Project ID- 489

Competitive Research Grant

Sub-Project Completion Report

on

Development of Quality Value Added Fish Products and Utilization of By-products

Project Duration

May 2017 to September 2018

Department of Fisheries Technology and Quality Control
Sylhet Agricultural University, Sylhet-3100

Submitted to
Project Implementation Unit-BARC, NATP 2
Bangladesh Agricultural Research Council
Farmgate, Dhaka-1215

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Bangladesh Agricultural Research Council (BARC)
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Acronyms

Sign	Abbreviation	Sign	Abbreviation
ANOVA	Analysis of Variance	MSG	Monosodium Glutamate
AOAC	Association of Analytical Chemists	Nacl	Sodium Chloride
APC	Aerobic Plate Count	NATP	National Agricultural Technology Program
APHA	American Public Health Association	No.	Number
BARC	Bangladesh Agricultural Research Council	PCA	Plate Count Agar
BBS	Bangladesh Bureau of Statistics	PCR	Polymerase Chain Reaction
BDT	Bangladeshi Taka	Ph.D	Doctor of Philosophy
CFU	Colony Forming Unit	Phy	Physical
cm	Centimeter	PI	Principal Investigator
C-PI	Co- Principal Investigator	PIU	Project Implementation Unit
CRG	Competitive Research Grant	PP	Project proposal
df	Degree of Freedom	SAU	Sylhet Agricultural University
DMRT	Duncan's Multiple Range Test	SD	Standard Deviation
et al.	Associated	Sig	Signature
Fig.	Figure	Sp	Species
Fin.	Financial	SPSS	Statistical Package for the Social Sciences
FRL	Food-related life style	SSC	Secondary School Certificate
GLM	General Linear Model	Tk.	Taka
gm	Gram	TPC	Total plate count
GOB	Government of Bangladesh	US	United States
Govt.	Government	USAID	United States Agency for International Development
hr	Hour	USD	United States Dollar
HSC	Higher Secondary School Certificate	USDA	United States Department for Agriculture
ICMSF	International Commission on Microbiological Specifications for Foods	%	Percentage
Kg	Kilogram	/	per
KMO	Kaiser-Meyer-Olkin	<	Less than
mg	Milligram	>	Greater than
ml	Milliliter	±	Plus and Minus
mm	Millimeter	^{0}C	Degree Celsius

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Executive Summary

Studies were conducted to develop quality value added fish products from low cost fishes of both freshwater and marine sources. Besides, consumer's behaviour and attitudes towards the newly developed products were evaluated to find out the ways for marketing of the products. Low cost fish species easily found in the market such as Pangus *(Pangasius hypophthalamus)*, Poa *(Panna microdon),* Grass carp (*Ctenopharyngodon idella*) and Tilapia *(Tilapia niloticus)* were used to produce quality fish burger, fish ball and fish sticks. Products were developed considering the preferences and taste of the native consumer. Product preparation was continued for several months in several quarters using different ingredients to develop and establish a standard of quality product highly acceptable to the consumers for the commercial production of the value added fish products. Proximate composition, organoleptic and microbiological changes of the products were determined to evaluate the nutritional value, shelf life and storage stability. All types of the products were found rich in nutrient especially in case of protein content. Prepared fish burger, fish ball and fish sticks were kept at room temperature (with and without packaging) and at refrigeration temperature. At room temperature, the proximate composition and organoleptic attributes decreased significantly throughout the storage period for both packaged and non-packaged products. On the other hand, at refrigerated temperature the product were found more stable. The microbial load was also found to increase significantly at both temperatures but the growth was slower at refrigeration temperature. The products were found to be free from *Salmonella* but in some cases *Eschersia coli* were found in samples. After considering all the results, it was concluded that, the shelf life of the developed products were very short, in fact not more than 48 hours at 28^0C. On the other hand, the shelf life for storing the fish products in refrigerated condition was found as up to 4 days.

Consumer's behaviour and attitude analysis were done for fish sticks, fish ball, fish cutlet, fish finger and fish burger for better adaption in marketing of each type of foods in Bangladesh. Mean consumption frequency of fish stick, fish ball, fish cutlet, fish finger, and fish burger were studied following 544 questionnaire survey results. The used socio-demographic variables were gender, age, social class, number of family members in the home, the presence of minors less than 18-years in the home, educational level and geographical area. Frequency distribution, factor and cluster analysis, descriptive analysis, were performed to understand the food consumption frequency and food-related lifestyle segment respectively. The statistical significance test for differences between the mean values of different foods were tested by Snedecor's F-test. Fish burger had the probability of becoming the most frequently consumed, 77% consumer's were presumed to eat fish burger once daily. Fish ball was presumed to be the second most consumed (62%) whereas, fish stick was presumed to be in the third place (59%). Fish finger was supposed to be in the fourth place of consumption frequency level. However, 24% of consumer's response indicated that they did not consume fish finger. The same types of behavior observed for fish cutlet, which was least consumed (7%) once daily, 48% once every two weeks, whereas 13% did not consume any fish product. There was no specific consumer behaviour pattern for the socio-demographic variables and type of fish products studied here.

CRG Sub-Project Completion Report (PCR)

A. Sub-project Description

1. Title of the CRG sub-project**:** Development of Quality Value Added Fish Products and Utilization of By-products

2. **Implementing organization:** Sylhet Agricultural University, Sylhet-3100, Bangladesh

3. Name and full address with phone, cell and E-mail of PI/Co-PI (s):

Principal Investigator: Professor Dr. Md. Abu Sayeed
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4. **Sub-project budget (Tk**):
   4.1 Total: Tk. 4800000
   4.2 Revised (if any): Tk. 4800000

5. **Duration of the sub-project:**
   5.1 Start date (based on LoA signed): 07 May 2017
   5.2 End date: 30 September 2018

6. **Justification of undertaking the sub-project:**

Fish is a rich source of easily digestible protein that also provides polyunsaturated fatty acids, vitamins and minerals for human nutrition. There is an increasing demand for fish and fishery products around the world in recent years due to recognition of their nutritive value. In Bangladesh, a large proportion of total landed fish (particularly marine fish) remains unused due to problems related to inherent color, flavor, texture, size and high fat content. Most of these underutilized fish belong to the abundantly available pelagic species, which are landed as by-catch and some are unconventional species such as krill. Although some species are used industrially for fish meal production, a need for their conservation and utilization for human consumption has been recognized to prevent post-harvest fishery losses. Recovery of flesh by mechanical deboning and development of value added products are probably the most promising approaches in this aspect. This research will focus on reduction of post-harvest losses and the way of product development using mince from underutilized low-cost fishery resources. These include surimi and surimi based products, extruded products and biotechnological possibilities. The dual advantages of this approach, namely, finding ways for better utilization of low value fish species and providing protein-rich convenience foods, have been pointed out. However, the key to success of this approach depends largely on the market strategies utilized and on the consumer's behavior and attitudes on such approaches.

7. **Sub-project goal**: Development of value added fish products and better utilization of by-products.

8. Sub-project objective (s):

i) Development of new fish products and their quality assurance;
ii) Proper utilization of fish by-products; and
iii) Value addition to fish product and by-product.

9. **Implementing location (s):** Sylhet and Cox's Bazar, Bangladesh

10. Methodology followed:
The following activities were performed to achieve the goal(s) and objectives of the Project:

A. Development of quality fish burger using Grass carp (*C. idella*) available in Sylhet sadar

Objectives:
The objectives of the present attempt are:
a) Development of fish burger from Grass carp meat using different recipes;
b) Assessment of proximate composition;
c) Determination of microbiological and sensory changes of burger during different storage condition.

Materials and methods
Selection of fish species:
Low priced fish Grass carp available in the local fish market was selected as the source of raw material for the preparation of fish burger. This species is selected as it can provide a good taste as well as nutrition to the fish burger. This can be used as a nutritious food in a cheaper price to the young and outgoing people for combating the malnutrition problem of Bangladesh.

Collection of fish species:
Fresh Grass Carp was collected from Kazir Bazar Fish Market (Biggest fish market in Sylhet Division) and Major Tila Fish Market. Immediately after collection, the fish was preserved with crushed ice in an insulated box and brought to the Fish Processing Laboratory of the Department of Fisheries Technology and Quality Control, Faculty of Fisheries, Sylhet Agricultural University, Sylhet. The average weight of the fishes was 1.50 to 1.80 kg and the price of the fishes was Tk 130 ± 15 per Kg.

Preparation of Grass carp burger:
Two steps were followed for the preparation of fish burger. First, mince preparation from the raw fish and then fish burger preparation from the prepared mince.

Mince preparation from the fish species:
The fresh fish were weighed, washed with clean tap water and dressed to remove the unnecessary parts. After that, the fishes were filleted and deboned manually in iced condition. Then the mince was prepared by using a mechanical mincer. All the utensils used in the experiment were cleaned with adequate washing and all the process was done by maintaining proper hygiene and sanitation. After mincing, the mince was kept in a clean small bowl that was fixed in a big plastic bowl around ice. The ratio of mince and ice was 1:2. Scheme for the preparation of fish mince from the fish species is presented in Fig. 1.

For the preparation of Grass carp fish burger with new taste and flavor mince prepared from the raw fish was mixed and ground with necessary ingredients. Addition of different ingredients and spices were done considering the acceptance of local people. The mixing was done generally for 5-7 minutes. After that, dough was formed from where burger patty was prepared manually. Then the patties were left for some times for seasoning. After seasoning, the patties were dipped in a batter formulation. Then it was fried in dip- soybean oil and was ready to eat.

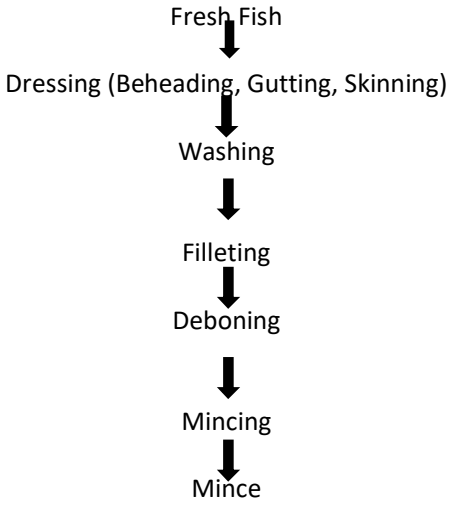

Fig. 1: Flow-diagram of fish minces preparation.

Scheme for the preparation of burger from the fish mince is presented in Fig. 2. After cooling, the burger patty was packaged in air tight polyethylene bag for different biochemical analyses.

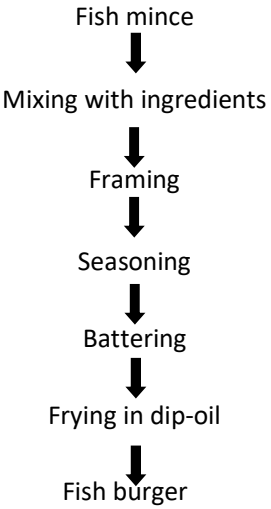

Fig. 2: Flow-diagram of fish burger preparation from the mince.

Combination of ingredients for burger preparation:

For the preparation of fish burger, Grass carp mince and potatoes were mixed with other additives and ingredients in different quantities which are given in Table 1.1. Three compositions were used to find out the best combination for the production of quality burger. In case of ingredients selection,

emphasis was given to native taste of the people so that the products could attract local consumer's acceptance.

Table 1. Ingredients used for the preparation of burger with their percentages

Ingredients	Quantity (%)		
	Formula 1	**Formula 2**	**Formula 3**
Minced meat	60	70	80
Potato	25	15	8
Onion	10	10	10
Cumin	0.3	0.3	0.3
Coriander	0.2	0.2	0.2
Garlic	1	1	1
Ginger	0.5	0.5	0.5
Green chili	1	1	1
Turmeric	1	1	1
Bread crumbles	As per need	As per need	As per need
Egg white	As per need	As per need	As per need
Wheat flour	As per need	As per need	As per need
Black seed	As per need	As per need	As per need
Common salt	As per need	As per need	As per need

Products were prepared for 5-6 quarters covering 2 in each month. Grass carp burger was randomly chosen and analyzed in triplicate for microbiological and sensory attributes as described in the following sections.

Quality assessment:

Proximate composition

Moisture, fat, protein and ash content were determined according to the method of AOAC (2000).

Microbial analysis

Microbiological examinations were carried out as per APHA (1992) methods. The microbial count was estimated by spread plate technique. One grams of sample was taken aseptically and homogenized in a sterilized mortar. The sample was homogenized with 10 ml of distilled water and then the homogenate was transferred into a sterile bottle. One ml sample was transferred with a micropipette to a test tube containing 9.0 ml of distilled water and test tube was shaken thoroughly on a vortex mixture in order to get 10^{-1} dilution of original sample solution. Preparation of sample and serial dilutions were done near the flame in a horizontal laminar flow apparatus that was pre-sterilized by ultraviolet radiation observing all possible aseptic precautions. Total plate count (TPC) was determined by spread plate method using Agar medium and were incubated at 37 °C for 24 to 48 h. *Salmonella* and *Eschersia coli* were analyzed by streaking method. The microbial count of *E. coli* and *Salmonella* spp. were estimated by spread plate technique.

Shelf life analysis

Prepared fish burgers were stored at 5°C and 28°C subsequently. Shelf life of the stored products was assessed by the determination of microbial load, chemical and sensory characteristics. Data was taken at 24 hours intervals.

Organoleptic analysis

Sensory characteristics of fish burger was evaluated by selected panel members of Sylhet Agricultural University who have experience in evaluation of similar products, on a five-point scale (Singh-Ackbarali *et al.,* 2014 and Tokur *et al.,* 2006) and scores were assigned with "1" being the least and "5" being the highest for attributes. The characteristics which were evaluated during sensory evaluation are appearance, colour, odour, flavor, taste, texture and over all acceptability.

Cost-benefit analysis

A simple cost and profit analysis was done on the basis of market survey. Total cost and net profit of 100 fish burger were calculated to identify the profitability and marketing feasibility of the product.

Statistical analysis

Factorial as well as one-way ANOVA Analysis was performed by SPSS 21 (SPSS, 2012). If the significant difference observed then Duncan's multiple range tests was done. Moreover, descriptive statistics were used in some cases.

Results and discussion:

Data were analyzed on the basis percentages, storage system, Quarter and Different time such as 24 hours, 48 hours and 72 hours duration.

Table 2 Multivariate tests results of the analyzed data

Effect	**Value**	**F**	**Hypothesis df**	**Error df**	**Sig.**
Percentage	1.19	131.36	8.00	716.00	0.00
Storage system	0.92	75.67	8.00	716.00	0.00
Quarter	0.83	23.61	16.00	1440.00	0.00
Time	0.89	37.66	12.00	1077.00	0.00
Percentage*Storage system	0.08	1.80	16.00	1440.00	0.03
Percentage*Quarter	1.52	27.62	32.00	1440.00	0.00
Percentage*Time	0.05	0.82	24.00	1440.00	0.72
Storage system*Quarter	0.15	1.79	32.00	1440.00	0.00
Storage system*Time	0.98	19.49	24.00	1440.00	0.00
Quarter* Time	0.11	0.83	48.00	1440.00	0.80
Percentage*Storage system* Quarter	0.30	1.84	64.00	1440.00	0.00
Percentage *Storage system*Time	0.08	0.64	48.00	1440.00	0.97
Percentage*Quarter*Time	0.22	0.89	96.00	1440.00	0.76
Storage system* Quarter*Time	0.17	0.66	96.00	1440.00	0.99
Percentage*Storage system*Quarter*Time	0.33	0.67	192.00	1440.00	1.00

Data revealed a significant result for most of the cases where the highest level of significance was observed in case of percentage, quarter, time and storage system. However, interaction between Percentage and Time, Quarter and Time, and among Percentage, Storage system and Time,

Percentage, Quarter and Time, Storage system, Quarter and Time, Percentage, Storage system, Quarter and Time showed no significant difference. Findings indicate quality of fish burger directly related with muscle percentage, quarter, storage time and storage systems. A composition of seventy percent muscle gave best result. The product was quite good up to two days when kept at ambient temperature. On the other hand, it was quite good condition for four days when it was stored at refrigerated temperature.

Evaluation of the quality of the burger
Proximate composition analyses

The proximate composition of the fish burger in different conditions is presented in Fig. 3,.4,.5 and 6.
In Fig. 3, mean value of moisture, lipid, protein and ash content of fish burger of different quarters are shown. Five lots of fish burger were prepared.

