SUITABILITY OF FIBROUS CASING FOR THE PRODUCTION OF FISH HAM

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CERTIFICATE

This is to certify that the thesis entitled "***Suitability of Fibrous Casing for the Production of Fish Ham***" submitted by **Ms. Rajalakshmi R., I.D. No. MFK-1005** in partial fulfillment of the requirements for the award of **Master of Fisheries Science** in **Fish Processing Technology** of the **Karnataka Veterinary, Animal and Fisheries Sciences University, Bidar** is a record of bonafide research work carried out by her during the period of her study in this University under my guidance and supervision and the thesis has not previously formed the basis for the award of any degree, diploma, associateship, fellowship or other similar titles.

Mangalore
January 2014

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Dedicated

to

My Parents and my grandmother…

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ABBREVIATIONS

A_{532}	:	Absorbance at 532nm
ANOVA	:	Analysis of variance
AR	:	Analytical reagents grades
Cfu	:	Colony forming unit
Dia	:	diameter
FC	:	Fibrous casing
FFA	:	Free Fatty Acid
G	:	gram
GR	:	Guaranteed reagents grades
GRAS	:	Generally recommended as safe
Hrs	:	hours
Mg	:	milligram
Min	:	minutes
Mm	:	millimeter
Mmt	:	Million metric ton
N	:	Normality
PV	:	Peroxide Value
S	:	second
SC	:	Synthetic casing
TBARS	:	Thiobarbituric acid reactive substances
TMA-N	:	Trimethylamine-nitrogen
TPC	:	Total Plate Count
TVB-N	:	Total volatile base nitrogen

Introduction

I. INTRODUCTION

The last six decades, the independent India has witnessed the evolution of fisheries from primarily being a capture based sector to one having almost equal shares in aquaculture and capture production. With an annual growth rate of 4.7 %, the fisheries sector has been contributing 1.1 % of the total GDP and 5.3 % of the agricultural GDP of the nation (Ayyappan and Jena, 2013). The sector has also been taking pride of providing nutritional security to about 700 million people of the country and livelihood to 14.5 million people, besides earnings of foreign exchange to the tune of US$ 3.41 billion. India is the third largest fish production nation and second largest aquaculture producer in the world (Ayyappan and Jena, 2013).

Fish and sea foods can play a great role in the nutritional picture. As a rich source of nutrients, fish provide a good balance of protein, vitamins and minerals, and relatively low calorie content. In addition fish are excellent sources of Omega-3 poly unsaturated fatty acids (PUFA) which appear to have beneficial effects in the reducing the risk of cardio-vascular diseases and are liked with positive benefits in many other pathological conditions particularly, certain types of cancer and arthritis. A high intake of fish is compatible with a reduction of both caloric and saturated fatty acids intakes, coronary heart diseases, hyper tension, cancer, obesity, iron deficiency, protein deficiency, osteoporosis and arthritis are contemporary health problems for which fish provide a number of nutritional advantages and therapeutic benefits (Rathod *et al.*, 2012).

Spoilage starts as soon as a fish dies as a result of a complex series of chemical, physical, bacteriological and histological changes that occur in the muscle tissue. These interrelated processes are usually accompanied by the gradual loss or development of different compound that affect fish quality. These quality changes are influenced by many factors, the most important of which is temperature. Exposure of fresh fish to temperature abuse can cause serious deterioration in fish quality. Commercially, icing or chilling continues to play a major role in slowing down bacterial and enzymatic activities in fish. However, this process is not designed to totally eliminate changes in quality, since it only offers protection for few weeks, depending on the species (Santos and Yap, 1985). Freezing is considered an excellent process for preserving the quality of fish for longer periods of time. The fishing industry is faced with problems, which arise from consumer demands, fluctuations in supply and seasonal factors. Freezing seems to be one method that can provide a solution to this problem by maintaining a balance between demand and supply. In addition, finds its application in a number of different products from different fish species (Lakshmisha, 2005).

Value addition is the most talked about word in food processing industry, particularly in export oriented fish processing industry because of the increased realization of valuable foreign exchange. Value can be added to fish and fishery products according to the requirements of different markets. These products range from live fish and shell fish to ready-to-serve convenience products. In general value addition means "any additional activity that in one way or another changes the nature of a product thus adding to its value at the time of sale". As for as fish processing industry is concerned value addition is one of the possible approaches to raise profitability since this industry is becoming highly competitive and increasingly expensive. (Joseph, 2003).

The Indian sea food industry is mainly export oriented and accordingly the fishing operations are carried out to catch the species of export value. Shrimps and squids are highly sought after by processors because of its economic value. While catching shrimps and squids, nearly more than 30 % of the catch is constituted by low value fishes (Joseph, 2003). This group of fishes includes perches, soles, ribbon fish, jaw fish, pink perch, bull's eyes, horse mackerels, lesser sardines and silver bellies. These fishes are normally derived to either salt curing or for reduction purposes. The nutritive values of these fishes are as good as any other quality fishes like seer, pomfret. There is an urgent need to utilize the shrimp-by-catch effectively for human consumption. Different group of fishes available in shrimp-by-catch can be effectively utilized for human consumption by preparing ready-to-serve, heat processed products.

It augur well for fish processing industry in India to introduce new heat processed ready-to-serve fish mince based products, which are stable, acceptable and nutritious The potential for acceptance especially from urban population is high. Fish ham is one such product where the technology is already available and needs to be adopted to Indian condition. Fish ham is a composite produce, where in, the separated fish flesh is mixed with different additives and animal meat and stuffed into suitable casings and heat processed. This boneless products having good flavour is very popular in Japan, U.S.A, Canada, Europe and some of Far East countries. The development of technology of fish ham has mainly come from Japanese workers (Amano, 1965).

Until recently the seafood industry looked at minced fish primarily as a source of low cost protein and as a potential material for fish by products. But with the extensive studies carried out in the recent past, the mince processing technology has come out as one of the most versatile techniques for fish utilization and marketing (Muraleedharan, 2003). An important use of fish mince is in the preparation of surimi, which is an intermediate product which, because of its characteristic ability to form gels, can be used to develop of variety of products conforming to consumer fancies, surimi is the myofibrillar protein concentrate produced by repeated washing of fish mince in order to remove water- soluble nitrogenous matter and flavour compounds. Washing enhances the gel forming capacity of the structural proteins. Surimi is used as a raw material for the preparation of sea food analogues, but in Japan, where the technology originated, surimi is mainly used to prepare the traditional kamaboko products like fish ham and fish sausage etc. (Joseph *et al.*, 2003).

In Japan fish ham is prepared by mixing cured cubes of red meat of tuna and whale meat with pork fat and adhesive binding meat. The adhesive binding meat is prepared by grinding fish meat having high elasticity and good binding capacity with the cured red meat, preservatives, seasoning materials and smoke flavour in a grinder for 15-20 min to get a fine paste of high viscosity. Normally for the preparation of fish ham is composite of 60 % fish paste and 40 % of cured red meat of tuna or land animals red meat. After heat treatment, when cut into slices the product exhibited mosaic pattern (Siddappaji and Prabhu, 2002).

Normally ham fish paste is stuffed into vinylidine chloride casing. It is in red colour synthetic casings, are used it is due to prevent oxidation of added fat and also cured red meat. One of the drawback uses of synthetic casing is due to its non biodegradable, it creates environmental pollution problems. Cured red meat normally treated by using potassium or sodium nitrite ($NaNo_3$). Consumption of cured meat along with fish ham it leads to toxicological health problems in long run periods. Hence there is a great need to use off alternative to synthetic casing for fish ham.

Once of the promising packaging material is fibrous casings. Fibrous casings are cellulose casing reinforced with strong cellulose fibres. These fibrous casings are resistant enough for large sausage calibres and used for a wide range of application including boneless hams and other pieces of meat as well as cooked or dried sausages. They are especially well suited to manufacture of products which are intended for slicing. (Vikase.com.2013). Hence in the present investigation an attempt has been made to use in fibrous casing and shrimps as a substitute to cured red meat of tuna or land animals with the following objectives:

1. To prepare fish ham by using low cost marine fish.
2. To evaluate the shelf life of fish ham in fibrous casings as a packaging material in comparison with synthetic casing under ambient and refrigerated conditions.
3. To study the frozen storage stability of sliced fish ham packed in polyethylene bags.

Review of literature

II. REVIEW OF LITERATURE

Fish and fishery products form a substantial part of human diet, both of poor and wealthy. Fishery products have to possess organoleptic (palatability) and visual characteristics (attractiveness) which will have a possible "upmarket" appeal.

2.1 Fish paste products

According to Suzuki (1981) any species of fish may serve as raw material for paste products, but elasticity forming ability called "ashi" varies with the species of fish. Those differences in elasticity forming ability are due to the quantitative and qualitative properties of salt soluble proteins in the fish meat (Miyake and Hayashi, 1957). In the past, only locally available fish having delicious taste were employed for the preparation of traditional paste products. More recently, since the taste of the final product could be improved by the addition of sodium glutamate, many species which were not used because of consumer unfamiliarity, boniness, and unpleasing look could make a major contribution to the increased utilisation of underutilized fishes, many of them contained excellent white flesh (Regenstein, 1980). As the catch grew with the development of trawl fishing, the first way to use the underutilized and underexploited fish species is to make minced meat. Minced meat is the flesh separated in a comminute form from the skin, bones, scales and fins of the washed whole fish. A common mechanical machine employed is drum or reciprocator type (Venugopal, 2006). The meat separated using reciprocatory machine require further reduction of meat particle size by mincing operation (Shamasundar, 2006). Nishiya *et al.* (1961) have developed the technology for processing and preservation of surimi from *Alaska pollack* by water washing to remove fat and water soluble components and blended with cryoprotectant, which possesses enhanced gel forming, water holding, fat binding and other functional properties. According to Suzuki (1981) surimi is semi-processed intermediate material used in the preparation of a wide range of finished paste products which are of higher economic value. There are two distin-etly coloured forms of muscle in fin fish, white and dark muscle (Love, 1988). White muscle, which usually forms the greater part of the muscle present in a fish. According to Tanikawa (1971), fishes like Sharks, Rays, Cod, Flatfish, Croaker, Pink perch, Lizard fish and *Alaska pollack* have white coloured meat with low fat content and the meat of such varieties containing high amount of salt soluble proteins, shows good elasticity formation could be used as a binding meat for fish ham and in the other type of paste products. Various workers have investigated the suitability of fatty fishes like sardines (Ishikawa, 1979; Satish, 1984), medium fatty fishes like lesser sardine (Prabhu *et al.*, 1988), mackerel (Fukada *et al.*, 1974; Kakehata, 1977; Shimizu, 1978) and Horse mackerel (Suzuki, *et al.*, 1969) and various deep sea fishes on the gel forming ability of the paste products. Iwata *et al.*(1977) have described kamaboko preparation using two different fishes like Mackerel and Red seabream. Borderias and Tejada (1981) have discussed the utilisation of small pelagic fishes in the preparation of paste products like kamaboko and fish finger. A product known as active fish protein powder (AFPP), that is easy to handle, transport and preserve, it can be used for preparing kamaboko and other food products has been developed by Niki *et al.* (1982). Burges (1975) and Bailey (1976) have reported about the preparation of different types of fish products using minced meat of grandier (*Coryphenoides*), smooth head (*Alepocephalus*) and Rabit fish (*Chimaera spp*). Other important fishes used to prepare minced products are Cat fish (*Aurics sp.*), Lizard fish (*Saurida tumbil*), thread fin bream (Bremner and Snell, 1978), Ribbon fish (Badonia and Devadasan,1980), Lactarius (Agarwal, *et al.,* 1986), *Priacanthus sp.* is a deep water fish, available in considerable quantities in Indian water (Joseph, 1984), is popularly known as 'Bull's eye' or 'big eye.' It is a red coloured fish of size range 13 - 21cm consisting of four main species namely *Priacanthus hamrur, Priacanthus tayemus. Priacanthus cruentaus Priacanthus arentus.* The Bull's eye were located on the south - west coast from Goa to Mandapam and from point Calimore to Visakhapatnam on the east coast at 50 - 400 m depth, with peak concentration at 100 - 200 m. The average catch per hour and maximum catch per hour for the bull's eye in different areas of availability as reported by Joseph (1984). For utilising this low cost fish and presenting acceptable products to the consumer, trials were carried out in canning, freezing and drying the fish in different styles as reported by (Samuel, *et al.,* 1987). Tilapia, Mullet (*Mugil*), Croaker (*Micropogan)* and Sand trout *Cynoscian* (Finne and Nickerson 1980). Carver and King (1971), have reported that by means of meat separating machines, significant quantities of edible flesh can be recovered from fish filleting waste. The yield of meat obtained from diverse species of fish processed under various conditions has been reported by Sen (2005).

2.2 Sub materials for paste products

2.2.1 Salt

Salt is a food additive with a ritual character. Salt is a GRAS substance, saltiness is readily detected on the sides and tip / edges in the front part of tongue. Salt has a tendency to stimulate meaty flavour (Luck, 1980). According to Tanikawa (1965) and Suzuki (1981), salt is one of the important ingredients used for the preparation of paste products, particularly for the extraction of muscle proteins from the fish meat. Tanikawa (1965) reported that the amount of added salt should generally be about 3 %. According to Suzuki (1981) fish paste product like kamaboko cannot be prepared without mixing sodium chloride to the fish meat. When salt is added to the minced meat and ground, the ground meat mix becomes a viscous paste called sol form (Suzuki, 1981). If added salt is at 3 % on the basis of the weight of ground minced fish meat, the elasticity is the strongest.

According to Suzuki (1981) a concentration between 1.2 M and 1.5 M of sodium chloride gives maximum extraction of myosin from fish muscle as well as the best resiliency in the finished products. Venugopal (2006) studied that myosin and actomyosin majorly influence the structural and functional properties of muscle based food. The function of salt is to solubilise these salt extractible proteins, facilitating the formation of a three dimensional structure during heating process of mince. Salt solubilisation allows the binding of water with salt ions that in turn increase the mobility and release of flavour compounds by decreasing the water availability for interaction and solubilisation these flavour compounds (Rabe *et al.*, 2003). Salt addition also influences the textural properties of foods (Desmond, 2006). The use of the salt at these concentrations however, would make the product too salty and 0.4 M to 0.6 M sodium chloride is used as optimum levels in the commercial products. However, Tanikawa (1971) has suggested 2.5 % salt as an optimum level for the preparation of paste products of good quality. If the salt concentration is too high, the protein will not dissolve in salt solution. Attempts to reduce salt level less than 2 % in minced meat products have resulted in reduced stability and textural firmness (Whiting and Jenkins, 1981). In India, Krishnaswamy *et al.* (1968), Cross (1984), Raju (1999), have prepared paste products using different concentration of salt between 2 to 2.5 % based on weight of minced meat. Common salt concentrations above 2 % show preservative action (Luck, 1980).

2.2.1.1 Sugar

Sugar enhances taste and flavour of paste products (Tanikawa, 1965). Sugar imparts sheen to the surface of high jelly products. Substances which give sweet taste such as sugar alcohols (sorbitol, malitol, etc.,) are also used in Japanese style fish paste products and to decrease water activity. Many authors like Tanikawa (1965); Krishnaswamy *et al.* (1968); Iwata *et al.* (1970); Gerrard (1969); Chandrasekhar *et al.*(1977); Raju (1999), have suggested addition of sugar in the range of 1-2 % in order to enhance the taste and flavour of the product. Addition of sugar to fish mince is to stabilise the proteins thermodynamically through their interaction with surrounding water (Venugopal, 2006). Addition of sugar increases sweetness as desired and decreases water activity, thus enhancing the shelf life of the product (Lee *et al.*, 1992). Sugars are also used to counter the unpleasant after taste of salt and spice powders. Dextrose have reducing groups, they have the ability to inhibit undesirable oxidative reactions in foods thus extend the shelf life of the product (Wurzburg, 1968).

2.2.1.2 Sodium tripolyphosphate

According to Amano (1965), polyphosphates are added to raw fish gel in order to get high resiliency in the Japanese style fish paste products. Okada and Yamazaki (1958) have shown that the concentrations from 0.2 to 0.5 % of the products were found to be very effective in improving the texture of fish sausage, with the higher concentration impairing taste and affect both palatability and meat binding. The use of polyphosphate in surimi reduces the viscosity of paste, providing better machinability (Park and Lin, 2005). Polyphosphates are used to enhance adhesive effect of the gel and elasticity of the paste products like kamaboko and sausage formed from such gels

(Tanikawa, 1971). As polyphosphates increase the pH of fish meat, which increase the solubility and water holding capacity of myosin group of protein sand thus help in swelling of the meat proteins. Okamura *et al.* (1958) have extensively studied the effect of various phosphates and combination of starch as well as other additives in improving the binding properties of kamaboko. Miyake *et al.*(1963) have found that the combination of polyphosphate and mg^{++} ‘ions" increase the gel strength and Ca^{++} ions found to decrease the gel strength. Polyphosphate are also known to act as antioxidant, antimicrobial and buffering agent (Ellinger, 1977). Polyphosphates or tripolyphosphates exert a weak bacteriostatic action, possibly due to its capability of chelating minerals (Vishniac, 1950; Dirheimer and Jean - Pierre, 1956). Uchiyama and Amano (1959) have shown that the addition of polyphosphates can partially arrest the growth of *Bacillus pantothenicus* which cause softening type of spoilage in fish sausage. Maragal (1979), Prabhu *et al.* (1988), Joshi (1990), Raju (1999) have tried sodium tripolyphosphate at 0.2 to 0.3 % to prepare good quality fish sausage. Proper addition of polyphosphate increases texture and water retention of the surimi reported by Park and Lin (2005).

2.2.1.3 Starch

Starch is one of the most abundant natural polysaccharide (Narayan, 1994). The world starch production in 2012 was 75 millions tons. Main agricultural sources for starch production include wheat, potato, and cassava Wurzburg (1968). As major ingredients or additives, native or modified starches have been playing important roles in the food industry. Nutritionally, starches supply more than 50 % of the calories consumed by the human population (Whistler, 1984). In addition, starches and their derivatives have been used to modify physical properties of food products such as soups, snacks, batters and meat products (Thomas and Atwell,1997), contributing mainly to texture, viscosity, gel formation, adhesion binding, film formation and product homogeneity (Thomas and Atwell, 1997).

Starch is deposited in the plant in the form of minute, cold water insoluble granules. Depending upon the plant source, the size of the granules range from about 3-100 microns in diameter. The shape of the granules is characteristics of a particular plant species. Starch is produced commercially by extraction, from the seeds of plants such as corn, wheat, sorghum or rice, the tubers or roots of plants like cassava, potato or arrow root and the pith of the sago palm (Chopra and Panesar, 2010). The characteristic of the starch varies with the plant source from which it is derived. Starch, in its native or modified form, is used throughout the food industry as a carbohydrate source, extender, texture modifier, stabilizer processing aid, and thickener. According to Wurzburg (1968), the major source for commercial starch is corn, due to the availability and relatively low cost. Corn starch granules are spherical to polygonal in shape with a diameter ranging from 2 to 30 microns. The smaller size starch granules have 2 - 10 microns and larger granules are 20 - 35 microns which may be flat, round or Elliptical granules in shape. Tapioca starch granules are round and truncated at one end to form kettle drum shapes where as rice starch are polygonal in shape. Most starch contains amylose and amylopectin polymers. According to Okada and Yamazaki (1957), amylose and amylopectin on gelatinisation enhance the jelly strength of paste products. According to Tanikawa (1965), Binder is an essential part of paste products. Starch is the most widely used filler ingredient in surimi based product (Lee, *et al*., 1992). Starch act as asimple filler of the myofibrillar protein gel (Lee, *et al.,*1992).Water soluble polysaccharides are long chain polymers that dissolve or disperse in water to give a thickening or viscosity binding effect. These polysaccharides play an important role in the stabilisation of food emulsions.

