

## RESEARCH ARTICLE

### DEVELOPMENT OF FISH BURGER PATTY FROM SPOTTED TRIGGERFISH (*Canthidermis maculate* L.) MINCE AND DETERMINATION OF ITS NUTRITIONAL PROPERTIES AND SHELF-LIFE

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#### ABSTRACT

Fish is a popular protein source, and consumer interest in value-added fish products is increasing. In addition to abundant underutilized fish species that are not in demand as fresh fish, this is a sustainable approach to fulfilling protein needs while minimizing the over-exploitation of highly demanded fish varieties such as yellow-fin tuna. Therefore, this study aimed to prepare a burger patty using Spotted triggerfish (*Canthidermis maculata* L.), a relatively abundant underutilized fish species in the Indo-Pacific region and determine its nutritional properties and shelf-life. Four different formulations of triggerfish mince (FM) and bread crumb (BC) (T1: 62% FM and 10% BC, T2: 58% FM and 14% BC, T3: 54% FM and 18% BC and T4: 50% FM and 22% BC w/w) were prepared while keeping other ingredients constant (28% w/w). Proximate composition, physico-chemical properties and shelf-life analysis were conducted using standard methods. Sensory analysis with 30 semi-trained panellists using a 5-point hedonic scale identified treatment T2 (58% FM:14% BC w/w) as the most preferred fish burger patty. Proximate analysis of T2 revealed 54.92±0.23 g/100 g moisture, 15.5±0.82 g/100 g crude protein, 3.11±0.14 g/100 g crude fat, and 2.28±0.02 g/100 g total ash content. T2 demonstrated significantly higher hardness ( $p<0.05$ ) than the control sample (commercially available fish burger patty). The total volatile base nitrogen (TVB-N) levels of T2 significantly increased from 14.68 to 26.73/100 g during the storage period. The total plate count was assessed biweekly during storage. The product was absent from *E. coli*, *Staphylococcus aureus*, and *Salmonella*, which were tested at the initial stage. It can be concluded that the developed burger patty, which contains 58% w/w triggerfish mince, is safe to consume after one month, and further studies are required to determine the shelf-life. This product offers a promising alternative to commercially available burger patties, with higher nutritional value in fat, fibre, protein, and energy.

Keywords: Fish burger, Nutritional value, Shelf-life, Triggerfish, Value addition

#### INTRODUCTION

The global demand for seafood is increasing due to its health benefits and unique taste. Fish is a great source of protein, vitamins, and minerals. In recent years, there has been a surge in the demand for fish burgers due to their convenience and affordability. However, most fish burgers are made from commonly available fish species such as cod, haddock, and pollock. The use of underutilized fish species can provide an opportunity to diversify the market and offer an alternative source of

income for fishermen while increasing food security in the country (De Silva *et al.*, 2021).

Spotted triggerfish (*Canthidermis maculate* L.) is a relatively underutilized and abundantly available fish species in the Indo-Pacific region. It is a reef-associated fish that feeds on crustaceans and Mollusca. The flesh of the triggerfish is firm in texture and has a slightly sweet taste. It is recognized as a good source of protein and essential amino acids while being relatively low in fat and calories (Rifat *et al.*, 2023). They typically contain

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beneficial omega-3 fatty acids, which are advantageous for cardiovascular health (Bowen *et al.*, 2016). The high protein content makes the triggerfish an ideal candidate for developing nutritious fish burger patties.

During January and September 2023, a harvest of 95,870 metric tons (Mt) from deep-sea fish production and 117,195 Mt from coastal fish production contributed to a total marine fish production of 213,065 Mt, highlighting the nation's dependence on marine resources for sustenance and economic growth (Ministry of Fisheries, 2023). Within this context, the inland and aquaculture production of triggerfish emerged as a notable component, contributing 1,340 Mt to the overall catch (Sri Lanka Export Development, 2024). This substantial figure underscores the significance of triggerfish within Sri Lanka's fisheries sector, highlighting its role in both traditional fishing practices and emerging aquaculture endeavours. As a valuable marine resource, triggerfish production provides food security and contributes to the country's socio-economic development through employment generation and export opportunities. Understanding and managing the sustainable exploitation of triggerfish resources remain essential for ensuring the long-term viability of Sri Lanka's fisheries sector and safeguarding the marine ecosystem (MFAR, 2020).

