 ± 2.8 cm for farm B, respectively. Fish samples were transported to fish processing lab., national institute of oceanography and fisheries (NIOF), Egypt. Samples were washed, glazed and packed in polyethylene bags then, stored for 180 days at -18°C. Fish samples (fresh and frozen) were deep-fried in oil and every 60 days for frozen samples.

Frying oil

Sunflower oil was used in frying process, it contained: polyunsaturated fatty acid (60%); saturated fat (12.85%); monounsaturated fatty acid (27.14%).

Frying process

Tilapia fish samples were prepared and rubbed with flour for 3-4 minutes, then put in pre-heated oil at 170°C -180°C for 10-15 minutes using Electrical Fryer pan (Moulinex brand). When the golden brown color was appeared on surfaces Fried fish samples were removed. Fried samples were placed in the basket to drain out the excess amounts of oil then cooled and kept for analysis.

Analytical methods

Tilapia samples (Raw, frozen and fried) were analyzed at 0, 60, 120 and 180 days of frozen storage. The results were triplicates and expressed as mean \pm SD.

Physicochemical Parameters

pH value

pH value was determined according to Pearson et al. [7]. Five grams of fresh, frozen and fried samples were homogenized with 50 ml of distilled water, and then filtered by filter paper. pH value was measured by using digital pH meter (adwa AD 1030).

Total Volatile Basic Nitrogen (TVB-N) content

Total volatile basic nitrogen (TVB-N) content was determined by using Macro-Kjeldahl distillation apparatus as described by Pearson et al. [7] as follows: 10 g of minced sample add to 100 ml of distilled water and mix, add 200 ml distilled water, 2 g of magnesium oxide and antifoaming agent. In 500 ml receiving flask add 25 ml of boric acid solution 2% and a few drops of mixed indicator (0.1 g of methylene blue and 0.1 g of methyl red to 100 ml of ethyl alcohol), the condenser terminal should be dipped in boric acid solution. After boiling by heating, the condenser was washed with distilled water and the distillate was titrated with sulfuric acid (0.1 N). Multiply the titration (minus blank) by 14 to obtain the TVB-N as mg N per 100 g sample.

Thiobarbituric Acid (TBA) value

Thiobarbituric acid (TBA) was determined in minced fish flesh samples according to the method described by Torres-Arreola et al. [8]. 10 g of minced sample were mixed with 50 ml of distilled water for 2 minutes, 47.5 ml of distilled water was used in washing into a distillation flask, add 2.5 ml of HCL (4 N). Collect 50 ml of distillate, and pipette 5 ml from distillate into glass coppered tube and added 5 ml of thiobarbituric acid reagent then mix the mixture. Use water bath to heat the mixture to boiling the mixture for 35 minutes. After that cooling the mixture; measure the optical density against the blank using spectrophotometer at 538 nm. This method based on the spectrophotometric quotation of the pink complex formed after reaction of one molecule of malondialdehyde (MDA) with two molecules of Thiobarbituric acid (TBA). Thiobarbituric acid value was expressed as mg MA/kg sample.

Statistical analysis of results

Results were statistically analysed using SPSS software program, version 16, 2007. Mean and SD measure by LSD at 5.0% level of significant.

Results and Discussion

Biochemical quality attributes of fresh Tilapia fish

Data illustrated in Table 1 shows the biochemical quality attributes of tilapia fish samples collected from A and B farms. The values of pH, TVB-N (mg/100 gm) and TBA (mg MA/kg) were 6.74, 10.64 mg/100 g and 0.21 mg/kg for farm A samples, while these values were 6.2, 12.04 mg/100 g and 0.23 mg MA/kg for farm B, respectively. These results are on harmony with Mohamed [9,10] who found that the TVB-N of tilapia was 17.5 and El-Sherif et al. [11] who found that the TVB-N and TBA of tilapia samples were 14.31 mg/100 g and 0.55 mg MA/kg, respectively. From the previous results it could be noticed that the TVB-N, TBA and pH values in the two farms samples are less than the