According to Tanikawa (1971), starch is added at the end of the grinding operation. The purpose of this is to adjust the final elasticity and bulkiness of the product. Starch is added as powder or aqueous suspension in the amount of 10 % of bulk weight. According to the author, Tanikawa (1971), addition of more starch (up to 20 % with added water at 5 % level) is necessary to improve the elasticity formation. Further, it has been found that potato starch as solution (because of its greater hydration activity) showed better effect than wheat starch. In our country, Chandrasekhar *et al.* (1977) have tried 16% starch where as Cross (1984), Prabhu *et al.* (1988), Joshi (1990) and Raju (1999), have tried 9 % starch for the preparation of fish sausage. Hegde (1989), reported that cooked potato is not suitable for sausage preparation but potato flour at 7 % level can be successfully used.

2.2.1.4 Oil

Oils are smooth, greasy substances that form emulsion with water. Oils and fats provide more than twice as much energy as do carbohydrates and proteins. Oils contribute to characteristic flavour to the paste product (Raju, 1999). Chandrasekhar (1970).Vegetable oil can be used to modify texture that is a product with a smoother texture. Addition of oil tends to weakness the gel texture, although it reduces rubbery and chewy texture. (Westerly, *et al.*, 1980). The addition of oil may improve freeze thaw stability by preventing the development of a sponge like texture resulting from freeze suppression (Lee and Toledo, 1979).

(Dallyn and Shorten, 1998). An equally important function of packaging is to protect the prepared product from physical, chemical or biological damage (Dallyn and Shorten, 1998). The most well-known packaging materials that meet these criteria are polyethylene or co - polymer based materials which have been in use by the food industry for over 50 years. These materials are not only safe, inexpensive, versatile, but also flexible (Tice, 2003). Global productions of packaging materials are estimated at more than 180 million tons per year, with growth and demand increasing annually (Tice, 2003). Within the plastic packaging market, food packaging is the largest growing sector (Comstock *et al.,* 2004). However, one of the limitations with plastic food packaging materials is that it is meant to be discarded, with very little being recycled (Comstock *et al.,* 2004). The presence of these types of packaging materials in landfills can be problematic on many fronts. First, if plastic is not recycled, these items end up in landfills, where they can last forever and never degrade. Secondly, many countries are faced with a decrease in landfill space, especially in densely populated areas (Comstock *et al.,* 2004). Thirdly, existing landfills may be unable to meet new regulatory guidelines set forth by the US environmental protection agency, and may end up in closing of the unit. (Environmental protection Agency, 2006). So, finding landfills for consumer and industrial waste may become more difficult in the future (Comstock *et al.,* 2004).

In addition to the above environmental issues, food packaging has been impacted by notable changes in food distribution, including globalization of the food supply, consumer trends for more fresh and convenient foods, as well as desire for safer and better quality foods. Consumers are demanding that food packaging materials be more natural, disposable, potentially biodegradable, as well as recyclable (Lopez - Rubio *et al.,* 2004).

To meet the growing demand of recyclable or natural packaging materials and consumer demands for safer and better quality foods, new and novel food grade packaging material or technologies have been and continue to be developed. Examples of these packaging materials include bio-based polymers, bioplastic or biopolymer packaging products made from raw materials originating from agricultural or marine sources (Cha and Chinnan, 2004). These types of packaging materials include, but are not limited to starch, cellulose, chitosan / chitin, protein (animal, plant based), or lipids (animal, plant derived, etc). Within this context of packaging, edible films, gels or coating many be considered biopolymers with some biodegradable properties. (Cha and Chinnan, 2004). Naturally derived resources are becoming more essential in the production of industrial products, and that bio - based packaging materials are beginning to replace petroleum based packaging materials in the food industry (Weber *et al.,* 2002). Researchers also have further categorized bio polymers based on the ability to be compostable or biodegradable (Comstock *et al.,* 2004). It is important to note that while some bio-based packaging materials may be biodegradable, not all biodegradable material are bio- based (Weber *et al.,* 2002).

Recent technological advances also have allowed bio polymers to be processed similarly to petroleum - based plastics, whether in sheets by extrusion, spinning, injection molding, or thermo foaming (Comstock *et al.,* 2004). Notable advances in biopolymer production, consumer demand for more environmentally - friendly packaging, and technologies that allow packaging to be more than just encompass the food are driving new and novel research and developments in the area of packaging for muscle foods. Bio-based materials have been shown to prevent moisture loss, drip, reduce lipid oxidation and improve attributes, as well as enhancing the handling properties, color retention, and microbial stability of foods

(Cutter, 2006). The author also has studied on opportunities for bio-based packaging technologies to improve the quality and safety of fresh and further processed muscle foods.

Research studies have demonstrated that cellulose based films applied to muscle foods can reduce oil up take during frying, minimize run-off during cooking, and reduce moisture loss when applied as glazes for poultry and sea food. (Baker *et al.,*1994, Cutter and Sumner, 2002, Meyers, 1990). Coatings made with ethyl cellulose and lipids were transparent and readily peelable, prevented desiccation, and extended the shelf life of prepared products (Ayers, 1959). While cellulose based films have demonstrated mechanical, oxygen barrier and oil barrier properties for foods such as pizza and ice cream cones. Very little information exists for application to fresh or further processed muscle foods (Gennadios *et al.,* 1997).

2.2.2.1 Natural casings

Natural casings are mainly derived from small and large intestines from sheep, goats and pigs, but also from cattle and horses. Chawla *et al.* (2006). They are strong enough to resist the pressure produced by filling them with sausage mix. Natural casings are permeable to water vapour and gases, thus allowing fillings to dry and absorb smoke for additional flavour and preservation / expand or shrink firmly attached to the sausage mix and can be closed at the ends by tying or clipping. Small intestines of sheep, goats and pigs are popular for small calibre natural casings. They are processed in a way that makes them tender (edible) and are mostly eaten with the sausage casing. Many other parts of the intestinal tract of slaughter animals can also be used for natural casings. Those casings are processed differently and have stronger and tougher casing walls. Due to their toughness they are generally not considered "edible" (although not unfit for human consumption) and are usually peeled off before consuming the sausages (Maragal, 1979). The development of high value added products with quality requisites is a challenge for the meat industry. The sausage packaging process and the technological characteristics of hog casings are important to maintain the functional properties, the quality attributes and the safety of sausage (Elisa *et al.,* 2008). Natural casings should present good barrier to water vapour, low cost, easy manufacturing and calibre regularity, producing uniform size and shapes. Natural casing present variability in calibre and elongation capacity. Studies to improve casings properties are essential to enlarge the stuffing efficiency, raising the sausage production has been reported by Bakker *et al.* (1999) and Benli *et al.* (2008).

Natural casings in sausage stuffing are frequently salted to reduce microbial activity, with no changes in their technological properties. Chemical preservatives such as lactic, tartaric and citric acid, hydrogen peroxide or ethanol are also applied (Chawla *et al.,* 2006; Byun *et al.,* 2001). These chemicals can generate adverse effects on casings technological characteristics, including an increase in water vapour permeability due to the pH reduction. Another compound extensively used in starch or protein based films is the plasticizer sorbitol (glucitol). This plasticizer can decrease inter and intra - molecular attraction forces and increase chain mobility, improving the film flexibility. Sorbitol also increases the film water vapour permeability (Byun *et al.,* 2001; Rodriguez *et al.,* 2006). From the point of economies of natural casings in sausage production, there is lack of infermation on wastage of casing during the stuffing of sausage batters. The exceptions regarding the use of chemicals on natural casings are the works presented by Houben *et al.* (2005). The application of trisodiumphosphate to improve the stuffing efficiency and Bakker *et al.*

(1999) have showed that food grade additives to develop the microbiological and mechanical properties of casings. The use of ozone to advance casing characteristics was also considered by Benli *et al.* (2008). Natural casing made from intestine of beef, pork, or lambare used to stuff meat products such as sausages, salami, and frankfurters. There is demand for natural casing due to consumer liking. Contamination of natural casing by enteric and exogenous microorganisms is inevitable. Microbial quality of natural casing depends on the hygiene of manufacturing produce, post processing handling and storage temperature (Trigo and Fraqueza, 1998). Natural casing are contaminated with bacteria of public health significance such as *Fecal streptococci*, Enterobacteriaceae, *Coliforms*, sulphite reducing *Clostridia* (Byun, *et al.,* 2001, Trigo and Fraqueza, 1998).

2.2.2.2 Artificial casings

(Gunter -Heinz and Peter Hautzinger, 2007). Artificial casings were developed at the beginning of 20^{th} century when, in some countries, the supply of natural casings could no longer cope with the demand for such natural casings from the growing meat industries. Following the development of highly automated sausage filling equipment, artificial casings proved to be better suited to those systems, mainly due to their uniformity. Also from the hygienic point of view, there are certain advantages to artificial casings as the microbial contamination is negligible, refrigeration is not needed and there are no spoilage problems during transport and storage. Nowadays, for wide sausage calibres, artificial casings are the material of choice, while for smaller calibre products, artificial and natural casings remain equally important.

The concept of biodegradability enjoys both user - friendly and eco-friendly attitudes, and the raw materials are essentially derived from either replenishable agricultural feed stocks or marine food processing industry wastes. An additional advantage of biodegradable packaging materials is that on biodegradation or disintegration and composting they may act as fertilizer and soil conditioner, facilitating better yield of the crops (Cutter, 2006).

In the mid 1930's fibrous casing were introduced as another casing alternative for the meat industry stuffing needs as consumer demand increased. Fibrous casings are manufactured using the viscose process, in this process Wood cellulose is solubilised, treated and viscose is formed. The viscose is then impregnated into a special filament paper as it is being formed into a tube. This coated tube moves into a bath which regenerated the original cellulose. Fibrous casings are manufactured on the base of long stapled square paper and reclaimed viscose. At the present day a wide range of fibrous casing with different diameters and of various colours are available. It is notable for its mechanical strength, moisture and smoke proofness, simple clip ability and resistance to high temperatures. The light casing is manufactured as in the standard regular type, but found an increased adhesion to the stuffing. The adhesion level can be different for uncooked smoked sausages manufactured in the traditional way with an aging time of 30 days and more. It is recommended for uncooked, smoked sausages manufactured under the utilization of various ageing casing type. One of the nicest looking casings is the fibrous casing with Visko - net. The net used on the casing may be shaped as diamonds or like hexagonal combs. The fibrous casing can be easily colour printed. Fibrous casing are made from with paper a protein coating on the inside. All synthetic fibrous casing are biodegradable. Fibrous casing are manufactured in many different sizes and colours, because they are so tough fibrous casing are very easy to stuff. The most common sausages made with fibrous casing are summer sausage and salami (Viskase.2013).

According to their structure and composition of material, artificial casings belong to either natural casing or synthetic casing. Natural casings are made of either organic plant material (cellulose) or animal by products (collagen). Synthetic casings are made using thermoplastic materials. Suitable materials are polyamide (PA), polyethylene (PE), polypropylene (PP), polyvinylidenchloride (PVDC) and polyester (PET). These synthetic casing belongs either to polymer casing or plastic casing. Simple thin cellulose casings are used as peeling casings for frankfurter type sausages. The batter is filled into such casings (calibre range 12 to 42 mm) and portioned, thereafter the products undergo smoking and cooking (at 74 ^{0}C), heat processing causes a firm layer of coagulated protein under the casing. After this heat treatment, the cellulose casings are removed and the sausages maintain their shape due to the firm external layer of coagulated protein. As ready-to-eat sausages do not have a casing, they are also known as “skinless sausages” (Gunter - Heinz and Peter Hautzinger, 2007).

2.2.2.3 Collagen casings

Collagen films are used for sausage casings (Krochta and De mulder - Johnston 1997). Cellulose esters such as carboxymethyl cellulose, hydroxypropyl cellulose and methylcellulose are used as ingredients in coatings for fruits, meat, fish and other agricultural products.

This type of casings is fabricated from collagen, which is obtained from the corium layer of selected split cattle hide (Hood, 1987). The collagen - rich tissue is homogenized under high pressure, ring-extruded (hose-shaped) and hardened. This process produces mechanically strong casing, which are permeable for smoke and water vapour. Wide calibres have a relatively thick casing wall due to increased stability. Small calibres can be made with relatively thin casing walls. As collagen is an animal tissue fit for human consumption, the thin collagen casings are easy to chew and "edible". They are an alternative to replace natural sheep, goat or thin pig casings. The advantages of collagen casings are their standard diameter and strength and that they can be "shirred" i.e. folded together, in long lengths and used for manual or automatic filling without pre-soaking in water.

The edible collagen casings are also used for fried sausages (including the typical breakfast sausages) and small calibre dry sausages such as beef sticks, etc. Collagen casings of 32 mm and above are not intended to be eaten as part of the sausage, they have to be peeled off. They can be used for most fresh sausages, raw - cooked and smoked sausages or raw - fermented sausages. Production of collagen sausage casings from the regenerated corium layer of food grade beef hides is a well established technology and has been discussed (Hood, 1987., Rust, 1987; Gennadios *et al.,* 1994). As an alternative to performed collagen casings, unilever has developed a technology where a collagen casing is co-extruded around the sausage meat batter (Smits, 1985). The author also point out that large processing plants with high volume lines use the co-extrusion technology for collagen casing production. Further more, use of proteins other than collagen, such as wheat gluten, corn zein, soy protein, peanut protein, and feather keratin, in manufacturing of sausage casings has been suggested (Mullen, 1971).

Collagen based edible coatings also have been proposed for use on meat products other than sausages (Jones and Whitmore, 1972), described a method where ground collagen was mixed with an aqueous mixture of lactic acid and glyceraldehydes, heated to about 75 ^{0}C, and neutralized to pH 7 to make a coating for hamburgers capable of withstanding cooking temperatures without melting. An edible, collagen film, intended for use on boneless hams, fish fillets, roast beef, and meat pastes, was commercialised in the US in the late 1980s. Farouk *et al.* (1990) reported that both refrigerated and frozen / thawed round beef steaks wrapped in coffi collagen film prior to standard retail packaging exhibited significantly less fluid exudates than unwrapped controls. The author also point out that based on thiobarbituric acid analysis and on instrumental and sensory color evaluation, the collagen film had no significant effect on meat oxidation and color. Researchers at the U.S. army Natick research, development and engineering centre have been investigating the potential of replacing plastic meat wrappings with collagen based edible films (Rice, 1994). Beef cubes wrapped in collagen- based films and stored at -18 ^{0}C for 20 weeks were not significantly different than plastic- wrapped controls in terms of oxidation, color, microbial growth and sensory attributes (Rice, 1994).

2.2.2.4 Fibrous casings

Fibrous casings are cellulose casings reinforced with strong cellulose fibers. These fibrous casings are resistant enough for large sausage calibers. These fibrous casings also suitable for large calibers and still suitable for smoking and are especially well suited to the manufacture of products which are intended for slicing (Viskase.com, 2007). In the manufacture of edible films, cellulose based films tends to be water soluble, resistant to fats and oils, tough, and flexible (Baldwin, *et al.,* 1997, Cutter and Sumner, 2002, Krumel and Lindsay, 1976). While not considered edible, cellulose casings used in this manner are removed from the meat product and the casings disposal in landfills or enzymatically treated to assist in disposal (Cumba and Bellmer, 2005). These casings can be used for a wide range of applications including pepperoni, salami, luncheon meats, boneless hams and other delicious-style processed meats. They are ideal for slicing products requiring precise diameter control. Fibrous Casings can also be used for processed and smoked cheeses (Viskase.com, 2013).

2.2.2.5 Types of Fibrous Casing

Most of the manufactured fibrous casings are permeable to smoke and for some extent to moisture. Pre-stuck or pre-drilled types of fibrous casing are small holes that allow air evacuation during stuffing and excess surface water / fat to escape during cooking. For specific applications requiring moisture and oxygen barrier properties, the casings are coated with thin plastic layer either inside or outside of the casing. To another type of fibrous casing internal treatment is given to promote easy release of the casing from the final product. These casing lifts away from the product quickly and cleanly, leaving no scarring of the product surface. To get smoked and caramel odour, fibrous casings are pre-moisturized with natural liquid smoke that will transfer smoke flavour and colour to the surface of the product. The fibrous casings are available in the form of shirred, cut / clipped or Rolls of desired lengths requiring soaking prior to use. Fibrous calibres size range from 32 to 245 mm. Fibrous casings which are not pre–moisturized must be soaked in water prior to use. The minimum recommended soaking time is 20 minutes in warm water at 80 to 100 °F (27 to 38 °C). Where as pre-moisturized casings do not require soaking prior to use. It is important to select the proper clip size and clipping pressure to assure a tight fit without causing damage to the casing. These fibrous casing are stored in a cool dry place away from steam pipes or hot storage areas. Keep the casing in original containers until it is ready for use. If casing are exposed to freezing temperatures during transport or storage they should be held at temperatures between (12 to 24 ºC) for at least two days before being used, in order to prevent break during stuffing. Wrinkle formation, splitting, meat separation from the casing and muddy odour were some of the problem reported with fibrous casing (Viskase.com, 2013).

2.2.3 Processing of Fish ham

Tanikawa (1971) described the methods of processing of fish ham. According to him, the raw material for fish ham is prepared by mixing cubes of red meat of tuna with the paste and binding meat thoroughly in a mixer, and stuffed into casings by means of a stuffer. To maintain the shape, the casings stuffed with mixed meat are put into a retainer made from stainless steel before processing. Fish hams are processed by immersing in water at 90 °C for 30 min and then at 85 °C for 60 min. Then fish ham is dried by passing through a drier and packed with red cellophane paper and the finished products are packed in cartons. The chemical composition of the product prepared in Japan generally contains moisture (61.5 %), fat (11.3 %), protein (17.5 %), ash (3.0 %) and sugar (1.7 %). Chandrasekhar and Desai (1972) have also reported about processing of fish ham. Siddappaji (1986) and Siddappaji and Prabhu, (2002) have reported a processing time of 88 to 90 °C for 105 min for fish ham.

2.3 Quality Characteristics and Standards for Japanese fish Paste Products

In 1962, the Japanese agricultural and forestry ministry had adopted quality standard for sausage and ham in the agriculture and forestry products standards law. Tanikawa (1971), has described the methods for total bacterial counts and chemical parameters like estimation of VBN, TMA, pH and PV for the estimation of quality of Japanese style fish paste products. Various grades and scores have been assigned to are directly related to shelf life extension and safety improvement. Ueno (1968) and Chandrasekhar and Desai (1972) have reported that the commercial fish ham and sausages produced in Japan, may not be spoiled within two or three months in summer seasons stored at 30 °C to 40 °C. Tanikawa (1971), has reported that the fish ham and sausages containing no preservatives and pH around 5.5, showed no indication of spoilage even after two weeks of storage at 38 °C. On the contrary spoilages by gas formation occurred during five days storage among all the control samples when their pH was 6.0. Tanikawa (1971) reports that by smoking and storing in cooler storage the shelf life can be increased up to 28 days. Cross (1984) has reported, that the fish sausages sterilized at 115 °C for 20 min, could be stored for 9 days at room temperature, for 70 days in cooler and refrigerated temperatures. Various scientists Chandrasekhar *et al.*(1988), Hegde (1989), Ravishankar (1990) Joshi (1990), Raju (1999), Srinivasa (1998) and Siddappa (2002), have reviewed different storage period for prepared different types of Japanese paste products.