Fresh Triggerfish lacks consumer acceptance due to its unappealing appearance and difficulty cleaning the fish. Consequently, demand and the price of fresh Triggerfish remain low in the local market, resulting in surplus production. Due to the considerable amount of Triggerfish used to make dry fish, it is leading to economic losses and environmental concerns. Therefore, finding innovative and sustainable solutions to utilize this excessive harvest efficiently and minimize the wastage of fresh triggerfish is crucial. Developing a fish burger emerges as a promising solution among the various preservation methods considered for Triggerfish. By transforming the fish into burger products, it can be preserved for a more extended period, allowing for strategic marketing and distribution throughout the year.

This is also a valuable initiative for developing value-added fish products currently lacking in Sri Lanka. Moreover, by creating innovative products like a burger patty, we can enhance the utilization of this sustainable fish species, contributing to more diverse and healthier diets. Comprehensive analyses and incorporation of this fish into new food products could also promote its broader acceptance and provide valuable insights into its full nutritional potential. Thus, integrating Spotted triggerfish into the food industry supports sustainable seafood practices and offers significant health benefits, such as contributing to fulfilling the community's protein needs, making it a valuable candidate for new food product development. Moreover, the scarcity of affordable fish burger products in the Sri Lankan market adversely impacts the fast-food sectors, leaving a significant market gap in the burger industry.

Therefore, this study aimed to develop a fish burger patty utilizing the mince from Spotted Triggerfish (*Canthidermis maculate L.*) and to systematically evaluate its nutritional composition and shelf-life under frozen storage conditions ( $-18 \pm 2$  °C). Additionally, it addresses the need for the value addition of spotted triggerfish, thereby promoting its broader acceptance and use.

## MATERIALS AND METHODS

### *Sample collection*

Triggerfish were purchased from the central fish market in Peliyagoda, Sri Lanka, after careful evaluation of physiological and sensory parameters, including aroma, texture, colour, scales, adhesion, and lustre. Selected samples were transported to the research centre in a cool box at  $-18 \pm 2$  °C.

### *Development of fish burger patty*

#### **Preparation of culinary blend**

Preliminary experiments were conducted to select the optimum amounts of different ingredients to prepare a culinary blend for fish burgers (Table 1).

**Table 1: Main ingredients in culinary blend mixture**

| Ingredient  | Weight (g/100 g) |
|-------------|------------------|
| Ice cube    | 12.00            |
| Corn starch | 4.45             |
| Salt        | 1.50             |
| Spice mix   | 0.95             |
| Soy protein | 0.80             |

**Preparation of mince and burger patty from the triggerfish**

Cleaned fish was deboned and minced using a meat mechanical mincer (XG-3000, China), maintaining the chilled condition ( $10 \pm 2$  °C) using ice cubes. Then, the mixer was blended with sodium tripolyphosphate (STPP) (0.25% w/w) and stored in frozen condition ( $-18 \pm 2$  °C). The mince obtained from triggerfish was used to develop the fish patties.

Four different treatments (changing the amount of fish mince and bread crumb, keeping the culinary blend % constant) were used to develop the various types of fish burger patties (Table 2).

**Table 2: Ingredients allocation according to weight for the four treatment combinations**

| Ingredient         | Treatments |    |    |    |
|--------------------|------------|----|----|----|
|                    | T1         | T2 | T3 | T4 |
| Fish Mince (%)     | 62         | 58 | 54 | 50 |
| Bread crumb (%)    | 10         | 14 | 18 | 22 |
| Culinary Blend (%) | 28         | 28 | 28 | 28 |

The batter solution was prepared by mixing water and wheat flour in 1:2.5 ratios. The fish burger patties were then dipped into the batter solution for 30 seconds and rolled in breadcrumbs until the surface had a uniform coating. Battered and breaded fish burger patties were flash fried at  $180 \pm 2$  °C temperature at 30s. Fifteen burger patties were prepared from each treatment and packed in high-density polyethylene (HDPE) bags followed by storing at  $-18 \pm 2$  °C for further analysis and shelf-life determination.

**Sensory analysis**

The sensory evaluation involved 30 in-house panellists. Colour, aroma, flavour, texture, and overall acceptability were analysed using a 5-point hedonic scale. Based on the sensory analysis, the most suitable fish burger product was selected for further studies.

