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ORIGINAL RESEARCH ARTICLE

Quality assessment and shelf-life of processed tilapia (*Oreochromis niloticus*) fish sticks: Laboratory based study

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ABSTRACT

Customers prefer tilapia (*Oreochromis niloticus*), one of the most popular freshwater fish species farmed in Bangladesh, because of its flavor and affordable market pricing. This study aimed to develop value-added tilapia fish sticks and evaluate the quality changes, shelf life, and storage stability of the developed tilapia fish sticks in order to investigate the possibilities of better utilizing low-value tilapia fish and to satisfy consumers' growing demand for quality ready-to-eat food products. For this regard, storage characteristics in room (28°C) and refrigerator (5°C) temperatures were assessed in terms of microbiological, chemical, proximate, and sensory attributes. The moisture, lipid, protein, and ash contents of the fish sticks were observed to be 56.23±0.62, 7.62±0.27, 26.01±0.39, and 2.93±0.23%, respectively, at fresh condition. As storage time increased, it was discovered that ash content at room temperature increased while moisture, lipid, and protein levels steadily declined. On the other hand, it was discovered that at refrigeration temperatures, ash and fat content increased while moisture and protein content decreased. Compared to fish sticks held at ambient temperature, changes in the proximate composition of fish sticks stored in a refrigerator were found to be more stable. TVB-N was initially measured as 12.38±0.45 mg/100 g. After 24 hours of room storage, the TVB-N value exceeded the acceptable level; however, after 72 hours of refrigeration, it did not exceed the acceptable limit and was deemed fit for consumption. TPC was observed in fresh fish sticks as 3.74±0.31 Log CFU/g. In 48 hours at room temperature, the bacterial load of tilapia fish sticks increased sharply ($p<0.05$) during the course of the storage period and went above the microbiological threshold for fishery products (7 Log CFU/g of flesh). The bacterial growth trend was slower and, after 72 hours, was within the permitted limit at refrigerated storage temperature. All fresh products had the highest initial sensory ratings. At ambient temperature, all of the products sensory qualities significantly declined with time ($p<0.05$), however at refrigeration temperature, the product was determined to be more stable. The overall acceptability score assessed for appearance, flavor, taste, and texture was within acceptable limits for up to 24 hours at room temperature, but not for 72 hours at refrigeration temperature. According to the study's findings, tilapia fish sticks have a very limited shelf life at room temperature (28°C), only lasting around 24 hours, whereas they can last up to 72 hours at 5°C in the refrigerator.

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INTRODUCTION

The preference of consumers has considerably focused on fast-food consumption these days since there has been rapid urbanization and an increase in the working women population. As a result, many fast-food shops have been opened in cities and industrial areas of the country (Ejaz *et al.*, 2013). Today's working people along with new-generation students and young people are now more dependent on convenient foods like ready-to-eat and/or ready-to-cook foods (Akter *et al.*, 2013). The demand for ready-to-eat and/or ready-to-cook products is gradually growing because of their convenience (Yerlikaya *et al.*, 2004). Along with the convenient foods that look and taste good, consumers now a days giving utmost priority to hygienically prepared and attractively packed foods having nutritional value. In this perspective, value addition and diversification of fish products might be helpful to satisfy the ever-changing and diverse demands of consumers. Worldwide, fish has already been recognized as a favorite foodstuff due to having high-quality proteins, nutritional vitamins, and essential omega-3 polyunsaturated fatty acids (PUFA) (Ehsani and Jasour, 2012; Shahidi and Ambigaipalan, 2018). Likewise, in Asian countries, due to increasing awareness of consumers on health issues, the consumption of fish and fishery products is increasing day by day (Akter *et al.*, 2013).