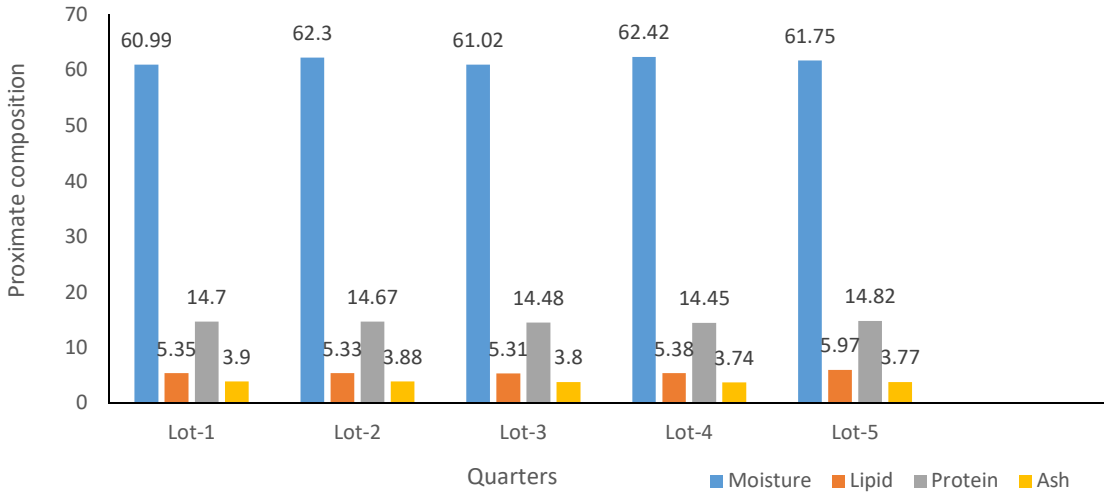

Fig. 3: Mean value of proximate composition of fish burgers at different compositions in different quarters

Proximate composition (mean value) of fish burgers of different compositions is shown in Fig. 1.4. Moisture, lipid and protein content found 67.39, 6.06 and 15.85%, respectively where 80% fish mince were used and lower 56.77, 4.78 and 13.28%, respectively where 60% fish mince were used. But ash content was found higher (4.79%) in burgers made with 60% fish mince and lower (2.94%) in burgers made with 80% fish mince. Higher amount of moisture in burgers made with 80% fish mince might be due to addition of lower amount of potatoes and other food additives. Higher amount of protein was found in burgers made with 80% fish mince as it is prepared with higher amount of fish mince. As this is a new idea, there is no reference regarding this finding.

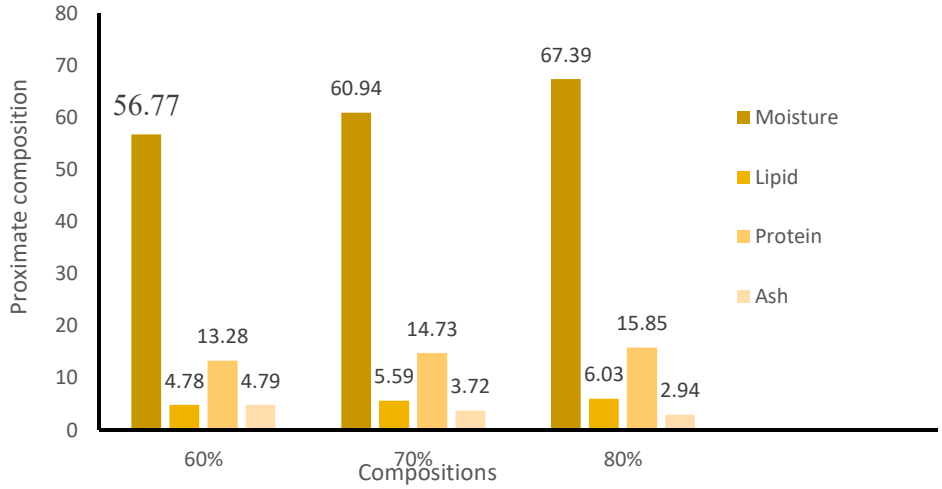

Fig.4: Proximate composition of fish burgers at different compositions

Proximate composition of fish burgers at different storage conditions are shown in Fig.5. Burgers kept in refrigerated condition showed lower amount of moisture (61.29%) and higher amount of lipid (6.66%), protein (15.08%) and ash (3.84%). Lower amount of lipid, protein and ash were found in burgers which were kept in with and without packaging. No significant difference observed in with and without packaging indicates that packaging has no effect on moisture level.

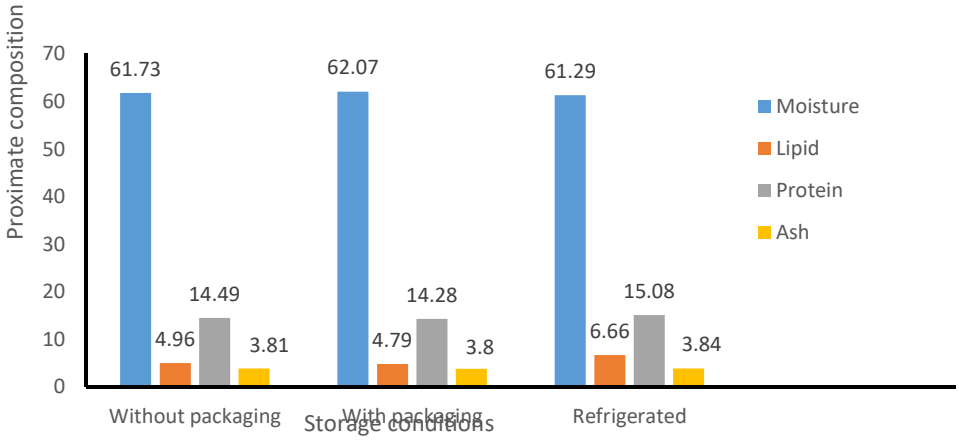

Fig.5: Proximate composition of fish burgers of different compositions at different storage conditions.

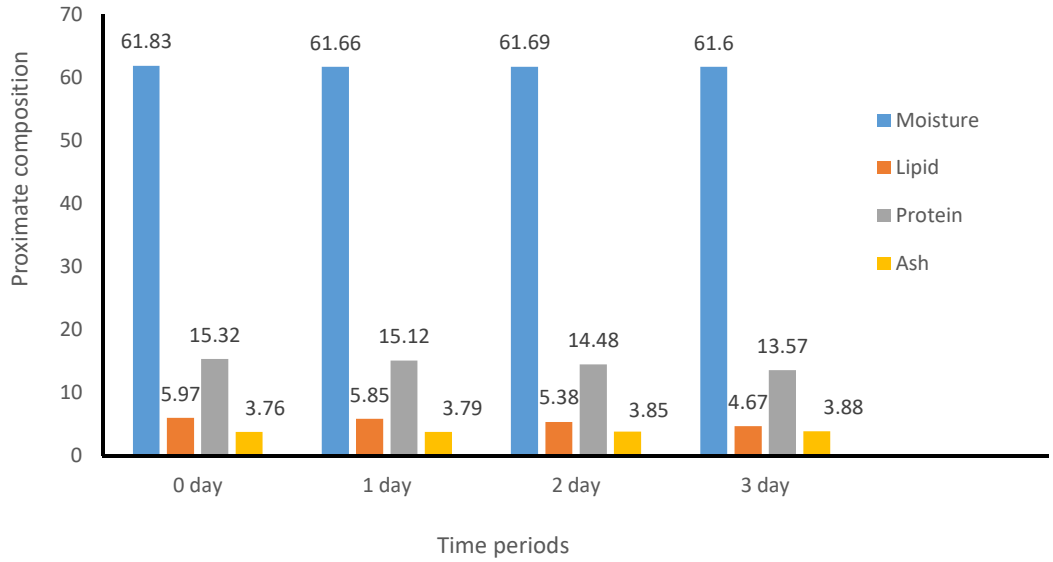

Fig.6: Proximate composition of fish burgers using different muscle level under different time periods

Lipid and protein content of the prepared burgers were found highest in fresh condition (Fig.6). Decrease of the lipid and protein content and increase of ash content were observed due to aging of products. Therefore, it is concluded that produced burgers kept in refrigerated temperature with wrapping could be a better option for increasing profitability as its proximate conditions remain in good condition.

Microbial analysis:

Changes in bacterial load (Log CFU/g) of Grass carp burger of different compositions in different storage periods and storage conditions are shown in Table 1.3. Bacterial load was found higher in burgers made of 70% fish mince and lower in 60% fish mince but TPC did not exceed the maximum levels (7 Log CFU/g of meat) of microbiological criteria for fresh and frozen fish given by the ICMSF (1978). TPC was found to increase throughout the storage period. Initial bacterial load of fish burger prepared from Grass carp fish mince was 3.36±.71Log CFU/g. Bacterial growth in fish burger kept at room temperature found to increase rapidly with the progress of storage time with a trend of increase to 4.51±1.06 Log CFU/g after 24 hours that reached to 6.80±1.42Log CFU/g after 72 hours.

Table 3. TPC (Mean ± SD) of fish burgers of different compositions at different time periods and storage conditions

Compositions (%)	60%	4.86±1.83 Log CFU/g
	70%	5.30±1.90Log CFU/g
	80%	5.14±1.72Log CFU/g
Storage periods (day)	day 0	3.36±.71Log CFU/g
	day 1	4.51±1.06Log CFU/g
	day 2	5.75±1.42Log CFU/g
	day 3	6.80±1.42Log CFU/g
Storage conditions	Without packaging	6.00±2.14Log CFU/g
	With packaging	5.24±1.60 Log CFU/g
	Refrigerated	4.07±0.99 Log CFU/g

The changes in the total plate count (TPC) during storage were enumerated and the results are presented in Table 3. Lower TPC was observed during refrigerated storage from the initial value of 6.00±2.14 Log CFU/g to 4.07±0.99 Log CFU/g. Liston (1980) observed that freezing generally causes a reduction in bacterial count and the number will continue to fall during storage. Neither bad smell nor fungal growths were apparent in the product during that period (Shammi, 2005).

Table 4. Status of *E. coli* and *Salmonella* in Grass carp fish burgers

Sample Source	**No of positive sample (%)**	
	E. coli	***Salmonella***
60% (n=25)	08 (32%)	0 (0%)
70% (n=25)	05 (20%)	0 (0%)
80% (n=25)	05 (20%)	0 (0%)

From Table 4, fish burger was found to be free from *Salmonella* but *E. coli* was found in the sample. Higher amount was observed in burgers made with 60% fish mince and lower amount found in burgers made with 70 and 80% fish mince.

Sensory and shelf life evaluation of the burger

The overall acceptability scores of fish burger of different compositions during different storage condition and temperature are presented in Table 5, 6 and 7. Freshly prepared products from 3 compositions received an initial score above 4.50 and were rated as good. But considering the flavor, taste and texture preferences of the selected panelists and some other consumer's fish burger made with 70% fish composition was rated the best and the preparation recipe was suggested for the commercial production of the burgers.

Changes in general appearance, flavor, taste and texture of burger both packaged and without packaged stored at room (28°C) and refrigerated temperatures (5°C) are shown in Table 5, 6 and 7. All the sensory attributes were decreased with the progress of storage period at room temperature. The initial prominent fresh sweet odour of fish burger became gradually pungent followed by a sour odour accompanied by moist slimy surface with fungal growth at 72 hours 28°C, which was indicative of large bacterial growth. Bad smell observed at 48 hours of storage time and fungal growth was visible at the storage time of 72 hours. Although a slight pungent odour was felt at 24 hours. According to the statistical analysis, there was no significant differences ($P>0.05$) in colour, odour and texture between 0 hour to 24 hours of storage period with exception of taste and general appearance properties.

On the other hand, at refrigerated temperature (5°C) the product was found more stable. Colour, taste and general appearance of the products did not changed markedly even after 72 hours ($P>0.05$). A slight pungent odour felt at 72 hours but it was not considered as unacceptable. Fungal growth also was not found after 72 hours. The textural quality changed significantly ($P<0.05$) at 48 and 72 hours but they also considered as acceptable. The results of the study are in agreement with the results of Shammi (2005) which is also supported by the results of Koelkar and Pagarkar (2013). They evaluated organoleptic characteristics such as color, taste, odor, appearance, texture and overall acceptability of fish ball prepared from *Catla Catla* kept in chilled storage (0 to -2°C) and observed a declined trend of organoleptic scores from 9.0 to 3.1 for the storage period of 0 to 12 days. Besides, they stated that fish ball in curry kept in chilled storage was not acceptable after 9 days. Ejaz *et al.* (2009) also reported that at room temperature, the sensory attributes decreased significantly ($P<0.05$) throughout the storage period. Storage estimated shelf life of fish burger were

about 10 days while correlating the sensory scores with the storage period of fish burger in the refrigerator.

Table 5. Changes of sensory attributes fish burger made with 60% fish stored at room and refrigerated temperatures in different storage conditions

Storage Condition	Storage Period (Day)	Appearance	Flavour	Taste	Texture	Overall
Without packaging (28^0c)	0	4.71±0.48	4.69±0.48	4.70±0.00	4.75±0.30	4.50±0.00
	1	3.85±0.37	3.42±0.53	4.00±0.00	3.71±0.48	3.57±0.53
	2	2.71±0.48	2.57±0.78	2.85±0.69	2.57±0.53	2.42±0.33
	3	1.14±0.37	1.00±0.57	1.42±0.53	1.85±0.37	1.00±0.57
With packaging (28^0c)	0	4.71±0.48	4.69±0.48	4.70±0.00	4.75±0.30	4.80±0.00
	1	3.85±0.37	3.71±0.48	3.85±0.37	3.71±0.48	3.71±0.48
	2	2.85±0.69	3.00±0.57	2.90±0.81	2.85±0.37	2.71±0.28
	3	2.28±0.48	1.14±0.69	1.57±0.53	2.28±0.75	1.14±0.37
Refrigerated (5^0c)	0	4.71±0.48	4.69±0.48	4.70±0.00	4.75±0.30	4.75±0.37
	1	4.24±0.37	4.28±0.48	4.00±0.00	3.42±0.53	4.57±0.53
	2	3.15±0.69	3.71±0.48	3.93±0.37	3.57±0.79	3.37±0.43
	3	3.04±0.37	3.54±0.69	3.71±0.48	3.43±0.57	3.20±0.57

1=Extremely dislike; 2=Dislike;3 =Average; 4=like; 5=Extremely like
Each value is represented as the mean ± SD of n=7

Table 6. Changes of sensory attributes fish burger made with 70% fish stored at room and refrigerated temperatures in different storage conditions

Storage Condition	Storage Period (Day)	Appearance	Flavour	Taste	Texture	Overall
Without packaging (28^0c)	0	4.57±0.53	4.71±0.48	4.92±0.53	5.00±0.00	4.61±0.24
	1	3.71±0.49	3.57±0.53	3.14±0.38	3.43±0.53	3.42±0.53
	2	2.29±0.49	2.00±0.00	2.00±0.00	2.57±0.53	2.00±0.00
	3	1.71±0.49	1.00±0.00	1.14±0.37	1.57±0.53	1.14±0.37
With packaging (28^0c)	0	4.57±0.53	4.71±0.48	4.92±0.53	5.00±0.00	4.82±0.28
	1	4.00±0.58	3.42±0.53	3.14±0.37	3.71±0.48	3.42±0.53
	2	2.71±0.49	2.00±0.00	2.29±0.49	2.42±0.53	2.28±0.48
	3	2.00±0.57	1.42±0.53	1.71±0.75	1.85±0.37	1.42±0.53
Refrigerated (5^0c)	0	4.57±0.53	4.71±0.48	4.92±0.53	5.00±0.00	4.85±0.45
	1	4.29±0.48	4.57±0.53	4.71±0.48	4.00±0.00	4.60±0.00
	2	3.71±0.48	3.57±0.78	3.89±0.37	3.14±0.37	3.42±0.53
	3	3.29±0.75	2.85±0.69	3.28±0.48	2.85±0.37	3.30±0.00

1=Extremely dislike; 2=Dislike; 3 =Average; 4=like; 5=Extremely like
Each value is represented as the mean ± SD of n=7

Table 7. Changes of sensory attributes fish burger made with 70% fish stored at room and refrigerated temperatures in different storage conditions

Storage Condition	Storage Period (Day)	Appearance	Flavour	Taste	Texture	Overall
Without packaging (28^0c)	0	5.00±0.00	4.57±0.53	4.74±0.00	4.85±0.37	4.71±0.48
	1	3.42±0.53	3.14±0.37	4.00±0.00	3.28±0.48	3.57±0.53
	2	2.57±0.53	2.14±0.37	3.14±0.37	2.57±0.53	2.71±0.48
	3	2.14±0.69	2.00±0.00	2.85±0.37	1.85±0.37	2.28±0.48
With packaging (28^0c)	0	5.00±0.00	4.57±0.53	4.74±0.00	4.85±0.37	4.79±0.33
	1	3.71±0.48	3.42±0.53	4.00±0.00	3.28±0.48	3.42±0.53
	2	2.57±0.53	2.71±0.48	3.14±0.37	2.28±0.48	2.57±0.78
	3	2.28±0.48	2.14±0.69	2.42±0.53	1.85±0.69	2.00±0.57
Refrigerated (5^0c)	0	5.00±0.00	4.57±0.53	4.74±0.00	4.85±0.37	4.88±0.27
	1	4.57±0.53	4.28±0.48	4.71±0.48	4.57±0.53	4.62±0.53
	2	3.28±0.48	3.78±0.48	3.70±0.48	3.11±0.48	3.56±0.69
	3	3.14±0.37	3.00±0.00	3.28±0.48	3.42±0.53	3.14±0.37

1=Extremely dislike; 2=Dislike; 3 =Average; 4=like; 5=Extremely like
Each value is represented as the mean ± SD of n=7

Cost-benefit analysis of Grass carp fish burger

A simple cost-benefit analysis was done on the basis of market survey shown in Table 8. It was done for 100 fish burger. About 3440g fish mince was required for the production of 100 fish burger, which was obtained from approximately 10 kg fish. The production cost/Grass carp fish burger was 21.60 taka. The maximum retail price for the product was set as 40 taka. A net profit of 1840 taka was obtained from the product in the analysis. The margin of profit was about 85.19%.

As the margin of profit of the Grass carp fish burger was very high and most of the consumer responded positively. Therefore, it can assumed that business with value added products like Grass carp fish burger in Bangladesh has very good prospect and it would be economic benefit to the producer. Therefore, it is concluded that considering the above facts the fish burger produced with 70% mince of grasp carp fish could be a good package.

