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Quality Assessment and Shelf Life Evaluation of Ready to Eat [Fish Cutlet] of Bycatch Anchovies during Frozen Storage

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A B S T R A C T

An attempt was undertaken to evaluate the shelf-life of Ready To Eat (fish cutlet) of anchovies (*Stolephorus commersonii*). Fish cutlet was prepared by following standardized recipe and it was subjected to proximate composition, biochemical, microbiological and organoleptic analysis at an interval of three days during 18 days of storage period. Results for protein, lipid, ash and moisture indicates decreasing trend significantly from 15.14% to 13.26, 12.33% to 10.55%, 2.86% to 2.47% and 60.59% to 57.22% ($P < 0.05$) respectively. However $_{P}H$, Peroxide value, free fatty acid and Total volatile base Nitrogen depicted an increasing trend significantly ($P < 0.05$) from 6.44 to 6.70, 6.07 to 11.87 meq O_2/kg of fat, 2.13 to 5.01% of oleic acid and 3.85 to 20.32 % mgN/100g of sample respectively. TPC of bacteria was increased during storage day from 5.70×10^2 to 7.1×10^5 cfu/g of sample. Similarly the psychrophilic count was also increased from 2.6×10^2 to 6.8×10^4 cfu/g of sample. The organoleptic score for appearance, colour, taste, odour, texture and overall acceptability reduced significantly ($p < 0.05$) with increase of storage period. Therefore an ideal shelflife for storing fish cutlet in refrigerated condition was found as 18 days.

Introduction

Consumers around the world have increased in consumption of fish and fishery product in recent years due to recognition of their nutritional value (Wang *et al.*, 2010). Ready to eat or serve fishery products gaining good consumer preference. When it is prepared from by catch and discards, it further contributes to the economic growth of producers and as well as nation (Sowmya Praneetha *et al.*, 2017). Shrimp trawl by catch like anchovies could be properly utilized by transforming low-valued fishes into value added product. During recent year's value addition have received more attention because of increased urbanization due to social and cultural changes (Pagarkar *et al.*, 2011). There is great demand for seafood based product especially battered and breaded products. Battered and breaded or coated systems have the benefits of versatility and familiarity because they enhance the flavour and texture of processed food products (Biswas *et al.*,

2004). The process of canning with batter and bread crumbs increases the bulk of the product, thereby reducing the content of costly fish and thus reducing the cost of products, coating enhances the appearance, colour, texture, and taste of food products and also the nutritional value of the product (Rathod *et al.*, 2012). However, there are almost no studies on fish cutlet developed from anchovies (*Stolephorus commersonii*). The aim of this study was to prepare fish cutlet from anchovies and to investigate quality changes for shelf-life evaluation viz., proximate, biochemical, microbiological and organoleptic analysis during refrigerated storage.

Materials and Methods

Sample preparation

Fresh by catches of anchovies (*Stolephorus commersonii*) with an average length and weight of 8-9 cm and 5-6 g were purchased from local market of Ponneri, Thiruvallur district. It was immediately transported to the processing lab within 20mins in polythene bags along with crushed ice. The head and visceral of anchovies were removed and cleaned fish was washed with large amount of water. The entire fish was boiled for 10 mins without removing bones, after that it was finally minced using domestic mincer to prepare fish cutlet.

Shaped cutlet was battered by dipping it in egg and rolled in bread crumbs powder. After battering that was flash fried at 180^{o}C for 30 sec. The product were later cooled, packed in HDPE pouches, sealed, labelled and stored at frozen storage for quality evaluation. Samples were drawn randomly in duplicates at 3 days intervals up to 18 days of frozen storage to analyse proximate, biochemical, microbiological and organoleptic parameters. All the parameters were analysed in triplicate from initial to final storage day.

Analysis

Proximate analysis

Proximate composition viz, moisture, crude protein, fat, ash of fish cutlet were analysed according to AOAC method (2005).

Bio chemical analysis

pH was determined by taking sample (5g) and added with 45ml of distilled water. Then filtered using a filtered paper. The pH of filtrate was recorded using pH meter AOAC method (2005). Peroxide value (PV) was measured using titrimetric determination of amount of peroxide or hydro peroxide groups, PV was expressed as milli equivalent of O_2 / kg fat AOAC method (2005). Free fatty acid (FFA) value was determined as per AOAC method (2005) and expressed as percent (%) of oleic acid. TVBN contents were determined by procedure given by Beatty and Gibbons (1937) using Conway micro-diffusion units and results were expressed in terms of nitrogen mg/100g.