Bangladesh is considered one of the most suitable countries in the world for freshwater aquaculture, because of its favorable resources and agro-climatic conditions. Freshwater inland aquaculture production in Bangladesh is the third highest in the world after China and India (FAO, 2018). A large number of low-cost fish species are found available in the market throughout the year. Henceforth, there is a need to develop some convenience products from the meat of low-cost fish to enhance their consumer acceptability and for the better utilization of the low-valued fish (Akter *et al.*, 2013). Development of different types of value-added ready-to-eat products (i.e., fish balls, fish fingers, fish sticks, fish cutlets, etc.) could be a better option. Earlier, some studies were undertaken to make value-added fish products from low-cost fish but it was not successful due to shortage of funds, unavailability of equipment's for processing underutilized species and a lack of entrepreneurship at the commercial level at that time (Akter *et al.*, 2013; Ejaz *et al.*, 2013). Now a days, value-added products prepared from beef and poultry are served to the fast-food shops in the market of Bangladesh, but unfortunately, value-added fish products are not produced commercially here. That's why, in this study, Tilapia- a low-valued and fast-growing aquaculture species were selected to make improved value-added ready to eat fish sticks yet has not been introduced in Bangladesh. The product attains a meaty texture and can conceal the fishy odor by incorporating local taste which can attract consumer's acceptance easily. However, Ready-to-cook/eat meat products are susceptible to spoilage and pathogenic microorganisms (Zhu *et al.*, 2004). Coating of these products with edible materials can provide better protection against oxidation and microbiological deterioration. It can also significantly enhance the sensory qualities of meat products and could be an effective method of value addition with better consumer acceptability (Duman and Peksezer, 2016). Besides, freezing and frozen storage are commonly used method for the storage of different types of value-added fish products because of the consistency, reliable quality, ease of transportation and the fact that they are very close to fresh equivalents (Bainy *et al.*, 2015; Sarma *et al.*, 2000).

Although there are many food processing techniques that can safeguard food, fish and fisheries products might experience unfavorable changes during storage, and deterioration may shorten the storage time (Duman and Peksezer, 2016). Therefore, efforts should also be undertaken to enhance and assess the quality and consistency of ready-to-eat foods. To the best of our knowledge, no researches have yet been conducted on the quality and storage stability of tilapia fish sticks. Given the aforementioned information, the purpose of this study was to establish the shelf-life of processed fish sticks made from tilapia fish as well as the quality changes that occur during the refrigerated and room storage settings using proximate composition, sensory and bacteriological assessments.

MATERIALS AND METHODS

Selection and collection of raw fish

Tilapia (*Oreochromis niloticus*) an exotic fish, was selected for the production of fish sticks as it is a good aquaculture species, available in the market throughout the year and low in cost. Fresh tilapia fish was collected from local fish markets of Sylhet Sadar in an insulated ice box and then transported to the laboratory of Fisheries Technology and Quality Control, Sylhet Agricultural University for further processing. The average size of the fish was 730 ± 0.85 gm and the price of the fish was BDT 160 ± 35 per kg.

Preparation of tilapia fish sticks

The fishes were washed with clean water, beheaded, eviscerated, skinned and then washed with chilled water. The skinned fishes were filleted, cut into strips of definite size and shape and deboned manually in iced condition. Then the strips were dipped into the previously prepared batter solution. The battering solution was prepared by mixing 34% wheat flour, 18% egg white, 2% spices, 1% salt, 1% Monosodium glutamate (MSG) and 1 teaspoon vinegar. After that, it was rolled in bread crumbs. Battered and breaded fish sticks were then dip-fried in soybean oil until the surface of the sticks becomes golden brown color. Finally, prepared fish sticks were then kept on the kitchen paper in order to soak the extra oil from the surface of the fried fish sticks. A portion of the fresh sample was analyzed immediately and the remaining portions were stored in room 28°C and refrigerated 5°C temperature for up to 72 hours to determine quality changes as well as the shelf life. Good Manufacturing Practices (GMP) established by International Standards Organization (ISO) was followed during the whole operation (Chen *et al.*, 2019). Local gel enhancing ingredients and spices were used for

Table 1. Amount of ingredients used to prepare 2% species for batter solution.

Ingredients	Percentage (%)
Garlic powder	15
Onion paste	25
Ginger powder	15
Green chili paste	15
Mixture of hot spices	30

the preparation of batter solution to ensure the products a Bangladeshi known taste so that the products could attract local consumer's acceptance.

Quality changes and shelf-life study

The shelf-life study is an important consideration in respect of business aspects and consumers health issues. Chemical, microbial and sensory qualities were evaluated for the study of quality changes and shelf life of tilapia fish sticks during room (28°C) and refrigerated (5°C) storage conditions for up to 72 hours at an interval of 24 hours. All the data were analyzed in triplicates.

