



Evaluation of Proximate Composition, Oxidative Stability and Sensory Characteristics of Ready to Eat Fish Balls Prepared from Rohu (*Labeo Rohita*) Mince Stored at Refrigeration Temperature

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ABSTRACT

Demand for value added fish products is increasing continuously and a wide range of 'Ready to Cook (RTC)' or 'Ready to Eat (RTE)' fish products are available in the market. To study the shelf life of ready to eat fish balls prepared from rohu (*Labeo rohita*) mince and stored at refrigerated temperature ($4\pm 1^\circ\text{C}$) under aerobic packaging fish on the basis of changes in proximate composition, oxidative stability and sensory characteristics. With the progress of time moisture content in fish balls reduced significantly ($p < 0.05$) from 45.56 to 41.59%, whereas protein content increased significantly from 21.43 to 23.52% on 21st day. No significant ($p < 0.05$) change in fat, carbohydrate and ash content recorded during the storage period. Significant ($p < 0.05$) change in biochemical parameters like pH (5.24 to 5.58), Titratable Acidity (0.38 to 0.63% lactic acid), Peroxide Value (1.52 to 3.7 meq/kg), free fatty acid (0.18 to 0.42%) and Thiobarbituric Acid Reactive Substances (TBARS) 0.86 to 1.32 mg malonaldehyde/kg recorded on 21st day. Textural changes as hardness, fracturability, springiness, chewiness, shear force and work of shear showed significant ($p < 0.05$) change with progress of storage period. Ready to Eat fish balls shows better acceptability upto 14th day at refrigerated temperature ($4\pm 1^\circ\text{C}$) under aerobic packaging.

Key Words: Fish balls, aerobic packaging, oxidative stability, sensory characteristics

INTRODUCTION

Fish is an excellent source of quality protein with high biological value (BV), contains essential amino acids, poly unsaturated fatty acids (PUFA) along with essential vitamins (A, B and D) and minerals like calcium, selenium, zinc, iodine, iron (FAO, 2020). In recent times, consumption of fish and fishery items has witnessed a steady increase due to various reasons including changes in lifestyle, perception of fish as a healthy food (Singh and Surasani, 2020) and it has the ability to lower the blood cholesterol level, reduce heart attack risks, reduces blood pressure, regulates heart rhythms and hence reducing possible heart failures (Martha, 2002). Further, Omega-3 fatty acids from fish supplements are helpful in preventing a number of diseases including Diabetes, Arthritis, Cancer,

Depression, Hyperactivity (Khora, 2013). Carps are the mainstay of Indian freshwater aqua farming system and rohu (*Labeo rohita*) is one of the major fish species belongs to carp. Due to excellent culinary properties, high production, throughout year availability, it always remains in demand. Rapid urbanization, more working hours and modern life style is acting as triggering force in food consumption pattern and preference, earlier people prefer to eat fresh food but now they are demanding customized products in the form of 'Ready to Cook (RTC)' or 'Ready to Eat (RTE)', and fish is also not an exception. Present trend of marketing reflects a sharp growth in demand of value added fish products such as fish fillets, fingers, nuggets, cutlets, patties balls, sausages and other texturized products as they are more convenient to handle,

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store and cook (Kolekar and Pagarkar, 2013).

In processed food, protecting the changes in biochemical characteristics, texture, culinary and sensorial properties and microbial growth are major challenges required to be maintained during its storage. Storage of fresh/ processed products for short duration under chilled conditions is most common practice to preserve the food including fish. Quality of food product is an important issue which declines with storage time and the consumer always show concern about it. The present study was conducted for 28 days to study the shelf life of ready to eat fish balls prepared from rohu (*Labeo rohita*) mince and stored at refrigerated temperature ($4\pm 1^\circ\text{C}$) under aerobic packaging fish on the basis of changes in proximate composition, oxidative stability and sensory characteristics.

MATERIALS AND METHODS

For present study, fish balls were prepared by mixing ingredients as per quantity given in Table 1 and fish balls were prepared as per process given in Fig. 1.