Sensory analysis

Organoleptic quality of fish cutlets were evaluated using nine point hedonic scale [1-dislike extremely to 9- like extremely]. Experienced members of the institute were indulged in the panel for evaluating the product acceptability. The characteristics covered under the panel were appearance, colour, odour, flavour, taste, texture and overall acceptability (Reddy, 1992).

Statistical analysis

Statistical package was used for analysis of experimental results. Sufficient number of samples was carried out for each analysis. The results were expressed as mean ± standard deviation(SD).The significant differences

were tested by 5% level of significance and are mentioned as ($P<0.05$) for significances difference by Snedecor and Cochran (1967).

Results and Discussion

Proximate composition

The percentage of proximate composition like moisture, protein, fat, and ash content of fresh anchovies (*Stolephorus commersonii*) were 78.39±0.38, 17.14±0.87, 2.34±0.30 and 1.98±0.25 respectively. Similarly with slight variation was reported by (Palanikumar *et al.,* 2014) as moisture (79.32±0.17), protein (16.32±0.93), lipid (2.41±0.17) and ash (1.31±0.02) contents in fresh anchovies, whereas (Chao minh le *et al.,* 2015) reported the moisture, protein, fat, and ash content in fresh anchovies were 78.3±0.7, 16.3±0.3, 2.4±0.3 and 2.5±0.4 respectively. The moisture, protein, fat, and ash of anchovies fish cutlet were found to be 60.59±1.66, 15.14±0.84, 12.33±0.28 and 2.86±0.72 respectively which at the end of storage period, it showed the variation in proximate composition of the moisture, protein, fat, and ash content of fish cutlet were decreased from 60.59±1.66 to 57.22±1.71%, 15.14±0.84% to 13.26±1.19%, 12.33±0.28 to 10.55±0.33% and 2.86±0.72% to 2.47±0.52% respectively. Reddy *et al.,* (1992) reported decrease in moisture content in fish cutlet prepared from reef cod mince. The reduction of protein is due to denaturation of fish muscle during chilled and frozen storage (Gopakumar, 2002). Sehgel *et al.,* (2010) suggested that cooking can be a possible reason for reduction in the protein content. Similarly, the decrease in lipid content has been reported by siddique et.al. (2011) in *Puntius spp*. Oxidation of fat in product led to reduction in fat content (Mc Gill *et al.,* 1974; Josephson, 1989). Also, the decrease in ash content could be due to drip loss resulting in loss of bulk and trace elements (Gandotra *et al.,* 2014). In the present study, it was observed that moisture, protein, fat, and ash content of fish cutlet put together as 91% and the remaining percentage might be contributed by carbohydrates (Takur *et al.,* 2006). Addition of coating materials like bread crumbs gives higher percentage of carbohydrate content to the product (Sayar, 2011).

Biochemical change

pH

The pH of fish cutlet increased from 6.44±0.09 to 6.70±0.09. It might be due to decomposition of nitrogenous components in post-mortem. Bett and Dionigi, (1977); Turhan *et al.,* (2001) reported pH increase is attributed due to release of CO_2 by microbial flora present in product. Enzymatic degradation increases the pH in fish muscle (Love, 1992 and Vareltiset *et al.,* 1997). Breakage of hydrogen bond and electrostatic interaction leads to increase in pH (Dhanpal *et al.,* 2012). Pawar (2011) reported increasing trend of pH for cutlet made from catla fish from 6.50 to 6.79 at -2 to -4^{o}C. Similar observation was also made by the Kilinc, (2007) during refrigeration storage of fish patties produced from anchovy. Increasing trend in pH has been observed by Rathod and Pagarkar (2013) in Pangasius fish cutlets.

Peroxide value

Peroxide value of fish cutlet during the initial storage period increased from 6.07±0.23 to 14.65±2.54 meq O_2/kg of fat at the end of 12th day storage and after that PV value subsequently decreased to 11.87±0.69 meq o_2/kg of fat at the end of 18th day of storage. (Kamat, 1999) reported that PV of mackerel mince cutlet was increased from an initial value of 3.4 to 40 meq O_2/kg at the end of 5th week of storage at -14^{o}C respectively. PV value of fish burger prepared from tilapia

(*Oreochromis niloticus)* was 0.18 meq/kg at the beginning of storage period and increased to 5.03 meq/kg at the 6th month and then decreased to 0.82 meq/kg at the end of 8th month (Tokur *et al.,* 2006). The unstable nature of PV is due to decomposition of peroxides and its products may internet with protein, which results in PV reduction (Yerlikaya *et al.,* 2005).