Proximate composition analysis

Proximate composition analysis of moisture, crude protein, lipid and ash were carried out according to the methods of the Association of Analytical Chemists (AOAC) with some modifications (AOAC, 2019). Moisture content was determined in a hot air oven at 105°C for 24 hours. Lipid content was extracted using acetone by the Soxhlet method with the Soxhlet apparatus. The Kjeldahl method was used to determine the total nitrogen content and then the crude protein level was subsequently calculated using nitrogen to protein conversion factor 6.25. The ash content of the sample was determined in a muffle furnace at 550°C for 6 hours.

Chemical evaluation

Total Volatile Base Nitrogen (TVB-N) was determined according to the methods given in AOAC (1997) with certain modifications. 10 gm of ground sample was homogenized for 2 minutes with 90 ml of 6% perchloric acid with a blender. Then Whatman no.1 filter paper was used to filtrate the homogenates. Before distillation, the filtrates were alkalized with 10 ml of 20% NaOH. After distillation, the collected distillate was titrated with 0.01N HCl. The TVB-N value was assessed to study the shelf-life of the sample and expressed as mg/100g of sample.

The pH of the sample was measured following the method described by AOAC (2006). Accurately weighted 5g of sample was homogenously mixed in 50 ml distilled water and then the pH of the homogenized sample was measured using an electronic pH meter (HANNA pH 211 Microprocessor pH Meter) at ambient temperature.

Bacterial load analysis

For evaluating microbiological changes, Total Plate Count (TPC) was performed using Plate Count Agar (PCA) by spread plate technique according to the methods of ICMSF (2012). At first, 10 g of the sample was diluted with 90 ml sterile 0.1% peptone water and then homogenized using a vortex mixer. Then, 0.1 ml samples of serial dilutions (1:10, diluent, 0.1% peptone water) of the homogenates were spread on the surface of dry plate count agar media (Difco, Haryana, India). The colonies were counted for total plate count having plate's 30-300 colonies after incubation at 30°C for 48 hours. All counts were converted to logarithms of the colony- forming units per gram (Log CFU/g).

Sensory evaluation

Sensory quality was evaluated by seven experienced panelists for the determination of the shelf-life of the sample. Panelists scored for sensory characteristics such as appearance, color, flavor, taste, texture and overall acceptability on a five-point scale (Tokur *et al.*, 2006) where scores were assigned with '1' being the least and '5' being the highest for attributes. A score of 3 (neutral) was regarded as the acceptable margin for the product. Samples were drawn four times within a period of 72 hours with a 24 hours interval between samplings.

Statistical analysis

Analysis of variance was performed by one-way ANOVA procedures with the application of Duncan's multiple range tests and descriptive statistics using SPSS version 20 to find the significant difference at a 5% level of significance. The results were presented as mean ± standard deviation (SD).

RESULTS AND DISCUSSION

Proximate composition analysis and pH measurement

Proximate composition was evaluated to find out the nutritional value of tilapia fish sticks. Moisture, lipid, protein and ash contents of fresh tilapia fish fillet at fresh condition were 77±0.98, 2.74±0.42, 19.28±0.65 and 2.25±0.31%, respectively (Table 2) which is comparable to the other freshwater fishes (Bhatta *et al.*, 2015; Panchavarnam *et al.*, 2003; Reddy *et al.*, 2012; Vanitha *et al.*, 2013). However, in fresh tilapia fish sticks those parameters were estimated as 56.23±0.62, 7.62±0.27, 26.01±0.39 and 2.93±0.23%, respectively which was comparatively higher (except the moisture content). The decrease in moisture content in tilapia fish sticks can be attributed to the frying and cooking processes (Rahman *et al.*, 2019). Lower moisture content in the final product than the raw material was also reported from the study of Ejaz *et al.* (2013), Ihm *et al.* (1992) and Taşkaya *et al.* (2003). An increase in protein value might be due to the addition of eggs and other food additives. Higher protein contents were also reported in fish balls after the cooking process as compared to the raw fish (Duman and Peksezer, 2016). Increased lipid content in the fried product may also be attributed to cooking the product in vegetable oil (Sehgal *et al.*, 2008; Tokur *et al.*, 2006). The ash content in the final product was higher than the fresh fish fillet, due to the addition of the species and other ingredients (i.e., NaCl, Potato) during the preparation. (Praneetha *et al.*, 2015).