Plate 1: Grass carp fish burger (without packaging) stored at room temperature in different storage periods (hr). a=ohr, b=24hrs, c=48 hrs and d=72 hrs

Table 8. Cost and benefit analysis of fish burger prepared from Grass carp

Cost				Profit				
Items	**Unit Cost (Tk.)**	**Amount**	**Total cost (Tk.) including vat and tax**	**Amount**	**Unit Price (Tk.) including vat and tax**	**Total Price (Tk.) including vat and tax**	**Net profit (Tk.)**	**% Profit**
Grass carp fish	130Tk/kg	10 kg	1365	100 pcs	40	4000	1732	76.37
Fish mince		3440g						
Ingredients and bread			903					
Total			2268					

B. Development and qualitative evaluation of fish ball using Poa *(Panna microdon)* under different storage conditions

Objectives of the study

The study was conducted to achieve the following objectives:

i) To diversify fish products by developing fish ball from Poa (*Panna microdon*); and
ii) To evaluate the quality of ball for offering a quality product to the consumers.

Materials and methods

Fish collection and fish ball preparation

Premium quality Poa fish (*Panna microdon*) was collected from the different fish markets of Chittagong, Bangladesh. The average size and weight of the fish were 22 ± 3 cm and 0.75 ± 0.10 kg, respectively. Immediately after collection, fish were preserved properly with crushed ice(fish: ice= 1:2)and transported to the laboratory of Fisheries Technology and Quality Control, Sylhet Agricultural University, Sylhet-3100, Bangladesh in a triple layered insulated box to ensure maximum freshness. Having been transferred to the laboratory, the fish was frozen and stored at -18° C for 36 hrs. Fish were then beheaded, eviscerated, skinned and washed with clean potable water. The skinned fish were filleted and deboned manually in iced condition. Then the mince was prepared by a mechanical mincer through a 1.0 mm orifice diameter to remove all bones and connective tissues from the muscles. Conventional standards were utilized for the preparation of fish mince according to the US Department of Agriculture (USDA, 2001). In addition to this, minced fish was washed in ice-cold water (2±1°C) with a ratio of 1.5:2 (mince-meat to water), to eliminate the mossy odor and afterward, the samples were dewatered by squeezing manually. The above-mentioned activities were performed in a chilled (<4^{o}C) room. All the utensils used in the experiment were cleaned with adequate washing and kept cool (4°C). Crushed ice was used to maintain adequate temperature throughout the product preparation.

Preparation of fish ball

The ingredients used for the preparation of fish balls are given in Table 2.1. At first ginger, garlic, chili powder, and onion are fried slightly with oil. Then the fried spicy mixed properly with minced meat and mashed potato and then again the whole mixture was fried again slightly. Then fried minced fish was coated with egg followed by biscuits crumbs made ball shaped. Fish ball thus prepared was fried in fresh soybean oil (Datta *et al.,* 2015). Then the products were kept at refrigerator (5°C) and at room temperature (28°C) as well. The whole procedure is of fish ball preparation is given in Fig. 2.1, 2.2; plate-02and Table 2.1. Potatoes were used as a source of good starch while egg was used as a binder as well as a source of protein. Biscuit crumbs were used according to Chowdhury *et al.,* 2017 and Datta *et al.,* 2015to make the fish ball crispy.

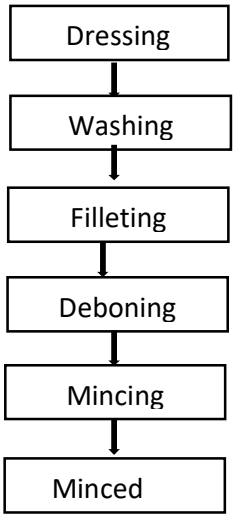

Fig. 7: Flow-diagram of fish minces preparation.

Table 9. Ingredients of fish ball with their percentage according to Datta *et al,* (2015) with slight modification

Name of ingredients	**Percentage (%)**
Minced fish	75
Egg	3.5
Biscuit crumbles	5
Ginger	1
Turmeric	1
Water	2.5
Soybean oil	3
Garlic powder	4
Onion flakes	2
Salt	2
Chili	1

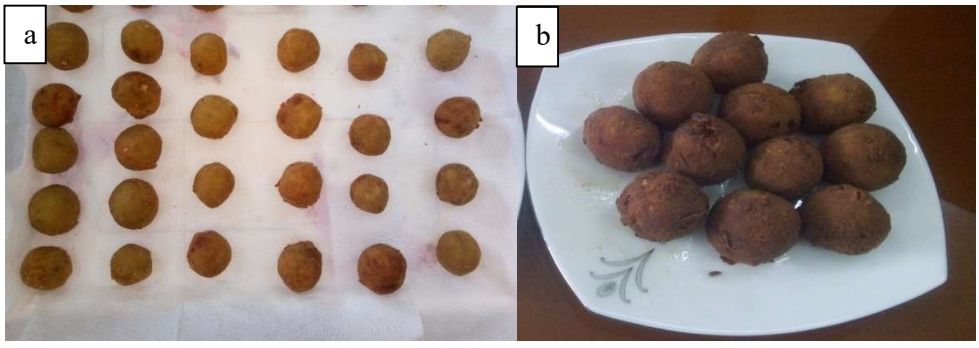

Plate 2: (a) Ready to eat fish ball; (b) Ready to serve fish ball

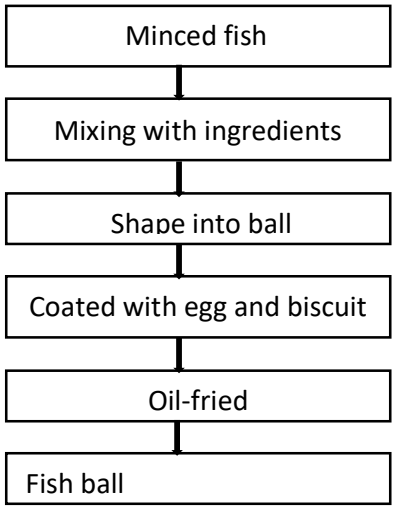

Fig. 8: Flow-diagram of fish ball preparation (Datta *et al.,* 2015).

Organoleptic analysis

A taste panel of 7 (seven) experienced panelists of the Department of Fisheries Technology and Quality Control, SAU formed for organoleptic test. From the stored products, fish balls were randomly chosen and from each packed sample were analyzed to evaluate organoleptic attributes. Samples were scored according to their quality. Samples were taken four times at a 24 hours interval for a period of 72 hours. All the samples were served to 7-panel members to evaluate the attributes like appearance, flavor, taste, texture and overall acceptability) of the samples by using 5-points descriptive scale (Singh-Ackbarali *et al*., 2014 and Tokur *et al.,* 2006). Scores were assigned with '1' being the least and '5' being the highest for attributes. Scores 5, 4, 3, 2, 1 were taken for like extremely, like, neither like nor dislike, dislike, dislike extremely respectively for each of the organoleptic characteristics.The observations were converted to equivalent numerical scores (M. Vanitha *et al*, 2015 and Tokur *et al.,* 2006).Another organoleptic evaluation was conducted by making a Likert scale based semi-structured questionnaire. For each attribute (appearance, taste, color, general acceptability) 50 questionnaire were solved by surveying different consumer groups (businessman as high-income consumers, service holder as middle-income consumers and student as no/ low-income consumers) and their opinion on each specific attribute was collected. The monthly income ranges of businessman, service holder and student were ≥40000 BDT, 20000-39000 BDT and ≤19000 BDT, respectfully. We choose service holder, businessman and student as consumers because in Bangladesh context these play a vital role in daily products market. Different question was asked to them like as what was the appearance (oily, grainy, wrinkle, shiny, crunchiness and other), flavor (rancid, egg smell, soapy, fishy, sweet note and other), taste (Salty, bitter, spicy, vinegary, pungent and other) and textural (elastic, spongy, juicy, sandy, compact and other) quality of the prepared fish ball, what was the overall acceptability of the products (accepted or rejected), did the quality of the fish ball satisfy them and whether it is necessary to developed fish ball from Poa fish. Their opinion about these questions was analyzed on the basis of age, gender and occupational status.

Table 10. Characteristic features of Demographic study

Parameter	Sub-parameter	Quantity	Percent (%)
Gender	Male	35	70
	Female	15	30
Age	20-25	11	22
	25-30	7	14
	30-35	6	12
	35-40	15	30
	>40	11	22
Marital Status	Married	18	36
	Single	32	64
Education	PhD	4	8
	Master	20	40
	Bachelor	21	42
	HSC	3	6
	SSC	2	4
Religion	Muslim	38	76
	Hindus	12	24
Occupation	Businessman	12	24
	Service holder	20	40
	Student	18	36
Price Preference	High	3	6
	Middle	32	64
	Low	15	30

Proximate composition analysis

Proximate composition analysis of moisture, crude protein, crude lipid and ash were carried out according to the methods of Association of Analytical Chemists (AOAC, 1980) with certain modification.

Bacterial load analysis

Standard plate count expressed as Colony Forming Units per gram (log cfu g-1) of dry fish were determined by using consecutive decimal dilution technique using spread plates (Fig. 2.3). One gram of sample was taken aseptically and homogenized in a sterilized mortar. The sample was homogenized with 10 ml of distill water and then the homogenate was transferred into a sterile bottle. One ml sample was transferred with a micropipette to a test tube containing 9.0 ml of distill water and test tube was shaken thoroughly on a vortex mixture in order to get 10^{-1} dilution of original sample solution. Using the similar process several dilutions of 10^{-2}, 10^{-3}, 10^{-4}, 10^{-5}, 10^{-6} and so on were made.

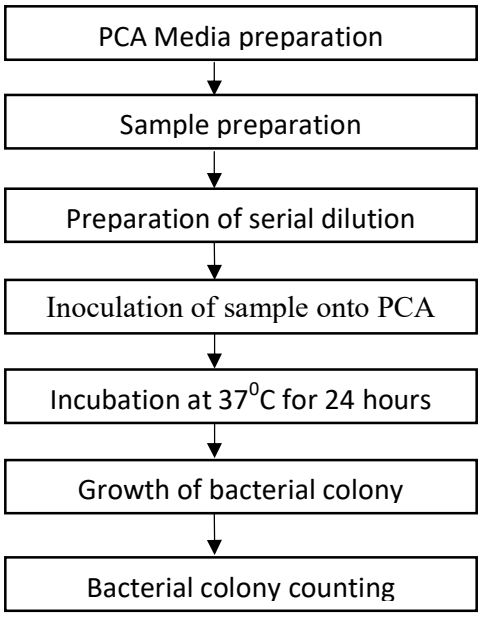

Fig. 9: Flow diagram showing the procedure of total bacterial count

All plates in duplicate on sterile Petri dish were done on sterile plate count agar media. From sample solution of different test tubes bearing varying dilution, 0.1 ml aliquot was taken by a micropipette and transferred aseptically in to the pre-prepared agar plates by raising the upper lid sufficient enough to enter the lip of the pipette. The samples were then spread homogenously and carefully by sterile flamed L-shaped glass rod throughout the surface of the media until the sample were dried out. The plates were incubated at 370C in an inverted position in an incubator. After 48 hours of incubation colonies developed on the Petri dish was counted following a standard method.

The concentration of bacteria in the original samples was calculated by multiplying the colony counts by the total dilution factor. Only plates that contain between 30 and 300 colonies were used to calculate the original concentration. Because, counts less than 30 are not considered to be statistically reliable, and if the number exceeds 300 colonies, it is likely that colony formation by some bacteria will be suppressed, leading to an underestimation of the actual bacterial numbers. Sometimes two plates had colony counts between 30 to 300, and for this, the concentrations calculated for each plate was averaged.

The formula for counting bacteria are as follows-

$$\textbf{Cfu g}^{-1} = \frac{\text{No.of colonies on Petri dish} \times 10 \times \text{dilution factor} \times \text{Volume of the total sample solution}}{\text{Weight of fish sample (g)}}$$

Statistical Analysis

One-way ANOVA for nutritional, microbiological and organoleptic analysis was performed by SPSS (IBM 2010 and Version 20) at 5% confidence level. If main effects differ significantly then Duncan's Multiple Range Test (DMRT) was done.

Results and Discussions

Prepared fish ball was analyzed for organoleptic attributes, bacterial load, and proximate composition. Total plate counts by using ten-fold serial dilution method were done for quantitative assessment of microbial status of the fish ball. The results are presented and summarized both in tabulated and graphical form.

Organoleptic analysis

Effect of storage periods on organoleptic attributes of fish ball prepared from Poa fish (*Panna microdon*) under different storage conditions is shown in Table 2.3. All the sensory qualities were found to decrease with the progress of the storage period ($p<0.05$) at room temperature. The initial prominent fresh sweet flavor of fish ball became gradually pungent followed by a sour odor accompanied by moist slimy surface with fungal growth after 72 hours at 28°C, which was indicative of large microbial growth. Bad smell observed at 48 hours of storage time and fungal growth was visible at the storage time of 72 hours, although a slightly pungent odor was felt during 24 hours. After 72 hours all the attributes appearance, flavor, taste, texture and overall acceptability of fish ball were considered as "extremely dislike" according to their scores and it was not acceptable. On the contrary, at refrigerated temperature (5°C) the product was more stable. The appearance, flavor, taste, texture and overall acceptability did not change considerably even after 72 hours ($p>0.05$). Fungal growth also was not observed after 72 hours. The textural quality changed significantly ($p<0.05$) at 48 and 72 hours but considered as acceptable. After 72 hours all the attributes appearance, flavor, taste, texture and overall acceptability of fish ball were considered as "averagely like" according to their scores and it was acceptable. The appearance, taste and overall acceptability of fish ball stored at 28° C were significantly different with the appearance, taste and overall acceptability of the same fish ball stored at 5° C after 24, 48 and 72 hours, respectively. Whereas the flavor and texture of the fish ball stored at 28° C were significantly different with the flavor and texture of the same fish ball stored at 5° C only after 48 and 72 hours, respectively.

The results of this study found consistent with the results of Koelkar and Pagarkar (2013). They evaluated organoleptic characteristics such as color, taste, odor, appearance, texture and overall acceptability of fish ball prepared from *Catla Catla* kept in chilled storage (0 to -2°C). They observed a declined trend of organoleptic scores from 9.0 to 3.1 for the storage period of 0 to 12 days. Besides, according to this, fish ball in curry kept in chilled storage was not acceptable after 9 days. Ejaz *et al*. (2009) also reported at room temperature, the attributes decreased significantly ($p<0.05$) throughout the storage period. This is also similar to the findings of the present study. With the increase in the population, there is a change in the people's attitude to consume foods that are ready to eat and have nutritional value. From the results, it was observed that fish ball was acceptable condition till the end of storage periods of 3 days.

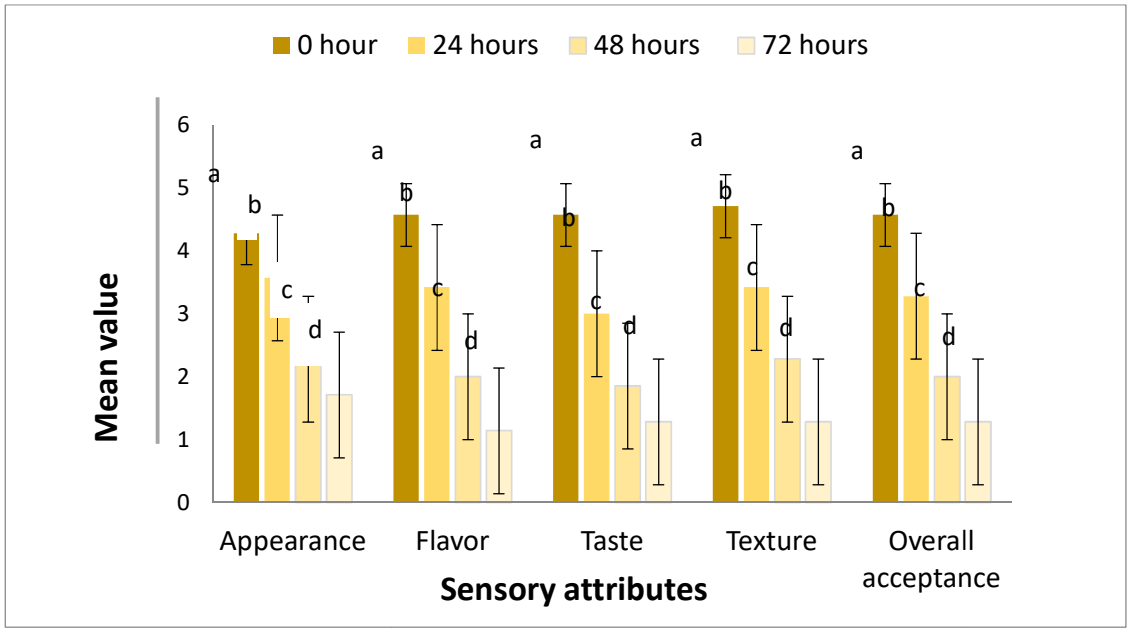

Fig. 10: Sensory attributes changes of fish ball stored at 28°C temperature.

Means followed by the same superscript within different time periods of the specific attribute (appearance, flavor, taste, texture and overall acceptance) are not significantly different ($p > 0.05$).