**Analysis of the proximate composition of the developed product**

Proximate composition of fish mince of developed fish patties and commercially available fish burger patty (burger patty developed from *Cyprinus carpio*) were tested according to the methods described by the Association of Official Analytical Chemists (AOAC) such as moisture (934.15), crude protein (2001.11), crude ash (942.05), crude fibre (978.10) (AOAC, 2019). Available carbohydrate content was determined by subtracting the sum of all other contents by 100 g (FOA, 2003).

**Determination of oil content**

Bligh and Dyer method (Bligh and Dyer, 1959) with some minor modifications was used to evaluate the oil content of the developed product. Ten grams of sample weighed out and mixed with 15 mL of chloroform and 30 mL of methanol were added (1:2 v/v). Then, the mixture was homogenised by a homogenizer (IKA, Germany). After that, 15 mL chloroform was added and shaken vigorously for 2 minutes. Then, 15 mL of distilled water was added and remixed for 30 seconds. The content was centrifuged (MEGAFUGE 16R, Germany) for 15 min at 3000 rpm. After centrifuging, the water/methanol phase sucked. From the separated layers from the centrifuge, the upper aqueous phase was eliminated, and the lower chloroformic phase and solidified solids remaining in the centrifuge tube were poured into 50 mL volumetric flasks through filter paper. The residue remaining on the filter paper was rinsed with 20 mL chloroform. Then, chloroform was added to the mark of 50 mL and placed in a fume cupboard until the chloroform evaporated. Lipid content (wet basis) was determined gravimetrically after keeping samples in an oven at 105 °C for 1 hr (Pérez-Palacios *et al.*, 2008).

$$\text{Lipid content (g/100g)} = \frac{\text{weight of lipid aliquot} \times \text{volume of chloroform layer}}{\text{volume of aliquot} \times \text{sample size}}$$

... Equation 01

### Determination of the energy

The energy content of both the developed product and the control (commercially available fish burger) sample was calculated by using the following equation (FOA, 2003).

$$\text{Energy} \left( \frac{\text{Kcal}}{100\text{g}} \right) = \left[ \frac{(\text{Carbohydrate} \times 4 \text{ Kcal}) + ((\text{Fat} \times 9 \text{ Kcal}) + (\text{Protien} \times 4 \text{ Kcal}))}{100} \right]$$

... Equation 02

### Analysis of the hardness of the developed product and control sample

The hardness of the samples was analysed using a Brookfield texture analyser (Texture Pro CT V1.8 Build 31, USA). A compression test was conducted using the probe TA39 with the settings of Target 1.0 mm, Load Cell 10000 g, Hold Time 3 s, Trigger Load 4 g, Test Speed 2.50 mm/s, Return Speed 2.5 mm/s, and Number of Cycles 2. The hardness of the burger patty and the control sample (commercially available fish burger) were analysed according to the above settings. Analysis was conducted with a two-week interval until a one-month storage period.

### Analysis of the Shelf-life of the developed product

The shelf-life of both the selected sample and the control sample (a commercially used fish burger patty) stored under freezing conditions ( $-18 \pm 2$  °C) was analysed within a two-week interval during a one-month storage period, considering both microbiological (Aerobic Plate Count, E coli *Staphylococcus aureus*) and chemical parameters (pH, Moisture Content, Total Volatile Base Nitrogen) parameters.

### Microbiological analysis

#### Preparation of the sample and dilution series

One gram of sample was used to prepare the dilution series. A volume of 9 mL of peptone water was added to each test tube, and the 6<sup>th</sup> dilution series was used for the analysis.

#### Determination of aerobic plate count (SPC) Preparation of nutrient agar media

The 15.12 g of nutrient agar powder was accurately weighed and transferred into a 1 L volume of the conical flask, to which 540 mL of distilled water was added. The media was sterilized in the autoclave at 121 °C for 20 minutes and cooled to 45 °C under the laminar flow cabinet.

#### Culturing

The pour method was used for the culturing, and plates were sealed after the agar solidified. Then, samples were incubated at room temperature ( $30 \pm 2$  °C). Observations were taken using a colony counter for 24 hours.

#### Determination of E-coli

One millilitre from each dilution was inoculated to the *Escherichia coli* petri film. The petri films were then placed in an incubator at 33-35 °C for 24 - 48 hours, and colonies were counted.