2.3.3 pH

pH is one of the important parameter used to assess the quality of fish and fishery products. In the of the present experiments changes in pH during storage at ambient, refrigerated and frozen storage. During storage time, the pH values decrease and then increased gradually (Song *et al.,* 2010). The increase in pH is caused by the enzymatic degradation of fish muscle (Love, 1992 and Vareltzis *et al*., 1997).The decrease in pH of fish product during refrigerated storage is attributed to the breakdown of starch by bacterial action have been observed by Raju *et al.* (2003). Rathod and Pagarkar (2013) showed slightly increased in pH of the cutlet prepared from *Pangasius hypophthalmus*. Pawar (2011) reported the cutlet made from catla fish showed increasing trend of pH from 6.50 to 6.79 when stored at -2 to - 4 °C. Low pH may decrease water holding capacity of myofibrillar proteins and promote oxidation of lipids (Haard, 2002).

2.3.4 Peroxide Value (PV)

The peroxide value provides a measure of the degree of lipid oxidation and indicates the amount of oxidized substances. Hydroperoxides are the primary products of auto-oxidation, which themselves are odourless. However, their decay leads to the formation of a wide range of carbonyl compounds, hydrocarbons, furans and other products that contribute to the rancid taste of decaying food has been studied by Yanishlieva and Marinova (2001) and development of off-flavors. The PV tends to increase during early stages of oxidation, when the rate of formation of hydroperoxides is higher than the rate of decomposition. Suttamit and Ahromrit (2011) have observed that, after the maximum value is reached, the value then decreases as a result of the lower availability of substrate and instability of peroxide molecules, which leads to the formation rate of decomposition.

2.3.5 Thiobarbituric acid value (TBA)

Suttamit and Ahromrit (2011) reported that determination of TBARS is a general method for estimation of secondary oxidation products. Malonaldehyde can interact with other compounds such as amines, nucleosides and nucleic acid, proteins, amino acids and phospholipids, or other aldehydes that are end-products of lipid oxidation, to form polymers that decrease the quality of fish (Nazemroaya, *et al.*, 2009). The increasing of the TBA value during refrigerated storage of rainbow trout fillet, for fish fingers (made from lizard fish, threadfin bream and barracuda), fish balls made from carp, minced pond-bred flesh of silver carp, has been demonstrated by Jasour *et al.* (2011); Tokur *et al.* (2006); Yanar and Fenercioglu (1998); Gelman and Benjamin,(1988). Aubourg *et al.* (2007) reported the concentrations of thio-barbituric acid (TBARS) in the control blue shark samples increased constantly during the storage study. The significant increase in TBARS values indicated oxidative deterioration. During a study on effect of tannic acid and kiam wood extract on lipid oxidation of fish sausage TBARS values showed a sharp increasing trend up to 12 days of refrigerated storage which was followed by slight decrement until the end of storage period (Maqsood *et al.,* 2012). Addition of refined tuna oil at 6 - 10 % to emulsion fish sausage accelerated lipid oxidation (Panpipat and Yongsawatdigul, 2008). High content of unsaturated fatty acid (EPA and DHA) were reported to be the reason for such increment.

2.3.6 Free Fatty Acid (FFA)

The FFA is a result of enzymatic decomposition of lipid during storage (Tokur *et al.,* 2006). The FFA content in the lipid of a fish is an indication of lipid hydrolysis. As the freshness quality of fish gets reduced, the FFA content in the lipids of fish increases due to the action of lipases Reddy *et al.* (2012). Lipids and proteins do not react to form complexes unless the fat or fatty acids are oxidized (Bhattacharya *et al.,* 2008). Aubourg *et al.* (2007) documented the change in quality of lipid oxidation in fish to the action of endogenous enzymes in each species. Endogenous fish enzymes may be active during refrigerated storage, even at -20 °C (Suttamit and Ahromrit, 2011). These enzymes may be influenced by a wide range of internal and external factors, which must be considered in the commercialization and storage of fish products. Free fatty acids (FFA) are triacylglyceride products formed either by chemical or enzyme-mediated hydrolysis (Barthet *et al.,* 2008). Lipid hydrolysis obviously occurred throughout refrigerated storage and generally increased progressively over time. Formation of FFA does not entail loss of nutritional value but its accumulation has been associated with a decrease in the acceptability of food by the consumer (Aubourg *et al.,* 2005). The decrease in consumer acceptance of refrigerated product has been related to accumulation of FFA because these compounds have deleterious effects on ATPase activity, protein solubility and cause disagreeable flavour, discoloration, viscosity - related deterioration of the texture (due to interactions between proteins), and oxidize more rapidly to form higher molecular weight lipids (triglycerides and phospholipids), thus providing greater accessibility (lower steric effect) to oxygen molecules and other pro-oxidant molecules. FFA is sometimes quantified to establish whether or not a food product is organoleptically acceptable (Barthet *et al.,* 2008). Free fatty acid (FFA) of Pangasius fish cutlet stored in refrigerated display unit showed an increasing trend from 1.26 to 4.83 mg/100g (Rathod and Pagarkar, 2013). Joseph *et al.* (1988) reported FFA content in flashed fried and raw cutlet in the range of 0.98 to 1.49 and 2.03 to 2.82 mg/100g respectively at 4°C. Reddy *et al.* (1992) reported increasing FFA in fish finger developed from croaker and pink perch meat up to 6^{th} week and 10^{th} week respectively and then decreased slightly up to 14^{th} week and remained almost stable at -20 °C. Tokur *et al.* (2006) reported increased FFA from the beginning of the storage up to 8^{th} month. The result shows that FFA increased with the duration of storage.

2.3.7 Total Volatile Base Nitrogen (TVB-N)

Volatile base nitrogen (TVB-N) primarily composed of ammonia; trimethylamine (TMA-N) and dimethylamine (DMA) originated from the breakdown of nucleo-tides and as a result of deamination of amino acids by microorganisms have been reviewed by Contreras - Guzman, (2002). Quantity of TVB-N value present in fish and fishery products, widely used as an index of spoilage (Mahmoudzadeh *et al.*, 2010). The TVB-N in freshwater fish and their products comes from ammonia as reported by Rathod and Pagarkar (2013). TVB-N of pangasius fish cutlet stored in refrigerated display unit increased from 2.52 to 22.4 mg / 100 g during the study period observed by Rathod and Pagarkar (2013). Bao *et al.* (2007) reported the increasing trend of TVB-N in fish product at super chilling (-2°C) and chilling (3°C) storage temperature. TVB-N value is not stable during refrigerated storage and could be changed according to species, processing methods, and storage temperature has been well documented by Tokur *et al.* (2006). TVB-N continuously increases in anchovy fish patties during the refrigerated storage (Turhan *et al.*, 2001). The increase of TVB-N content during storage is related to bacterial spoilage and activity of endogenous enzymes (Chomnawang *et al.*, 2007). Irregular changes on TVB-N values in the study of Kilina *et al.* (2003) during the storage period of sardine (*Sardina pilchardus*) were due to the elimination of dissolved volatile constituents through drip. Leaching out phenomenon of volatile bases could be caused decreasing of TVB-N values if samples were stored in loose closed bags (Ozogul and Ozogul, 2000) also decreasing of TVB-N value may be the result of hypothesis that mentioned for reduction of pH value. Enzymatic and microbial activities led to the slight increment in TVB-N values for sausages prepared from rohu both at ambient as well as refrigerated storage has been studied (Sini *et al.*, 2008) whereas some authors have reported no significant variation in TVB-N values for sausages prepared from Nile tilapia during 40 days of storage period (Oliveira *et al.*, 2010).

2.3.8 Sensory Quality

The Sensory evaluation is a scientific discipline that evokes, measures, analyses and interprets reactions to those characteristics of food and materials as they are perceived by the senses of vision, odour, taste, touch and hearing (Sidel and Stone, 2006). It is a quantitative science which collects numerical data and establishes specific relationship between characteristics of product and human perception (Heymann and Lawless, 1998). Sensory characteristics like texture, flavour, aroma, shape and colour (Hogan *et al.*, 2005) are highly related with consumer preference and satisfaction with foods (Tuorila and Monteleone, 2009). Measurement of sensory quality is influenced by many factors including sample under investigation, the assessment method and the judges / panel (Hanna, 1992). Fish product acceptability, the degree of liking and disliking is commonly estimated using a scalar method, the nine point structural hedonic scale (consumer acceptability). Hedonic scale assumes the continuum existence of consumer preferences and these references can be cate-gorised by responses based on liking and disliking (Martinsdottir *et al.*, 2009).

2.4 Microbiological Quality

Gram and Huss (1996) have proposed spoilage of fish and fishery products is due to the production of off odour and off flavour that is generally caused by bacterial metabolites. Chang *et al.* (1998) reported that 6.0 to 7.0 log cfu / g was set as the maxi-mum acceptable load for maintaining a good shelf - life in fresh water fish products. It is possible to predict the shelf life of fish product based on knowledge of microbial load (Ozogul, 2010).

2.4.1 Total Plate Count (TPC)

Serial dilution - agar plating method (APM) is the universally used method for counting living viable cells in different samples has been investigated by Khuntia (2011). Total plate count represents the number of bacteria that are capable enough of forming visible colonies on a culture media at a given temperature. This is a good indicator of the sensory quality or expected shelf life of the product has been suggested by Huss *et al.* (1974). The increasing of TPC in fish products during refrigerated storage has been demonstrated by Leung *et al.* (1992), Lyon and Reddmann (2000).

2.4.2 Aerobic Spore former

Aerobic spore former e.g. from the Bacillus groups develop in fish sausage *via* spices as well as during handling and transportation of raw fish (Amano, 1965). Gradual increase in Aerobic spore formers was reported in the sausages prepared from threadfin bream stored at ambient and refrigerated temperatures has been reviewed thoroughly by Raju *et al.* (2003).

2.5 Spoilages

Simidu and Also (1964), have reviewed the bacteriology of the deteriorations of fish sausages. Yokoseki (1957) has pointed out the respective typical species of bacteria which cause the various types of spoilages. Fish ham is highly susceptible to spoilage when stored at elevated temperatures. In case of properly sealed product, any spoilage which occurs is attributed to microorganisms originally present in the fish meat and other materials. Several types of spoilages in fish ham and sausages have been observed in which there was formation of gas, changes in pH and volatile base nitrogen, bacterial count, colour, flavour and taste. Ogasawara *et al.* (1957) reported a good quality sausage with 2.82 % of total nitrogen, 32.83 mg % of volatile base nitrogen and 19.42 mg % of trimethylamine nitrogen, without the presence of histamine and coliforms. Further, mold and yeast had not grown, bacterial count was 45×10^{7} cfu / g for aerobic bacteria, and 67×10^{3} cfu / g for anaerobic bacteria. Generally the types of spoilages occurring in fish ham and sausages are gas and acid formation, putrefactive odour, Softening in texture, discolouration, liquification and black or brown spots.

2.6 Gas formation

The yellowish white discolouration at the surface of fish ham or sausages is caused by low heat resistant bacteria, where as discolouration developed internally is caused by the heat resisting bacteria. Apart from bacteria, air which gained entrance through the mouth opening also plays important role. Swelling of the casing owing to the generation of gases such as ammonia and carbon dioxide gives unpleasant odours (Tanikawa, 1971). Yokoseki (1957) reported that these types of spoilage are due to the growth of *Lactobacillus Sp* and *Clostridium Sp.*

2.7 Acid formation

According to Yermal *et al.* (1972), acid formation is characterised by change in colour, taste and texture of the meat without the development of swelling which may be caused by non gas forming and thermotolerant bacteria originating from the raw materials example. *Lactobacillius Sp.*

2.8 Putrefactive spoilage

This occurs with or without gas formation caused by bacteria originating from the raw materials.

2.9 Softening spoilage

Softening of the meat to mud - like consistency is due to the action of Proteolytic bacteria on product. The softening spoilage is due to the growth of *Bacillus panthothenticus* and *B. Circulans* (Yokoseki and Oskawa 1964) *B. Licheniformes*, *B. coagulance*, *B. subtilish* and *B. sphaericus* and *B. putrifaciens* (Yamagato and Nagaoka, 1966).

2.9.1 Discolouration

The discolouration begins around the meat at the mouth of the casing and spreads to all the visible surface of the contents, resulting in the swelling of casing. When actual spoilage is not evident, the discolouration of fish ham may be due to the oxidation of pork fat in the meat. According to Yokoseki and Oskawa (1964), *Streptococcus Sp.* Caused discolouration. The bacterium responsible for brown discolouration in kamaboko and other paste products was *Pseudomonas Sp.* (Mori, *et al.,* 1974).

2.9.2 Spot spoilage

Spot spoilage occurs, when the product is stored at higher temperatures for long time. Yokoseki and Okawa (1964) reported *B. Coagulance* as a causative agent for spot spoilage. Further, 10^{5} to 10^{7} cfu / g viable bacterial counts were obtained in spot spoiled products. Tanikawa (1965) studied the chemical properties and mechanism of black spot discolouration. A sulphur containing compound in onion, is decomposed by bacterium to hydrogen sulphide which further reacts with iron component present in the fish paste to from iron sulphide which finally results in the black spot discolouration.

2.9.3 Slime formation

Uchiyama and Amano (1958) found slime formation in kamaboko. Slime formation was encountered only if sucrose or riffinose was incorporated in kamaboko products (Uchiyama and Amano, 1959). Jensen (1954) has indicated that the spiecies of *Pseudomonas* have a relatively greater importance in the formation of slime.

Indian workers have reported all the above mentioned spoilage in different types of Japanese paste products such has fish sausage, fish ham, fish cutlet, Chikuwa, kamaboko and other product.

Material and methods

III. MATERIALS AND METHODS

3.1 Materials

3.1.1 Fish

Fresh Bull's eye (*Priacanthus hamrur*) (Plate.1) were procured from the fish landing centre, Mangalore. The fishes were washed in fresh water, packed in insulated box with sufficient quantity of ice (1:1) and transported to the laboratory. The fish was dressed so as to remove scale, fins, head and entrails. The dressed fish was thoroughly washed with chilled water and excess water was drained. The fish meat separation was carried out by reciprocatory type meat picking machine (SG model of Toyo Seikan Kaisha Ltd, Japan). The separated meat was minced in order to reduce the particle size using meat mincer (M - 3, Type - 42, supplied by Toyo Seikan Kaisha Limited, Japan). The mincing operation were carried out in two stages, the first stage using bigger perforations disc (5 - 6 mm dia) and second stage in smaller perforated disc (2 mm dia).

At the beginning of processing, the physical characteristics such as total length, total weight of 55 individual fishes was randomly noted. The average length and weight of fishes selected for the experiments were found to be 18.96 cm and 100 g respectively.

3.1.2. Shrimps

Fresh shrimps (*Penaeus indicus*) were procured in iced condition from the Mangalore fish landing centre, washed in potable water, repacked in insulated box with sufficient quantity of fresh ice (1:1) and transported to the laboratory. In the processing hall shrimps were thoroughly washed and dressed (peeled and deveined)

Plate: 1. Bull's eye (*Priacanthus hamrur*)

and washed in chilled water and stored at cooler storage (0 ± 2 °C) until further use (Plate 2).

3.1.3 Sub Materials

All other food grade additives / ingredients used in this experiment such as salt, sugar, chilli powder, coriander powder, pepper powder, ginger - garlic paste, corn starch powder and double refined sunflower oil were purchased from local market in fresh condition. Sodium tripolyphosphate (STPP), manufactured and supplied by BDH India, Ltd. Crushed ice made from chilled potable water was purchased from a local ice factory.

3.1.4 Chemicals

All the chemicals and reagents used in the present study were AR or GR grade. Sulphuric acid, hydrochloric acid, Sodium chloride, bromocresol green, methyl red, potassium carbonate, thiobarbituric acid, sodium hydroxide pellet, sodium dihydrogen phosphate 2 - hydrate crystal, pure di sodium hydrogen phosphate dihydrate, boric acid, copper sulphate - penta hydrate, potassium sodium tartarate, petroleum benzine (40 - 60 °C), petroleum ether, trichloro acetic acid (TCA) were procured from E - Merck India limited. Sodium carbonate was procured from Qualigens Fine Chemicals Glaxo India Ltd. Total plate count agar and 2 - thiobarbituric acid was ob- tained from Hi - media laboratories, India. Selenium dioxide and Folin's ciocaleau's phenol reagent, 1 - butanol, glycerol, acetic acid and glycine were procured from Merck specialties private limited. The cellulose thimbles and whatman filter paper were obtained from whatman Limited, U.K.

3.1.5 Media and broth:

All the media and broths were prepared according to the "Compedium of methods for the microbiological examination of foods (APHA, 1992)." The media and broths were prepared in distilled water and pH was adjusted to 7.0 ± 0.2 before sterilization. Hugh - leifson glucose broth (Liston - Barross), hydrogen peroxide, ammonium oxalate, crystal violet, lugols iodine, safranin, kovac's reagent, methyl red indicator, nutrient broth, tetra methyl paraphenylene diamine and plate count agar were procured from Hi - media Limited, India.

3.2 Casings

3.2.1 Fibrous casing

Fibrous casing reinforced with strong cellulose fibres which are suitable for large sausage calibres (80 mm x 320mm) and intended for slicing, were manufactured and supplied by Viskase companies' inc. Illinois, USA. and marketed by Vee tech Ltd, New Delhi. Fibrous casing were used as an alternative casing to synthetic casing.

3.2.2 Synthetic Casing

Krehalon synthetic casing (copolymers of viny chloride and vinylidine chloride) measuring 100 mm x 320 mm, manufactured and supplied by Kureha Chemical Industry Company Limited, Japan was used for stuffing the ground fish paste.

3.2.3 Equipments

3.2.4 Fish Meat Picking Machine

Model SG / S - 6 Type -1 “CAN” Mark, power driven reciprocatory type, fish meat picking machine, manufactured and supplied by Toyo Seikan Kaisha Ltd. Japan was used for meat separation from bone and skin. The separation disc has average perforation diameter of 6.5 mm.

3.2.5 Meat Mincer

Model M - 3, Type 42 : S - 2 ,“CAN” Mark supplied by Toyo Seikan Kaisha Ltd., Japan was used to reduce the size of picked meat of fish, fish meat was minced first with the disc having perforation of 1/8 ” diameter and then with second disc having perforation of 1/16 ” diameter.

3.3 Silent Cutter

Chopper (model SPL / Type: S - 4 “CAN” Mark, Toyo Seikan Kaisha Ltd., Japan) was used for grinding of various additives with minced meat.

3.3.1 Meat Stuffer

Locally designed hand stuffer was used for stuffing of fish ham into casings. The ends of the casings stuffed with fish ham were closed by folding and the tightly tied with thread.

3.3.2 Air blast freezer

Air blast freezer manufactured by Arm Field Ltd., Ringwood Hampshire, England was used for freezing sliced ham pieces. Temperature during freezing was maintained at - 35 °C for 3 hrs.

3.3.3 Deep freezer

Blue Star Co. Bombay, India. One ton capacity deep freezer was used for storage of frozen sliced fish ham at - 18 ± 2 °C.