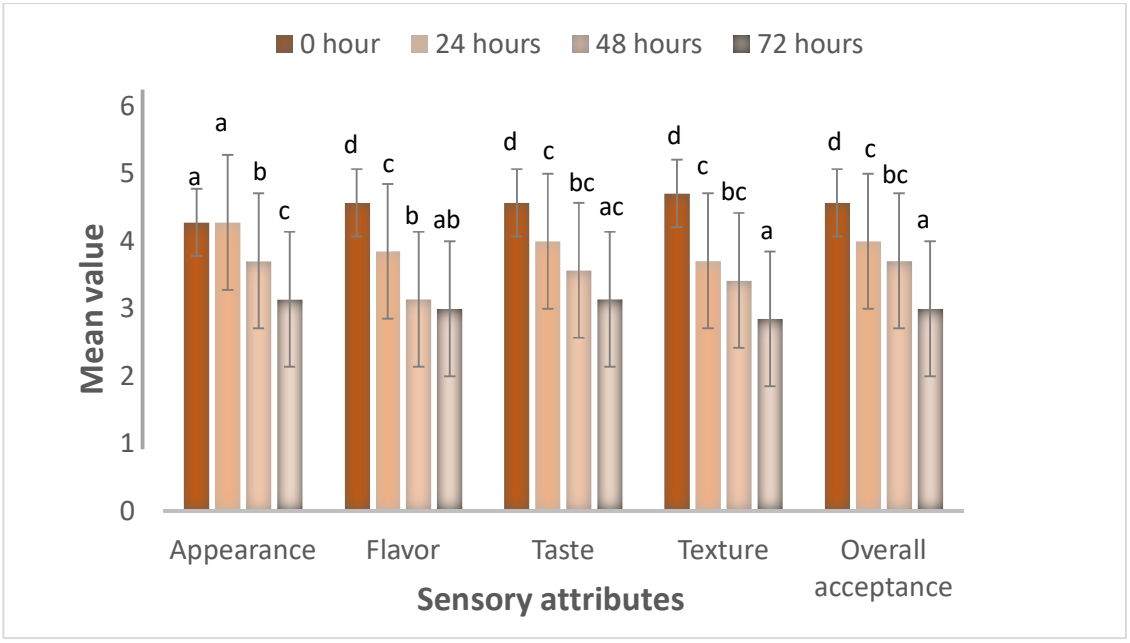

Fig. 11: Sensory attributes changes of fish ball stored at 5°C temperature.

Means followed by the same superscript within different time periods of the specific sensory attribute (appearance, flavor, taste, texture and overall acceptance) are not significantly different ($p > 0.05$).

Table 11. Effect of storage periods on sensory attributes of fish ball prepared from Poa fish (*Panna microdon*) during different storage conditions

Sensory attributes	Storage periods	Storage condition	
		Room temperature (28° C)	**Refrigerated temperature (5° C)**
Appearance	0	4.28± 0.48^{a}	4.28±0.48^{ad}
	24	3.57±0.53^{b}	4.28±0.48^{c}
	48	2.28±0.48^{c}	3.71±0.48^{b}
	72	1.71±0.48^{d}	3.14±0.37^{a}
Flavor	0	4.28± 0.48^{a}	4.28±0.48^{ad}
	24	3.57±0.53^{b}	3.85±0.37^{bc}
	48	2.28±0.48^{c}	3.14±0.37^{b}
	72	1.71±0.48^{d}	3.00±0.00^{a}
Taste	0	4.57±0.53^{a}	4.57±0.53^{ad}
	24	3.00±0.00^{b}	4.00±0.00^{c}
	48	1.85±0.37^{c}	3.57±0.53^{b}
	72	1.28±0.48^{d}	3.14±0.37^{a}
Texture	0	4.71±0.48^{a}	4.71±0.48^{ad}
	24	3.42±0.53^{b}	3.71±0.48^{bc}
	48	2.28±0.48^{c}	3.42±0.53^{b}
	72	1.28±0.48^{d}	2.85±0.37^{a}
Overall acceptability	0	4.57±0.53^{a}	4.57±0.53^{ad}
	24	3.28±0.48^{b}	4.00±0.00^{c}
	48	2.00±0.57^{c}	3.71±0.48^{b}
	72	1.28±0.48^{d}	3.00±0.00^{a}

1= Extremely dislike; 2= Dislike; 3 = Neither like nor dislike; 4 = like; 5 = Extremely like
Each value is represented as the mean ± SD of n = 7
Means followed by the same superscript within a row are not significantly different ($P > 0.05$).

Table 12. Terms generated by the participants to describe the sensations perceived during consumption of fish ball grouped by frequency of mention

Sensory attributes	Frequency (50)	Percentage (%)
Appearance		
Oily	11	22.9
Grainy	6	12.5
Wrinkle	4	8.3
Shiny	14	29.2
Crunchiness	6	12.5
Other (Pale, intense and dark)	7	14.6
Texture		
Elastic	5	10.4
Spongy	7	14.5
Juicy	8	16.7
Sandy	2	4.2
Compact	16	29.2
Others (Easy to chew, pasty, soft, gummy)	12	25
Flavor		
Rancid	1	2.1
Egg smell	14	29.2
Soapy	3	6.3
Fishy	17	35.4
Sweet note	2	4.2
Others (Tasty, Intense odor, Undercooked)	11	22.9
Taste		
Salty	13	27.1
Bitter	3	6.3
Spicy	13	27.1
Vinegary	5	10.4
Pungent	3	6.3
Others (Fish taste, Egg taste, Tasteless)	11	22.9
Overall acceptance		
Accepted	40	83.3
Rejected	8	16.7

A question “did the quality of the prepared fish ball satisfy them and is it necessary to developed & marketed fish ball from Poa fish” (Yes/No) was asked to the surveyed consumers based on gender, age and occupational status. The frequency of their opinion is presented below-

The frequency of gender surveyed is given in Table 2.5. Survey reveals that 16.7% (75 – 58.3) of the male gender was more satisfied with fish ball and wanted the product into the market.

Table 13. Frequency of gender surveyed

Gender	Evaluation	Frequency	Percent	Valid percent	Cumulative Percent
Male	Yes	18	75	75	75
	No	6	25	25	100
	Total	24	100	100	
Female	Yes	14	58.3	58.3	58.3
	No	10	41.7	41.7	100.0
	Total	24	100.0	100.0	

Age based survey for product satisfaction is given in Table 2.6. Findings reflects that 20 to 25 age people were more satisfied with our products quality and wanted fish ball in the market. It is because of people belongs to age group between 20 and 25 tends to consume more fast food on the basis of prevailing trend and food habits. People in this age group prefer more convenient and easily available food.

Similiar occupation based status survey is for product satisfaction (Table 2.7). Data reveals that mostly service holders (75%) were more agreed with the product quality and they want these products in the market more than student and businessman category. It is because of working class people in their work breaks grab quick food at fast food joints and students closely follow service holders.

Table 14. Age based product satisfaction

Age	Evaluation	Frequency	Percent	Valid percent	Cumulative percent
20-25	Yes	13	81.3	81.3	81.3
	No	3	18.8	18.8	100.0
	Total	16	100.0	100.0	
25-30	Yes	11	68.8	68.8	68.8
	No	5	31.3	31.3	100.0
	Total	16	100.0	100.0	
30-35	Yes	10	62.5	62.5	62.5
	No	6	37.5	37.5	100.0
	Total	16	100.0	100.0	
35-40	Yes	9	56.3	56.3	56.3
	No	7	43.8	43.8	100.0
	Total	16	100.0	100.0	
> 40	Yes	8	50	50	50
	No	8	50	50	100.0
	Total	16	100.0	100.0	

Table 15. Occupation based product satisfaction

Occupation	Evaluation	Frequency	Percent	Valid percent	Cumulative percent
Businessman	Yes	7	43.8	43.8	43.3
	No	9	56.3	56.3	100.0
	Total	16	100.0	100.0	
Service holder	Yes	12	75	75	75
	No	4	25	25	100
	Total	16	100	100	
Student	Yes	10	62.5	62.5	62.5
	No	6	37.5	37.5	100.0
	Total	16	100.0	100.0	

Proximate composition analysis

Effect of storage periods on proximate composition of fish ball made by Poa fish (*Panna microdon*) under storage conditions is presented in Table 2.8. At room temperature (28^0C) the moisture and protein content found to decrease gradually which can be attributed to the leaching out of the water-soluble nitrogenous components, during storage along with moisture (Vanitha *et al.*, 2015). Moisture content was 67.93±0.16 at 0 hours which decreased to 66.23±0.20after 72 hours. Moisture content at 0 hours was significantly different ($p<0.05$) with the moisture content after 48 hours (66.92±0.07) and 72 hours (66.23±0.20) but not significantly differed ($p<0.05$) with moisture content after 24 hours (67.48±0.36). Protein content at 0 hours was 18.35±0.33 and after 72 hours was 17.29±0.08. The protein content at 0 hours (18.35±0.33) significantly differed ($p<0.05$) with protein content after 48 hours (17.62±0.22) and 72 hours (17.29±0.08).

In the case of lipid and ash, both were found to increase with the progress of storage time. Such increase in fat can be attributed to the decrease in moisture content as they are inversely proportional (Vanitha *et al.*, 2015). Lipid content at 0 hours (4.28±0.07) was significantly different ($p<0.05$) with lipid content after 24 hours (5.05±0.11),48 hours (5.39±0.09) and 72 hours (5.70±0.04) respectively. Whereas, ash content at 0 hours (3.86±0.03) was significantly different with ash content after 48 hours (4.44±0.09) and 72 hours (4.87±0.14) but not 24 hours (4.03±0.09).

In case of refrigerated temperature (5^0C), moisture content was found decreasing manner but in the other three cases protein, lipid and ash content were found to increase slowly. The moisture, lipid, protein and ash content of fish ball at 0 hours were 67.93±0.16, 4.28±0.07, 18.35±0.33 and 3.86±0.03, respectively. After 72 hours the lipid, protein and ash content were observed 5.83±0.07, 19.00±0.16 and 4.29±0.08, respectively. Only protein content after 24, 48 and 72 hours at 28°C were found significantly different with the protein content at 5°C after 24, 48 and 72 hours, respectively. The others (Lipid, ash and moisture) content at 28°C were not significantly different with lipid, ash and moisture content at 5° C after 0, 24, 48 and 72 hours, respectively. Sandhya Rani *et al* (2017) observed an increase in lipid, protein and ash content of fish finger produced from mince of Mrigal (*Cirrhinus Mrigala)* during frozen storage. Authors also reported that the moisture content of fish finger was gradually decreased during frozen storage. So, the result of the present study is in agreement with the results of Sandhya Rani *et al* (2017). In this study, the moisture content was found decreased. Sandhya Rani *et al* (2017) and Vanitha *et al* (2015) reported that the moisture content of fish fillet produced from Mrigal (*Cirrhinus Mrigala) and fish burger producd from Catla (Catla Catla) was decreased during frozen storage is supports the present finding.*

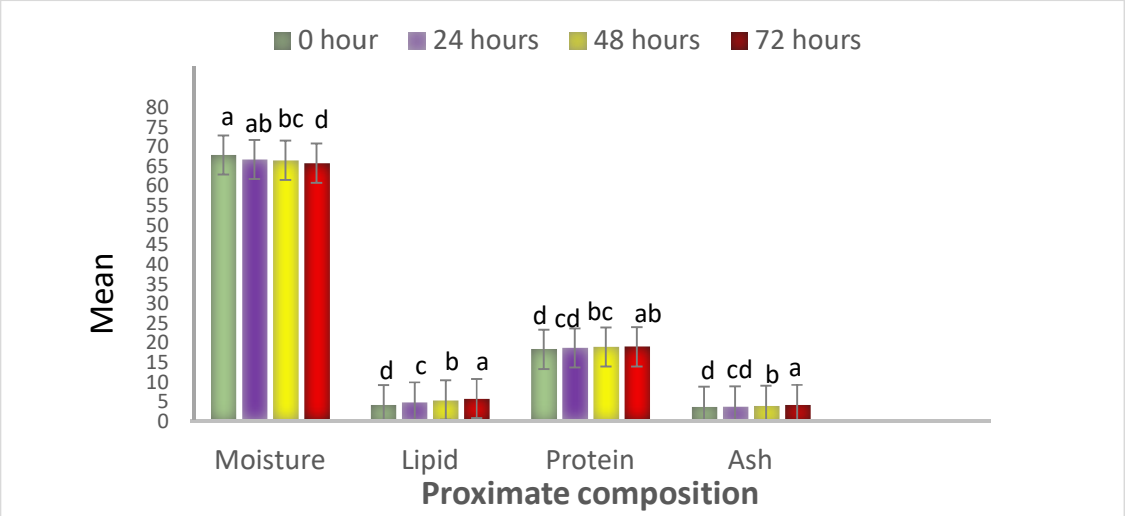

Figure 12: *Proximate composition changes of fish ball stored at 28°C*

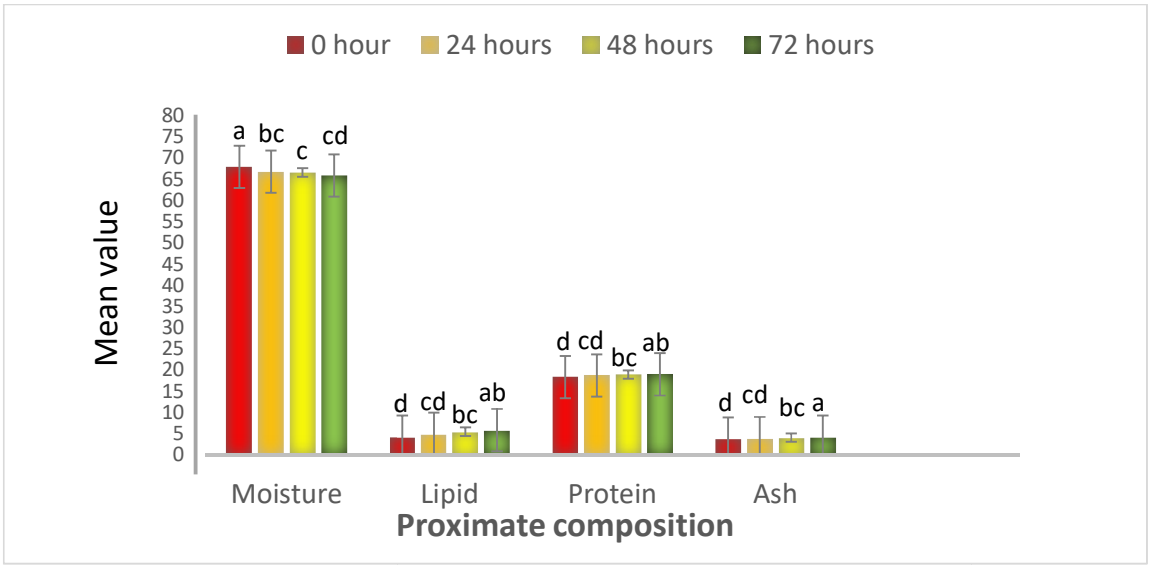

Figure 13: Proximate composition changes of fish ball stored at 5°C temperature.

Table 16. Effect of storage periods on proximate composition of fish ball prepared from Poa fish (*Panna microdon*) under different storage conditions.

Proximate composition	Storage periods (hours)	Storage conditions	
		Room temperature (28°C)	Refrigerated (5°C)
Moisture	0	67.93±0.16^{a}	67.93±0.16^{da}
	24	67.48±0.36^{b}	66.78±0.03^{cb}
	48	66.92±0.07^{c}	66.60±0.72^{bc}
	72	66.23±0.20^{d}	65.87±0.31^{ad}
Lipid	0	4.28±0.07^{a}	4.28±0.07^{da}
	24	5.05±0.11^{b}	4.94±0.41^{cb}
	48	5.39±0.09^{c}	5.46±0.41^{bc}
	72	5.70±0.04^{d}	5.83±0.07^{ad}
Protein	0	18.35±0.33^{a}	18.35±0.33^{da}
	24	17.93±0.13^{b}	18.71±0.08^{c}
	48	17.62±0.22^{c}	18.95±0.02^{b}
	72	17.29±0.08^{d}	19.00±0.16^{a}
Ash	0	3.86±0.03^{a}	3.86±0.03^{da}
	24	4.03±0.09^{b}	3.94±0.03^{cb}
	48	4.44±0.09^{c}	4.06±0.07^{bc}
	72	4.87±0.14^{d}	4.29±0.08^{ad}

1= Extremely dislike; 2=Dislike; 3 =Neither like nor dislike; 4=like; 5=Extremely like. Each value is represented as the mean ± SD of n=8. Means followed by the same superscript within a row are not significantly different (P > 0.05).

Bacterial load analysis

Effect of storage periods on growth and survival of bacterial load of fish ball prepared from Poa fish (*Panna microdon*) under different storage conditions is presented in Table 17. In both temperatures, the TPC (Total Plate Count) was found to increase significantly ($p<0.05$) throughout the storage period. Preliminary bacterial load of fish ball was 3.39±0.53 Log CFU/g. Bacterial growth in fish ball kept at room temperature (28^0 C) rapidly found to increase ($p<0.05$) with the progress of storage time and within 24 hours TPC increased to 5.43±0.55 Log CFU/g, after 48 hours and 72 hours the bacterial load was 7.17±0.56 Log CFU/g and 8.79±0.59 Log CFU/g, respectively.