spoilage values or unaccepted values reported by various authors and in the range of fish freshness values, so the tilapia fish samples used in this study were in fresh state. Abbas et al. [9] stated that pH can act as an indicator of fish freshness as pH is low at the early stages of storage when the nutritional state is still good and increases after storage for a certain period of time. Buchtova et al. [10-13] showed that pH in fresh fish muscle is ranged from 6.0 to 6.5 [14,15]. Total volatile basic nitrogen (TVB-N) levels have been recognized as useful indicators of seafood spoilage; under EU directive 95/149/EEC, the European Commission has specified that TVB-N levels be used if sensory methods raise doubts about the freshness of seafood species [16]. A maximum thiobarbituric acid value, indicating a good quality of fish, is 5 mg malonaldehyde (MA/kg), while the fish can be consumed up to a TBA value of 8 mg MA/kg [17]. The threshold of TBA values for detecting rancidity varies from 1.98 to 4.40 MA/kg in fish flesh [18].

Biochemical Criteria	Farm A	Farm B
pH value	6.74 ± 0.04	6.2 ± 0.06
TVB-N (mg/100 g)	10.64 ± 0.65	12.04 ± 0.22
TBA(mg MA\ kg)	0.21 ± 0.11	0.23 ± 0.05

Farm A=Al-Batts Drain; Farm B=El-Wadi Drain
M: Mean; SD: Standard Deviation

Table 1: Biochemical quality attributes of fresh Tilapia samples (M ± SD).

pH value

Table 2 shows the pH values tilapia fish samples stored at -18°C and their fried products. pH values decreased after 60 days of storage for farm A samples, while increased gradually after 120 and 180 days of frozen storage. pH values gradually increased from 6.2 at zero time to 7.02 after 180 days for farm B samples. After deep frying the values of pH increased from 6.76 for raw material to 6.8 after frying at zero time for farm A, on other hand farm B fresh tilapia samples have pH values 6.2 increased after deep oil frying to 6.76 at zero time. From the results it noticed that pH values of tilapia fish were high which may be due to the high in protein fraction of tilapia fish. The values of pH were 6.61 and 6.53 after 60 days for samples stored at -18°C and increased to 6.87 and 6.92 after frying for fish samples collected from A and B farms, respectively. similar results were found by Lakshmanan et al. [17] who reported a decline in pH value after 36 weeks in whole, gutted and fillet frozen rock cod (*Epinephelus spp.*) at -20°C. Same trend observed after 120 days of frozen storage (Tables 3 and 4) at the end of frozen storage the pH values increased to 6.86, 7.02 for frozen samples and 6.88, 7.05 for fried samples from farms A and B, respectively. The increase in pH value in farm B samples was higher than farm B samples. These results agreed with Olgunoglu et al. [18] who found that pH value increased from 6.80 to 7.02 in frozen pike perch fillets frozen stored at -20°C throughout 7 months. The same trend was found by El-Sherif et al. [11] who reported that the pH values of fresh tilapia and mullet fish were 6.07 and 5.94 and increased to 6.21 and 6.12 after frying for tilapia and mullet fish samples respectively. They noticed that the pH value of frozen tilapia fish increased markedly as the time of frozen storage extended. This increase in the pH value during frozen storage could be attributed to proteolysis and breakdown of protein fraction leading particularly to free some ammonia and other basic products. After 6 months, (end of storage) the frozen raw

tilapia and mullet were fried and the pH value tilapia samples were 6.58, 6.37 for raw and 6.81, 6.61 for fried samples respectively. This increase may be due to the formation of some basic compounds as a result of amino acid degradation [1]. El-Badry et al., [19] reported that the pH values of raw and fried tilapia were 6.46 and 6.79 respectively. The same finding was obtained by Abo-Zeid [20] and Ibrahim [19-22] and El-Sherif [23]. Also our results agreed with Buchtova et al. [10] who reported that the pH of fresh silver carp samples was measured by 6.29. In the first three months of storage, pH decreased insignificantly to 6.25 and increased again after month six. The initial decrease in pH values may be due to the dissolution of CO₂ in fish muscle, and the subsequent increase in pH values depends on the production of volatile nitrogen bases (ammonia, Trimethylamine and others) in dependence of enzymatic activity [12,24,25].