3.3.4 METHODS

3.3.5 Preparation of fish ham in synthetic and fibrous casings

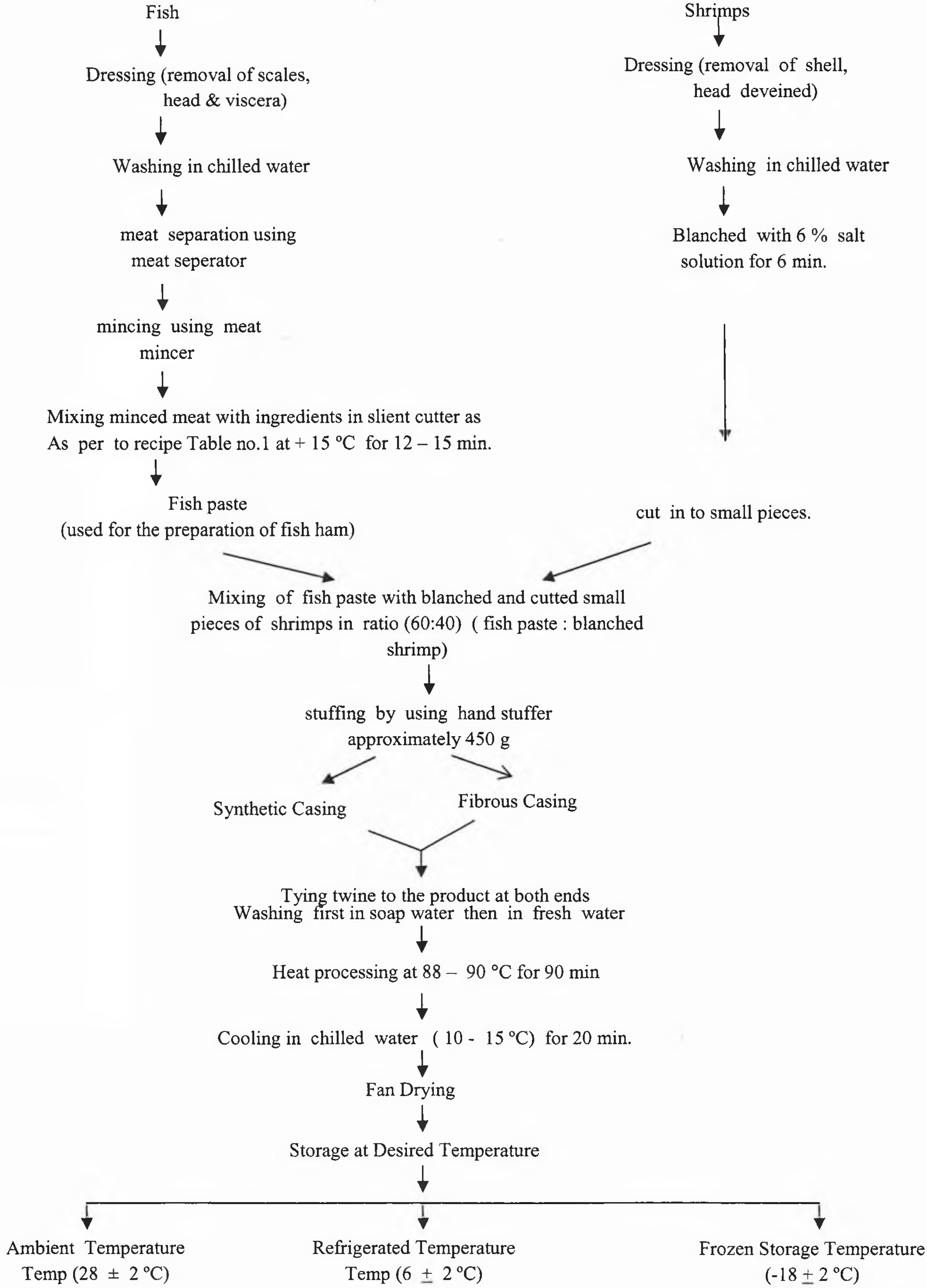
The method of preparation of fish ham is given in Fig.1. The fish was dressed so as to remove scale, fins, head and entrails. The dressed fish was thoroughly washed with chilled water and excess water was drained. The fish meat separation was carried out by reciprocatory meat picking machine SG, modle of Toyo Seikna Kaisha Limited, Japan. The separated meat was minced in order to reduce the particle size using meat mincer (M-3, Type-42, supplied by Toyo Seikan Kaisha Limited, Japan). The mincing operation were carried out in two stages, the first stage using bigger perforations disc (5-6 mm dia) and second stage in smaller perforated disc (2 mm dia). The addition of ingredients to the fish mince was carried out in silent cutter, SG, Model of Seikan Kaisha Limited, Japan. The capacity of silent cutter was 3 kg. The peeled and deveined shrimp from the chilled storage was taken out and rinsed in chilled water. Then shrimp was blanched in 6 % salt solution for 6 min, cooled and cut into small size uniformly. Blanched shrimp pieces were mixed with fish paste in

Table 1: Final recipe used for preparation of fish ham.

Sl. No	Ingredients	Percent *
1	Fish minced meat	70.00
2	Salt	2.50
3	Sugar	1.50
4	Polyphosphate	0.20
5	**Spice mixture**	
A	Chilli powder	0.60
B	Pepper powder	0.50
C	Coriander powder	0.45
D	Ginger	0.10
E	Garlic	0.15
6	Corn Starch	9.00
7	Crushed ice	10.0
8	Sundrop oil	5.00

Note * : Percentage based on fish paste weight

Fig 1:The following flow diagram gives the various stages of the preparation of fish ham.



Fish Ham Prepared in Fibrous Casing

Fish Ham Prepared in Synthetic Casing

Fish ham in slicedform with shrimp chunks

Plate: 2

the ratio 60 : 40 (Fish paste : blanched shrimps) in the bowl chopper. The composition of additives along with percentage of weight is given in the Table 1.

The duration of mixing operation was restricted to 12 min and any increase in temperature was compensated by the addition of crushed ice. The temperature of fish paste was maintained at 15 - 16 °C during mixing operation. The paste thus obtained was used for the preparation of fish ham. The paste was stuffed into Krehalon casing of 100 mm x 320 mm and fibrous casing of 80 mm x 320 mm (diameter x length) using hand stuffer and both the ends were tied using a twine tightly followed by tying around the product to maintain a attractive shape. The product was heat processed at 90 ± 2 °C for 90 min in a constant temperature water bath and then cooled in chilled water (10 - 15 °C) for 20 min and then stored at different temperatures viz., ambient temperature (28 ± 2 °C), refrigerated temperature (6 ± 2 °C) and frozen temperature (-18 ± 2 °C) for further analyses.

3.3.5.1 ANALYSIS

Samples were drawn regularly to study the changes in quality during storage. The acceptability and shelf life of the products were assessed by drawing samples once daily for products stored at ambient temperature. The samples stored at the refrigerated and frozen storage temperature were drawn once in a week and once in a month respectively. All the parameters were analyzed by using the standard methods.

3.3.5.2 Analysis of Chemical and Biochemical Characteristics

3.3.5.3 Proximate composition.

The proximate composition of fish meat and final products prepared were estimated. Moisture, crude protein, crude fat and ash contents of fresh fish and final products were determined according to the method described in AOAC, (2005).

3.3.5.4 Moisture content

The moisture content of ham was determined by hot air oven method AOAC, (2005). About 10 g of fish ham was taken in moisture bottle in triplicate and dried in a hot air oven with mechanical convection at 100 ± 2 °C for 10 - 16 hrs. The samples were cooled to room temperature in a desiccator containing silica gel. Drying and cooling was repeated till constant weight was obtained. The weight loss was expressed as percentage of the moisture content of the sample. The mean of three values were taken as moisture percentage of sample by using the formula as follows:

$$\text{Moisture (\%)} = \frac{W_1 - W_2}{W_1} \times 100$$

Where,

W_1 = initial weight of sample

W_2 = final weight of sample

3.3.5.5 Crude protein

The crude protein content of the fish ham was determined by estimating total nitrogen content by Kjeldahl method (AOAC, 2005). About 1.0 g sample was weighed accurately and digested with 10 ml of concentrated sulphuric acid and a pinch of digestion mixture (K_2SO_4: $CuSO_4$: SeO_2 in the ratio of 10:1:0.25) in a 250 ml digestion flask. Few glass beads were added to the digestion flask to avoid bumping.

The contents in the digestion flask were heated in the digestion chamber. The digestion was continued till the solution became clear and heating was stopped. The digested solution was cooled and made up to a known volume with distilled water. 2 ml of aliquot was transferred to kjeldahl distillation unit with 10 ml of 40 % sodium hydroxide. 10 ml of 2 % boric acid with mixed indicator (0.06 % bromocresol green and 0.06 % methyl red in 95 % ethyl alcohol) was taken in a conical flask in which liberated ammonia was absorbed. The ratio of bromocresol green and methyl red was 5:1 (5 parts of bromocresol green and 1 part of methyl red). The absorption of ammonia was indicated by change in colour of boric acid with mixed indicator from pink to green. Back washing was followed after distillation of each sample. The collected distillates were then titrated against 0.02 N H_2SO_4, till the pink colour was developed. Crude protein of meat samples was calculated by multiplying the nitrogen value obtained by a factor of 6.25. The protein content was expressed as percentage of total sample.

Calculation:

$$\text{Total nitrogen \%} = \frac{14 \times N \times X \times 250 \times 100}{1000 \times V \times W}$$

Where,

N = Normality of H_2SO_4

X = ml of standard H_2SO_4 required for titration of samples

V = Aliquot (ml) of digested extract taken for distillation

W = Weight (g) of sample

Since average nitrogen content of the fish protein is 16 %, so

1g nitrogen = 100 / 16

= 6.25 g protein

Hence, crude protein % = total nitrogen % × 6.25 (conversion factor)

3.3.6 Crude fat

The crude fat content of sample was estimated by Soxhlet extraction method (AOAC, 2005). Dried ham sample was used for fat estimation. About 1.0 g of sample was taken in Whatman thimble. The thimble was loosely plugged with cotton and placed in a soxhlet extraction unit using petroleum benzene or petroleum ether (40 - 60 °C) as solvent. The extraction of fat was carried out on a constant temperature controlled mantle set at 40 - 60 °C for about 16 hrs. The receiver flask containing extracted fat and solvent was evaporated on a water bath at 80 °C so as to remove most of the solvent and further the residual solvent was dried by placing the flask in hot air oven at the temperature of 60 °C. The flask was cooled in a desiccator and weight was recorded. The drying and cooling was repeated till constant weight was obtained. The fat content in the sample was calculated using the following formula

$$\text{Crude fat \%} = \frac{W_1 - W_2}{S} \times 100$$

Where,

S = Weight of dried sample

W_1 = Weight of the empty flask with glass beads

W_2 = Weight of the flask with glass beads after evaporation

3.3.6.1 Ash content

Ash content of fish ham was determined by the method as described in AOAC (2005). About 1.0 g of moisture free meat sample was taken in a pre - weighed silica crucible. Preliminary slow heating of the dried sample was done to allow smoking off fat on a flame without burning. The charred sample was then incinerated in a muffle furnace (Rotek, Kerala, India) at 550 ± 10 °C for 5 - 6 hrs. The crucibles were removed and cooled in desiccator and weighed. Ash content was calculated from the weight difference of crucible and expressed as the ash content in percentage by using following formula:

$$\text{Ash content (\%)} = \frac{W_1 - W_2}{W_1} \times 100$$

Where,

W_1 = Weight of crucible with sample (initial weight)

W_2 = Weight of crucible with ash (final weight)

3.3.6.2 Biochemical analysis

3.3.6.3 pH

The pH of the fish ham slurry was measured using pH meter (Systronix μ pH system 361) in the temperature range of 27 °C - 29 °C. About 5.0 g of meat sample was macerated with 45 ml of distilled water and pH was measured. Prior to pH measurement of the sample, pH meter was calibrated with standard buffer solution of pH 4.2 and 9.2 prepared using buffer capsule.

3.3.6.4 Peroxide value (PV)

The method of Tarr (1947) was employed for estimating the PV and expressed as milliequivalent of oxygen per kilogram of fat. 20 g of sample was ground well using a pestle and mortar with 20 g anhydrous sodium sulphate. Then it was transferred to a 100 ml stoppered flask. 70 ml chloroform was added and stirred well, kept in dark place for about 20 min with occasional shaking to extract crude fat.

10 ml of chloroform extract were taken into a 250 ml distilled conical flask to which 1ml of saturated potassium iodide was added and titrated against 0.02 N sodium thiosulphate solution using starch as an indicator. Peroxide value in the sample was calculated by the following formula,

$$\text{Peroxide value (milli. eq. of } O_2 \text{ / kg of fat)} = \frac{1000\,(V - X)\,N}{W}$$

Where, W = Weight of the sample (g)

V = Volume of sodium thiosulphate used (ml)

X = Volume of sodium thiosulphate used for blank (ml)

N = Normality of standard sodium thiosulphate

3.3.6.5 Thiobarbituric acid (TBA)

TBA number of samples was determined by the method of Sinnhuber and Yu, (1977). 10 g of sample was mascerated with 50ml of distilled water and transferred into a distillation flask using 47.5 distil water. 2ml of 4 N HCL was added to bring that pH to 1.5. The flask was heated so as to collect the 50 ml distillate in 10 min from the time boiling commenced. 5 ml of TBA reagent was added to 5 ml of distillate in a glass stoppered test tube, mixed well, heated in a boiling water bath for 20 min, cooled in running water for 10 min and the colour developed was measured at wave length of 532 nm using spectronic photometer. The TBA number was calculated from a standard graph based on the hydrolysis of 1, 1, 3.3 - tetra ethoxy propane (TEP), and expressed as mg of malonaldehyde per kg of fat.

3.3.6.6 Determination of free fatty acids (FFA)

The method of Dyer and Morten (1956) was employed for estimating FFA, after making a chloroform extract of the tissue, and expressed as percentage of Oleic acid. 10 g of sample was taken into a mortar, and added equal amount of anhydrous sodium sulphate, grinded well, then transferred to a conical flask. 70 ml of chloroform was added and kept in dark place for 15 min. The slurry was filtered and 10 ml of filtrate was taken in conical flask, dried on water bath to evaporate of chloroform.

50 ml of hot neutral alcohol was added, and titrated against standard alkali using phenolphthalein as indicator. Free fatty acid in the same was calculated by the following formula:

$$\text{FFA as Oleic acid \%} = \frac{\text{Titrate value x N NaOH x 28.2 x100}}{\text{Weight of fat}}$$

3.3.7 Total volatile base nitrogen (TVB-N)

Total volatile base nitrogen (TVB-N) of fish meat used for product preparation and final product during storage was analyzed.

10 g of sample was ground with 50 ml distilled water and proteins were precipitated by the addition of 15 ml of 20 % trichloroacetic acid (TCA) and filtered through whatman no. 4. Filter paper. The filtrate was made up to a known volume. Total volatile base nitrogen from the trichloroacetic acid extract thus prepared for fresh fish and final products were determined by the procedure of Beaty and Gibbons, (1937) using Conways micro diffusion units. The results were expressed as mg N percent of sample by using the following formula.

$$\text{TVB-N (mg / 100 g)} = \frac{14 \times N \times (X - Y) \times 100 \times 100}{S}$$

Where,

N = Normality of sulphuric acid

X = ml of sulphuric acid required for titration of sample

Y = ml of sulphuric acid required for titration of blank

S = weight of sample

3.3.7.1 Trimethylamine nitrogen (TMA-N)

Total volatile base nitrogen (TVB-N) of fish meat used for product preparation and final product during storage was analyzed.

10 g of the meat sample was macerated with 50 ml of distilled water using pestle and mortar and allowed to stand in refrigerator for 30 min. 10 ml of 20 % trichloro acetic acid (TCA) was added to the above solution and allowed to stand for 10 min. The slurry was filtered with coarse filter paper and was made up to 100 ml with distilled water.

2 ml of boric acid containing mixed indicator (0.066 % methyl red and 0.066 % bromocresol green solution in alcohol in the ratio of 1:1 was pipetted into the inner chamber and 1 ml of sample into outer chamber of Conway micro diffusion unit. The unit was covered with covering glass with application of grease leaving a small gap for addition of saturated potassium carbonate and 1ml of formaldehyde in the outer chamber. The content of outer chamber was mixed gently with rotation. Solution was incubated at 37 °C overnight. After the incubation, inner chamber content was titrated against 0.02 N sulphuric acid. A blank was conducted using 2 % TCA solution instead of sample. TVB-N was calculated using following formula and expressed in mg %.

$$\text{TMA-N (mg / 100 g)} = \frac{14 \times N \times (X - Y) \times 100 \times 100}{S}$$

Where,

N = Normality of sulfuric acid

X = ml of sulfuric acid required for titration of sample

Y = ml of sulfuric acid required for titration of blank

S = weight of sample

3.3.7.2 Microbial analysis

Total plate count and aerobic spore former from fresh fish, additives used for product preparation and prepared fish ham were enumerated according to the procedure described in APHA (1992).

3.3.7.3 Total plate count (TPC)

Total plate count was estimated according to the procedure as described in APHA (1992). Bacterial counts were determined by Pour plate method over the storage time. 10 g of fish ham sample was aseptically taken and homogenised in 90 ml of saline water using pestle and mortar. Adequate serial dilutions were prepared and 1 ml of each dilution was inoculated in Total plate count agar media. Under aseptic conditions, plates were rotated clockwise and anticlockwise for proper mixing and incubated at 37 ± 2 °C for 48 hrs. All estimations were done in triplicates and results were expressed as colony forming unit per gram (cfu / g).

3.3.7.4 Aerobic spore former

Aerobic spore formers were estimated according to the procedures described in APHA (1992). Spore counts were determined by Pour plate method over the storage time. 10 g of fish ham sample was homogenised in 90 ml of saline water using pestle and mortar. Adequate serial dilutions were prepared by using saline and subjected to heat treatment at 80 °C for 10 min to kill all vegetative cells. The tubes were cooled and 1ml of inoculums was plated in Total plate count agar media. Under aseptic conditions, plates were rotated clockwise and anticlockwise for proper mixing and incubated at 37 °C for 48 hrs and aerobic spore count per gram of sample were expressed.

3.3.7.5 Sensory evaluation

The sensory evaluation of fish ham in synthetic and fibrous casing was carried out for based on a 9 point hedonic scale. The minimum number of panellists for each evaluation was 12. The procedure of evaluation followed by using the method described by Rathod and Pagarkar (2013). The trained panel of judges were asked to rate the products based on the attributes like appearance, colour, odour, texture, taste and flavour. The score sheet provided to the panellists is given in Appendix 1.The samples were warmed in boiling water for 5 min at 100 $^{\circ}$C before subjecting to sensory evaluation. The score obtained by panellists were analysed statistically to draw meaningful conclusion.

3.4 Statistical analysis

All the analysis were conducted in triplicates. The data was analysed using analysis of variance (ANOVA) using SPSS (2000) and the means were separated with Duncan's multiple ranges test.

Experimental results

IV. EXPERIMENTAL RESULTS.

4.1 Raw material characteristics

The quality characteristics of fresh Bull's eye (*Priacanthus hamrur*) and the proximate composition of shrimp are presented in Table 2 and 3 respectively. Mean sensory score for standardization of cooking time of fish ham in both fibrous and synthetic casing are presented in Table 4.

4.1.1 Physical characteristics

The physical characteristics of fish namely the average total length and average weight of the fish were found to be 18.96 cm, and 84.35 g respectively. Dressing yield from whole fish was 55.85 %. From dressed fish to picked meat yield percentage was 58.04 % and minced meat yield is 55.21 %. Further meat yield percentage from the cleaned whole fish to picked meat and minced meat were found to be 32.42 % and 30.84 % respectively.

4.1.2 Proximate composition of fresh shrimp

The proximate composition of fresh shrimp used in fish ham are presented in Table 3. Moisture, protein, fat and ash contents of the shrimp were found to be 81.40 %, 16.40 %, 1.12 % and 1.56 % respectively.