FFA

FFA of anchovies fish cutlet showed an increasing trend from 2.13±0.36 to 5.01±0.21 mg/100g. Similar results with slight variation were reported by Nikheel Rathod and Asif Pagarkar (2013) with increase in FFA of pangasius fish cutlet from 1.26 to 4.83 mg/100g. FFA is a result of enzymatic decomposition of lipid during storage (Tokur *et al.,* 2006). Cooking of minced meat might have deactivated the lipolytic enzymes and subsequent flash frying of product led to increase in FFA (Ninan *et al.,* 2011). FFA content showed variation in fresh fried and raw cutlet in the range of 0.98 to 1.49 and 2.03 to 2.82 mg/100g respectively at 4ºC (Joseph *et al.,* 1984). Tokur *et al.,* (2006) also reported increased FAA from the begining of the storage period up to 8th month.

TVBN (Total Volatile base Nitrogen)

TVBN is known as a product of bacterial spoilage and is a commonly used chemical method to determine spoilage of fish (Tokur *et al.,* 2004). TVBN value of anchovies cutlet showed increasing trend from 3.85±0.19 to 20.32±0.95 during storage period. Increase of TVBN content during storage is due to bacterial spoilage, activity of endogenous enzymes and degradation of tissue proteins (Chomnawang *et al.,* 2007) or production of ammonia (Adebona, 1978). TVBN value of pangasius fish cutlet showed increased trend from 2.52 to 22.4 mg/100g Nikheel Rathod and Asif Pagarkar, (2013). Bao *et al.,* (2007) reported increasing trend of TVBN in arctic charr (*Salvelinus alpines*) fillets at chilled storage temperature. TVBN value of tilapia fish cutlet was increased in the range of 12.4 to 20.2 mg% respectively (Ninan *et al.,* 2008).

Table.1 Using standardized recipe the cutlets were prepared (Pawar *et al.,* 2011)

INGREDIENTS	QUANTITY(g)
Cooked fish meat	100
Table salt	3
Green chilly	5
Coriander leaves	5
Ginger	5
Garlic	5
Onion	25
Potato cooked	70
Pepper powder	0.3
Clove powder	0.3
Cinnamon powder	0.2
Turmeric powder	0.2
Bread powder	20

Oil – Sunflower oil was used (10ml) for heating purpose

Table.2 proximate composition of raw meat, initial and final day of cutlet

Attributes	Raw meat	Cutlet Initial day	Cutlet Final day
Moisture	78.39±0.38	60.59±1.66	57.22±1.71
Protein	17.14±0.87	15.14±0.84	13.26±1.19
Fat	2.34±0.30	12.33±0.28	10.55±0.33
Ash	1.98±0.25	2.86±0.72	2.47±0.52

Table.3 proximate composition of cutlet during storage days

Storage duration (days)	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0	60.59±2.04^{a}	15.14±1.03^{a}	12.33±0.34^{e}	2.86±0.89^{a}
3	60.11±2.07^{a}	14.84±1.12^{a}	12.06±0.25d^{e}	2.80±0.87^{a}
6	59.66±2.02^{a}	14.64±1.21^{a}	11.75±0.20^{cd}	2.72±0.88^{a}
9	59.14±1.66^{a}	14.30±1.23^{a}	11.48±0.23^{bc}	2.66±0.82^{a}
12	58.77±1.70^{a}	13.83±1.47^{a}	11.08±0.26^{ab}	2.62±0.79^{a}
15	57.93±2.06^{a}	13.55±1.60^{a}	10.98±0.33^{a}	2.59±0.77^{a}
18	57.22±2.09^{a}	13.26±1.46^{a}	10.55±0.41^{a}	2.47±0.64^{a}

Each value is represented as the mean ± SD of n=3
^{abcde}Means followed by the same superscript within a column

Table.4 Biochemical changes in cutlet during storage

Storage duration (days)	pH	PV(meq O2/kg of fat)	TVBN(mg/100g of sample)	FFA (% of oleic acid)
0	6.44±0.09^{a}	6.07±0.23^{a}	3.85±0.19^{a}	2.13±0.36^{a}
3	6.48±0.10^{ab}	7.78±1.08^{b}	5.04±1.13^{a}	2.55±0.32^{a}
6	6.52±0.10^{abc}	9.16±0.78^{c}	8.45±1.80^{b}	3.24±0.41^{b}
9	6.56±0.10^{abc}	11.76±0.87^{d}	11.22±2.41^{bc}	3.88±0.44^{b}
12	6.61±0.10^{abc}	15.26±0.61^{e}	14.65±2.54^{cd}	4.33±0.48^{c}
15	6.64±0.10^{bc}	14.09±0.74^{e}	17.22±1.62^{d}	4.59±0.37^{c}
18	6.70±0.09^{c}	11.87±0.69^{d}	20.32±0.95^{d}	5.01±0.21^{d}