After cooling, cooked fish balls were packed in High Density Polyethylene (HDPE) clear bags and sealed with the help of hand sealer for aerobic packaging and stored in refrigerator. For analysis, samples were withdrawn at every 7 days intervals viz; 1,7,14,21 and 28th day to evaluate proximate composition (moisture, protein, fat and ash) as per the method describe by AOAC, 2000. The

Table 1. Quantity of ingredients for fish ball preparation.

Sr. No.	Ingredient	Quantity
1	Fish Mince	800g
2	Refined flour	120g
3	Breadcrumb	55g
4	Red chilies powder	2g
5	Black pepper Powder	2g
6	Clove powder	0.5g
7	Common salt	20g
8	Vinegar (Acetic acid)	25ml

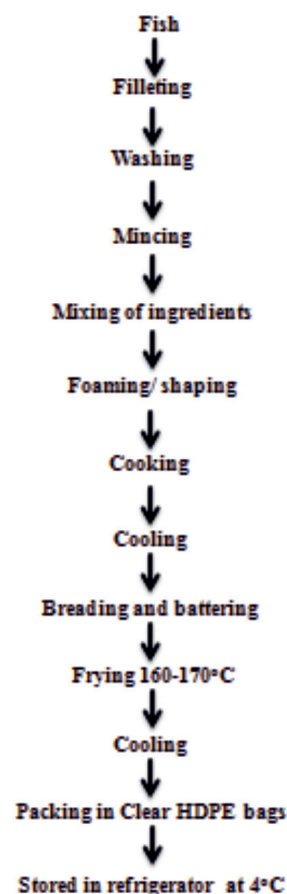


Fig. 1. Fish ball preparation process

carbohydrate content was calculated by numerical formula [Carbohydrate (%) = 100 - (Moisture % + Protein % + Fat % + Ash %)]. Biochemical characteristics like pH (Trout *et al*, 1992), Titratable Acidity (TA, % lactic acid) (Shelef and Jay, 1970), Peroxide Value (PV, meq/kg), Free Fatty Acid (FFA, %) (Koniecko, 1979), Thiobarbituric Acid Reactive Substances (TBARS, mg malonaldehyde/kg) (Witte *et al*, 1970), textural quality attributes as Hardness (kg), Fracturability (kg), Springiness (cm), Chewiness (kg), Shear Force (kg) and Work of Shear (kg/s) and sensory properties using nine point 'Hedonic Scale' as per likings suggested by Popper *et al* (2004).

STATISTICAL ANALYSIS

Samples were taken in triplicate and the data obtained during the study were statistically

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analysed by Duncan -t-test under SPSS 16.0 by one-way ANOVA. The results were expressed as Mean± Standard Error (SE) along with statistical differences.

RESULTS AND DISCUSSION

Proximate Composition

Changes in proximate composition of fish balls stored at refrigerated temperature ($4\pm 1^\circ\text{C}$) under aerobic packaging were evaluated to observe changes in proximate composition w.r.t. progress of storage period. During storage period, on 21st day significant decrease ($p < 0.05$) in moisture recorded which was reduced from 45.56 to 41.59%, whereas protein content increased significant ($p < 0.05$) from 21.43 to 23.52%. However, no significant change in fat, ash and carbohydrate content (Table 2) recorded throughout the study period. Mugale (2015) also reported reduction in moisture of treated fish balls when stored under chilled storage conditions. In present study, increase in protein content during storage may be the effect of loss of moisture in fish balls during storage. Reddy and Kahirnar (2015) and Parnetha *et al* (2017) studied and reported proximate composition of rohu (*Labeo rohita*) mince incorporated fish cutlets and our result also almost equivalent to their findings. Pawar *et al* (2013) reported the loss of moisture during frying and results in higher protein content in fried fish cutlets as compared to fresh fish cutlets, prepared from Catla fish meat. They also suggested that the decrease in protein content in fish cutlet

during the storage period could be connected with denaturation of fish proteins during frozen storage. According to them, in raw catla fish cutlet showed slight reduction in moisture (65.71 to 64.86%) and protein (16.57 to 15.86%) content during its frozen storage.