4.1.3 Biochemical and microbiological characteristics of the fresh fish.

Biochemical and microbiological characteristics of fresh fish are given in Table 2. Initial pH of fish meat was found to be 6.84. TVB-N and TMA-N content of fresh fish muscle were observed to be 7.44 and 2.82 mg % respectively. The PV of fresh fish was found to be 6.58 milli. eq O_2 / kg of fat. It was found that FFA was 2.85

Table 2 : Raw material characteristics of fresh Fish bull's eye (*Priacanthus hamrur*)

Sl. No.	Characteristics	Composition
1	**Physical characteristics ***	
	Average total length (cm)	18.96 (± 0.65)
	Average standard length (cm)	15.59 (± 0.52)
	Average total weight (g)	84.35 (± 5.16)
	Average dressed yield (%)	55.85 (± 0.12)
	Average picked meat yield (%)	32.42 (± 0.85)
	Average minced yield (%)	30.84 (± 0.76)
2	**Proximate composition ****	
	Moisture (%)	77.80 (± 0.16)
	Protein (%)	18.53 (± 0.24)
	Fat (%)	1.38 (± 0.08)
	Ash (%)	1.45 (± 0.18)
3	**Non – protein nitrogenous compounds**	
	pH	6.84 (± 0.18)
	TVB-N (mg %)	7.44 (± 1.70)
	TMA-N (mg %)	2.82 (± 1.15)
4	**Lipid characteristics**	
	PV (milli eq. of O_2 / kg of fat)	6.58 (± 0.60)
	FFA (% of Oleic acid)	2.85 (± 0.14)
	TBA (mg of malonaldehyde / kg of fat)	0.56 (± 0.20)
5	**Microbiological characteristics**	
	Total plate count (cfu / g)	2.6×10^4 (± 0.05)
	Spore counts (No / g)	-Nil-

Note * : Mean values of 20 fishes

** : Mean values of triplicates

( ) : Values in parenthesis indicate standard deviation.

Table 3 : Proximate Composition biochemical and microbiological parameters of Fresh Shrimps

Sl. No.	Proximate composition *	Percentage
1	Moisture	81.40 (± 2.14)
2	Protein	16.40 (± 0.24)
3	Fat	1.12 (± 0.06)
4	Ash	1.56 (± 0.08)
5	**Non – protein nitrogenous compounds**	
6	pH	7.18 (± 0.13)
7	TVB-N (mg %)	4.48 (± 1.12)
8	TMA-N (mg %)	2.80 (± 0.36)
9	**Lipid characteristics**	
10	PV (milli eq. of O_2 / kg of fat)	2.30 (± 0.09)
11	FFA (% of Oleic acid)	1.05 (± 0.16)
12	TBA (mg of malonaldehyde / kg of fat)	0.44 (± 0.02)
13	**Microbiological characteristics**	
14	Total plate count (cfu / g)	6.4×10^5 (± 1.24)
15	Spore counts (No / g)	Nil

Note * : Mean values of triplicates.
( ) : Values in parenthesis indicate standard deviation

Table 4 : Mean sensory scores for standardization of fish ham cooked time for synthetic and fibrous casings.

Time (min) / Attributes	70		80		90		100	
	SC	FC	SC	FC	SC	FC	SC	FC
Appearance	6.50	6.00	7.80	7.50	8.75	8.40	8.00	7.50
Color	7.00	6.70	8.00	7.50	8.75	8.50	8.50	8.00
Odor	6.00	6.00	6.50	6.50	8.50	8.25	8.00	8.25
Taste	5.60	5.80	7.00	7.25	8.75	8.10	8.25	8.00
Texture	5.80	6.00	7.00	6.50	8.80	8.50	8.35	8.00
Flavor	6.00	5.50	7.15	6.20	8.75	8.50	8.50	7.50
Overall acceptability	6.50	6.00	7.25	7.00	8.75	8.25	8.00	7.75

Note : SC : Synthetic casing

FC: Fibrous casing

Table 5 : Proximate Composition of fish ham before storage

Parameters*	S C	F C
Moisture (%)	71.39 (± 0.14)	67.63 (± 0.08)
Protein (%)	18.15 (± 0.18)	19.20 (± 0.24)
Fat (%)	5.66 (± 0.12)	6.33 (± 0.08)
Ash (%)	2.50 (± 0.04)	2.21 (± 1.01)

Note * : Mean values of triplicates on (w/w).
( ) : Values in parenthesis indicate standard deviation.

SC : Synthetic casing

FC : Fibrous casing

synthetic and fibrous casing respectively. The protein content of fish ham packed in synthetic casing was 18.15 % and in fibrous casing was 19.20 %. The ash content was reported to be 2.5 % and 2.21 % for products in synthetic casing and fibrous casing respectively.

4.2 Changes in chemical quality characteristics of fish ham during ambient temperature storage at (28 ± 2 °C)

4.2.1 pH

The changes in pH of fish ham stored at ambient temperature are presented in Table 6 and Fig. 2. The initial pH of the ham product soon after preparation was 6.82 in both synthetic and fibrous casing. As the storage period increased. The values of pH in both the products gradually decreased to 6.25 and 6.10 at the end of storage period of 4th and 3rd day in synthetic casing and fibrous casing ham respectively.

4.2.2 Total volatile base nitrogen (TVB-N)

The results of the changes in TVB-N are presented in Table 7 and Fig.3. The initial TVB-N of fibrous casing ham was 1.5 mg % which had increased to 12.78 mg % after 1 day and reached a value of 41.84 mg % at the end 3rd day. The fish ham packed in synthetic casing has also gradually increased showing 28.28 mg % on 3rd day from its initial value of 1.16 mg %.

4.2.3 Trimethylamine nitrogen (TMA-N)

The results of the changes TMA-N are presented in Table 8 Fig. 4. The recorded values of TMA-N in fibrous casing fish ham indicated a higher value from the beginning till the end of the storage period with its initial value of 2.80 mg % to

18.67 mg % at the end of 3rd day. The fish ham stuffed in synthetic casing showed a gradual increase in TMA-N value from 2.40 mg % to 16.25 mg % on 4th day.

4.2.4 Peroxide value (PV)

The results of the changes in PV are presented in Table 9 and Fig. 5. The initial PV of both the products recorded a gradual increase with the increase in storage days to 20.23 milli eq. of O_2 / kg of fat from 1.38 milli eq.of O_2 / kg of fat in synthetic casing fish ham on 4th day where as a PV value of 25.58 milli eq. of O_2 / kg of fat from 2.45 milli eq.of O_2 / kg of fat has been recorded in fibrous casing product on 3rd day.

4.2.5 Free fatty acid (FFA) value

The results of FFA presented in Table 10 and Fig. 6. The free fatty acid content of both the products increased to 8.5 and 8.23 % of oleic acid on 4th and 3rd day from its initial values of 2.39 and 2.9 oleic acid % in synthetic and fibrous casing fish ham respectively.

4.2.6 Thiobarbituric acid (TBA)

The TBA content of the samples are given in the Table 11 and Fig. 7. A very slow increase in its TBA content in both the products were observed from initial values of 0.11 to final value of 2.01 and 0.21 to 2.94 mg. of malonaldehyde initial values to final value per kg of meat on 4th day in synthetic casing fish ham and on 3rd day in fibrous casing fish ham respectively.

Table 6 : Changes in pH of fish ham stored at ambient temperature (28 ± 2 ^{0}C)

Storage days	**pH**	
	S C	**F C**
0	6.82 (± 0.18)	6.82 (± 0.04)
1	6.63 (± 0.14)	6.60 (± 0.15)
2	6.40 (± 0.16)	6.20 (± 1.08)
3	6.30 (± 0.12)	6.10 (± 0.04)
4	6.25 (± 0.02)	*
5	*	

Note: * : Spoiled **SC :** Synthetic casing **FC :** Fibrous casing

( ) : values in parenthesis indicate standard deviation, n = 3

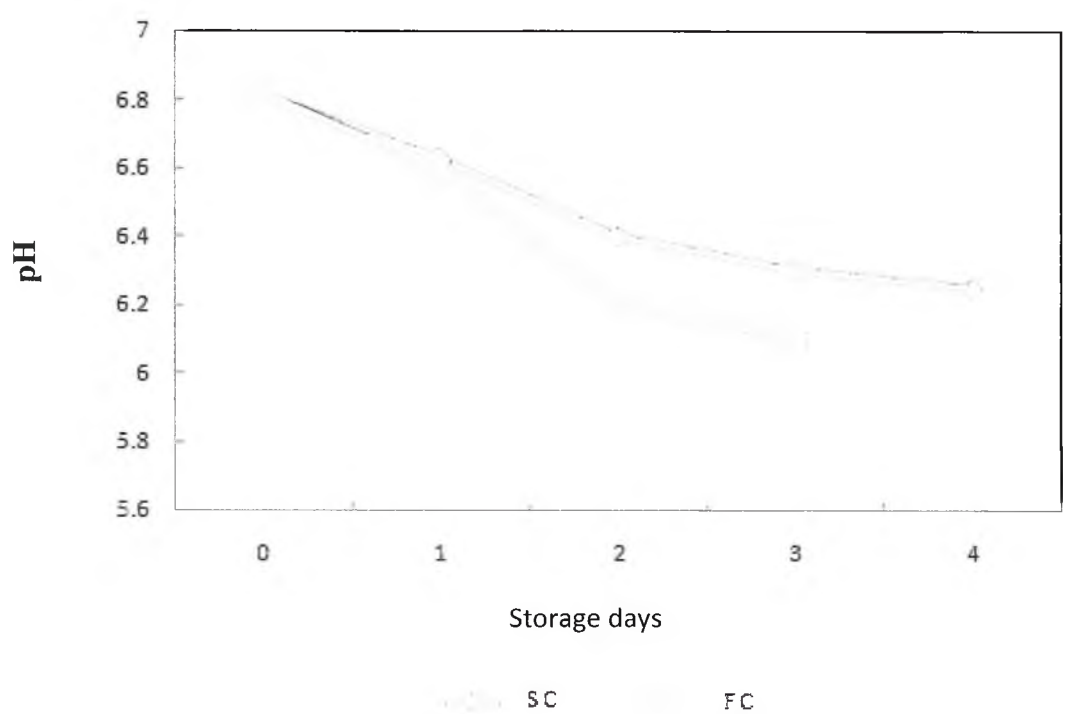


Fig : 2 Changes in pH of fish ham stored at ambient temperature (28 ± 2 ^{0}C)

SC : Synthetic casing **FC :** Fibrous casing

Table 7 : Changes in Total volatile base nitrogen (TVB-N) content of fish ham stored at ambient temperature (28 ± 2 ^{0}C)

Storage days	TVB-N (mg / 100 g)	
	S C	**F C**
0	1.16 (± 0.14)	1.50 (± 0.74)
1	5.56 (± 1.12)	12.78 (± 0.44)
2	15.93 (± 0.42)	26.64 (± 0.18)
3	28.28 (± 1.83)	41.84 (± 1.64)
4	41.40 (± 1.74)	*
5	*	

Note : * : Spoiled **SC :** Synthetic casing **FC :** Fibrous casing.

( ) : Values in parenthesis indicate standard deviation, n = 3.

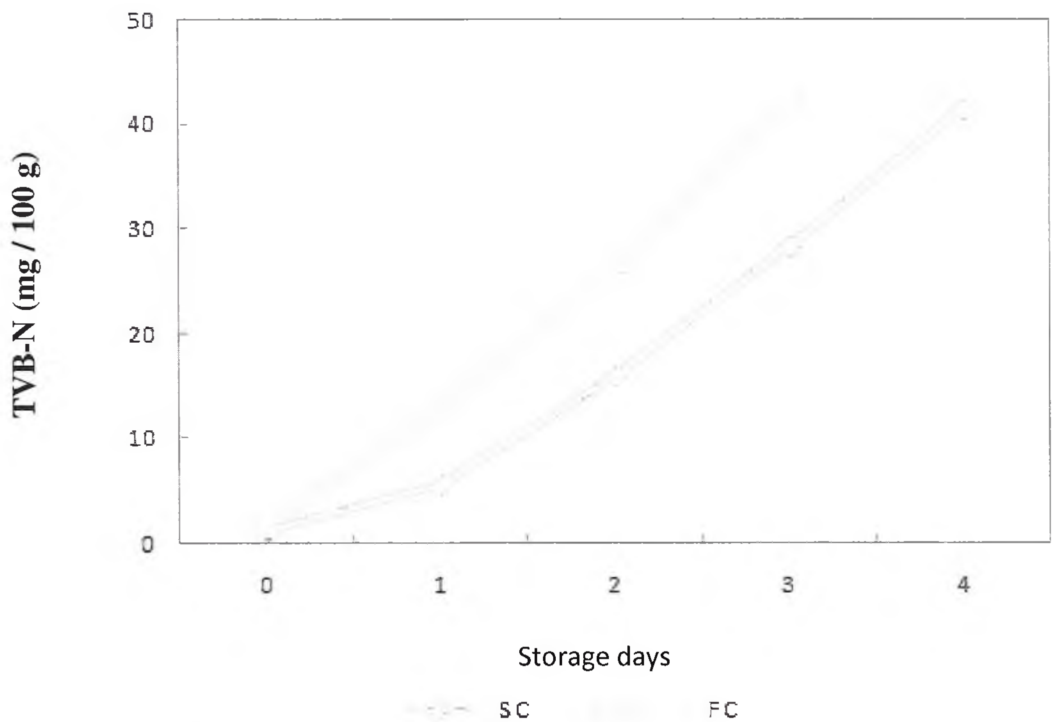


Fig : 3 Changes in Total volatile base nitrogen (TVB-N) content of fish ham stored at ambient temperature (28 ± 2 ^{0}C)

SC : Synthetic casing **FC :** Fibrous casing.

Table 8 : Changes in Trimethylamine nitrogen (TMA-N) content of fish ham stored at ambient temperature (28 ± 2 ^{0}C)

Storage days	TMA-N (mg / 100 g)	
	S C	**F C**
0	2.40 (± 0.34)	2.80 (± 0.36)
1	3.97 (± 0.84)	5.96 (± 1.02)
2	6.80 (± 1.34)	12.30 (± 0.18)
3	8.50 (± 1.74)	18.67 (± 1.24)
4	16.25 (± 0.12)	*
5	*	

Note : * : Spoiled **SC :** Synthetic casing **FC :** Fibrous casing

( ) : Values in parenthesis indicate standard deviation, n = 3

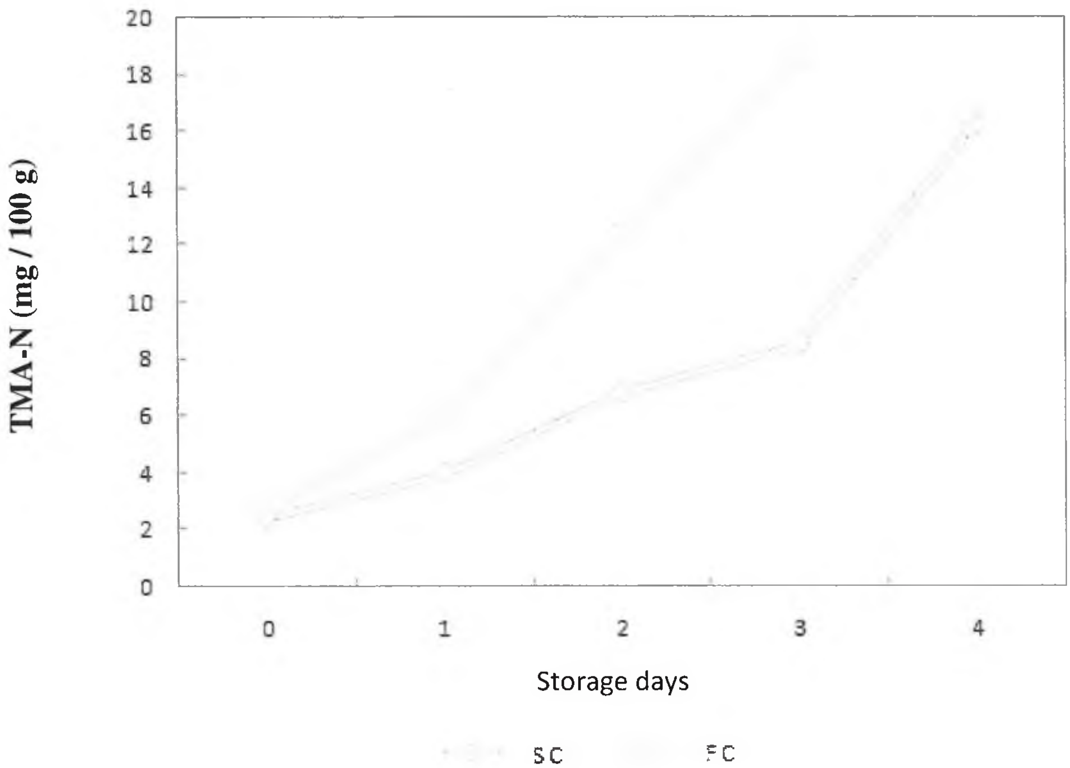


Fig : 4 Changes in Trimethylamine nitrogen (TMA-N) content of fish ham stored at ambient temperature (28 ± 2 ^{0}C)

SC : Synthetic casing **FC :** Fibrous casing.

Table 9 : Changes in peroxide value of fish ham stored at ambient temperature (28 ± 2 ^{0}C)

Storage days	P V (milli eq O_2 / kg of fat)	
	S C	F C
0	1.38 (± 0.08)	2.45 (± 0.09)
1	10.31 (± 0.034)	11.01 (± 0.28)
2	12.44 (± 1.24)	14.31 (± 0.56)
3	14.58 (± 0.45)	25.58 (± 0.84)
4	20.23 (± 0.08)	*
5	*	

Note : * **:** Spoiled **SC :** Synthetic casing **FC :** Fibrous casing.

( ) : Values in parenthesis indicate standard deviation, n = 3

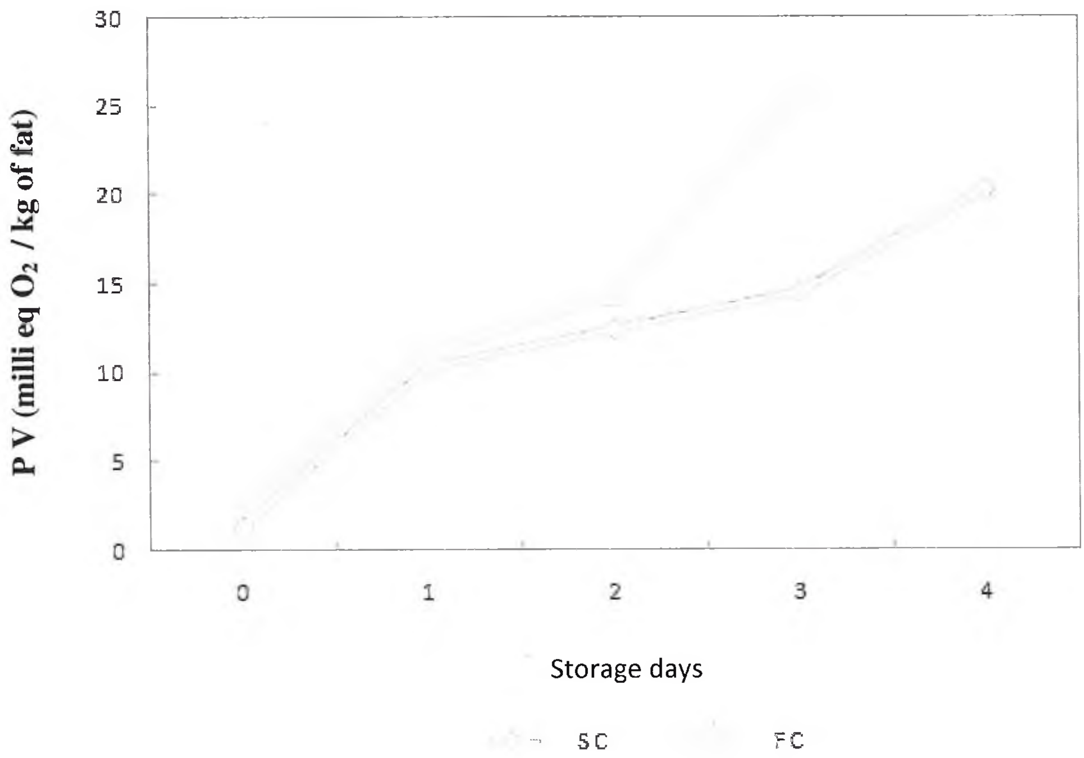


Fig : 5 Changes in peroxide value of fish ham stored at ambient temperature ($28 \pm 2\ ^0$C)

SC : Synthetic casing **FC :** Fibrous casing.