#### Determination of *Staphylococcus aureus*

Baird Parker medium was used to detect *Staphylococcus aureus*. Media was put into the beaker, heated until the solids dissolved, and autoclaved at 121 °C for 15 min. Then, 50 mL of egg yolk was cooled, and 50 mL of egg yolk was added aseptically. It was stored in a refrigerator at 4 °C. The 100 mL of sterilized buffered peptone water was taken into a stomacher bag, and 10 g of the test sample was aseptically added. The sample was blended using a stomacher blender (stomacher 3500 Jumbo, UK) and dilutions of  $10^{-2}$  and  $10^{-3}$  were prepared. The volume of 0.1 mL of the sample was aseptically introduced to the paired Parker plate using a pipette. Then, that quantity was carefully spread as quickly as possible over the surface of the Baird Parker plates. Plates were incubated at  $35 \pm 0.5$  for 24

to 48 hours. Colonies were counted and appeared in the media using the colony counter.

### Analysis of chemical properties

#### Determination of pH

The pH was determined using a calibrated pH meter (HI 99161, Romania) at room temperature ( $27 \pm 2$  °C). Five grams of sample were cut and ground well. They were added to a beaker, and 50 mL of distilled water was added to it. Then, they were stirred well using a glass rod.

#### Determination of Moisture content

According to the method described in AOAC method 934.15, the moisture content of the samples was analysed in two-week intervals until 60 days (AOAC, 2019).

#### Determination of Total Volatile Base Nitrogen (TVB-N)

Total volatile base nitrogen content was determined using the method described in Jinadasa (2014) with some modifications. The fish burger patty (100 g) was cut into pieces and transferred to the blending jar. Then 200 mL of 7.5% Trichloroacetic acid (TCA) was added and blended for 1-2 min. Then, 25 mL of filtrated solution was taken into the distillation apparatus, followed by the addition of 6 mL of 10% (w/v) NaOH. A 150 mL conical flask containing 10 mL of 4% (v/v) boric acid and a few drops of indicator was placed below the condenser with the delivery tube below the liquid level in the flask. The distillation was started and continued until a final volume of 50 mL was obtained from the beaker (40 mL of distillate). Then, the collected solution was titrated with standard 0.02%  $\text{H}_2\text{SO}_4$ .

$$\text{TVB} - \text{N}(\text{g}/100 \text{ g}) =$$

$$\frac{14 \text{ mgmol}^{-1} \times a \times b \times 300}{25 \text{ mL}}$$

... Equation 03

Where;

a = mL of Sulfuric acids

b= normality of Sulfuric acid

### DATA ANALYSIS

All the data were presented as mean  $\pm$  standard deviation. Kruskal Wallis test was used to analyse the sensory data using MINITAB software version 17.0 and an independent student *t*-test was applied to determine the statistically significant ( $p < 0.05$ ) of the developed product and the control product using MINITAB software version 17.0 for Windows.

### RESULTS AND DISCUSSION

The sensory properties of burger patties, including appearance, texture, and flavour, played crucial roles in consumers' preferences and overall product acceptability. Sensory evaluation was conducted to select the best combination of fish mince and bread crumb to develop a burger patty. There were four treatments according to the fish mince and bread crumb combination ratios. All treatments were in different ratios of fish mince (FM) and bread crumb (BC), and culinary blend (CB) was added the same amount for each treatment. There was a significant difference ( $P < 0.05$ ) that can be observed among the treatments for appearance, aroma, texture and overall acceptability (Table 3).

**Table 3: Mean rank values of sensory evaluation of four treatment combinations**

| Attribute     | Treatments                   |                              |                              |                              |
|---------------|------------------------------|------------------------------|------------------------------|------------------------------|
|               | T1                           | T2                           | T3                           | T4                           |
| Appearance    | 60.3 $\pm$ 4.80 <sup>c</sup> | 95.7 $\pm$ 3.78 <sup>a</sup> | 64.8 $\pm$ 5.11 <sup>b</sup> | 24.4 $\pm$ 3.96 <sup>d</sup> |
| Aroma         | 67.7 $\pm$ 3.50 <sup>b</sup> | 90.9 $\pm$ 1.15 <sup>a</sup> | 61.7 $\pm$ 1.22 <sup>c</sup> | 24.9 $\pm$ 2.01 <sup>d</sup> |
| Texture       | 63.1 $\pm$ 0.89 <sup>c</sup> | 88.2 $\pm$ 0.48 <sup>a</sup> | 67.1 $\pm$ 0.85 <sup>b</sup> | 26.5 $\pm$ 0.79 <sup>d</sup> |
| Flavour       | 66.4 $\pm$ 2.57 <sup>b</sup> | 94.3 $\pm$ 2.65 <sup>a</sup> | 66.4 $\pm$ 2.11 <sup>b</sup> | 28.9 $\pm$ 2.75 <sup>c</sup> |
| Overall       | 63.8 $\pm$ 2.98 <sup>b</sup> | 95.5 $\pm$ 3.12 <sup>a</sup> | 57.1 $\pm$ 3.21 <sup>c</sup> | 26.8 $\pm$ 3.35 <sup>d</sup> |
| Acceptability |                              |                              |                              |                              |