Biochemical Parameters

Biochemical parameters of stored fish balls stored at refrigeration temperature ($4\pm 1^\circ\text{C}$) under aerobic packaging conditions were performed to observe changes in pH, Titratable Acidity, Peroxide Value, Free Fatty Acid and TBARS. During storage, the pH reduced from 5.58 to 5.24 and recorded significant ($p < 0.05$) change on 21st day of storage, which may be the effect of oxidative or hydrolytic activity of nutritional components. Singh *et al* (2014) reported pH 5.23-5.48 in fish cutlets prepared from common carp meat, while Gupta *et al* (2015) reported pH value in the range of 6.3 – 7.4 in fish cutlets of *Wallago attu* meat. Titratable Acidity in fish balls increased significantly ($p < 0.05$) on 21st day of storage it's value ranged between 0.38 to 0.63% lactic acid. In present study, change in peroxide value recorded in between 1.52 to 3.72 meq/kg which may be due to unstable nature of nutrients which leads towards spoilage. Ninan *et al*, 2011 reported peroxide value in tilapia (*Oreocromis mossambicus*) fish cutlet gradually increased upto 12-15 weeks, thereafter decreased under frozen storage condition. According to them spices added in the product act as antioxidant, breeding and

Table 2. Changes in Proximate Composition (%) of Fish Balls during Storage at $4\pm 1^\circ\text{C}$ Under Aerobic Packaging (Mean ±SE).

Day	Parameters (%)				
	Moisture (%)	Protein (%)	Fat (%)	Ash (%)	Carbohydrate (%)
1	45.56 ^a ± 0.29	21.43 ^b ±0.12	14.85 ^a ±0.19	2.86 ^a ±0.13	14.90 ^a ±0.26
7	45.60 ^a ±0.26	21.59 ^b ±0.23	14.78 ^a ±0.47	2.98 ^a ±0.25	15.19 ^a ±0.25
14	45.83 ^a ±0.22	21.88 ^b ±0.29	14.93 ^a ±0.29	3.09 ^a ±0.03	15.12 ^a ±0.52
21	42.77 ^a ±0.24	23.34 ^a ±0.23	14.83 ^a ±0.19	3.13 ^a ±0.24	15.73 ^a ±0.51
28	41.59 ^a ±0.17	23.52 ^a ±0.11	14.93 ^a ±0.13	3.32 ^a ±0.20	16.44 ^a ±0.60

$p < 0.05$

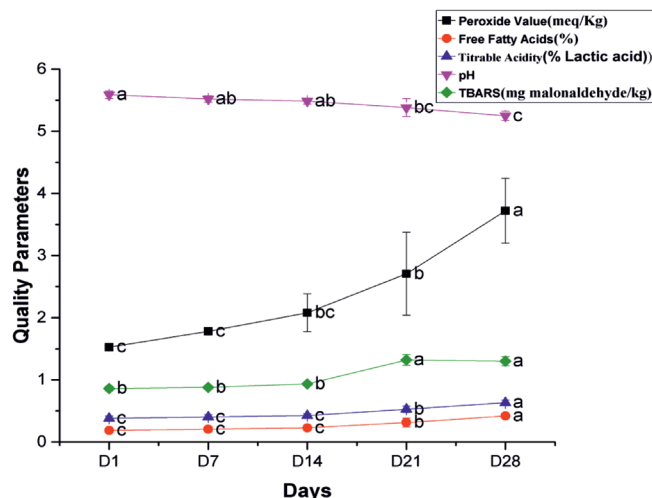


Fig 2. Changes in Biochemical Parameters of Fish Balls during Storage at 4±1°C Under Aerobic Packaging (Mean ±SE)