Table 10 : Changes in Free fatty acid of fish ham stored at ambient temperature ($28 \pm 2\ ^0C$)

Storage days	FFA (% of Oleic acid)	
	S C	**F C**
0	2.39 (± 0.36)	2.90 (± 0.94)
1	5.29 (± 0.09)	5.29 (± 1.04)
2	6.41 (± 1.04)	6.49 (± 0.84)
3	7.49 (± 1.14)	8.23 (± 1.12)
4	8.50 (± 0.14)	*
5	*	

Note : * **:** Spoiled **SC :** Synthetic casing **FC :** Fibrous casing.

( ) **:** Values in parenthesis indicate standard deviation, n= 3.

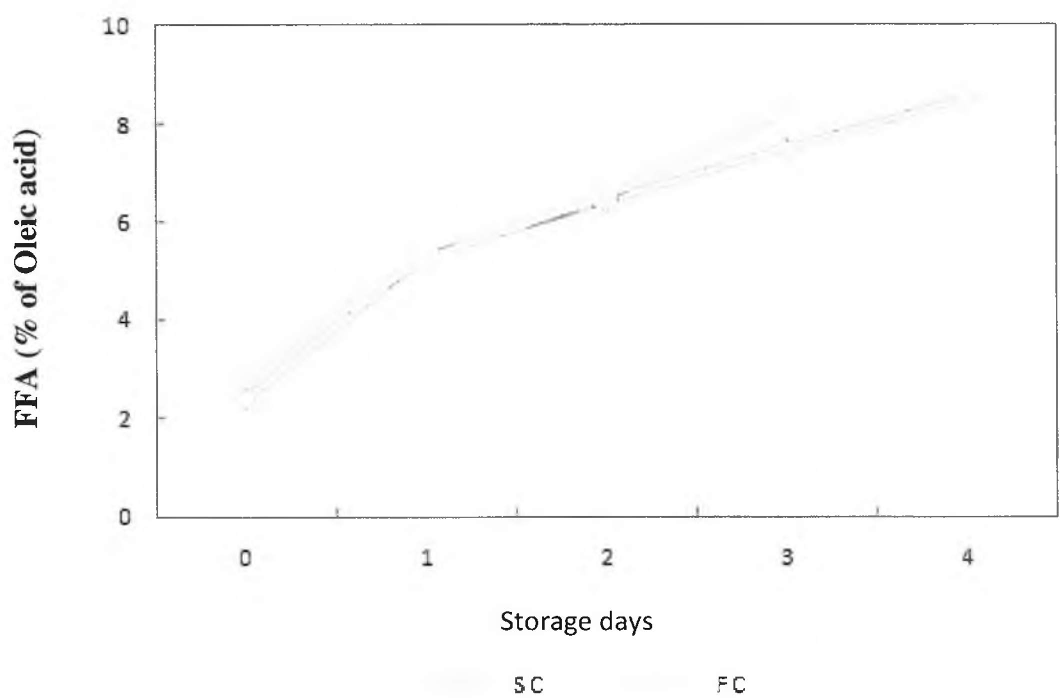


Fig : 6 Changes in Free fatty acid of fish ham stored at ambient temperature (28 ± 2 ^{0}C)

SC : Synthetic casing **FC :** Fibrous casing.

Table 11 : Changes in TBA value of fish ham stored at ambient temperature (28 ± 2 ^{0}C)

Storage days	TBA (mg malonaldehyde / kg meat)	
	S C	**F C**
0	0.11 (± 0.12)	0.21 (± 0.10)
1	0.23 (± 0.02)	0.80 (± 0.02)
2	1.59 (± 0.13)	1.95 (± 1.07)
3	1.97 (± 0.45)	2.94 (± 0.04)
4	2.01 (± 0.08)	*
5	*	

Note : * : Spoiled **SC :** Synthetic casing **FC :** Fibrous casing

( ) : Values in parenthesis indicate standard deviation, n= 3.

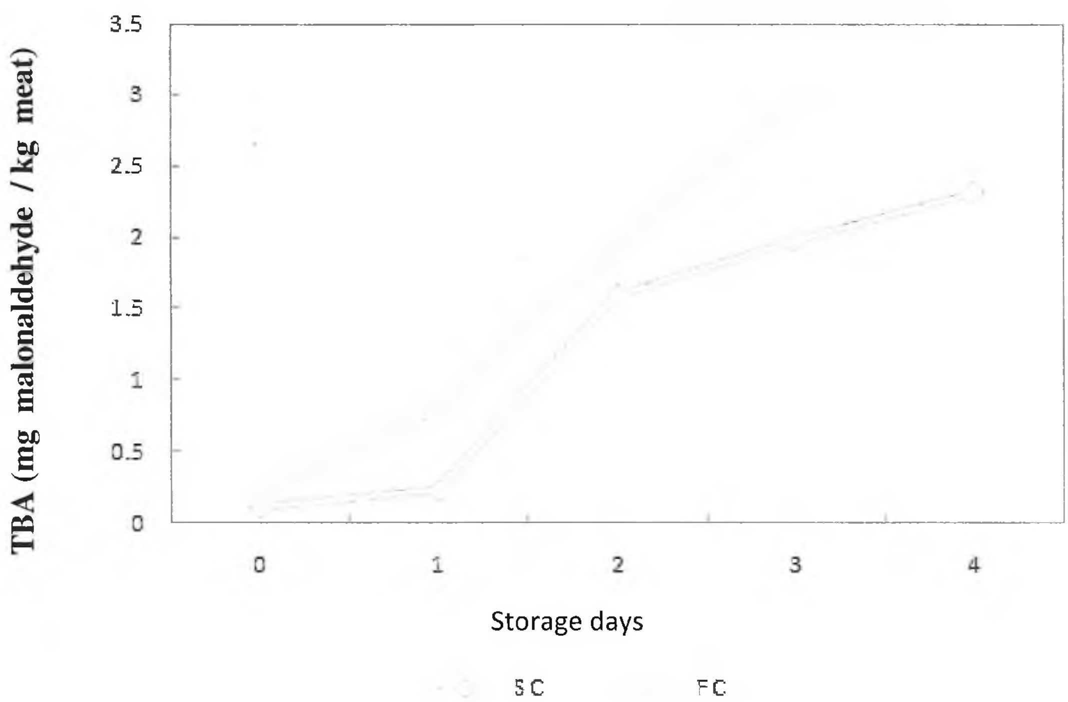


Fig : 7 Changes in TBA value of fish ham stored at ambient temperature (28 $\pm$ 2 ^{0}C)

SC : Synthetic casing **FC :** Fibrous casing.

Table 12 : Changes in microbiological quality characteristics of fish ham stored at ambient temperature (28 ± 2 ^{0}C)

Storage days	TPC (c f u /g)		Spores (No / g)	
	S C	F C	S C	F C
0	1.15×10^1 (1.24)	1.35×10^1 (1.48)	0.12×10^1 (0.28)	0.94×10^1 (0.40)
1	6.40×10^2 (3.80)	5.30×10^3 (2.18)	3.6×10^2 (1.12)	4.2×10^2 (2.02)
2	3.25×10^5 (5.40)	4.10×10^4 (1.18)	3.4×10^2 (2.18)	8.2×10^3 (4.16)
3	8.40×10^5 (6.24)	3.18×10^6 (5.02)	4.5×10^3 (1.10)	6.10×10^4 (5.08)
4	3.40×10^6 (5.10)	*	6.4×10^3 (3.45)	*
5	*		*	

Note : * : Spoiled **SC** : Synthetic casing **FC** : Fibrous casing

( ) : Values in parenthesis indicate standard deviation, n= 3.

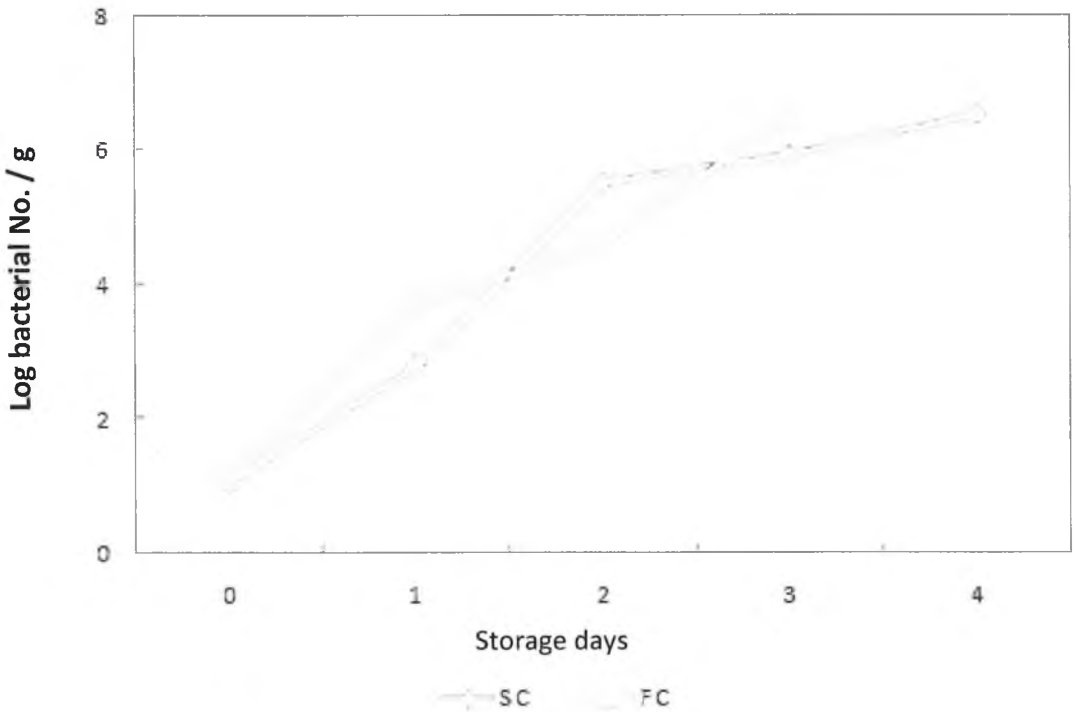


Fig : 8 Changes in microbiological quality characteristics of fish ham stored at ambient temperature (28 ± 2 ^{0}C)

SC : Synthetic casing **FC :** Fibrous casing

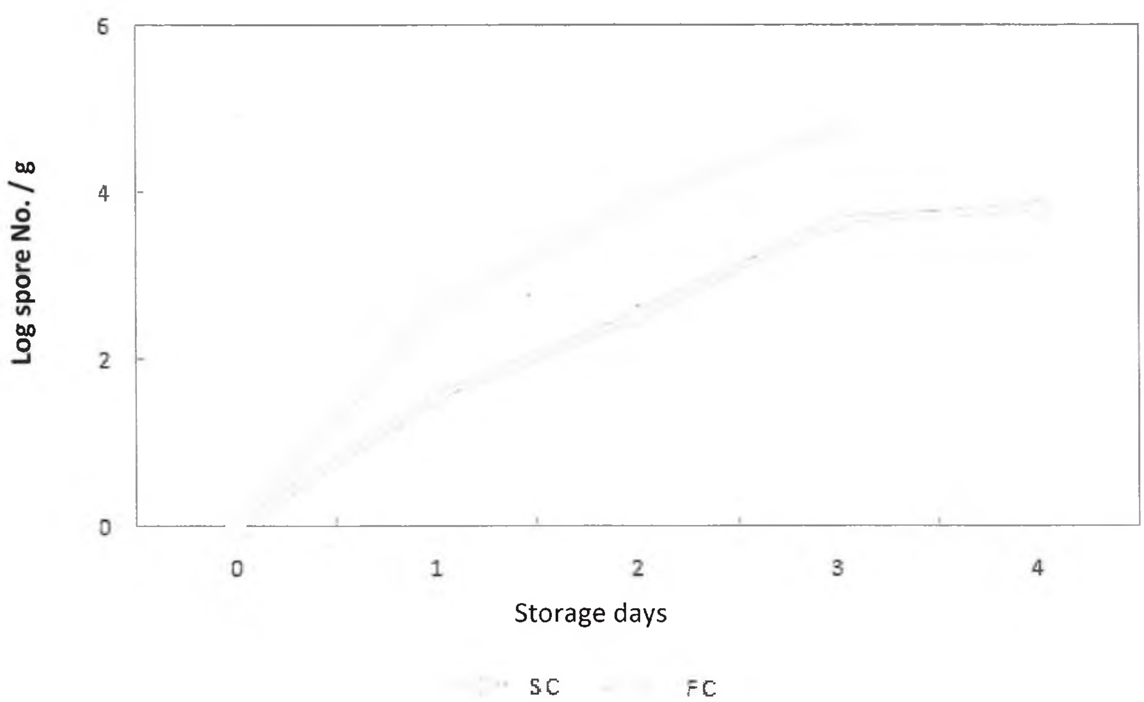


Fig : 9 Changes in microbiological quality characteristics of fish ham stored at ambient temperature (28 ± 2 ^{0}C)

SC : Synthetic casing **FC :** Fibrous casing

Table 13 : Changes in Oragnoleptic quality characteristics of fish ham stored at ambient temperatu

Attributes / Storage days	Appearance		Colour		Odour		Texture		Taste	
	S C	**F C**	**S C**	**F C**	**S C**	**F C**	**S C**	**F C**	**S C**	**F C**
0	8.00	7.70	8.00	7.90	8.13	8.13	8.06	8.06	7.80	7.94
1	8.00	7.40	7.90	7.60	8.10	8.10	8.00	7.50	7.70	7.60
2	7.80	6.50	7.50	6.50	7.80	6.40	8.00	5.50	7.50	5.20
3	7.00	*	7.10	*	6.50	*	6.00	*	6.00	*
4	*		*		*				*	

Note : * : Spoiled **SC** : Synthetic casing **FC** : Fibrous casing

4.2.7 Microbiological quality changes in fish ham stored at ambient temperature (28 ± 2 °C)

Changes in total plate counts and spore counts of fish ham in synthetic and fibrous casing during storage at ambient temperature were given in Table 12 and Fig. 8 and 9 indicate the changes in total plate count and spore counts. The TPC of synthetic casing fish ham was 1.15×10^1 cfu / g and 1.35×10^1 cfu / g in fibrous casing ham. As the storage days increased there was rise in TPC to 3.4×10^6 and 3.18×10^6 on day 4 and 3 in synthetic casing ham and fibrous casing fish ham respectively. Spore formers were observed to increase gradually, on 4th day spore counts were found to be 6.40×10^3 in synthetic casing fish ham and 6.10×10^4 in fibrous casing fish ham on 3rd day.

4.2.8 Sensory changes of fish ham stored at ambient temperature (28 ± 2 °C)

The change in organoleptic quality characteristics of fish ham stored at ambient temperature (28 ± 2 °C) is presented in Table 13 and Fig. 10. The overall acceptability score is the mean of the attributes such as colour, texture, appearance, flavour and taste. The overall acceptability scores of 8.0 and 7.90 for fish ham in both synthetic and fibrous casing stored at ambient temperature was observed to be decreased to 6.35 on 3rd day and 5.86 on 2nd day respectively.

4.3 Changes in chemical quality characteristics of fish ham at refrigerated temperature (6 ± 2 °C)

4.3.1 pH

The changes in pH in both type of fish ham products stored at refrigerated temperature were very marginal, which is presented in the Table 15 and Fig. 11. An increase in pH from its initial value of 6.82 to 6.96 and 7.00 at the end of 14^{th} and 28^{th} day for synthetic casing and 7.30 for fibrous casing at the end of 21 days.

4.3.2 Total volatile base nitrogen (TVB-N)

According to Table 16 and Fig. 12 the TVB-N values of fish ham in fibrous casing during its storage at refrigerated temperature has gradually increased to 28.64 mg % after 21 days from 16.60 mg % recorded on 7^{th} day. TVB-N content of 18.02 mg % was reached on 21^{st} day but the product was spoiled at the end of 28 days in fibrous casing fish ham. In case of fish ham in synthetic casing the increase was gradual and reached 26.44 mg % on 28^{th} day from of 15.6 mg % on 14^{th} day of storage at refrigerated temperature indicated that the product is in acceptable condition.

4.3.3 Trimethylamine nitrogen (TMA-N)

The TMA-N content of fish ham in synthetic and fibrous casing products were 2.40 and 2.80 mg % in the beginning but gradually increased to 6.01 mg % in synthetic casing fish ham at the end of 28 days and to 9.80 mg % on 21^{st} day in fibrous casing fish ham (Table 17 and Fig. 13). The TMA-N content for fibrous casing ham was found to be more at the end of 28 days when compared to synthetic casing ham.

4.3.4 Peroxide value (PV)

The results of changes in peroxide value are presented in the Table 18 and Fig 14. The peroxide value content of fish ham in synthetic casing and fibrous casing stored at refrigerated temperature showed increase with the increase of storage period from 2.34 to 15.01 milli eq. of O_2 / kg of fat on 7^{th} day and 28^{th} day in synthetic casing fish ham, where as peroxide value in fibrous casing fish ham also showed value increased from 3.48 to 22.77 milli eq. of O_2 / kg of fat on 7^{th} and 21^{st} day respectively. At the end of 28 days the ham in fibrous casing was observed to be spoiled.

4.3.5 Free fatty acid (FFA) value

The free fatty acid content of fish ham synthetic casing stored at refrigerated temperature showed increasing trend with increase of storage period from 2.39 to 7.88 % of oleic acid on 28^{th} day. The free fatty acid of the same in fibrous casing also increased from 2.90 to 8.05 % of oleic acid on 21^{st} day (Table 19 and Fig. 15).

4.3.6 Thiobarbutric acid (TBA)

The TBA content of fish ham is presented in the Table 20 and Fig. 16 for fish ham in synthetic casing stored at refrigerated condition showed a gradual increase in its content with increase in storage days from 1.18 on 7^{th} day to 1.97 mg malanoaldehyde / kg of meat on 28^{th} day. Where TBA of fish ham in fibrous casing increased from its initial value of 0.21 to 1.13 and 3.94 mg maloaldehyde / kg of meat at the end of 7^{th} and 21^{st} day indicating the oxidative spoilage has occurred.

4.3.7 Microbiological quality changes in fish ham stored at ambient temperature ($6 \pm 2\ ^{o}$ C)

The initial TPC of fish ham soon after processing was found to be 1.35×10^{1} and 1.15×10^{1} cfu /g in products packed in fibrous casing and synthetic casing respectively. However the spore counts for the same were very less. Microbiological counts of both the samples increased gradually during the storage period to, 4.5×10^{6} on 21^{st} day in fibrous casing and 2.9×10^{4} cfu /g on 28^{th} day in synthetic casing fish

Table 14 : Changes in proximate composition of fish ham stored at refrigerated temperature (6 ± 2 ^{0}C)

Storage days / Components	0		7		14		21	
	S C	F C	S C	F C	S C	F C	S C	F C
Moisture (%)	71.39 (± 0.14)	67.63 (± 0.08)	68.67 (± 0.09)	63.89 (± 0.12)	67.74 (± 0.14)	63.15 (± 0.06)	68.47 (± 0.09)	59.88 (± 0.12
Protein (%)	18.15 (± 0.18)	18.15 (± 0.24)	17.70 (± 0.19)	16.70 (± 0.06)	17.20 (± 0.12)	16.80 (± 0.06)	16.24 (± 0.04)	15.40 (± 0.17
Crude fat (%)	5.66 (± 0.12)	5.33 (± 0.08)	4.17 (± 0.14)	5.13 (± 0.09)	4.65 (±0.16)	5.62 (± 0.04)	6.14 (± 0.02)	7.03 (± 0.16
Ash (%)	2.50 (± 0.04)	2.21 (± 1.01)	2.75 (± 0.10)	3.09 (± 0.19)	3.15 (± 0.15)	3.64 (± 0.12)	3.64 (± 0.02)	3.95 (± 0.06

Note : * : Mean values of triplicates **SC :** Synthetic casing **FC :** Fibrous casing

Table 15 : Changes in pH of fish ham stored at refrigerated temperature (6 ± 2 ^{0}C)

Storage days	pH	
	S C	**F C**
0	6.82 (± 0.18)	6.82 (± 0.04)
7	6.92 (± 0.16)	6.98 (± 0.15)
14	6.96 (± 0.13)	7.00 (± 0.13)
21	6.98 (± 0.12)	7.30 (± 0.04)
28	7.00 (± 0.18)	*
35	*	

Note : * : Spoiled **SC :** Synthetic **FC :** Fibrous casing

( ) : Values in parenthesis indicate standard deviation, n = 3.