The pH of the fresh tilapia fillet was 6.7±0.04. A slight increase

Table 2. Proximate composition (%) and pH value of fresh tilapia fish and fish sticks.

Type of product	Proximate composition				
	Moisture	Lipid	Protein	Ash	pH
Fish Fillet	77±0.98	2.74±0.42	19.28±0.65	2.25±0.31	6.7±0.04
Fish Sticks	56.23±0.62	7.62±0.27	26.01±0.39	2.93±0.23	6.8±0.02

* Mean value ± standard deviation of 3 individual measurements

in pH value was observed in the tilapia sticks than raw fillet (Table 2). In the case of raw and deep-fried tilapia, a similar change in pH value was also reported in the study of Mohamed *et al.* (2019). A slight decrease in pH in the pangus burger than the mincewas observed in the study of Ejaz *et al.* (2013). The pH is considered as a determining factor for higher gel-forming ability. A good quality product can be prepared from the mince with around neutral pH (Azad, 2001). The pH of the fresh tilapia fillet used to develop tilapia fish sticks was about neutral because of the use of pre-rigor prime quality fresh fish. Changes in the proximate composition of tilapia fish sticks stored in room and refrigerated temperature were significantly different. At room temperature (28°C), moisture, lipid and protein content gradually decreased while ash content was found to increase with the progress of storage time (Figure 1). There wasn't any significant difference ($p<0.05$) in the moisture and ash content of fish sticks at 0, 24 and 48 hours of storage at room temperature. After a storage period of 72 hours, moisture, lipid, protein and ash content were found as 55.14±0.56, 5.39±0.32, 23.96±0.42 and 3.02±0.09%, respectively compared to the value of moisture, lipid, protein and ash content 56.23±0.62, 7.62±0.27, 26.01±0.39 and 2.93±0.23%, respectively of fish sticks at fresh condition. On the other hand, at refrigerated temperature (5°C), lipid and ash content were found increasing while moisture and protein content were in a decreasing trend with the progress of the storage period (Figure 2). After a storage period of 72 hours, moisture, lipid, protein and ash content were found as 54.95±0.65, 9.0±0.23, 25.47±0.36 and 2.99±0.22%, respectively. Similar results were reported from the study of (Praneetha *et al.*, 2015) during refrigerated storage of fish fingers where the moisture and protein content was found decreasing while fat and ash content was found increasing. The findings of this study are more or less parallel with the results of Rani *et al.* (2017) where an increase in lipid, protein and ash content and a gradual decrease in moisture content of Mrigal (*Cirrhinus mrigala)* fish finger were observed during frozen storage. The decrease in moisture content in the stored fish sticks can be due to the dehydration that occurs at refrigeration temperature (Shikha *et al.*, 2019). Vanitha *et al.* (2015) also observed decreased moisture content of fish fillets produced from Mrigal (*Cirrhinus mrigala)* and fish burgers produced from Catla (*Catla catla*) during frozen storage. Denaturation of protein and leaching out of water-soluble proteins might be one of the reasons for the decrement in protein content (Arannilewa *et al.*, 2006; Gandotra *et al.*, 2012; Siddique *et al.*, 2011). The increase in lipid content can be attributed due to the decrease in moisture content as they are inversely proportional (Vanitha *et al.*, 2015). In the case of ash, values were increased due to moisture loss during processing (Kocatepe *et al.*, 2011; Pawar *et al.*, 2013, 2019). From the findings of the present study, it is evident that changes in the proximate compositions of fish sticks stored in refrigerated condition are more stable than fish sticks stored at room temperature.

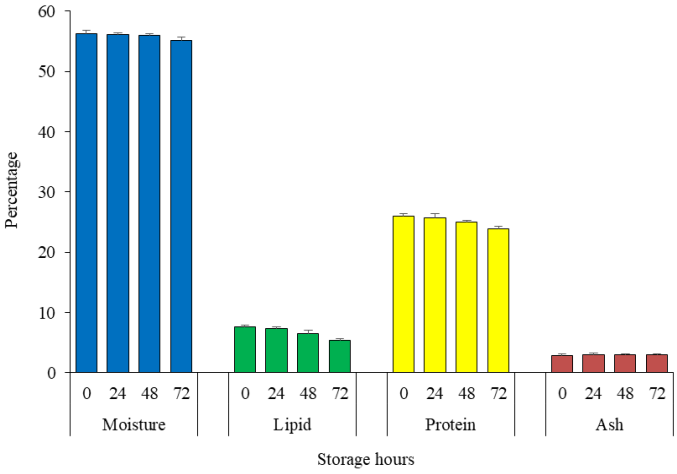

Figure 1. *Changes in Proximate composition (%) of tilapia fish sticks (n=3) stored at room temperature (28°C).*