Period of storage (days)	Farm A		Farm B	
	Frozen fish	Fried fish	Frozen fish	Fried fish
0	6.76 ± 0.04	6.8 ± 0.01	6.2 ± 0.06	6.76 ± 0.04
60	6.61 ± 0.04	6.87 ± 0.07	6.53 ± 0.01	6.92 ± 0.01
120	6.68 ± 0.00	6.76 ± 0.08	6.73 ± 0.08	6.88 ± 0.02
180	6.86 ± 0.014	6.88 ± 0.03	7.02 ± 0.04	7.05 ± 0.02

Farm A=Al-Batts Drain; Farm B=El-Wadi Drain
M: Mean; SD: Standard Deviation

Table 2: pH values (M±SD) of tilapia fish during frozen storage at -18°C for 180 days and their fried products.

Period of storage (days)	TVB-N (mg/100 g)			
	Farm A		Farm B	
	Frozen fish	Fried fish	Frozen fish	Fried fish
0	10.64 ± 0.65	19.04 ± 0.53	12.04 ± 0.22	14 ± 0.62
60	19.32 ± 0.45	24.36 ± 0.90	20.16 ± 0.65	21.84 ± 0.76
120	23.8 ± 0.77	24.64 ± 0.63	22.12 ± 0.49	22.4 ± 0.66
180	22.73 ± 0.53	24.3 ± 0.57	19.48 ± 0.33	22.7 ± 0.69

Farm A=Al-Batts Drain; Farm B=El-Wadi Drain.
M: Mean; SD: Standard Deviation

Table 3: TVB—N (mg/100 g) values (M±SD) of Tilapia fish stored at -18°C for 180 days and their fried products.

Total Volatile Basic Nitrogen (TVB-N) value

TVB-N considered an index to the putrefaction degree, decomposition and the degree of proteinous constituent's breakdown) Kung et al., reported that the values of total volatile basic nitrogen (TVB-N) are affected by species, season, catching region, sex and age of fish [25-27]. TVB-N includes TMA, DMA, ammonia and other volatile basic nitrogenous compounds related to seafood spoilage. Ozogul et al. reported that the total volatile basic nitrogen can be used

only as an indicator of fitness for consumption rather than as an index of freshness throughout the storage life of fish [26]. Data presented in Table 3 shows the TVB-N values of frozen fish at -18 for 180 days and their fried products. Data showed that fresh tilapia fish samples contain 10.64 and 12.04 mg/100 g TVB-N for farms A and B, respectively. These values increased to 19.04 and 14 mg/100 g after deep frying using sunflower oil at 170°C for 10 min. our results indicated that the tilapia fish samples were in fresh state according to Shen et al. [27] who reported that the fresh fish have TVB-N<15 mg/100 g. The fish is generally considered to be fresh if the TVB-N is less than 20 mg N/100 g sample [28]. Same trend was found by El-Badry et al., [19] who reported that the TVB-N of raw and fried tilapia fish were 11.02 and 12.86 mg/mg respectively. The values of TVB-N increased after 60 days from frozen storage, reached to 19.32 and 20.16 mg/100 g for samples from farms A and B, respectively. After deep frying, TVB-N values increased to 24.36 and 21.84 mg/100 g. The same trend was observed after 120 days of frozen storage. The values were 23.8 and 22.12 for frozen samples and increased to 24.64 and 22.4 mg/100 g after frying for samples from farms A and B, respectively. El-lahamy et al., [10] found that the initial TVB-N contents of raw, fried and grilled Mullet fish steaks were determined by 13.25 ± 0.144, 12.48 ± 0.277 and 12.96 ± 0.554 mg/100 g, respectively. Also same auther reported that TVB-N content of raw Mullet fish steaks increased to 14.46 ± 0.265, 18.8 ± 0.461 and 26.75 ± 0.433 mg/100 g after 60,120,180 days of frozen storage, respectively.

Storage period (days)	TBA (mg MA/kg)			
	Farm A		Farm B	
	Frozen fish	Fried fish	Frozen fish	Fried fish
0	0.21 ± 0.11	1.35 ± 0.11	0.23 ± 0.05	1.20 ± 0.21
60	0.35 ± 0.25	0.50 ± 0.21	0.33 ± 0.08	0.40 ± 0.18
120	0.36 ± 0.28	0.95 ± 0.69	0.38 ± 0.11	0.49 ± 0.06
180	0.58 ± 0.69	0.77 ± 0.15	0.60 ± 0.42	0.63 ± 0.06

Farm A=Al-Batts Drain. Farm B=El-Wadi Drain
M: Mean; SD: Standard Deviation

Table 4: Thiobarbituric acid (mg MA /kg) values (M±SD) of frozen Tilapia fish stored at -18°C for 180 days and their fried products.