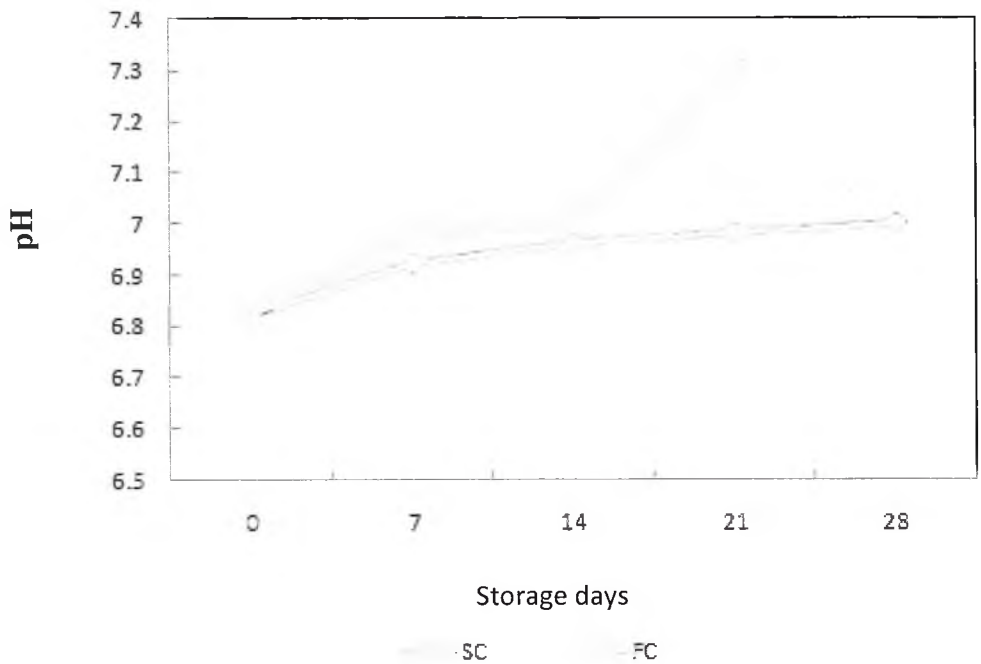


Fig : 11 Changes in pH of fish ham stored at refrigerated temperature (6 ± 2 ^{0}C)

SC : Synthetic casing **FC :** Fibrous casing

Table 16 : Changes in Total volatile base nitrogen (TVB-N) content of fish ham stored at refrigerated temperature ($6 \pm 2\ ^0C$)

Storage days	TVB-N (mg / 100 g)	
	S C	F C
0	1.16 (± 0.14)	1.50 (± 0.74)
7	14.2 (± 0. 42)	16.60 (± 0.42)
14	15.6 (± 1.04)	18.02 (± 2.58)
21	20.64 (± 1.84)	28.64 (± 0.18)
28	26.44 (± 1.73)	*
35	*	

Note : * : Spoiled **SC** : Synthetic casing **FC :** Fibrous casing

( ) : Values in parenthesis indicate standard deviation, n = 3.

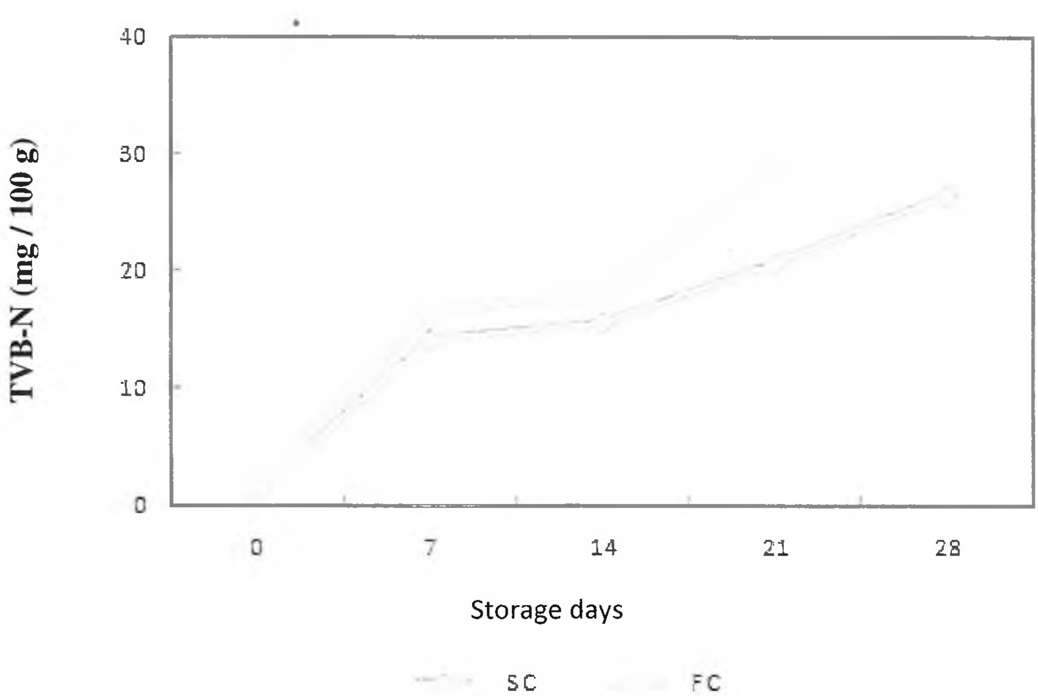


Fig : 12 Changes in Total volatile base nitrogen (TVB-N) content of fish ham stored at refrigerated temperature ($6 \pm 2\ ^0C$)

SC : Synthetic casing **FC :** Fibrous casing

Table 17 : Changes in Trimethylamine nitrogen (TMA–N) content of fish ham stored at refrigerated temperature (6 ± 2 ^{0}C)

Storage days	TMA-N (mg / 100 g)	
	S C	F C
0	2.40 (± 0.34)	2.80 (± 0.36)
7	2.80 (± 0.84)	3.92 (± 0.22)
14	3.64 (± 0.18)	4.48 (± 0.41)
21	4.40 (± 1.72)	9.80 (± 0.14)
28	6.01 (± 0.12)	*
35	*	

Note : * : Spoiled **SC :** Synthetic casing **FC :** Fibrous casing

( ) : Values in parenthesis indicate standard deviation, n = 3.

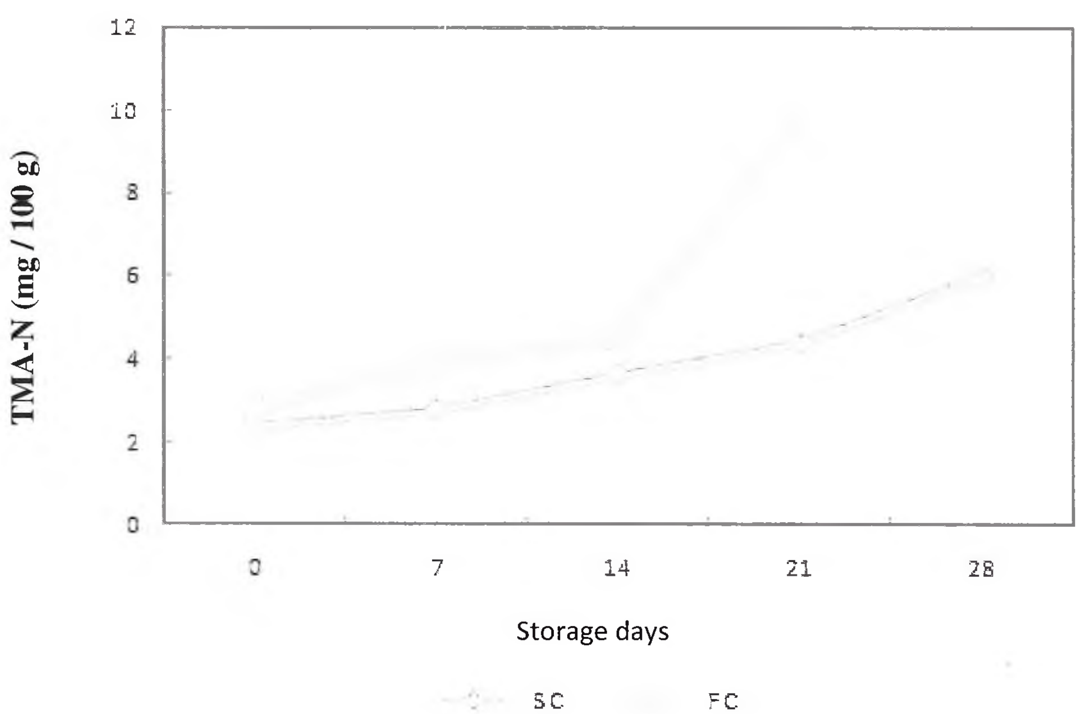


Fig : 13 Changes in Trimethylamine nitrogen (TMA–N) content of fish ham stored refrigerated temperature (6 ± 2 ^{0}C)

SC : Synthetic casing **FC :** Fibrous casing

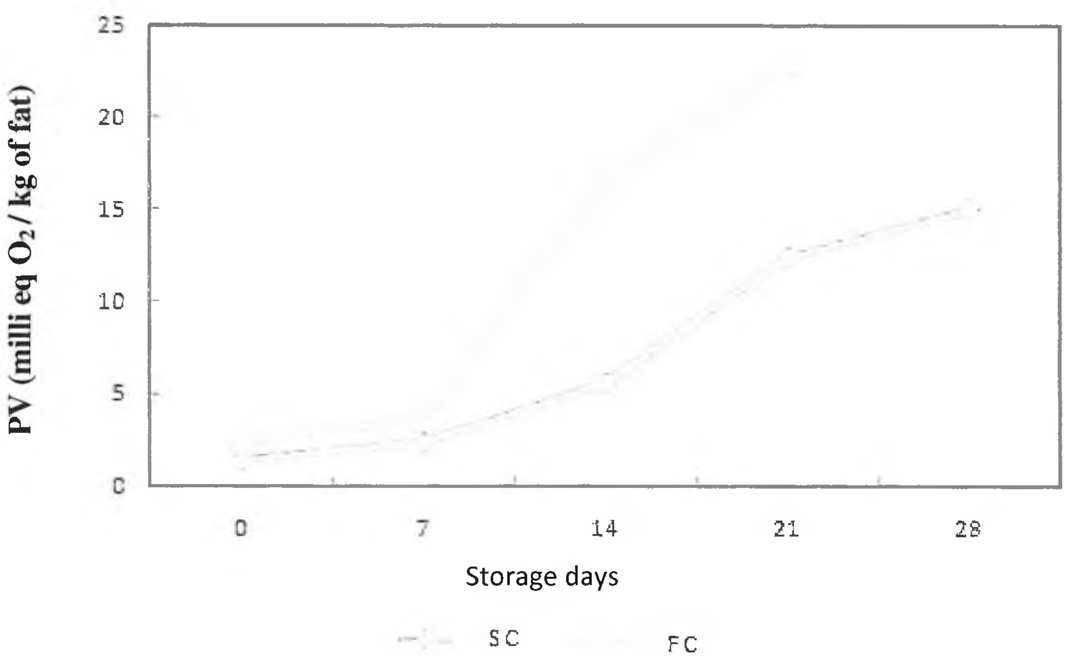


Fig : 14 Changes in Peroxide value of fish ham stored at refrigerated temperature (6 ± 2 ^{0}C)

SC : Synthetic casing **FC :** Fibrous casing

Table 19 : Changes in Free fatty acid of fish ham stored at refrigerated temperature ($6 \pm 2\ ^0C$)

Storage days	FFA (% of Oleic acid)	
	S C	**F C**
0	2.39 (± 0.36)	2.90 (± 0.94)
7	1.98 (± 0.16)	4.23 (± 0.20)
14	3.16 (± 0.12)	5.33 (± 1.02)
21	4.75 (± 1.12)	8.05 (± 1.12)
28	7.88 (± 0.92)	*
35	*	

Note : * : Spoiled **SC :** Synthetic casing **FC :** Fibrous casing

( ) : Values in parenthesis indicate standard deviation, n = 3.

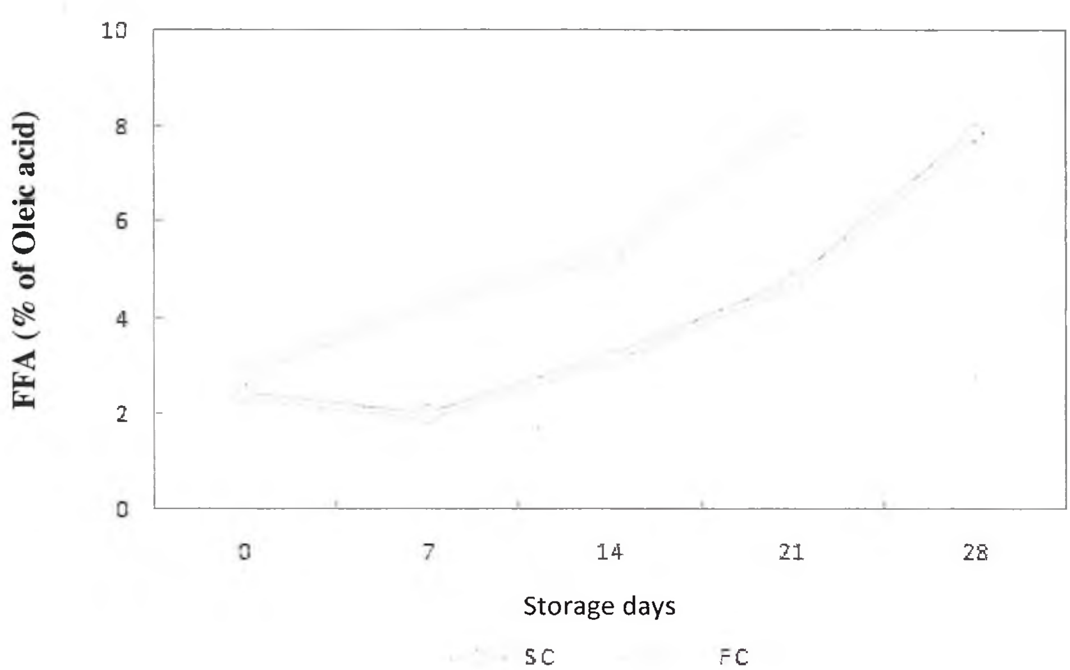


Fig : 15 Changes in Free fatty acid of fish ham stored at refrigerated temperature (6 ± 2 ^{0}C)

SC : Synthetic casing **FC :** Fibrous casing

Table 20 : Changes in TBA value of fish ham stored at refrigerated temperature ($6 \pm 2\ ^0C$)

Storage days	TBA (mg malonaldehyde / kg meat)	
	S C	F C
0	0.11 (± 0.12)	0.21 (± 0.10)
7	1.18 (± 0.04)	1.13 (± 0.80)
14	1.24 (± 1.02)	2.14 (± 0.14)
21	1.60 (± 0.06)	3.94 (± 0.92)
28	1.97 (± 1.12)	*
35	*	

Note : * : Spoiled **SC :** Synthetic casing **FC :** Fibrous casing

( ) : Values in parenthesis indicate standard deviation, n = 3.

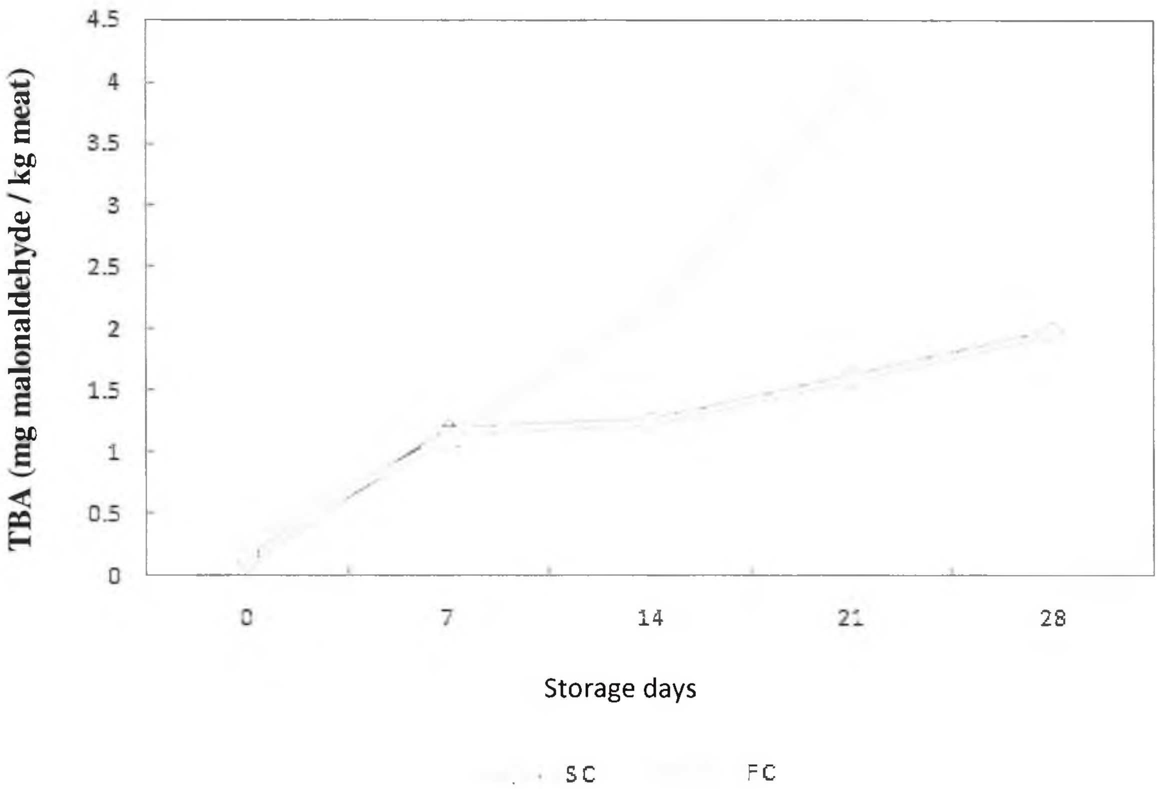


Fig : 16 Changes in TBA value of fish ham stored at refrigerated temperature (6 ± 2 ^{0}C)

SC : Synthetic casing **FC :** Fibrous casing

Table 21 : Changes in microbiological quality characteristics of fish ham stored at refrigerated temperature ($6 \pm 2\ ^0C$)

Storage days	TPC (c f u /g)		Spores (No / g)	
	S C	F C	S C	F C
0	1.15×10^1 (1.24)	1.35×10^1 (1.48)	0.12×10^1 (0.28)	0.94×10^1 (0.40)
7	2.1×10^2 (1.84)	7.0×10^2 (2.62)	1.20×10^2 (2.64)	8.5×10^2 (2.50)
14	1.65×10^3 (2.13)	3.9×10^6 (3.43)	2.58×10^2 (8.20)	4.5×10^2 (2.10)
21	4.5×10^3 (2.20)	4.5×10^6 (2.94)	1.85×10^3 (4.30)	6.4×10^3 (3.14)
28	2.9×10^4 (2.20)	*	2.5×10^3 (4.06)	*
35	*		*	

Note : * : Spoiled **SC :** Synthetic casing **FC :** Fibrous casing

( ) : Values in parenthesis indicate standard deviation, n = 3.