The sample T2 (58% FM and 14% BC w/w) emerged as the most visually appealing, aroma, texture, flavour, and overall accepted developed product with the highest mean ranks (Table 3). In contrast, the T4 sample (50% FM and 22% BC w/w) showed the lowest mean ranks with the lowest overall acceptability. According to the results, the T2 sample was selected and used for further analysis. This finding highlights the importance of ingredient ratios, as the optimal ratio of fish mince to bread crumb (5.4:1.4) aligns closely with previous recommendations for achieving favourable texture in burger patties (Rohall *et al.*, 2009).

Proximate composition of selected product (T2) and commercially available burger patty

Analyzing the proximate composition of fish burgers is essential for understanding their nutritional value, as it provides insights into key components such as protein, fat, moisture, and ash content. This information is crucial for both product development and labelling, ensuring that consumers are informed about the health benefits of the product. There was a significant difference ( $P < 0.05$ ) in moisture content between the selected sample (T2) ( $54.92 \pm 0.23$  g/100 g wet basis) and the control sample ( $70.08 \pm 0.71$  g/100 g wet basis) seen, and it may be due to several factors inherent to their respective compositions and processing. Breadcrumbs are the source of carbohydrates for both developed and controlled sample products. Carbohydrates have water retention abilities due to their hygroscopic nature. They attract and hold water molecules, contributing to increased water content in the foods. This property enhances texture, moistness, and palatability (Parafati *et al.*, 2019). The difference in the moisture contents between the two samples may be the use of varying amounts of breadcrumbs (14% developed product and 21% control sample (w/w)) in the formula. Breadcrumbs can contribute differently to the moisture content of the two products.

Moisture retention in fish burger patties offers several advantages and disadvantages. On the positive side, retaining moisture ensures that

the patties remain juicy and succulent, enhancing their overall texture and flavour. Additionally, it contributes to maintaining the freshness of the product by preventing the patties from drying out. Moisture retention also improves cooking properties, facilitating even cooking and consistent results. Ultimately, this leads to greater customer satisfaction and an increased likelihood of repeat purchases. However, excessive moisture retention can result in soggy patties and reduced crispiness, detracting from the overall dining experience. Moreover, it may create a conducive environment for bacterial growth, posing a risk of spoilage if not properly managed. Balancing moisture retention is essential to ensure optimal texture, flavour, and shelf-life of fish burger patties (Parafati *et al.*, 2019; Rajaretnam and Malik, 2023).

There was no significant difference ( $P > 0.05$ ) found in the protein content of the selected product (T2) and control sample (Table 4). This may be due to the lower crude protein content of the fish types used to develop burger products. The control sample used the Sword (*Sappara*) fish (*Xiphias gladius*), and the developed product used triggerfish, which contain 22 g/100 g and 18.5 g/100 g crude protein content on a wet basis, respectively (Sufiat *et al.*, 2022). Comparing the fat content of the samples, the selected sample showed significantly higher ( $P < 0.05$ ) fat content ( $3.11 \pm 0.14$  g/100 g) compared to the control sample ( $1.93 \pm 0.04$  g/100 g). This difference may be due to the contribution of other ingredients and the quality of the other ingredients used in the two different products. Ash content was significantly higher ( $P < 0.05$ ) in the control sample compared to the selected sample (T2). The main ingredient of the control sample was the total mineral content of *Sapparu* fish ( $968 \pm 0.15$  mg/100 g), and the selected sample showed a total mineral content of  $643 \pm 0.12$  mg/100 g (Sufiat *et al.*, 2022). Considering the energy value, a higher energy intake ( $166.31 \pm 0.87$  Kcal/100 g) can be obtained by consuming the developed burger product compared to commercially available products. This suggests that newly formulated burger product provides more

energy-dense options, which may be beneficial for consumers who need higher-energy (Wang, 2014).