Our results agreed with Begum et al., [29] who showed that The TVN value increased from 18 to 30 mg/100 g of shrimp with the increase of storage time. Also, Bilgin et al., [30] reported that TVB-N value of raw rainbow trout was 22.96 mg/100 g increased to 23.80 after frying in sunflower oil. At the end of storage, TVB-N values decreased to 22.73 and 19.48 mg/100 g for samples from farms A and B, respectively. After frying the values of TVB-N increased to 24.3 and 22.7 mg/100 g respectively. Similar results were observed by Ježek and Buchtova et al. [10] who reported that the TVBN value of fresh silver carp fish samples was 15.99 mg/100 g. This value decreased to 15.73 mg/100 g after 12 months of freezen storage. The same trend was found by Orak et al. [31] and El-Sherif et al. [11] who reported that tilapia fish containing 14.31 mg per 100 g flesh of total volatile basic nitrogen (TVB-N) at zero time of freezing. These values increased with advancing of frozen storage period, it was noticed that these values reached to 18.08, 21.2 and 29.31 mg/100 gm after 60, 120, 180 days of frozen storage at -18°C. This increase may be attributed to the activity

of proteolytic enzymes of microbial origin which breakdown nitrogenous substances.

TBA value

TBA (thiobarbituric acid) is used as an indicator for lipid oxidation degree, processing operations of seafood such as salting, cooking and mincing promotes oxidation. However, smoking and freezing are retarding oxidation. TBA considered a good technique to monitoring the oxidation processes in fish after conversion to malonaldehyde equivalents [32]. Table 4 shows the TBA values of frozen and fried products of tilapia fish from two farms (A and B). From the Table the TBA values of fresh tilapia fish were 0.21 and 0.23 mg MA/kg for fresh fish samples collected from A and B farms, respectively. After frying the values were increased to 1.35 and 1.20 mg MA/kg for A and B farms, respectively. After 2 months of frozen storage the values of TBA increased to 0.35 and 0.33 mg MA/kg for fish samples collected from A and B, respectively. TBA values increased to 0.50 and 0.40 mg/kg after frying for samples from farms A and B, respectively. Same trend observed after 120 days from frozen storage, values were 0.36 and 0.38 mg MA/kg for fish collected from A and B farms, respectively. After deep frying these, values increased to 0.95 and 0.49 mg MA/kg for samples from farms A and B, respectively. With respect to the relation between frying process and TBA, the obtained results showed that frying of tilapia fish either fresh or frozen caused a noticeable increase in the TBA value for processed sample at any given time of storage. The same trend was found by Stephen et al., [32] and Koizumi et al. [33]. Also Pikul et al. found the increasing in TVB values after cooking could be resulted from water evaporation and loss of fish juiciness during cooking. After 180 days of storage the TBA values increased to 0.58 and 0.60 mg/kg for fish samples collected from A and B farms, respectively. After frying the TBA values increased to 0.77 and 0.63 mg MA/kg for samples from farms A and B, respectively. Similar results were observed by Orak and Kayisoglu, [31] who found the TBA value of Gutted gray mullet were 0.19 and 0.33 mg MA/kg after 4 and 7 months of frozen storage. Also TBA value of whiting fish was 0.16 mg MA/kg and increased to 0.33 and 0.47 mg MA/kg after 4 and 7 months of frozen storage at -26C respectively. From the Table 4 it can be said that the thermal processing caused an increase in the TBA value of fish as a successful criterion for the secondary oxidation of lipid, in accordance with these by Seet et al. [34-39]. Also the TBA of fresh, frozen and fried tilapia fish were under or low the ratios of rancidity reported by several author such as Connell [5] throughout frozen storage periods [40-42].

Conclusion

In the study, it is necessary to state that, frozen Tilapia fish maintained their quality during 6-month storage period according to physic- chemical quality criteria. Raw and fired tilapia fish were found to be good quality fish end of storage as to general chemical means.

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