battering act as oxygen barriers helps to prevent the product from peroxide formation. In present study, free fatty acid value increased from 0.18 – 0.42% however the values recorded were almost within the range reported by Ninan *et al* (2011) (0.19-0.68 mg% oleic acid) in freeze dried fish balls prepared from rohu meat. Pawar (2013) also reported increase in FFA value during storage period at -18°C in fish cutlet prepared from catla fish. In present study, no significant ($p < 0.05$) change in TBARS value recorded till 14th day however on 21st day significant increase recorded. TBARS value recorded initially 0.86 and it increased 1.32 mg malonaldehyde/kg on 21st day. TBARS value is an indicator of degree of lipid oxidation (Aubourg, 1999). Gupta *et al* (2015) reported value of TBARS 0.15 mg MA/kg on day one, while 2.02 on 30th day in cooked fish cutlets stored at -12°C. In present study, lower TBARS value in fish balls could be due to the peroxide scavenging activity of added spices which possess antioxidant properties (Coban, 2013 and Frank *et al*, 2014). TBARS is present in the second stage auto-oxidation when peroxides are decomposed to aldehydes and ketones (Gokoglu, 2012). Details of changes in pH, Titratable Acidity, Peroxide Value, Free Fatty Acid and TBARS value is given in Fig. 2.

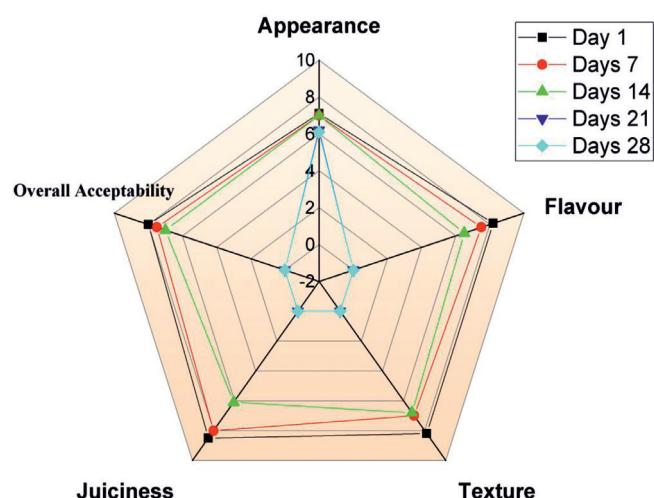


Fig. 3. Sensory Evaluation of Fish Balls for Consumers' Acceptability

Texture Properties

The textural properties of fish balls were tested as Texture profile analysis (TPA) (*two-cycle compression test*) using a TA.XT Plus Texture Analyzer (Stable Micro Systems Ltd., Surrey, UK), with a 50-kg load cell. A 75-mm-diameter compression platen was used with a speed of 1 mm/s throughout the process. A blade set (HDP/BSW) was used with a speed of 2 mm/s throughout the process. Hardness, fracturability, springiness, chewiness, shear force and work of shear was measured during the study period and on 21st day hardness and work of shear was increased significantly ($p < 0.05$), which may be due to loss of moisture and reflected in terms of increase in hardness, values for chewiness decreased significantly on 28th day after storage, whereas no change in fracturability, springiness and shear force recorded in fish balls (Table 3).

Sensory Evaluation

Sensory evaluation of fish balls was performed using nine point 'Hedonic Scale' (1-dislike extremely to 9-like extremely) for product acceptability suggested by Popper *et al* (2004). Till 14th day evaluators accepted the product with high likings but on 21st day evaluators refused for sensory evaluation. Details of sensory evaluation is

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Table 3. Changes in Textural Properties of Fish Balls during Storage at 4±1°C Under Aerobic Packaging (Mean ±SE).

Day	Parameters					
	Hardness (kg)	Fracturability (kg)	Springiness (cm)	Chewiness (kg)	Shear force (kg)	Work of shear (kg/s)
1	10.79 ^c ±0.11	5.41 ^a ±0.11	1.25 ^b ±0.01	5.43 ^a ±0.11	7.88 ^a ±0.99	96.24 ^a ±0.13
7	10.64 ^c ±0.05	5.59 ^a ±0.06	1.25 ^b ±0.01	5.64 ^a ±0.11	6.57 ^a ±0.14	94.95 ^a ±0.92
14	10.68 ^c ±0.21	5.63 ^a ±0.09	1.30 ^b ±0.02	5.25 ^{ab} ±0.22	6.32 ^a ±0.05	96.23 ^a ±0.15
21	11.42 ^b ±0.11	5.67 ^a ±0.10	1.31 ^b ±0.10	4.86 ^b ±0.13	6.55 ^a ±0.15	95.43 ^a ±0.59
28	12.06 ^a ±0.09	5.56 ^a ±0.15	1.59 ^a ±0.15	4.35 ^c ±0.14	6.50 ^a ±0.20	95.14 ^a ±0.13