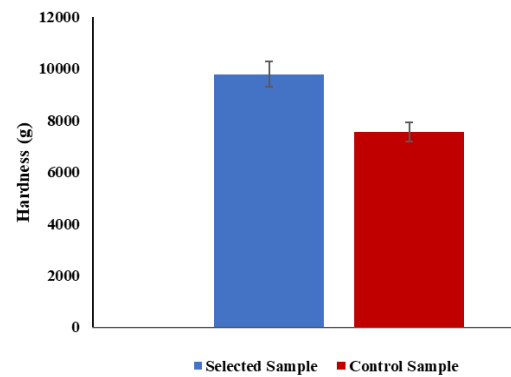
**Table 4: Proximate composition and Energy of the selected sample (T2) and control sample (commercially used fish burger patty)**

| Variable               | Selected sample (T2)       | Control sample             |
|------------------------|----------------------------|----------------------------|
| Moisture (g/100 g)     | 54.92 ± 0.23 <sup>b</sup>  | 70.08 ± 0.71 <sup>a</sup>  |
| Protein (g/100 g)      | 15.50 ± 0.82 <sup>a</sup>  | 15.51 ± 0.10 <sup>a</sup>  |
| Fat (g/100 g)          | 3.11 ± 0.14 <sup>a</sup>   | 1.93 ± 0.04 <sup>b</sup>   |
| Fiber (g/100 g)        | 2.11 ± 0.25 <sup>a</sup>   | 1.01 ± 0.25 <sup>b</sup>   |
| Ash (g/100 g)          | 2.28 ± 0.02 <sup>b</sup>   | 3.53 ± 0.15 <sup>a</sup>   |
| Available carbohydrate | 19.08 ± 0.11 <sup>b</sup>  | 7.94 ± 0.02 <sup>a</sup>   |
| Energy (Kcal/100g)     | 166.31 ± 0.87 <sup>a</sup> | 111.17 ± 5.38 <sup>b</sup> |

Values are mean ± standard deviation (n=3), while different letters for values in each row indicate significant differences (P < 0.05)

### Hardness of the developed product and control sample

Texture profile analysis provides a comprehensive understanding of a product's behaviour under applied forces, offering valuable insights into its sensory properties and overall quality. This analysis is widely employed in the food industry for product development, quality control, and ensuring consistency in texture across different batches (Ghaly *et al.*, 2010). There was a significant difference (P<0.05) between the two samples for hardness (Figure 1), where higher hardness was given by the selected sample (T2) (9789 ± 1702 g) in contrast to the control sample (7545 ± 424 g). This can be attributed to the nature of breadcrumbs, which introduce a lighter and more porous structure. The increased amount of breadcrumbs contributes to a less dense product and softer texture (Ghaly *et al.*, 2010; Wickrama, *et al.*, 2023).



**Figure 1: Texture profile analysis for hardness for selected (T2) sample and control (commercially used fish burger patty) sample**

independent sample t-test was used values are mean ± standard deviation (n=3), while different letters for values in each row indicate significant differences (P < 0.05).

### Self-life analysis of the developed product (T2) and control sample

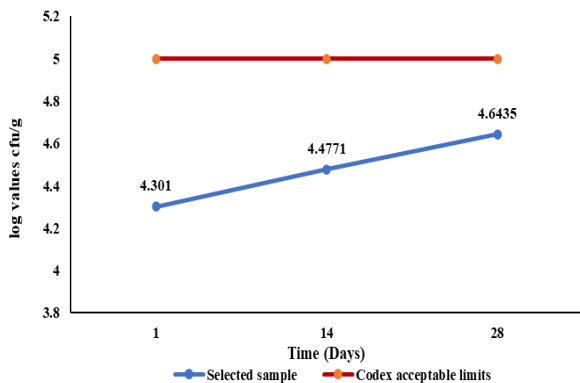
#### Changes in Microbial properties

The purpose of conducting microbial tests for food products is to ensure the safety and quality of the food before it reaches consumers. These tests are conducted to detect and quantify the presence of harmful microorganisms that can contaminate food and cause foodborne illnesses (Ghaly *et al.*, 2010). Food manufacturers and regulatory agencies can take appropriate measures to prevent the distribution of unsafe food products by identifying microbial contamination. Additionally, microbial testing helps to monitor the effectiveness of food processing, storage, and handling practices, ensuring compliance with food safety regulations and standards (Mahmud *et al.*, 2018; Parafati *et al.*, 2019). The developed product was initially tested for the presence of several pathogenic microorganisms, and it showed negative results for the tested microorganisms (Table 5).