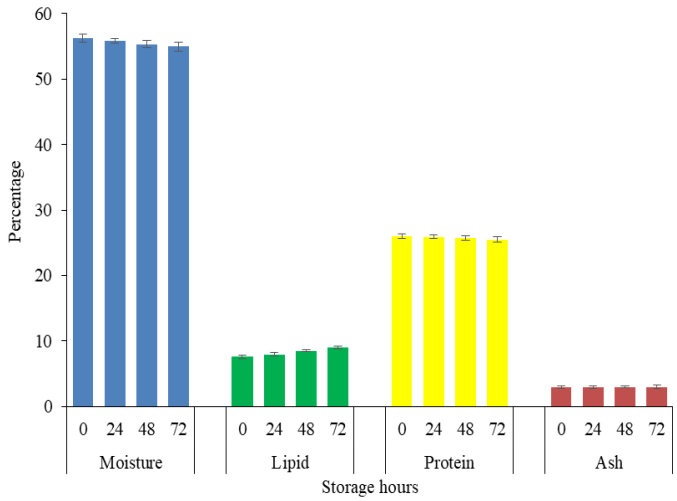

Figure 2. *Changes in Proximate composition (%) of tilapia fish sticks (n=3) stored at refrigerated temperature (5°C).*

Changes in TVBN value

Total Volatile Basic Nitrogen (TVB-N) is most widely used for the evaluation of the degree of spoilage in seafood. TVB-N is often used as an index to assess the keeping quality and shelf life of seafood products (Bouletis *et al.*, 2017). The initial TVB-N value was 12.38±0.45 mg/100 g and it increased progressively with storage time in the room (28°C) and refrigerated (5°C) temperature (Figure 3). Chomnawang *et al.* (2007) stated that the increasing of TVB-N value during storage is related to bacterial spoilage, activity of endogenous enzymes and degradation of tissue proteins. But in the case of room temperature, the increase was very rapid than the refrigerated temperature. At

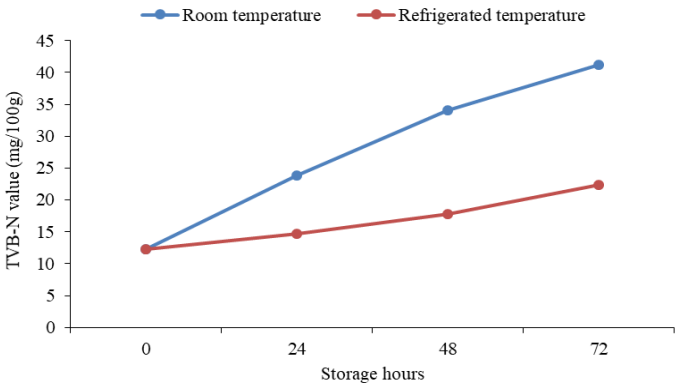

Figure 3. *Changes in TVB-N value of tilapia fish sticks stored at room (28° C) and refrigerated (5°C) temperature.*

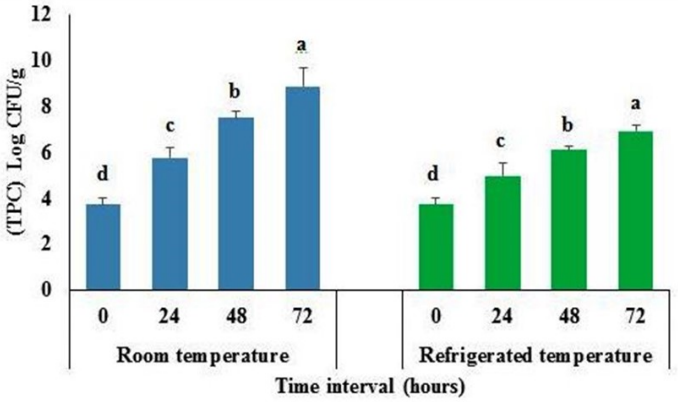

Figure 4. *Changes in microbial load of tilapia fish sticks (n=3) at room (28°C) and refrigerated (5°C) temperature. The different superscripts denote that the values are significantly different (p<0.05).*