Whereas, same fish ball kept at refrigeration temperature (5^0 C) showed bacterial growth pattern of somewhat different type. In this case, the rate of bacterial growth was slower and during the 24 hours storage time and no marked change were observed in TPC, after that bacterial growth gradually increased ($p<0.05$) and after 72 hours the TPC reached to 4.43±0.43 Log CFU/g in fish ball which is significantly different ($p<0.05$) with the bacterial load of 0 hours but not significantly different ($p<0.05$) with the bacterial load of 24 hours and 48 hours, respectively. The TPC after 24, 48 and 72 hours were significantly different at 28° C with the TPC of 5° C after 24, 48 and 72 hours, respectively. Microbial analysis of fish products did not exceed the maximum levels (7 Log CFU/g of meat) of microbiological criteria for fresh and frozen fish given by the ICMSF (1978). This finding is in agreement with ICMSF and the findings of Kamat (1999). The author reported TPC of fish ball in curry prepared from mackerel stored in chilled temperature was increased from 4.49 to 7.61 CFU/g during a storage period of 14 days. But, Koelkar and Pagarkar (2013) recorded the TPC of the fish ball in curry prepared from *Catla Catla* in chilled storage was increased from 2.55 to 4.38 log CFU/g during a storage period of 12 days. Mote (2001) found the TPC values of the chilled stored fish ball in spinach curry ranging from 4.23 to 7.75 log CFU/g. Taskaya et al (2003) observed an increase from the initial count of 4.04 Log CFU/g to 8.91 Log CFU/g in fish burger stored at a refrigerated

temperature over a period of 21 days. Kilinc (2007) observed an increase in total viable count from 2.75 logCFU/g of meat to 6.20 log CFU/g of meat in fish patties from anchovies at the end of 5 days which is also consistent with results of the current study.

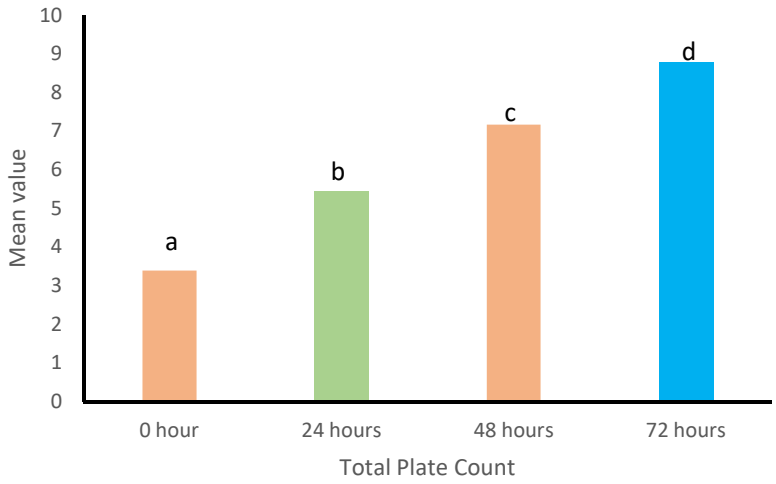

Fig. 14: Bacterial load changes of fish ball stored at 28°C temperature (Means followed by the same superscript within different time periods of Total Plate Count (TPC) are not significantly different ($p > 0.05$).

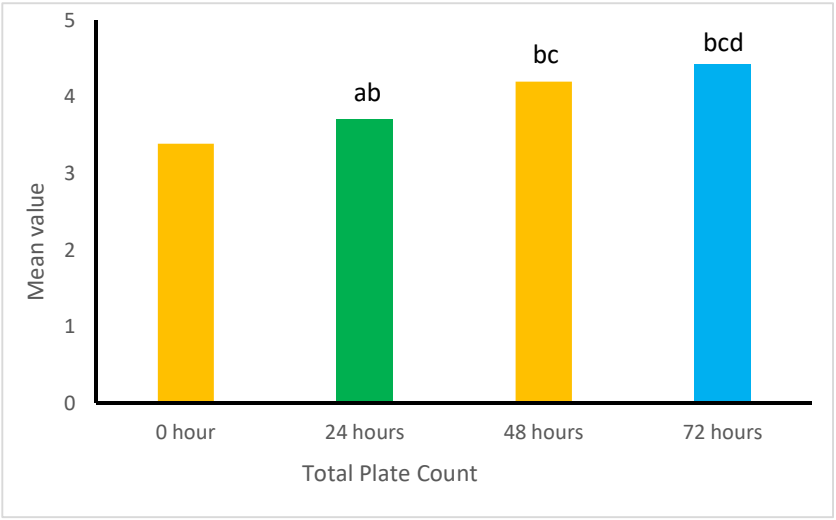

Fig. 15: Bacterial load changes of fish ball stored at 5°C temperature (Means followed by the same superscript within different time periods of Total Plate Count (TPC) are not significantly different ($P > 0.05$)).

Table 17. Effect of storage periods on bacterial load of fish ball prepared from Poa fish *(Panna microdon)* during different storage conditions

Term	Storage periods	Storage conditions	
		Room temperature (28° C)	Refrigerated temperature (5° C)
Total Plate Count (TPC)	0	3.39±0.53[a]	3.39±0.53[ad]
	24	5.43±0.55[b]	3.71±0.08[c]
	48	7.17±0.56[c]	4.20±0.41[b]
	72	8.79±0.59[d]	4.43±0.43[a]
	Average	**6.19±2.14**	**3.39±0.54**

Each value is represented as the mean ± SD of n=12; Means followed by the same superscript within a column are not significantly different (P > 0.05).

C. Development of fish burger using low cost Pangus *(Pangasius hypophthalamus)* fish

Materials and methods
Experimental site and period

The whole research work was performed in the different laboratories of Sylhet Agricultural University (SAU), Sylhet, during the period of July-October 2018 in the laboratory of the Department of Fisheries Technology and Quality Control, SAU.

Preparation of value-added products
Collection of fish

Fresh Thai-Pangus (*Pangasius hypophthalamus*) fish was collected from the Shibgang Bazar and Majortila Bazar of Sylhet Sadar. Immediately after purchasing, the fish was preserved properly with crushed ice in an insulated box and transported to the laboratory of the Department of Fisheries Technology and Quality Control, Sylhet Agricultural University. The average size of the fish was 42 ± 2.50 cm, 1.65 ± 0.35 Kg and the price of the fishes were Tk 100 ± 15 per Kg.

Preparation of fish mince

The fishes were weighed and then washed with clean water, beheaded, eviscerated, skinned and washed. The skinned fishes were filleted and deboned manually in iced condition. Then the mince was prepared by a mechanical mincer through a 1 mm orifice diameter so that all bones and connective tissues were removed from the muscles. All the utensils used in the experiment were cleaned with adequate washing and kept cool (5°C). Crushed ice was used to maintain adequate temperature throughout the product preparation (Fig. 3.1).

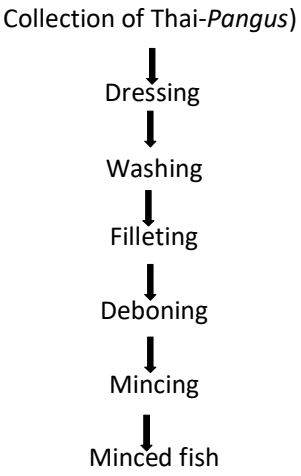

Fig. 16: Flow-diagram for preparation of fish mince.

Formulation of fish burger

List of ingredients used for the preparation of fish burgers are given in Table 3.1. The mince obtained from *Thai Pangus* fish muscle was ground with 2% NaCl, 2% oil, 0.6% sugar, 2% spices (onion, garlic, ginger, green chili paste and hot spices. The mixing was done generally for 5-7 minutes. The whole dough was stuffed into a steel frame. The size of each burger patty was (6.5 × 6 × 0.5cm). The steel frame was set on a wooden plate. Another wooden plate was fixed on the frame and held tightly with nuts and bolts to compress the material kept in the steel frame in between the two wooden plates. Then patties left for some times for seasoning. After seasoning, the patties were separated from the steel frame and dipped in a batter formulation. Then it was fried in dip-oil. Freshly prepared fish burgers were packed with polyethylene bag and kept at refrigerator (5^{o}C) and at room temperature (28^{o}C). The whole procedure is shown diagrammatically in Fig. 2.

Table 18. Ingredients of fish burger with their percentage

Sl. No.	Materials	Amount (%)
1	Minced fish	55.0
2	Chilled water	35.0
3	Chopped onion	2.5
4	Onion flakes	4.0
5	Vegetable oil	2.5
6	Salt	1

Preparation procedure of fish burger are given in the following diagram-

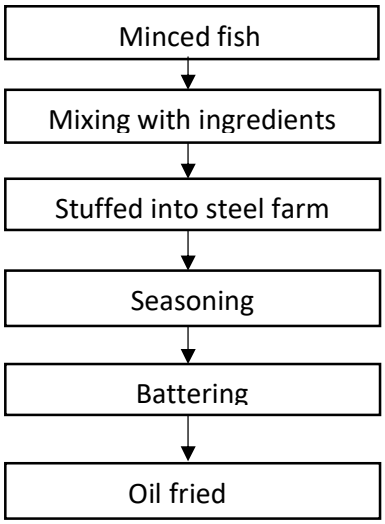

Fig.17: Diagram of fish burger preparation.

Determination of proximate composition

Proximate composition of freshly prepared fish burger was done in triplicates. Proximate analysis of moisture, crude protein, lipid and ash were carried out according to the methods given in AOAC (1990) with certain modifications as described below:

Organoleptic analysis

Stored lot fish burgers were randomly chosen and each packed sample was analyzed to evaluate organoleptic attributes. Samples were drawn four times within a period of 72 hours with 24 hours interval. A panel of 7 members of teachers conducted sensory assessments of the products as described by Nowsad *et al.* (2000). Prior to testing, panel members were familiarized with the properties of fish burger. All prepared fish burger were served to 7 panel members to evaluate the organoleptic attributes (Appearance, flavor, taste, texture and overall acceptability) of the samples by using 5-points descriptive scale. According to the table, scores were assigned with '1' being the least and '5' being the highest for attributes. Scores 5, 4, 3, 2, 1 were taken for likeextremely, like, neitherlike nor dislike, dislike, dislike extremely respectively for each of theorganoleptic characteristics. According to the scoring table, the scores from 1 to 2 indicate the negative evaluations and scores 4 to 5 indicate the positive evaluation. The middle of 3 reflects the ambivalent feelings in evaluation.

Determination of microbial load

Sample preparation

Standard plate count expressed as Colony Forming Units per gram (CFU/g) of dry fish were determined by using consecutive decimal dilution technique using spread plates. One grams of sample was taken aseptically and homogenized in a sterilized mortar. The sample was homogenized with 10 ml of distilled water and then the homogenate was transferred into a sterile bottle. One ml sample was transferred with a micropipette to a test tube containing 9.0 ml of distilled water and test tube was shaken thoroughly on a vortex mixture in order to get 10^{-1} dilution of original sample solution. Using the similar process several dilutions of 10^{-2}, 10^{-3}, 10^{-4}, 10^{-5}, and 10^{-6} were made.

Bacteria culture and counting

From sample solution of different test tubes bearing varying dilution 0.1 ml aliquot was taken by a micropipette and transferred aseptically in to the pre-prepared agar plates by raising the upper lid sufficient enough to enter the lip of the pipette. The samples were then spread homogenously and carefully by sterile flamed L-shaped glass rod throughout the surface of the media until the sample dried out. The plates were incubated at 37^{o}C in an inverted position in an incubator. After 48 hours of incubation colonies developed on the Petri dish was counted following a standard method. Only plates having 30 to 300 colonies were considered for counting in order to get acceptable values. Number of bacteria per gram of the sample (CFU/g) was calculated by using the following formula:

CFU/g=

No. of colonies on petridish ×10 × dilution factor × Volume of total sample solution / Wt. of fish sample (g)

Data analysis

ANOVA was performed using SPSS 20 with 5% confidence level.

Proximate composition analysis

Proximate composition of fish burger prepared from minced of Thai Pangus (*Pangasius hypophthalamus*) are presented in Table 3.2. In 28°C the initial moisture, lipid, protein and ash content were 59.34±0.02, 8.79±0.04, 19.92±0.07 and 5.22±0.06%, respectively. After 72 hours, moisture, lipid, protein and ash content were reached at 58.94±0.04, 8.79±0.04, 19.92±0.07 and 5.22±0.06%, respectively.

In case of 5°C, initial moisture, lipid, protein and ash content were 59.34±0.02, 8.79±0.04, 19.92±0.07 and 5.22±0.06%, respectively. After 72 hours the moisture, lipid, protein and ash content were reached at 59.19±0.01, 9.01±0.02, 19.11±0.02 and 5.46±0.01%, respectively. In storage condition lipid and ash content were gradually found to increase and on the contrary the moisture and protein content were found to decrease gradually which is parallel to the findings of Sandhya Rani et al (2017) and Vanitha et al (2015). Authors reported that the moisture content and protein content of fish fillet produced from Mrigal (*Cirrhinus Mrigala)* and fish burger produced from Catla (*Catla Catla*) were in decreasing manner during frozen storage.

Table 19. Effect of storage temperature on proximate composition of fish burger prepared from Thai-Pangus (*pangasius hypophthalamus*)

Storage condition	Storage period (Hours)	Moisture	Lipid	Protein	Ash
Room temperature (28^{0} C)	0	59.34±0.02	8.79±0.04	19.92±0.07	5.22±0.06
	24	59.17±0.04	8.95±0.05	19.63±0.09	5.35±0.02
	48	59.06±0.05	9.10±0.04	19.49±0.09	5.47±0.06
	72	58.94±0.04	9.23±0.04	18.96±0.12	5.62±0.04
Refrigerated (5^{0} C)	0	59.34±0.02	8.79±0.04	19.92±0.07	5.22±0.06
	24	59.30±0.01	8.88±0.04	19.90±0.04	5.33±0.01
	48	59.24±0.01	8.95±0.02	19.11±0.04	5.39±0.02
	72	59.19±0.01	9.01±0.02	19.04±0.02	5.46±0.01

* Each value is represented as the mean ± SD of n=8.

Organoleptic analysis

Changes in appearance, flavor, taste, texture and overall acceptance of fish burger formulated from Thai-Pangus stored at room temperature (28°C) and refrigerated temperature (5°C) are show nin Table 3.3. Data revealed that all the organoleptic qualities were decreased with the progress of storage period ($P<0.05$) at room temperature. The initial prominent fresh sweet odor of fish ball became gradually pungent followed by a sour odor accompanied by moist slimy surface with fungal growth at 72 hours at 28°C, which was the indicative of large bacterial growth. Bad smell observed at 48 hours of storage time and fungal growth was visible at the storage time of 72 hours. Although a mild pungent odor was felt at 24 hours. On the contrary, at refrigerated temperature (5°C) the product was found more stable. The appearance, flavor, taste, texture and overall acceptance did not change remarkably even after 72 hours ($P>0.05$). Fungal growth also was not observed after 72 hours. The textural quality changed significantly ($P<0.05$) at 48 and 72 hours but at acceptable level. The results of this study are in accordance with the results of Koelkar and Pagarkar (2013). They evaluated organoleptic characteristics such as color, taste, odor, appearance, texture and overall acceptability of fish ball prepared from *Catla Catla* kept in chilled storage (0 to -2°C). Authors observed a declined trend of organoleptic scores from 9.0 to 3.1 for the storage period of 0 to 12 days. Besides, they also stated that fish ball in curry kept in chilled storage was not acceptable after 9 days. The finding of the present study is found in agreement with the findings of Ejaz *et al.* (2009). They assessed organoleptic quality of fish burger formulated from *Pangasius sutchi* kept at room (28°C) and refrigeration (5°C) temperature. They reported that at room temperature, the organoleptic attributes decreased significantly ($P<0.05$) throughout the storage period. In contrast, at refrigerated temperature (5°C) the product was found more stable.

Table 20. Effect of storage temperature on sensory attributes of Thai-Pangus fish burger

Storage condition	Storage period (Hours)	Appearance	Flavor	Taste	Texture	Overall acceptance
Room temperature (28^{o} C)	0	4.57±0.53	4.42±0.53	4.28±0.48	4.14±0.37	4.57±0.53
	24	3.57±0.53	3.85±0.37	3.71±0.48	3.57±0.53	3.71±0.48
	48	2.85±0.69	2.71±0.48	2.85±0.69	3.00±0.57	2.85±0.69
	72	2.00±0.57	2.00±0.57	1.85±0.69	1.85±0.37	1.85±0.69
Refrigerated (5^{0} C)	0	4.57±0.53	4.42±0.53	4.28±0.48	4.14±0.37	4.57±0.53
	24	4.14±0.69	3.71±0.48	3.85±0.37	3.57±0.53	3.85±0.37
	48	3.57±0.53	3.14±0.37	3.42±0.53	3.28±0.48	3.57±0.53
	72	3.14±0.37	3.14±0.37	3.00±0.57	2.71±0.48	3.14±0.37

* Each value is represented as the mean ± SD of n=7.

Bacterial load analysis

Changes in bacterial load (CFU/g) of fish burger prepared from Thai Pangus both at room (28°C) and refrigerated temperature (5°C) are presented in Table 3.4. In both condition, the TPC (Total Plate Count) was found to increase significantly ($P<0.05$) throughout the storage period. Preliminary bacterial load of fish burger was 2.69±0.27 log CFU/g. Bacterial growth in fish burger kept at room temperature found to increase rapidly ($P<0.05$) with the progress of storage time and within 24 hours TPC increased to 6.68±1.08 logCFU/g, after 48 hours it was 7.88±0.59 log CFU/g and after 72 hours these values reached to 8.72±0.08 logCFU/g.

While, identical fish burger kept at 5°C the bacterial growth trend was slightly altered where bacterial development rate was slower and during the 24 hours storage time minor change were observed in TPC, after that bacterial growth gradually increased significantly (P<0.05) and after 72 hours the TPC reached to 5.35±0.27 log CFU/g in fish burger. This result coincides with the results of Ejaz *et al.* (2009). They estimated bacterial load of fish burger developed from *Pangasius sutchi* stored at room and refrigeration temperature. They reported significant (P<0.05) increase in bacterial load at both temperatures but the growth pattern was slower at refrigeration temperature. They recorded bacterial load of fresh fish burger 4.19 log CFU/g and after 72 hours at room temperature it increased to 9.77 log CFU/g while at refrigeration temperature it increased to 6.32 log CFU/g.