**Table 5: Initially tested results for developed product aerobic plate count (SPC), E-coli, and Staphylococcus aureus**

| Microbial test               | Result   |
|------------------------------|----------|
| <i>Escherichia coli</i>      | Negative |
| <i>Staphylococcus aureus</i> | Negative |
| <i>Salmonella</i>            | Negative |

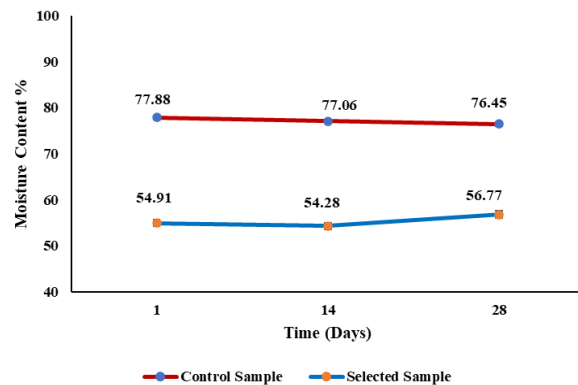
Considering the Total Colony Count, until the second week, the growth rate of colony-forming units showed a slightly increasing trend, and after the second week, it gradually increased (Figure 2). However, according to the Codex standard, the counts are within acceptable limits (acceptable limit  $\leq 1 \times 10^5$  cfu/g) (Codex Alimentarius, 2003).



**Figure 2: Total colony count change over the observed time period for selected (T2) sample and control (commercially used fish burger patty).**

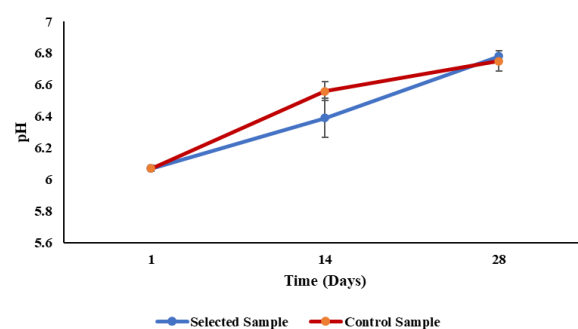
### Changes of Chemical properties

Measuring moisture content during the storage period is also crucial for several reasons. It helps to monitor the stability and quality of the product over time, and changes in moisture content can indicate potential microbial growth, chemical reactions, or physical changes that may affect the product's safety and quality. Controlling moisture content is essential for preserving the product's texture, flavour, and overall sensory properties. The initial moisture content of the developed product was  $54.91 \pm 0.03\%$ , and it experienced a slight reduction on the 14<sup>th</sup> day and subsequently increased up  $56.77 \pm 0.35\%$  on the 28<sup>th</sup> day, which may indicate some chemical and physical changes may occur in the developed product (Figure 3). Similarly, the control sample also demonstrated a gradual reduction in moisture content over the same period. The variation in moisture levels between the developed product and control samples may be attributed to the different amounts of specific ingredients used in the formulation and the storage conditions.



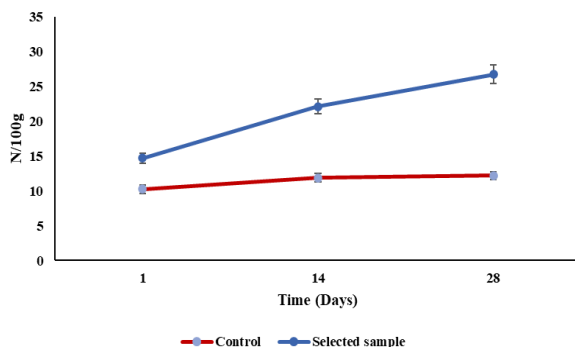
**Figure 3: Deviation in moisture content during storage period for selected (T2) sample and control (commercially used fish burger patty) sample, values are mean  $\pm$  standard deviation (n=3).**

No significant difference ( $P > 0.05$ ) was found in pH change over the observed period in both samples. The results showed higher pH levels in both developed and control samples, which were observed on the 28<sup>th</sup> day (Figure 4). The observed patterns may be due to differences in composition, microbial activity, or chemical reactions that influence the pH changes in each sample over time. Further analysis is needed to identify the specific factors causing these pH changes and to understand their impact on the quality and stability of the samples.