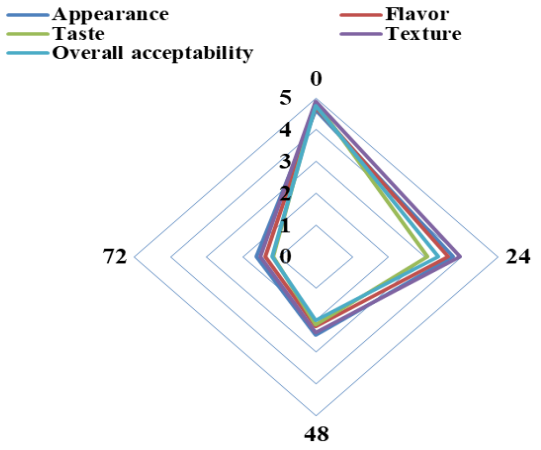

Figure 5. *Changes in sensory quality attributes of tilapia fish sticks at room temperature (28°C).*

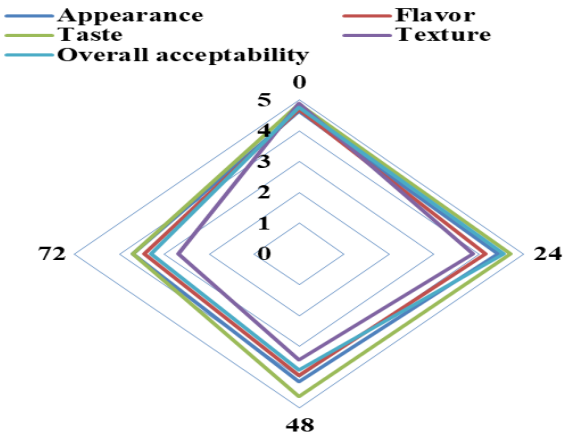

Figure 6. *Changes in sensory quality attributes of tilapia fish sticks at refrigerated temperature (5°C).*

room temperature, the TVB-N value at 48 hours was 34.02±0.49 mg/100g, higher than the acceptable limits reported by different researchers. Giménez *et al.* (2002) proposed a level of more than 25 mg N/100 g as an unacceptable value in fish and fishery products. A level of 30 mg muscle TVB-N/100 g has been considered by Gökodlu *et al.* (1998) as the upper limit above which some fishing products are considered spoiled and unfit for human consumption. The acceptability limits of TVB-N were reported at 30 mg/100g meat for silver carp (*Hypophthalmichthys molitrix*) by Bhatta *et al.* (2015) and Osman and Zidan (2014). At 72 hours the value of fish sticks stored at room temperature reached 41.16±0.76 mg/100g, which indicates that the product is completely spoiled. On the other hand, at refrigerated temperature, the TVB-N values did not exceed the upper acceptability limit even after 72 hours of frozen storage and were considered fit for consumption.

Bacterial load analysis

Total viable count is a useful tool for the evaluation of shelf-life and post-processing contamination in fishery products (Duman and Özpolat, 2012). Changes in TPC of fish sticks kept at room (28°C) and refrigerated (5°C) temperature was determined at every 24 hours interval (Figure 4). In fresh fish sticks, TPC (Total Plate Count) was found as 3.74±0.31 Log CFU/g, indicated that high quality raw materials were used and good hygiene conditions were implemented to develop the fish sticks. In both storage temperatures, the TPC was increased significantly ($p<0.05$) throughout the storage period. Bacterial growth in fish sticks kept at room temperature increased quickly ($p<0.05$) with the progress of the storage period and within 24 hours TPC was found 5.77±0.43 Log CFU/g and after 72 hours it went to 8.88±0.78 Log CFU/g. The rate of bacterial growth of the same fish sticks kept at refrigerated temperature was slower but a significant difference ($p<0.05$) in TPC was still found. After 72 hours the TPC of refrigerated stored fish sticks reached to 6.93±0.23 Log CFU/g. Similar changes in bacterial load were also reported in the study of Ejaz *et al.* (2013). Fish sticks kept at room temperature exceeded the maximum levels (7 Log CFU/g of meat) of microbiological criteria for fisheries products given by the ICMSF (2012) in 48 hours. Refrigerated stored fish sticks were in acceptable condition throughout the storage period. Neither bad smell nor fungal growths were apparent in the product during that period (Gates, 2012). Islam *et al.* (2004) reported fish mince-ingredient mix kept at room temperature exceeded the maximum levels within 6 hours and at refrigerated stored after 18 hours. Tural and Turhan (2017) investigated that the total viable bacterial count exceeded the critical limit on the 5th day during refrigerated storage of anchovy (*Engraulis encrasicholus*) patties. Considering the findings of the present study in regard to ICMSF acceptable microbial limit it can be concluded that the shelf life of tilapia fish sticks is not more than 24 hours at room temperature and up to 72 hours at refrigeration temperature. The findings are also in agreement with the findings of Ejaz *et al.* (2013) and Gates (2012).