Table 21. Effect of storage temperature on bacterial load of fish burger prepared from Thai-Pangus (*Pangasius hypophthalamus*)

Storage condition	Storage period (Hours)				Overall (Log CFU/gm)
	0	24	48	72	
Room temperature (28^0 C)	2.69±0.27	6.68±1.08	7.88±0.59	8.72±0.08	6.49±0.47
Refrigerated (5^0 C)	2.69±0.27	3.58±0.16	4.34±0.12	5.35±0.27	3.99±0.30

* Each value is represented as the mean ± SD of n=12

D. Quality changes of newly developed fish sticks from Tilapia (*Tilapia niloticus*) fish during different storage conditions

Objectives:

- Development of improved fish sticks from low cost Tilapia fish.
- Quality assessment of the developed fish sticks.

Materials and Methods

Selection of fish species

Tilapia (*Tilapia niloticus*) an exotic fish, was selected for the manufacture of fish sticks as it is a commonly used good aquaculture species, available in the market throughout the year, already have a good demand among the consumers at raw form and a low cost commodity.

Collection of fish species

Fresh Tilapia fish was collected from local fish markets of Sylhet district. Immediately after collection, the fish was iced properly (fish: ice=1:2) with crushed ice in an insulated box and transported to the laboratory of Fisheries Technology and Quality Control, SAU. The average size of the 730 ± .45 gm and the price of the fishes were tk 115 ± 35 per Kg.

Preparation of the product

After weighing fish were washed with clean water, beheaded, eviscerated, skinned and 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 previously prepared batter solution. The battering solution was prepared by mixing egg white, spices, salt and MSG (Table 22). After that, it was rolled in bread crumb. Battered and breaded fish sticks were then dip fried in soybean oil until the surface of the stick becomes golden brown color. Finally, prepared fish sticks were then kept on the kitchen paper in order to soak the extra oil form the surface of the fried fish sticks. All the

utensils used in the experiment were cleaned with adequate washing and kept cool (5°C). Scheme for the preparation of fish sticks from Tilapia fish is presented in Fig. 11.

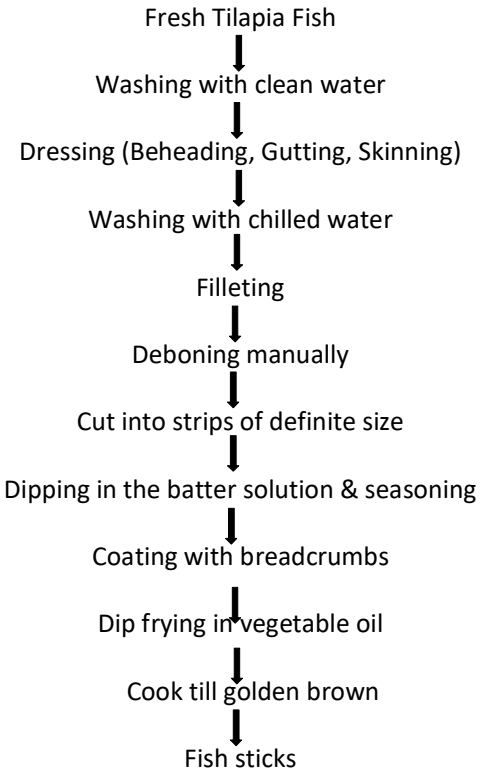

Fig. 18: Flow-diagram of stick preparation.

Table 22. Ingredients and the amounts used for the battering solution for fish stick

Ingredients	**Percentage (%)**
Wheat flour	34
Salt (NaC1)	1
Monosodium glutamate.	1
Spices (green pepper, ginger, garlic, cumin, onion paste, mixture of hot spices)	2
Egg	18
Vinegar	1 tea spoon
Water	44

Various local gel enhancing ingredients and spices were used for the preparation of batter solution for fish sticks from Tilapia fish to ensure the products a Bangladeshi known taste so that the products could attract local consumer's acceptance.

Table 23. Spices and the amounts used in the batter preparation

Spices	Percentage (%) out of 2% of all spices
Garlic powder	20
Onion paste	25
Ginger powder	15
Green chili paste	10
Mixture of hot spices	30

Quality analysis of Tilapia fish sticks

The quality of the fish sticks was analyzed by organoleptic proximate composition andmicrobial analysis.

Organoleptic evaluation

A seven members team of expert members of Fisheries Faculty, SAU carried out the responsibilities of organoleptic evaluation of the products, on a five-point scale (Singh-Ackbarali et al., 2014 and Tokur et al., 2006) and scores were assigned with "1" being the least and "5" being the highest for attributes. The characteristics evaluated during evaluation are appearance, colour, flavor, taste, texture and overall acceptability. Samples were drawn four times within a period of 72 hours with a 24 hours interval between samplings.

Proximate composition

Moisture, fat, protein and ash content were determined according to the method of AOAC (2000). In crude protein estimation, about 1g of comminuted sample was employed for Kjeldahl procedure. Lipid content was determined by extracting a given quantity of sample with acetone in Soxhlet Apparatus 3-4 hours. Moisture content was determined by drying a 5g sample in an oven at105°C for 24 hours. Ash content was determined by igniting the sample in a muffle furnace at 550°C for 6 hours.

Microbial analysis

Total plate count (TPC) was determined by consecutive decimal dilution technique. Samples for the TPC was accurately weighed and added with required amount of water and liquefied in a sterile blender jar and consecutive tenfold dilution were made in the test tubes. From all the dilutions spread plate cultures were made in duplicates and incubated at 37^0c for 24 to 48 hours. Colonies developed on the plates were counted in a colony counter and plates having 30 to 300 colonies were selected for TPC.

Statistical analysis

One-way analysis of variance and the general linear model were used to analyze the data. The Duncan's New Multiple Range Test (DMRT) was used to find the significant differences between storage periods.

Experimental design

The whole preparation was done into 4 quarters where each quarter comprises 15 days. Tilapia fish sticks was randomly chosen and analyzed in triplicate for proximate composition, microbiological and organoleptic attributes. Proximate composition, microbiological and organoleptic attributes were evaluated at different storage time and conditions.

Results and Discussions

Evaluation of the quality of the fish sticks

Proximate composition analysis

The proximate composition viz. moisture, protein, lipid and ash contents of fish sticks were determined and presented in Table 24 and 25. In fresh Tilapia fish sticks moisture, lipid, protein and ash content was found within the range of 54.42±1.05 to 56.84±0.63%, 7.42±0.39 to 8.04±0.37%, 29.76±0.33 to 30.90±0.61% and 2.68±0.37 to 3.19±0.22%, respectively considering all the 4 quarters. Latif Taskaya *et. al.* (2003) reported moister content 63.61% and crude protein 17.50% in fish burger prepared from rainbow trout. These results are in well agreement with the results of present study.

Moisture, lipid and protein content was found to decrease gradually in the packaged fish sticks kept at room temperature (28^0C). The decrease can be attributed to the leaching out of the water-soluble nitrogenous components during storage along with moisture (Vanitha *et al*., 2015) while in case of ash, it was found increasing with the progress of storage time.

In the case of refrigerated temperature (5^0C), moisture and protein content was found to decrease but the other two samples lipid and ash content were found increasing slowly. Sandhya Rani *et al* (2017) observed an increase in lipid, protein and ash content of fish finger produced from mince of Mrigal (*Cirrhinus Mrigala)* during frozen storage. The author also reported that the moisture content of fish finger produced from mince of Mrigal (*Cirrhinus Mrigala)* was found decreased gradually during frozen storage. The result of the current *study is more or less similar with the results of Sandhya Rani et al (2017). Sandhya Rani et* al (2017) and Vanitha *et al* (2015) reported that the moisture content of fish fillet produced from Mrigal (*Cirrhinus Mrigala)* and fish burger produced from Catla (*Catla Catla*) was decreased during frozen storage.

Table 24. Effect of storage periods on proximate composition of Tilapia fish sticks under different storage conditions

Proximate composition	Storage temperature	Storage period (hr)	Quarters			
			Q-01	Q-02	Q-03	Q-04
Moisture	Room temperature (without packaging)	0	56.84±0.63	55.63±0.97	56.10±0.85	54.42±1.05
		24	56.76±0.62	55.55±0.99	56.01±0.85	54.34±1.05
		48	56.62±0.62	55.41±0.99	55.87±0.87	54.20±1.06
		72	56.43±0.63	55.22±1.00	55.68±0.89	54.01±1.05
	Room temperature (with packaging)	0	56.84±0.63	55.63±0.97	56.10±0.85	54.42±1.05
		24	56.96±0.62	55.75±0.97	56.22±0.85	54.54±1.04
		48	57.29±0.63	56.09±0.95	56.55±0.83	54.85±1.03
		72	57.87±0.63	56.70±0.97	57.17±0.83	55.46±1.00
	Refrigerated temperature	0	56.84±0.63	55.63±0.97	56.10±0.85	54.42±1.05
		24	56.31±0.62	55.11±0.96	55.56±0.83	53.90±1.04
		48	56.02±0.61	54.80±0.96	55.25±0.86	53.59±1.04
		72	55.82±0.60	54.62±0.94	55.04±0.86	53.41±1.03
Lipid	Room temperature (without packaging)	0	7.52±0.40	7.93±0.35	7.42±0.39	8.04±0.37
		24	7.23±0.42	7.66±0.31	7.14±0.38	7.75±0.39
		48	6.37±0.49	6.85±0.36	6.49±0.36	7.09±0.36
		72	5.03±0.71	5.62±0.57	5.53±0.33	6.07±0.33
	Room temperature (with	0	7.52±0.40	7.93±0.35	7.42±0.39	8.04±0.37
		24	7.01±0.42	7.49±0.34	6.96±0.44	7.58±0.34
		48	5.78±0.43	6.50±0.40	6.15±0.34	6.71±0.33

	packaging)	72	4.48±0.45	5.33±0.50	5.12±0.30	5.67±0.38
	Refrigerated temperature	0	7.52±0.40	7.93±0.34	7.42±0.39	8.04±0.37
		24	7.85±0.39	8.22±0.34	7.73±0.37	8.30±0.34
		48	8.44±0.37	8.68±0.41	8.37±0.39	8.91±0.33
		72	8.93±0.30	9.26±0.55	8.81±0.44	9.30±0.27

Each value is represented as the mean ± SD of n=03

Table 25. Effect of storage periods on proximate composition of Tilapia fish sticks under different storage conditions

Proximate composition	Storage temperature	Storage period (hr)	Quarters			
			Lot-01	Lot-02	Lot-03	Lot-04
Protein	Room temperature (without packaging)	0	29.76±0.33	30.46±0.34	29.81±0.55	30.90±0.61
		24	29.52±0.36	30.22±0.32	29.59±0.60	30.61±0.63
		48	28.88±0.40	29.59±0.36	28.75±1.05	29.89±0.65
		72	27.73±0.44	28.44±0.41	27.70±1.15	28.70±0.56
	temperature (with packaging)	0	29.76±0.33	30.46±0.34	29.81±0.55	30.90±0.61
		24	29.60±0.37	30.28±0.36	29.66±0.58	30.66±0.62
		48	28.50±0.50	29.07±0.40	28.56±0.73	29.39±0.65
		72	27.23±0.62	27.71±0.42	27.37±0.86	28.02±0.70
	Refrigerated temperature	0	29.76±0.33	30.46±0.34	29.81±0.55	30.90±0.61
		24	29.63±0.33	30.31±0.33	29.69±0.53	30.76±0.63
		48	29.45±0.34	30.12±0.33	29.51±0.52	30.57±0.65
		72	29.23±0.34	29.89±0.34	29.30±0.50	30.34±0.66
Ash	Room temperature (without packaging)	0	2.77±0.31	2.82±0.21	3.19±0.22	2.68±0.37
		24	2.80±0.32	2.86±0.21	3.23±0.21	2.73±0.37
		48	2.82±0.33	2.88±0.21	3.25±0.22	2.75±0.37
		72	2.84±0.35	2.90±0.20	3.27±0.22	2.76±0.36
	temperature (with packaging)	0	2.77±0.31	2.82±0.21	3.18±0.22	2.68±0.37
		24	2.79±0.32	2.85±0.21	3.21±0.21	2.71±0.37
		48	2.81±0.32	2.87±0.21	3.24±0.21	2.74±0.37
		72	2.83±0.32	2.88±0.21	3.25±0.21	2.76±0.37
	Refrigerated temperature	0	2.77±0.31	2.82±0.21	3.19±0.22	2.68±0.37
		24	2.78±0.31	2.84±0.21	3.21±0.22	2.70±0.37
		48	2.81±0.32	2.87±0.21	3.22±0.21	2.73±0.37
		72	2.84±0.32	2.90±0.20	3.24±0.20	2.76±0.37

Each value is represented as the mean ± SD of n=03

Microbiological analysis:

TPC (Mean ± SD) of fish sticks at different time periods and storage conditions are presented in Table 26. In fresh fish sticks TPC was found within the range of 3.39±0.53 to 3.98±0.54 Log CFU/g .TPC was found to increase throughout the storage period. Bacterial growth in fish sticks kept at room temperature found to increase rapidly with the progress of storage time and within 24 hours TPC was found to ranging from 5.44±0.12 to 6.25±0.57Log CFU/g and after 72 hours these values reached 8.80±0.58 to 8.93±0.72 Log CFU/g for samples without packaging. In case of fish sticks kept in room temperature with packaging, TPC was found increasing. Lower TPC was observed during refrigerated storage. Liston (1980) observed that freezing generally causes a reduction in bacterial count and the number continue to fall during storage. Bashar (2004) observed that the bacterial

growth in fish sticks prepared from washed Queen fish mince kept at room temperature steadily increased with the progress of storage time. In addition, the author observed that in refrigeration temperature bacterial growth become slower and even after 24 hours no appreciable change in APC was observed. Shammi (2005) found that bacterial growth in fish ball, fish stick and fish sausage was gradually increased throughout the storage period, both at room and refrigeration temperature. The author concluded that the shelf life of the products is very short and it is not more than 24 hours at room temperature and 48 hours at refrigeration temperature.

Table 26. TPC (Mean ± SD) of fish sticks at different time periods and storage conditions
Each value is represented as the mean ± SD of n=03

Product	Storage temperature	Storage period (hr)	Quarters			
			Q-01	Q-02	Q-03	Q-04
Fish sticks(Log CFU/g)	Room temperature (without packaging)	0	3.39±0.53	3.84±0.49	3.98±0.54	3.65±0.09
		24	5.44±0.55	5.62±1.10	6.25±0.57	5.44±0.12
		48	7.17±0.50	7.65±0.88	7.69±0.96	7.69±0.06
		72	8.80±0.58	8.93±0.72	8.92±0.77	8.91±0.56
	Room temperature (with packaging)	0	3.39±0.53	3.84±0.49	3.98±0.54	3.65±0.09
		24	4.69±0.72	4.76±0.78	5.50±0.13	4.86±0.42
		48	6.34±0.67	6.61±1.02	6.38±0.44	6.01±0.43
		72	7.67±0.04	7.44±1.12	7.22±0.48	6.71±0.59
	Refrigerated temperature	0	3.39±0.53	3.84±0.49	3.98±0.54	3.65±0.09
		24	3.72±0.09	4.13±0.52	4.29±0.43	3.87±0.18
		48	4.20±0.42	4.78±0.53	5.03±0.46	4.41±0.21
		72	4.44±0.43	5.88±1.31	5.71±0.97	4.64±0.14

Organoleptic evaluation

Evaluation of the shelf life of the fish sticks at room and refrigeration temperature

The overall acceptability scores of fish sticks under different storage condition and temperature are presented in Table 27.Changes in general appearance, flavor, taste and texture of Tilapia fish sticks at both packaged and without packaged from stored at room (28°C) and refrigerated temperatures (5°C) are shown in Table 26. All the organoleptic attributes were found to decrease with the progress of storage period at room temperature. The initial prominent fresh sweet odour of fish sticks became gradually pungent followed by a sour odour accompanied by moist slimy surface with fungal growth at 72 hours, which was indicative of large bacterial growth. Bad smell observed at 48 hours of storage time and fungal growth was visible at the storage time of 72 hours. Although a slight pungent odour was felt at 24 hours.

On the other hand, at refrigerated temperature (5°C) the product was found more stable. Colour, taste and general appearance of the sample did not found to change remarkably even after 72 hours. A slight pungent odour felt at 72 hours but it was not considered as unacceptable. Fungal growth also not observed after 72 hours. The textural quality changed significantly at 48 and 72 hours but considered as acceptable. The results of this study are in accordance with the results of Shammi (2005) and Koelkar and Pagarkar (2013).