$p < 0.05$

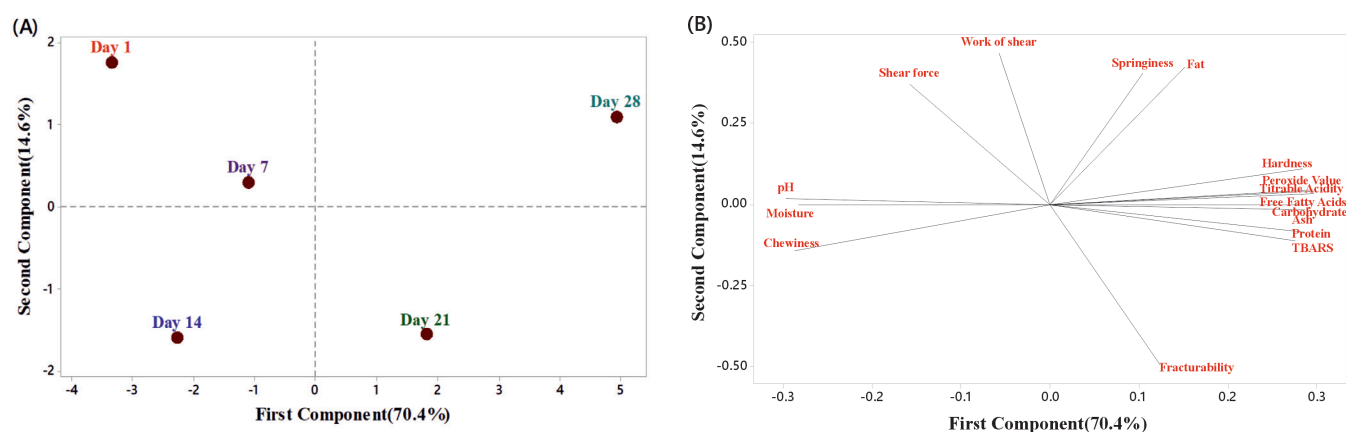


Fig. 4 A & B Showing principal component analysis (PCA) to estimate the relationships between quality parameters and storage days of fish ball samples. The arrangement of storage days along the PCA dimensions is depicted in Fig. 4A, while the distribution of quality attributes within the PCA-defined space is illustrated in Fig. 4B.

given in Fig. 3.

Principal Component Analysis (PCA)

Principal component analysis (PCA) was performed to assess the relationships between quality parameters and storage days of fish ball samples. Fig. 4A depicts the arrangement of storage days, while Fig. 4B illustrates the distribution of quality attributes within the PCA-defined space. The combined variance explained by PC1 and PC2 accounted for 85% of the variations among the samples. The PCA analysis provided valuable insights into the changes occurring in the quality attributes during the 28 days storage period, revealing a discernible pattern of variation along

the storage duration. Notably, no significant difference was observed between the 1st and 7th day of storage. The results of the PCA analysis indicated a significant relationship between quality parameters and storage days. PC1 explained 70.4% of the variations, while PC2 explained 14.6%. Overall, the quality of the fish balls deteriorated with increasing storage time, as evidenced by a decrease in pH, moisture, chewiness, and shear force, and an increase in springiness, fat, hardness, PV, titratable acidity, FFA, carbohydrate, ash, protein, TBARS, and fracturability.

CONCLUSION

On the basis of proximate composition,

biochemical parameters, textural and sensory characteristics analysis of ready to eat fish balls prepared from rohu mince and stored at refrigeration temperature ($4\pm 1^\circ\text{C}$) under aerobic packaging clearly indicate that the balls possess better acceptability and can be consumed up to 14th day of storage period when stored at refrigerated temperature ($4\pm 1^\circ\text{C}$) under aerobic packaging.

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