Sensory analysis

Sensory evaluation was done to study the shelf life of the prepared fish sticks. Changes in sensory attributes of fish sticks stored in room and refrigerated temperature were significantly different. The overall acceptability score of tilapia fish sticks (judged for appearance, flavor, taste and texture) was within the acceptable limit till 24 hours at room temperature (Figure 5), whereas at refrigerated temperature even after 72 hours (Figure 6). Freshly prepared tilapia fish sticks received an initial score of 4.77±0.53 and were rated as high quality. All the sensory attributes were found reduced prominently with the increase of storage time at room temperature which is in agreement with the study of Ejaz *et al.* (2013). Sensory analysis showed that at 24 hours of storage at room temperature, the sticks were acceptable for consumption according to their scores. A bad smell was observed at 48 hours of storage time and fungal growth was visible at the storage time of 72 hours which was indicative of large bacterial growth. After 72 hours, all the attributes i.e., appearance, flavor, taste, texture and overall acceptability of fish sticks were considered as "extremely disliked" according to their scores and it was unacceptable. According to the statistical analysis, significant differences ($p<0.05$) were found among the sensory attributes during the storage period. At refrigerated temperature, the product was found more stable. Neither pungent odor nor fungal growth was seen even after 72 hours. Additions of various ingredients that have antioxidant properties such as polyphosphate (Huffman *et al*., 1987), egg white (Yetim and Ockerman, 1995) and dry spices and ginger (Van Hecke *et al*., 2017) might have protected the products from development of rancidity during the storage period. The textural quality changed significantly at 48 and 72 hours but was considered acceptable. From the sensory results, it was seen that the shelf life of the tilapia fish sticks stored at room temperature (28°C) is not more than 24 hours but it was acceptable even after 72 hours at refrigerated temperature (5°C). Similar findings were also reported by Ejaz *et al.* (2013) and Gates *et al.* (2012). Many researchers observed that the shelf-life of various fish products stored in refrigerated conditions is 9 to 11 days (Boran *et al*., 2007; Taşkaya *et al*., 2003). According to Yerlikaya *et al.* (2004), the limit of acceptability was 6 days for anchovy patties in refrigerated storage. Turhan *et al.* (2001) also estimated the shelf life of refrigerated raw anchovy (*E. encrasicholus*) patties for 6 days at 4°C. These findings show very similar results to the present study.

Conclusion

Value-added fish products, such as fish sticks, can help meet the customer need for ready-to-eat foods, as well as satisfy hunger, supply important nutrients, and save time and money. The purpose of the study was to evaluate the quality and shelf life of tilapia (*Oreochromis niloticus*) fish sticks treated under controlled laboratory conditions. According to all of the microbiological, chemical, proximate, and sensory quality analyses conducted for this study, tilapia fish sticks can be stored for up to 24 hours at room temperature and 72 hours in a refrigerator (5°C) without experiencing any unfavorable changes to their microbiological, chemical, or sensory quality compared to when they are stored at room temperature (28°C). Given that all the ingredients required to prepare fish sticks are readily available locally, commercial production of this product under these storage conditions could present a significant opportunity for new small- and large-scale entrepreneurship, the creation of a new food market, and economic independence.

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Conflict of interest

The authors declare that they have no conflicts of interest.

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