Table 27. Organoleptic attributes changes of fish sticks stored at room and refrigerated temperatures under different storage conditions

Sensory attributes	Storage periods (hours)	Storage condition	
		Room temperature (28° C)	Refrigerated temperature (5° C)
Appearance	0	4.63± 0.37[a]	4.63±0.37[a]
	24	3.77±0.53[b]	4.12±0.48[b]
	48	2.48±0.45[c]	3.64±0.50[c]
	72	1.65±0.24[d]	3.10±0.37[d]
Flavor	0	4.70± 0.44[a]	4.70±0.44[a]
	24	3.63±0.43[b]	4.15±0.20[b]
	48	2.20±0.65[c]	3.44±0.34[c]
	72	1.39±0.54[d]	2.85±0.50[d]
Taste	0	4.86±0.43[a]	4.86±0.43[a]
	24	3.06±0.70[b]	4.20±0.50[b]
	48	2.15±0.64[c]	3.82±0.58[c]
	72	1.18±0.40[d]	3.10±0.09[d]
Texture	0	4.90±0.53[a]	4.90±0.53[a]
	24	3.95±0.53[b]	3.86±0.48[b]
	48	2.38±0.37[c]	3.33±0.20[b]
	72	1.52±0.43[d]	2.69±0.37[c]
Overall acceptability	0	4.49±0.48[a]	4.77±0.53[a]
	24	3.37±0.40[b]	4.45±0.30[a]
	48	2.00±0.00[c]	3.66±0.50[b]
	72	1.20±0.48[d]	3.00±0.00[c]

1=Extremely dislike; 2=Dislike;3 =Average; 4=like; 5=Extremely like
Each value is represented as the mean ± SD of n=7

E. Title: Consumers behaviour and attitudes towards newly developed fish products

Objectives of the study

- To investigate people's perceptions and attitude toward newly developed fish products;
- To attract consumers toward newly developed fish products; and
- To prepare a small-medium entrepreneurship for the newly developed products.

Methodology
Study area and sample selection

Sample collection area covered all the 64 districts under seven divisions. Total sample size was 560 (at least 8 from each districts), for an error of ≤2.86%, confidence level 95% (two sigmas; p=q=0.5). The error was below the desirable 4% limit suggested by Escriba-Perez *et al.*, (2017). Data were purged using a univariate exploration procedure by means of the frequency table. This table revealed the presence of 18 cases with missing data, thereby undermining the quality of information. As the number of cases (2.86%) did not exceed 10% of the total, we assumed that there is no underlying reason which tends to skew the data. Therefore, ruling them out for those options chosen for the missing cases. Finally, we analysed a total of 544 valid cases.

Survey procedure

Survey was conducted in Bangladesh on a convenience sample size of 544. The respondents were personally interviewed at their residence or work place and to complete a questionnaire it required about 30 min.

Sample demographics

Considering the socio-demographic variables (Table 28), the respondents show the predominant presence of male (62%), aged between 36-45 year old (30%), married (80%) who live in families having between of 4-5 members (52%), holding a Bachelor's degree (30%), private sector employees (26%), Muslim (62%), Upper middle (56%), live in peri-urban area (64%) and medium price preferred (65%).

Variables

The variables examined to determine the average frequency of consumers consumption of the studied food. This mean frequency is obtained from the consumption frequency scale considering Once daily=1, 2 to 5 times a week=2, Once every two weeks =3, Once a month =4, No consumption=5. Therefore, an average frequency around 1 means that respondent consumes this type of fish foods once a week or more, i.e. very often. The question on consumer food lifestyles is based on 18 parameters measured by means of a 5-point Likert scale range, from 1 "Strongly disagree" to 5 "Strongly agree", with a neutral midpoint at 3 "Neither agree nor disagree" (Table 29).

Table 28. State of socio-demographic scenario

Parameter	Sub-parameter	Quantity	Percentage (%)
Sex	Male	340	62
	Female	204	38
Age (years)	16-25	99	18
	26-35	157	29
	36-45	162	30
	46-55	89	16
	56-65 more	37	7
Marital status	Single	88	16
	Married	437	80
	Separated	19	4
Family size	≤3	167	31
	4 to 5	284	52
	>5	93	17
Education	Primary school	6	1
	High school	46	8
	SSC	58	11
	HSC	158	29
	Diploma	44	8
	Bachelor's	160	30
	Masters	59	11
	PhD	13	2

Table 28 Cont'd.....

Parameter	Sub-parameter	Quantity	Percentage (%)
Occupation	Student	95	18
	Govt. job	92	17
	Private job	143	26
	Housewife	106	19
	Business	108	20
Religion	Muslims	340	62
	Hindus	199	37
	Others	5	1
Social class	Lower	49	9
	Lower middle	32	6
	Middle	129	24
	Upper middle	307	56
	Upper	27	5
Place of living	City	81	15
	Peri-urban	346	64
	Rural	117	21
Price Preferences	Low	161	30
	High	28	5
	Medium	355	65

Parameters with highest Percentage (%) loadings are marked with underline in the table.

Table 29. Descriptive statistics for the parameters measuring food-related lifestyle (FRL)

Sl.	Parameters	Average	Std. Dev.
1	My family involved in cooking and preparing the meal	4.26	1.01
2	I always try to buy best quality food with the lowest price	4.14	1.07
3	I prefer organic food than inorganic food	4.10	0.99
4	I like shopping food for my family	4.00	0.94
5	I prefer fresh food than the processed food	3.91	1.02
6	I prefer organic fish than cultured fish	3.76	1.23
7	I like to buy food without preservatives	3.68	0.71
8	I believe food nutrition is more important than taste	3.47	0.67
9	I like to go the restaurant with family and friends	3.38	1.23
10	I prefer to keep fish stick in the occasion	3.29	0.93
11	Prefer to keep the fish burger in the snack	3.21	0.62
12	I prefer fish than meat	3.12	0.76
13	I prefer to keep the fish cutlets in the occasion	2.92	1.03
14	I prefer to keep fish ball in the snack	2.71	1.55
15	I like to cook new recipes	2.39	1.37
16	I prefer to keep fish ball in the snack	1.67	0.83
17	I like to buy new food items	1.38	0.53
18	I Prefer to keep only vegetables in the meal	1.21	0.48

The items getting the highest score was "My family involved in cooking and preparing meal" (with an average of 4.26) and "I always try to buy best quality food with the lowest price" (4.14). The lowest points were found to the items "I prefer to keep only vegetables in the meal" (1.21). Moreover, the question of fish stick, fish ball, fish cutlet, fish finger and fish burger consumers preferences were

measured by means of an another 5-point Likert scale (Scale: 1 = Once Daily; 2 = 2 to 5 times a week; 3 = Once every two weeks; 4 = Once a month; 5 = No consumption).

The socio-demographic variables analysed were “sex”: male or female; “age”: the ranks considered were (years ranging from) 16–25, 26–35, 36–45 , 46–55, 56–more; “educational level”: Primary school, High school, secondary school certificate, Higher school certificate, Diploma, Bachelors of science, Master of Science, Doctor of Philosophy; “social class”: lower, lower middle, middle, upper middle or upper; “presence of children younger than 18 years in the home”: if there are minors or not; “geographical area”: covering all the areas of mainland Bangladesh (Dhaka, Chittagong, Rajshahi, Khulna, Rangpur, Sylhet, Barisal); and salary/income. For the salary/income variable the stratifications were as BDT: 10000-19999,20000-29999, 30000-39999, 40000-49999, 50000-59999, 60000-69999, 70000-79999, 80000-89999, 90000 and more (1 USD=82 BDT). Social classes were obtained and determined according to the parameters reported in BBS, (2003). Social classes were also cross-checked by the educational level as suggested by Marginson, (2016).

Statistical analysis

Different statistical techniques were applied to analyses the results. First, we performed a frequency distribution analysis to describe the fish consumption of the sample.

A frequency distribution analyses was performed to understand the fish stick, fish ball, fish cutlet, fish finger and fish burger consumption. To find food-related lifestyle segment factor analysis and cluster analysis were used (Baglin, 2014). To ensure minimal information loss, Bartlett’s sphericity test and the Kaiser-Meyer-Olkin measure of sampling adequacy (KMO) were used. The principal component analysis method was used as the extraction method (Bishnoi and Yadav, 2017; Escriba-Perez *et al.,* 2017). The cluster analysis was configured by the hierarchical procedure.

In most of the cases 5-point Likert scale and semantic differential scales were used. However, some true false questions were also used. Developed by Rensis Likert, the Likert scale “requires the respondents to indicate a degree of agreement or disagreement with each of a series of statements about the stimulus objects” (Malhotra, 2006); whereas, the Semantic Differential (SD) measures people's reactions to stimulus words, this type of scale have endpoints with bipolar labels that have semantic meaning (Malhotra, 2006). The measurement items used in this study were usually either taken or adopted from previous scientific research.

Finally to analyze and compare the average animal originated food consumption frequency among the different studied variables, cross-tabulation of mean values with standard deviation were used. The statistical significance test for differences between mean values of different foods was tested by Snedecor’s F-test. Data were analysed using SPSS (Version 21).

11. Results and Discussions

Sample products consumption

The consumption frequency of fish stick, fish ball, fish cutlet, fish finger and fish burger has been shown in Table 30 Data revealed fish burger as the most frequently consumed item where 77% respondents ate the fish burger once daily. Fish ball was the second most consumed (62%) whereas, fish stick in the third place (59%). Fish finger was in the fourth place of consumption frequency level. However, 24% of respondent’s response indicates that they did not consume fish finger. The hygiene, price, taste, flavor might have influence not to consume fish finger. The same types of behavior observed for fish cutlet, which was least consumed (7%) once daily, 48% Once every two weeks, whereas 13% did not consume.

Table 30. Consumption frequencies of fish stick, fish ball, fish cutlet, fish finger and fish Burger

Frequency	Fish stick (%)	Fish ball (%)	Fish cutlet (%)	Fish finger (%)	Fish burger (%)
Once daily	59	62	7	30	77
2 to 5 times a week	39	24	26	23	13
Once every two weeks	1	8	48	18	6
Once a month	1	6	6	5	4
No consumption	0	0	13	24	0
Total	100	100	100	100	100

Consumption according to food-related lifestyle (FRL) segments

Factorial analysis of "five factors" explained 57.89% of the total variance. The Kaiser, Meyer and Olkin (KMO) measure of sampling adequacy resulted in a value of 0.720, which was greater than 0.50 (the minimum acceptable value) as suggested by Hair & Jnr, (2010). Bartlett's sphericity test verified the appropriateness of the factor analysis, resulting in a p-value of 0.000 at a significance level of 0.05.

The factorial analysis yielded five factors: (i) 'Food shopping', which encompasses matters related to the act of shopping, assessing not only the aspects associated with buying food but also those linked to shopping as an enjoyable and pleasing activity. (ii) 'Quality/price ratio', covering items associated with the freshness concept, natural (wild), pricing and quality, proactive attitude towards purchasing food. (iii) 'Eating with family', consisting of items communication and wellbeing, strengthening family bonding-staying connected. (iv) 'Eating at restaurants, out of home', includes taking meal as the social activity with or without a plan (v) 'Nutrition and new food innovations', encompassing consumers perception to food nutrition and willingness to purchase and prepare new food (Table-31).

From the above mentioned 5 factors, we performed the cluster analysis and obtained the following results (Table 32).

Table 31. Factor analysis for food-related life style (FRL)

Sl.	Items	Food shopping	Quality/ price ratio	Eating with family	Eating at restaurants, out of home	Nutrition and new food innovations
1	My family involved in cooking and preparing the meal	0.812	0.238	0.182	-0.369	0.141
2	I always try to buy best quality food with the lowest price	0.479	0.718	0.461	-0.128	0.143
3	I prefer organic food than inorganic food	0.478	0.471	-0.131	-0.109	0.771
4	I like shopping food for my family	0.568	0.312	0.185	-0.148	-0.119
5	I prefer fresh food than the processed food	0.147	0.661	-0.493	0.142	0.213
6	I prefer organic fish than cultured fish	0.132	0.763	0.237	0.143	-0.167
7	I like to buy food without preservatives	-0.119	0.781	0.194	0.142	0.133
8	I believe food nutrition is more important than taste	0.325	-0.174	0.209	-0.177	0.584
9	I like to go the restaurant with family and friends	-0.207	0.362	0.134	0.837	0.121
10	I prefer to keep fish stick in the occasion	0.734	0.443	0.428	-0.134	0.124

Table 31 Cont'd....

Sl.	Items	Food shopping	Quality/ price ratio	Eating with family	Eating at restaurants, out of home	Nutrition and new food innovations
11	Prefer to keep the fish burger in the snack	0.143	0.469	0.744	-0.228	-0.182
12	I prefer fish than meat	0.844	0.201	-0.162	0.428	-0.119
13	I prefer to keep the fish cutlets in the occasion	-0.224	0.232	0.309	0.576	0.122
14	I prefer to keep fish ball in the snack	0.169	0.116	-0.126	0.789	-0.187
15	I like to cook new recipes	0.342	-0.031	-0.062	0.147	0.576
16	I prefer to keep fish ball in the snack	0.187	-0.373	0.622	-0.201	0.059
17	I like to buy new food items	-0.155	0.333	0.354	0.129	0.528
18	I Prefer to keep only vegetables in the meal	-0.234	-0.248	0.587	0.075	0.015

Parameters with highest factor loadings are marked with underline in the table.

Table 32 Average consumption offish stick, fish ball, fish cutlet, fish finger and fish burger by food-related lifestyle (FRL)

Food Type	Not involved (8%)	Shopping (48%)	Restaurant consumer (12%)	Family priorities and social awareness (32%)	Total (100%)
Fish stick*	1.41	1.57	1.59	1.34	1.44
Fish ball**	1.39	1.83	2.28	1.26	1.59
Fish cutlet*	3.70	2.83	2.41	3.63	2.83
Fish finger	3.67	3.77	1.86	3.28	2.77
Fish burger**	1.24	1.92	1.40	1.23	1.39

Values are Mean± Standard Deviation. Scale: 1 = Once Daily; 2 = 2 to 5 times a week; 3 = Once every two weeks; 4 = Once a month; 5 = No consumption. *Significance differences ($p<0.05$); ** Significance differences ($p<0.01$)

Consumption of product according to socio-demographic variables

Consumption of product according to gender

Survey to understand the consumption pattern of fish product was carried out on 62% men and 38% women (Table 33). The food types that showed significant differences in the average consumption frequency according to the "gender" variable were fish stick, fish ball, fish cutlet, fish finger and fish burger. However, the consumption behaviour pattern for men and women was observed different. Fish cutlet, fish finger were the most frequently consumed product by men than women. In contrast, women consumed fish burger and fish ball more often than the men do (Table-33).

Table 33. Average consumption of fish stick, fish ball, fish cutlet, fish finger and fish burger by gender

Food Type	Male (62%)	Female (38%)	Total
Fish stick *	1.41±0.52	1.50±0.56	1.44±0.53
Fish ball *	1.68±0.97	1.45±0.72	1.59±0.89
Fish cutlet **	2.53±0.77	3.34±1.19	2.83±1.03
Fish finger **	1.96±0.95	4.10±1.45	2.77±1.56
Fish burger *	1.44±0.87	1.30±0.62	1.39±0.79

Values are Mean± Standard Deviation. Scale: 1 = Once Daily; 2 = 2 to 5 times a week; 3 = Once every two weeks; 4 = Once a month; 5 = No consumption.

*Significance differences ($p<0.05$); ** Significance differences ($p<0.01$)

Consumption frequency according to age group

The food types that showed significant differences in the average consumption according to the "age" were fish stick, fish ball, fish cutlet, fish finger and fish burger. Fish stick and fish burger were found most often consumed by the age group from 16 to 25 years followed by 26 to 35 years and 36 to 45 years group. In other age group, consumption of these products is less (Table 34). Fish stick alone was most preferred and consumed by 56 years and above age group. As whole fish stick, fish ball, and fish burger are mostly consumed by all age groups.

Table 34. Average consumption of fish stick, fish ball, fish cutlet, fish finger and fish burger by age (years)

Food Type	16-25 (18%)	26-35 (29%)	36-45 (30%)	46-55 (16%)	56 & above (7%)	Total
Fish stick	1.48±0.54	1.42±0.52	1.44±0.50	1.50±0.62	1.27±0.45	1.44±0.53
Fish ball *	1.56±0.79	1.43±0.73	1.69±1.01	1.72±0.99	1.62±0.92	1.59±0.89
Fish cutlet	2.83±0.95	2.89±0.87	2.85±1.13	2.71±1.09	2.89±1.15	2.83±1.03
Fish finger **	3.03±1.60	3.31±1.69	2.46±1.39	2.21±1.30	2.43±1.37	2.77±1.56
Fish burger	1.31±0.57	1.32±0.69	1.40±0.82	1.50±1.01	1.51±0.87	1.39±0.79

Values are Mean± Standard Deviation. Scale: 1 = Once Daily; 2 = 2 to 5 times a week; 3 = Once every two weeks; 4 = Once a month; 5 = No consumption.
*Significance differences ($p<0.05$); ** Significance differences ($p<0.01$)

Consumption frequency according to social class

For the "social class", all the product items showed significant differences in average consumption among the classes (Table 35). Fish cutlet consumption was found the maximum in the middle social class, followed by lower middle class. In contrast, the average consumption of fish stick, fish ball and fish burger consumption was found higher in the all social class. Interestingly, the fish burger was the most consumed item among all classes of society.