Table.5 Microbiological changes in cutlet during storage

Storage period (days)	Total plate count(cfu/g of sample)	Psychrophilic count (cfu/g of sample)
0	5.7×10^{2}(2.75)	2.6×10^{2}(2.41)
3	2.8×10^{3}(3.44)	5.3×10^{2}(2.72)
6	4.0×10^{3}(3.60)	7.9×10^{2}(2.89)
9	1.23×10^{4}(4.08)	1.4×10^{3}(3.14)
12	5.8×10^{4}(4.76)	6.5×10^{3}(3.81)
15	3.6×10^{5}(5.55)	2.3×10^{4}(4.36)
18	7.1×10^{5}(5.85)	6.8×10^{4}(4.83)

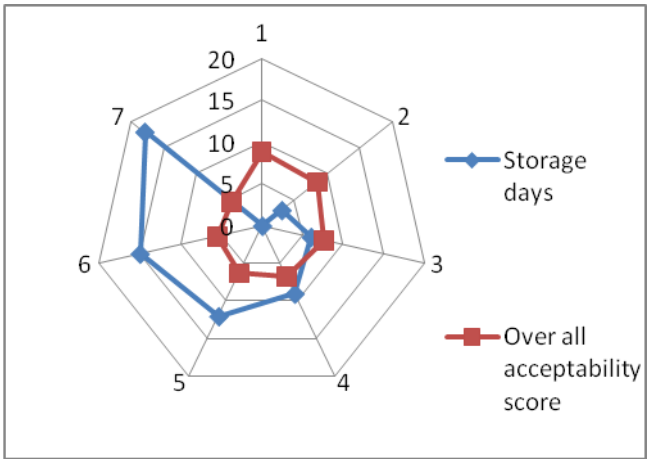
Table.6 Organoleptic changes in cutlet during storage

Storage period (days)	Overall acceptability score
0	8.89 ± 0.11^{g}
3	8.45 ± 0.14^{f}
6	7.62 ± 0.16^{e}
9	6.71 ± 0.18^{d}
12	6.23 ± 0.17^{c}
15	5.48 ± 0.24^{b}
18	4.63 ± 0.23^{a}

Each value is represented as the mean ± SD of n=3

^{abcdefg}Means followed by the same superscript with in a column

Chart.1 Organoleptic evaluation of fish cutlet



Microbiological changes

The changes in total plate count and psychrophilic bacterial count were enumerated for fish cutlet during storage period. TPC was increased from 5.7 $\times10^{2}$ (2.75) to 7.1 $\times$ 10^{5} (5.85) cfu/g. Similarly the psychrophilic count was also increased from 2.6 $\times10^{2}$(2.41) to 6.8 $\times$ 10^{4}(4.83) cfu/g during storage. The same increase of bacterial count was observed by Joseph *et al.*, (1984) in fish cutlet during storage at 4^{0}C. Similar increasing trend of bacterial count was reported by (Sowmya praneetha *et al.*, 2017), However, this level did not exceed the maximum limit (7 log cfu/g for meat) as described by ICMSF (1978). Pathogenic and spoilage bacterial like *salmonella spp.*, *vibrio spp*. and *Escherichia coli* were not detected in fish cutlet, this could be due to flash frying of the product.

Organoleptic evaluation

Score of sensory attributes like appearance, colour, taste, texture and odour were assessed and the mean value for overall acceptability score (OAS) was determined. It could be observed that overall mean acceptability score reduced significantly with increase of storage period. The ideal shelf life for storing anchovies fish cutlet at frozen storage condition was found to be 18 days.

It is concluded that, in recent years, increase of world population as well as various socio

economic changes led to an increased consumer preference for ready to eat food. From this attempt, quality and shelf life of fish cutlet were assessed, where all the quality criteria mentioned above are well below the maximum limit for the acceptability fish cutlets. This study will give an idea on shelf life stability and quality of fish cutlet during frozen storage and it will be useful for retailers and large scale food marketers.

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