**Figure 4: Changes of pH during storage period for selected (T2) sample and control (commercially used fish burger patty) sample, values are mean  $\pm$  standard deviation (n=3).**

Measuring total volatile nitrogen (TVB-N) levels in fish burger patties may provide information about the freshness and quality of the product. An increase in TVB-N levels typically indicates microbial activity and spoilage, reflecting the breakdown of proteins into nitrogenous compounds. Monitoring TVB-N helps assess the shelf-life and safety of the fish burger patties. The total volatile base nitrogen levels in the developed sample showed higher values than the control samples during storage (Figure 5). These observed variations in TVB-N levels may be attributed to the higher initial crude protein level in the sample. Protein contains nitrogen, so higher protein content would result in higher nitrogen levels. Therefore, it is plausible that the selected sample started with higher nitrogen content due to its higher protein content. This hypothesis is supported by our protein analysis, which indicates differences in protein levels between the selected and control samples, justifying the observed variations in nitrogen content (Walters and Samways, 2001).

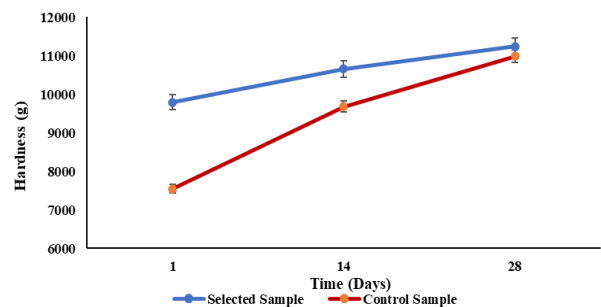


**Figure 5: Changes of Total volatile base nitrogen (TVB-N) levels during the storage period for the selected (T2) sample and control (commercially used fish burger patty) sample, values are mean  $\pm$  standard deviation (n=3).**

### Changes of hardness

Increased hardness was observed over storage time in both developed and control samples, which can be linked to moisture loss and protein denaturation, factors commonly responsible for the texture changes in stored fish products (Figure 6) (Christensen *et al.*, 2017). The higher initial hardness of the

newly developed patty may be attributed to its formulation, possibly due to the presence of binding agents or increased protein content, both of which can contribute to initial firmness. In contrast, a faster rate of hardness increase in the control sample suggests it may lack stabilizing ingredients or textural modifiers found in the developed patty, which can help retain moisture and mitigate rapid protein aggregation over time (Nawaz *et al.*, 2021). This difference indicates that the developed patty's unique formulation may contribute to a slower rate of hardness increase, potentially enhancing its shelf life and quality compared to the control (Badii & Howell, 2002; You *et al.*, 2022).



**Figure 6: Changes of Hardness during storage period for selected (T2) sample and control (commercially used fish burger patty) sample, values are mean  $\pm$  standard deviation (n=3).**

### CONCLUSIONS

The development of bugger patty using spotted triggerfish has demonstrated promising results in both sensory and nutritional quality. Among various formulations tested, bugger patty with 58% (w/w) triggerfish mince and 14% (w/w) breadcrumbs (T2) was found to be the most preferred by the consumers based on sensory analysis. This formulation offers a favourable balance of nutritional content, including 54.92 g/100 g moisture, 15.5 g/100 g crude protein, 2.11 g/100 g crude fibre, and 2.28 g/100 g ash. This development can be an alternative to high-cost fish products, potentially improving consumer access to protein-rich foods. However, we recommend continuing the

study to further evaluate the product's shelf-life. During the one-month study, all monitored parameters stayed within acceptable limits. Although the study was limited to a month, the frozen product is expected to maintain its quality for at least a year with proper storage ( $-18 \pm 2$  °C). By evaluating the nutritional profile and storage stability of the fish burger, the study aims to offer insights into its potential health benefits for consumers and its viability as a long-term, sustainable food product. The fish burger has proven an effective preservation method for spotted triggerfish, providing consumers with a convenient and tasty seafood option.

### AUTHOR CONTRIBUTION

MJPA designed the study; MWEMN conducted the experiment; MWEMN, GSNF, and HMJ analysed the data and wrote the original draft; GSNF and HMJ edited the manuscript.

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