Table 35. Average consumption of fish stick, fish ball, fish cutlet, fish finger and fish burger by social class

	Social class					
Food Type	**Lower (9%)**	**Lower middle (6%)**	**Middle (24%)**	**Upper Middle (56%)**	**Upper (5%)**	**Total**
Fish stick**	1.39±0.49	1.62±0.49	1.59±0.60	1.37±0.50	1.37±0.49	1.44±0.53
Fish ball **	3.38±0.93	3.00±0.00	2.06±0.35	1.00±0.11	1.11±0.58	1.59±0.89
Fish cutlet**	1.27±0.67	2.38±1.00	2.33±0.99	3.24±0.69	4.00±0.00	2.83±1.03
Fish finger **	1.67±1.49	2.44±1.92	1.96±1.70	3.18±1.23	4.25±0.44	2.77±1.56
Fish burger**	3.08±0.86	2.06±0.35	1.40±0.70	1.07±0.37	1.00±0.00	1.39±0.79

Values are Mean± Standard Deviation. Scale: 1 = Once Daily; 2 = 2 to 5 times a week; 3 = Once every two weeks; 4 = Once a month; 5 = No consumption.
*Significance differences ($p<0.05$); ** Significance differences ($p<0.01$)

Consumption according to the number of family member and presence of minors (<18 years old) in home

Fish stick and fish cutlet showed significant variation in the average consumption according to the "number of family member in the home" (Table 36). Fish stick, fish ball, and fish burger presented a higher mean consumption frequency. When the number of people in the home was higher, people mostly preferred fish stick, fish burger, and fish ball. The fish product types that showed significant differences in the average consumption for the "presence of minors younger than 18 years in the home" variable was fish stick, fish ball, and fish burger (Table 36). Fish stick, fish ball, and fish burger were found to consume at a higher mean consumption frequency when there were children younger than 18 years in the home.

Table 36. Average consumption of fish stick, fish ball, fish cutlet, fish finger and fish burger by the number of family member and by the presence of minors (<18years old) in home

	Number of family members					Presence of minors (<18 years)		
Food Type	**≤3 (31%)**	**4 to 5 (52%)**	**≥5 (17%)**	**Total**	**Food Type**	**Yes (69%)**	**No (31%)**	**Total**
Fish stick*	1.38±0.50	1.44±0.51	1.55±0.63	1.44±0.53	Fish stick*	1.43±0.53	1.46±0.55	1.44±0.53
Fish ball	1.54±0.88	1.57±0.89	1.76±0.90	1.59±0.89	Fish ball *	1.61±0.93	1.55±0.81	1.59±0.89
Fish cutlet*	2.96±1.04	2.82±1.01	2.63±1.01	2.83±1.03	Fish cutlet*	2.84±1.06	2.81±0.95	2.83±1.03
Fish finger	2.68±1.48	2.84±1.56	2.71±1.70	2.77±1.56	Fish finger	2.73±1.57	2.84±1.54	2.77±1.56
Fish burger	1.34±0.75	1.40±0.83	1.42±0.70	1.39±0.79	Fish burger*	1.40±0.83	1.36±0.68	1.39±0.79

Values are Mean± Standard Deviation. Scale: 1 = Once Daily; 2 = 2 to 5 times a week; 3 = Once every two weeks; 4 = Once a month; 5 = No consumption.
*Significance differences ($p<0.05$); **Significance differences ($p<0.01$)

Consumption level according to educational level

Significant variation in average consumptions of fish stick, fish ball, fish cutlets, fish finger and fish burger according to the education level were observed (Table 37). For all cases, highest consumption was observed in PhD level. For the all level groups fish stick, fish ball and fish burger were found on the most preferred items. In general, the lower the educational qualification, the lower the mean fish cutlets and fish finger consumption, whereas the average consumption frequency of fish burger was higher.

Consumption pattern according to the geographical area

According to the "geographical area" variable, all types of food items showed significant differences in average consumption pattern (Table 38). Highest average fish stick consumption found in the Sylhet followed by Barisal, Khulna, Rangpur, Rajshahi and Dhaka regions. Areas with the highest fish cutlets and fish finger consumption were Chittagong and Sylhet. Fish finger is consumed most often at Dhaka. Finally, the fish burger was consumed more frequently followed by fish stick, fish ball in Bangladesh.

Table 37. Average consumption of fish stick, fish ball, fish cutlet, fish finger and fish burgeraccording to education level

Food Type	Primary school (1%)	High school (8%)	Secondary (11%)	Higher Secondary (29%)	Diploma (8%)	Bachelor's (30%)	Masters (11%)	PhD (2%)	Total
Fish stick*	2.00±0.00	1.54±0.50	1.28±0.44	1.42±0.50	1.50±0.73	1.42±0.54	1.51±0.50	1.46±0.52	1.44±0.53
Fish ball	1.00±0.00	1.45±0.75	1.55±0.80	1.44±0.78	1.20±0.41	1.61±0.89	2.12±1.11	3.08±1.04	1.59±0.89
Fish cutlet*	3.33±0.52	3.70±1.21	3.26±1.23	2.94±0.95	2.84±0.47	2.65±0.92	2.15±0.76	1.62±0.77	2.83±1.03
Fish finger	4.00±0.89	2.80±1.51	3.12±1.16	3.38±1.56	3.05±1.31	2.24±1.37	2.15±1.47	1.15±0.38	2.77±1.56
Fish burger	1.33±0.82	1.26±0.68	1.34±0.64	1.29±0.65	1.18±0.50	1.35±0.75	1.73±1.20	2.77±1.17	1.39±0.79

Values are Mean± Standard Deviation. Scale: 1 = Once Daily; 2 = 2 to 5 times a week; 3 = Once every two weeks; 4 = Once a month; 5 = No consumption.
*Significance differences ($p<0.05$)
** Significance differences ($p<0.01$)

Table 38. Average consumption according to geographical area.

Food Type	Dhaka (22%)	Chittagong (20%)	Rajshahi (15%)	Khulna (18%)	Rangpur (17%)	Sylhet (5%)	Barisal (3%)	Total
Fish stick**	1.46±0.50	1.56±0.50	1.55±0.55	1.30±0.46	1.37±0.64	1.30±0.46	1.50±0.53	1.44±0.53
Fish ball*	1.47±0.72	1.88±0.95	1.58±0.89	1.51±0.90	1.43±0.81	1.70±1.14	1.87±0.36	1.59±0.89
Fish cutlet**	3.72±1.29	2.34±0.64	2.74±0.88	2.62±0.74	2.65±0.70	2.65±0.86	2.37±0.91	2.83±1.03
Fish finger**	2.07±0.96	2.84±1.80	3.07±1.62	3.02±1.52	2.93±1.61	3.00±1.59	2.75±1.66	2.77±1.56
Fish burger*	1.18±0.50	1.53±0.84	1.40±0.81	1.40±0.84	1.36±0.78	1.55±0.99	1.87±1.24	1.39±0.79

Scale: 1 = Once Daily; 2 = 2 to 5 times a week; 3 = Once every two weeks; 4 = Once a month; 5 = No consumption.
*Significance differences ($p<0.05$)** Significance differences ($p<0.01$)

Discussion

Consumption according to food-related lifestyle (FRL) segments

Factorial analysis of "five factors" explained 57.89% of the total variance. The Kaiser, Meyer and Olkin (KMO) measure of sampling adequacy resulted in a value of 0.720, which was greater than 0.50 (the minimum acceptable value) as suggested by Hair & Jnr, (2010). To best of my knowledge from the literature search, no information available for Bangladesh to support this study result. However, the study conducted by Bernués, Ripoll, & Panea, (2012) provides experiential support for my study. Food-related lifestyle (FRL) model is a powerful tool to determine and characterize consumers segments (Borgogno et al., 2015; Escriba-Perez *et al*., 2017).

From the table-5, we performed the cluster analysis in 5 factors following as (i) Not involved: this is the smallest portion (8%) characterized by those individuals who do not care freshness/quality, prefer to eat at home and depends on other family members decision on food. (ii) Shopping: this portion represents the largest portion (48%) of the sample, characterized by high level of awareness about quality and price ratio, food freshness. Highly concern about food, cuisine preference of other family members. (iii) Restaurant consumer: this portion of consumers comprises (12%) of the sample, prefers to consume outside of the home, maintain a very active social life. Important to note that, this behaviour does not prevent them being interested in the food shopping, quality and price ratio. (iv) Family priorities and social awareness: this is the second largest portion of the sample (32%). It is characterized by individuals who value the family bonding, make active purchases or link consumption to social activities, have high attention to the food preference of other family members, prioritize to consume at home.

Consumption frequencies according to socio-demographic variables

Consumption frequency according to gender

The present survey result was composed of 62% men and 38% women (Table 5.6). The consumption behaviour pattern for men and women was different. As Head et al. (2015) stated, household food purchasing is still performed by the men in Bangladesh. Women have limited or no access to markets (especially in rural Bangladesh) or finance (Coates et al., 2010). Males consistently consumed more proteins and calories than females at all ages, irrespective nutrient requirements due to varying body weight, pregnancy, lactation, and activity levels were considered (Merchant, 2014). However, household food insecurity construct is not as useful in places like Bangladesh where certain food insecurity-related manifestations are not collectively or similarly shared by members of the same living space (Coates et al., 2010).

Consumption frequency according to age

The present survey result was composed of 18% from 16 to 25 years, 29% from 26 years to 35, 30% from 36 to 45 years, 16% from 46 to 55 years, 7% from 56 to more years in age group in showing the Table 5.7. The price of fish stick, fish burger, and fish ball within the range of purchasing capacity of low (er) income people in Bangladesh (Mottaleb *et al.,* 2018). In contrast, fish cutlet, and fish finger were least consumed items for all age groups. Bangladesh is the lowest ten fish product consuming country (Smith, 2017; Speedy, 2003). However, this is compensated to some extent in Bangladesh by higher fish consumption (Bogard *et al*., 2017).

Consumption frequency according to social class

The present survey result was composed 9% lower, 6% lower middle, 24% middle, 56% upper middle and 5% upper in social class showing the table-8. The fish burger is the most consumed item among all class of society. This is may be because of lowest price food in Bangladesh (Waid

et al., 2017), therefore, influencing to buy more economical food items (Sununtnasuk and Fiedler, 2017).

Consumption frequency according to the number of family member and presence of minors (<18 years old) in home

The present survey result was composed 31% ≤3 years, 52% from 4 to 5 years, 17% ≥5 years and another part presence of minor (<18 years old) was composed 69% yes and 31% no in showing table-9. The number of people in the home was higher, people mostly preferred fish stick, fish burger, and fish ball. This is due to the cheapest fish product food items, easy to distribute into the meal (Mottaleb *et al.,* 2018). Fish stick, fish ball, and fish burger were found to consume at a higher mean consumption frequency when there were children younger than 18 years in the home. In contrast, the opposite behaviour was found for fish cutlet and fish finger.

Consumption frequency according to educational level

The present survey result was composed 1% Primary school, 8% High school, 11% Secondary, 29% Higher Secondary, 8% Diploma, 30% Bachelor's, 11% Masters and 2% PhD in educational level showing in Table 5.10. The lower the educational qualification, the lower the mean fish cutlets and fish finger consumption, whereas the average consumption frequency of fish burger was higher. This type of consumption patterns have been observed because of higher educated peoples, generally paid higher salary (Sununtnasuk and Fiedler, 2017). Thus, the consumption frequencies increased among the higher educated people (Davidson and Kropp, 2017; Lin *et al.,* 2017).

Consumption frequency according to the geographical area

The present survey result was composed 22% Dhaka, 20% Chittagong, 15% Rajshahi, 18% Khulna, 17% Rangpur, 5% Sylhet, and 3% Barisal in geographical area showing in the Table 5.11. The highest average fish stick consumption frequencies were found in the Sylhet. The areas with the highest mean fish cutlets and fish finger consumption were Chittagong and Sylhet. Fish finger is consumed most often at Dhaka. Finally, the fish burger was consumed more frequently followed by fish stick, fish ball in Bangladesh. These variations in consumption patterns may, therefore, be related to the consumer behaviors, habits (Molitor *et al*., 2017) and traditional recipes of these geographic areas (Escriba-Perez et al., 2017).

12. Research highlight/findings:

- Reduction of post-harvest losses and the way of product development using mince from low cost fishes.
- Nutrient rich value added fish products such as fish ball, fish burger and fish sticks are developed.
- Standard recipes for the development of value added fish products such as fish burger and fish balls have been established.
- Shelf life of the developed products both in room and refrigerated temperatures have been analyzed and sorted out.
- Consumer's behavior, attitudes and preferences towards different types of value added fish products have been evaluated.

B. Implementation Position

1. Procurement:

Description of equipment and capital items	PP Target		Achievement		Remarks
	Phy (#)	Fin (Tk)	Phy (#)	Fin (Tk)	
(a) Office equipment (Chair, Table, Desk Top Computer, laptop etc.)	08 pics	2.735 lacs	100%	2.735 lacs	-
(b) Lab &field equipment (Soxlet Apparatus, Laminar Air Flow, Freeze, Kjeldahl Apparatus, Hot Air Oven, Incubator, Microoven, Vortex Mixture, Blender and Kitchen Utensils)	12 pics	9.0 lacs	100%	9.0 lacs	-
(c) Camera	1 pic	0.25 lacs	100%	2.25 lacs	-
(d) Other capital items (Chemicals etc)	.	2.00 lacs			

2. Establishment/renovation facilities:

Description of facilities	Newly established		Upgraded/refurbished		Remarks
	PP Target	Achievement	PP Target	Achievement	
Establishment of a fish processing laboratory with some facilities for microbiological analysis	Chair-04, Table-01 Desk Top Computer-01 Laptop-01, Soxlet Apparatus-01 Laminar Air Flow-01, Freeze-02, Kjeldahl Apparatus-01, Hot Air Oven-01, Incubator-01,Micro oven-01, Vortex Mixture-01 Blender -01 , Kitchen Utensils-01 set, Camera-01 and Chemicals	100%	N/A	N/A	

3. Training/study tour/ seminar/workshop/conference organized: Not applicable

C. Financial and physical progress

Fig in Tk

Items of expenditure/activities	Total approved budget	Fund received	Actual expenditure	Balance/ unspent	Physical progress (%)	Reasons for deviation
A. Contractual staff salary	936435	936435	936435	0	100	Rest fund yet not been disbursed
B. Field research/lab expenses and supplies	1895500	1616161	1619124	-2963	100	
C. Operating expenses	566010	563510	563010	500	100	
D. Vehicle hire and fuel, oil & maintenance	55000	50500	46750	3750	100	
E. Training/workshop/seminar	0	0	0	0	100	

etc.						
F. Publications and printing	63155	59405	63155	-3750	100	
G. Miscellaneous	85400	41320	38857	2463	100	
H. Capital expenses	1198500	1198500	1198500	0	100	

D. Achievement of Sub-project by objectives: (Tangible form)

Specific objectives of the sub-project	Major technical activities performed in respect of the set objectives	Output(i.e. product obtained, visible, measurable)	Outcome(short term effect of the research)
To develop fish products and their quality assurance	Production of fish burger, fish balls by using low-cost Pungus, Grass carp and Poa fishes	85% value addition have been achieved	The poor people will be able to get increased availability of nutrition in fish products at cheap rate. Protein intake of the poorer section will be increased
To assess consumers' attitude towards different fish products.)	An questionnaire interview were performed taking people of different strata and different age, sex etc.	A clear idea has been understood for the profitable marketing considering peoples need and capacity	A profitable recipe for the utilization low priced fish developed

E. Materials Development/Publication made under the Sub-project:

Publication	Number of publication		Remarks (e.g. paper title, name of journal, conference name, etc.)
	Under preparation	Completed and published	
Technology bulletin/ booklet/leaflet/flyer etc.	A leaf let is ready		Fund yet no available
Journal publication	04	**01**	Paper title: Consumers profile analysis towards chicken, beef, mutton, fish and egg consumption in Bangladesh Name of Journal: British Food Journal
Information development			A package of fish burger preparation has been developed along with package of fish ball
Other publications, if any	-	-	-

F. Technology/Knowledge generation/Policy Support (as applied):

i. Generation of technology (Commodity & Non-commodity)

Technology for the utilization of different types of low cost fish species through the development of different types of value added fish products have been developed. Standard recipe for the development of Grass carp fish burger has been established.

ii. **Generation of new knowledge that help in developing more technology in future**

Consumer's behavior, attitudes and preferences towards different types of value added fish products have been evaluated. This knowledge will surely help to develop new technologies for the preparation of new products of consumer interest and will create opportunities for the proper utilization of different types of fish and fishery products. This will ultimately contribute in post-harvest loss reduction.

iii. Technology transferred that help increased agricultural productivity and farmers' income

For the dissemination of the newly developed technology to the farmers' level, a training program was conducted. Fast food producer groups were trained theoretically about the procedure for preparation of different types of value added fish products. Details information about the procedure, ingredients, equipment's and other necessary aspects for the preparation and marketing of the value added fish products was presented. The training was conducted in combination of oral presentation and pictorial demonstration. Total 40 participants were participated in the session. Among them, there were 23 females and 17 males. Female participants were the wives, sisters and relatives of the selected group members Women were more interested in preparation of the product than the males.

iv. Policy Support

Not applicable

G. Information regarding Desk and Field Monitoring

i) Desk /Monitoring [description & output of consultation meeting, monitoring workshops/seminars etc.):

Monitoring team	Date(s) of visit	Total visit till date (No.)	Output
Sylhet Agricultural University Research System	25.01.2018	01	Suggestion for immediate starting of laboratory work.
Do	17.09.18	01	Suggestion for immediate completion of PCR.

ii) Field Monitoring (time& No. of visit, Team visit and output):

Monitoring team	Date(s) of visit	Total visit till date (No.)	Output
Technical Division, BARC	05.02.2018	01	Suggested for immediate completion of procurement
PIU-BARC, NATP-2	07.04.18	01	Suggested for immediate staring of lab-based work
Internal Monitoring Team	08.05.18	01	Suggested for more lab-based work

H. Lesson Learned (if any)

Collecting premium quality marine fishes and its management.

I. Challenges (if any)

i) Maintenance of proper storage temperature.
ii) Maintenance of high temperature in muffle furnace.

Signature of the Principal Investigator
Date …….

Seal

Counter signature of the Head of the organization/authorized representative
Date …….

Seal

J. References:

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