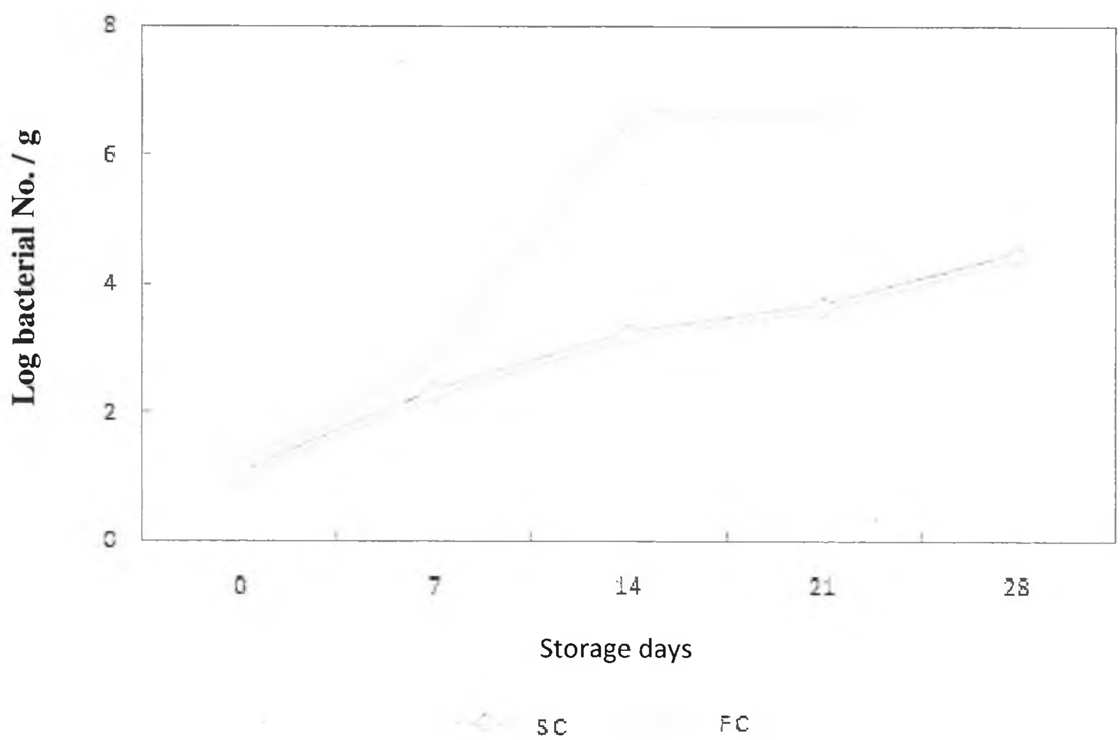


Fig : 17 Changes in microbiological quality characteristics of fish ham stored at refrigerated temperature (6 ± 2 ^{0}C)

SC : Synthetic casing **FC :** Fibrous casing

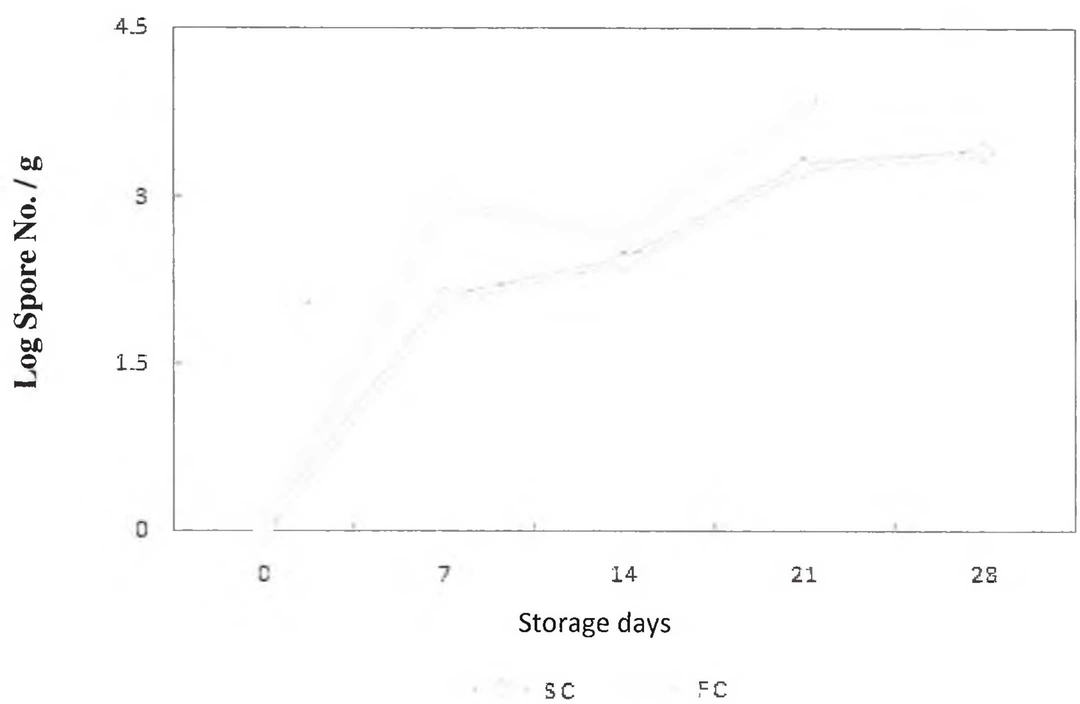


Fig : 18 Changes in microbiological quality characteristics of fish ham stored at refrigerated temperature (6 ± 2 ^{0}C)

SC : Synthetic casing **FC :** Fibrous casing

Table 22 : Changes in Organoleptic quality characteristics of fish ham stored at refrigerated tempera

Attributes / Storage days	Appearance		Colour		Odour		Texture		Taste	
	S C	**F C**	**S C**	**F C**	**S C**	**F C**	**S C**	**F C**	**S C**	**F C**
0	8.00	7.70	8.00	7.90	8.13	8.13	8.06	8.06	7.80	7.94
7	8.00	8.10	8.00	8.10	8.10	7.60	7.90	7.50	7.80	7.30
14	7.90	7.80	8.00	7.50	7.70	7.40	7.90	6.90	8.20	7.50
21	6.40	5.70	6.40	5.90	6.10	5.80	5.90	5.10	5.70	5.20
28	5.50	*	5.20	*	5.40	*	5.00	*	4.00	*
35	*		*		*		*		*	

Note : * : Spoiled   **SC :** Synthetic casing   **FC :** Fibrous casing

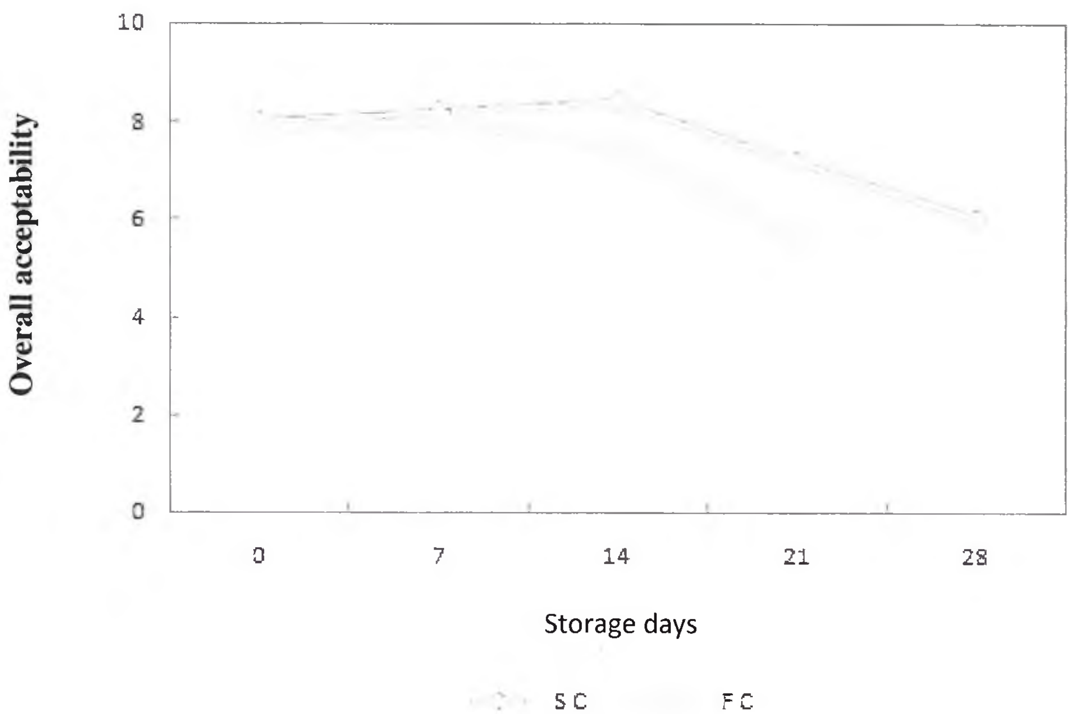


Fig :19 Changes in Organoleptic quality characteristics of fish ham stored at refrigerated temperature (6 ± 2 ^{0}C)

SC : Synthetic casing **FC :** Fibrous casing

ham. Similarly the spore counts were increased in fibrous casing to 6.4×10^3 No./g at the end of 21 days and 2.5×10^3 No./g on 28 days for synthetic casing fish ham. (Table 21 and Fig. 17 and 18).

4.3.8 Sensory changes of fish ham stored at refrigerated temperature (6 ± 2 °C)

The mean organoleptic scores obtained for appearance, colour, odour, taste, texture and overall acceptability of fish hams stored at refrigerated temperature for different durations are presented in Table 22 and Fig 19. The overall acceptability score is the mean of the attributes such as colour, texture, appearance, flavor and taste. The overall acceptability scores of fish ham in both synthetic and fibrous casing stored at refrigerated temperature was observed to be 5.22 on 28^{th}day and 5.48 on 21^{st} day respectively indicating slight quality deterioration of fish ham in fibrous casing when compared to synthetic casing ham.

4.4 Changes in chemical quality characteristics of fish ham during frozen storage temperature at (-18 ± 2 °C)

4.4.1 pH

Fresh fish hams in synthetic casing and fibrous casing have reported a pH value of 6.82. The changes in pH in both type of products stored at frozen temperature were very marginal, which is presented in the Table 24 and Fig. 20. pH increased from its initial value of 6.82 to 6.90 and 6.98 at the end of 90 days for both synthetic casing and fibrous casing respectively.

4.4.2 Total volatile base nitrogen (TVB-N)

The changes in TVB-N values of fish ham in fibrous casing, during its storage at frozen temperature has gradually increased to 12.28 mg % after 90 days, from 1.16

mg % recorded on 0^{th} day. TVB-N content of 12.94 mg % was recorded on 90^{th} fibrous casing fish ham is presented in the Table 25 and Fig. 21.

4.4.3 Trimethylamine nitrogen (TMA-N)

The TMA-N content for both the product were 2.40 and 2.80 mg % in the beginning but gradually increased to 15.90 mg % for synthetic casing at the end of 90 days and 17.19 mg % on 90^{th} day for fibrous casing ham (Table 26 and Fig. 22).

4.4.4 Peroxide value (PV)

The peroxide value content of both the ham in synthetic casing and fibrous casing stored at frozen temperature showed increase with the increase of storage period. The PV has increased fish ham 1.38 to 15.16 milli eq. of O_2 / kg of fat on 0^{th} and 90^{th} day for synthetic casing, whereas peroxide value for fibrous casing fish ham also increased from 2.45 to 16.57 milli eq. of O_2 / kg of fat on 0^{th} and 90^{th} day respectively. But at the end of 90 days the ham in fibrous casing was observed to be spoiled, which is presented in the Table 27 and Fig. 23.

4.4.5 Free fatty acid (FFA)

The free fatty acid content of fish ham packed in synthetic casing stored at frozen temperature showed increasing trend with increase of storage period from 2.39 to 6.02 % as oleic acid on 90^{th} day. The free fatty acid in fibrous casing fish ham also increased from 2.90 to 6.29 as oleic acid % on 90^{th} day (Table 28 and Fig. 24).

4.4.6 Thiobarbituric acid (TBA)

The TBA content of fish ham which is presented in the Table 29 and Fig. 25. Synthetic casing fish ham stored at frozen condition showed a gradual increase in its content with increase in storage days from 1.15 on 30th day to 2.09 on 90th day mg malanoaldehyde / kg of meat, whereas TBA in fibrous casing fish ham also increased from 2.54 to 2.86 mg maloaldehyde / kg of meat at the end of 60th and 90th day indicating the occurrence of oxidative spoilage at the end of 60 days.

4.4.7 Microbiological quality changes in fish ham stored at frozen temperature (-18 ± 2 °C)

The initial TPC of fish ham soon after processing was found to be 1.35×10^1 and 1.15×10^1 cfu /g in products packed in fibrous casing and synthetic casing respectively. However the spore counts were found to be very low. Microbiological counts of both the samples at -18 ± 2 °C increased gradually during its storage period to 7.0×10^4 cfu /g fibrous casing fish ham and 2.9×10^3 cfu /g on 90th day in synthetic casing fish ham. Similarly the spore counts were increased in fibrous casing fish ham to 2.6×10^2 No. /g at the end of 90 days and 1.8×10^2 No./ g on 90th day for synthetic casing fish ham, which is presented in the Table 30 and Fig. 26 and 27.

4.4.8 Sensory quality changes of fish ham stored at frozen temperature (-18 ± 2 °C)

The mean organoleptic scores obtained for appearance, colour, odour, taste, texture and overall acceptability of fish hams stored at frozen temperature for different durations are presented in Table 31 and Fig. 28. The overall acceptability score is the mean of the attributes such as colour, texture, appearance, flavour and taste. The overall acceptability scores of fish ham in both synthetic and fibrous casing stored at frozen temperature was observed to be 5.0 on and 4.0 at the end of 90 day respectively from its initial values of 8.0 and 7.9 indicating faster quality deterioration of fish ham in fibrous casing when compared to synthetic casing fish ham.

Table 23 : Changes in proximate composition of fish ham stored at frozen temperature (-18 ± 2 ^{0}C)

Storage days / Components	0		30		60	
	S C	**F C**	**S C**	**F C**	**S C**	**F C**
Moisture (%)	71.39 (± 0.14)	67.63 (± 0.06)	69.23 (± 0.08)	67.56 (± 0.14)	68.94 (± 0.09)	66.55 (± 0.1
Protein (%)	18.15 (± 0.18)	18.15 (± 0.24)	17.50 (± 0.16)	18.80 (± 0.06)	16.40 (± 1.40)	19.68 (± 1.1
Crude fat (%)	5.66 (± 0.12)	5.33 (± 0.08)	3.35 (± 0.14)	4.03 (± 1.14)	2.62 (± 0.06)	2.42 (± 0.6
Ash (%)	2.50 (± 0.04)	2.21 (± 1.01)	2.29 (± 0.16)	2.12 (± 0.14)	2.37 (± 0.15)	2.27 (± 0.9

Note : ( ) : Values in parenthesis indicate standard deviation n = 3.

SC : Synthetic casing   FC : Fibrous casing

Table 24 : Changes in pH of fish ham stored at frozen temperature (-18 ± 2 ^{0}C)

Storage days	pH	
	S C	**F C**
0	6.82 (± 0.18)	6.82 (± 0.04)
30	6.70 (± 0.18)	6.76 (± 0. 20)
60	6.80 (± 1.14)	6.64 (± 0.08)
90	6.90 (± 0.04)	6.98 (± 1.14)

Note : * : Spoiled **SC :** Synthetic casing **FC :** Fibrous casing

( ) : Values in parenthesis indicate standard deviation, n = 3.

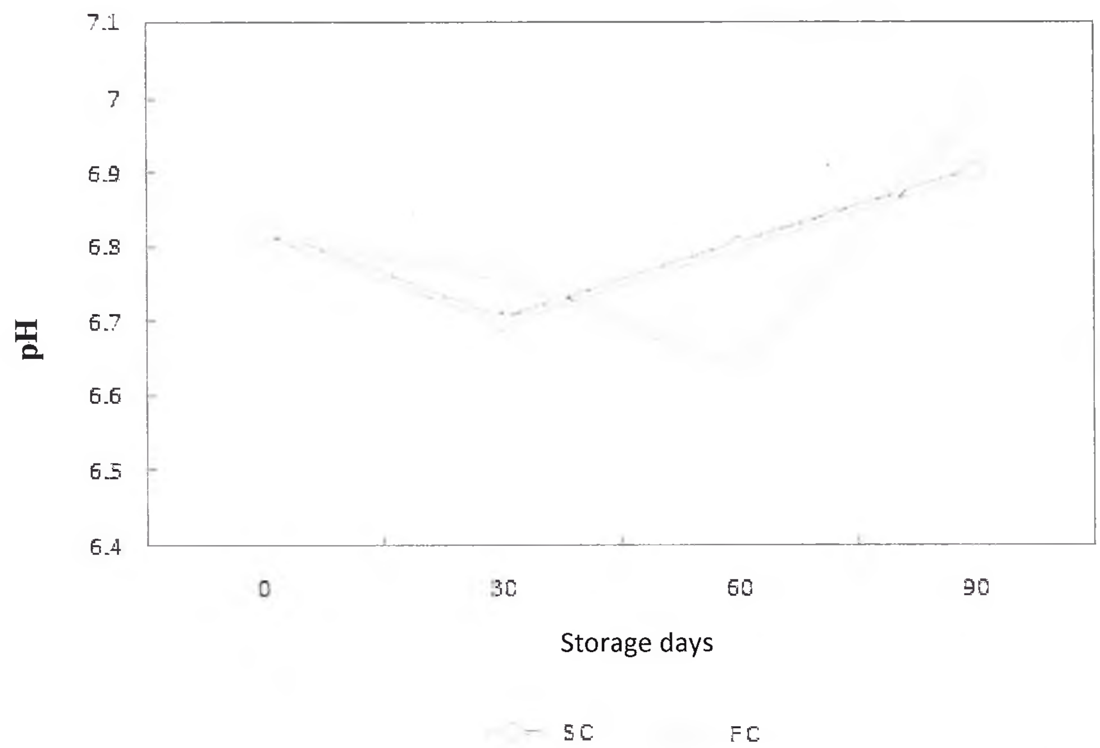


Fig : 20 Changes in pH of fish ham stored at frozen temperature (- 18 ± 2 ^{0}C)

SC : Synthetic casing **FC :** Fibrous casing

Table 25 : Changes in Total volatile base nitrogen (TVB-N) content of fish ham stored at frozen temperature (-18 ± 2 ^{0}C)

Storage days	**TVB-N (mg / 100 g)**	
	S C	**F C**
0	1.16 (± 0.14)	1.50 (± 0.74)
30	6.80 (± 0.94)	6.20 (± 1.14)
60	9.28 (± 1.54)	9.80 (± 2.14)
90	12.28 (± 0.18)	12.94 (± 0.18)

Note : * : Spoiled **SC :** Synthetic casing **FC :** Fibrous casing

( ) : Values in parenthesis indicate standard deviation, n = 3.

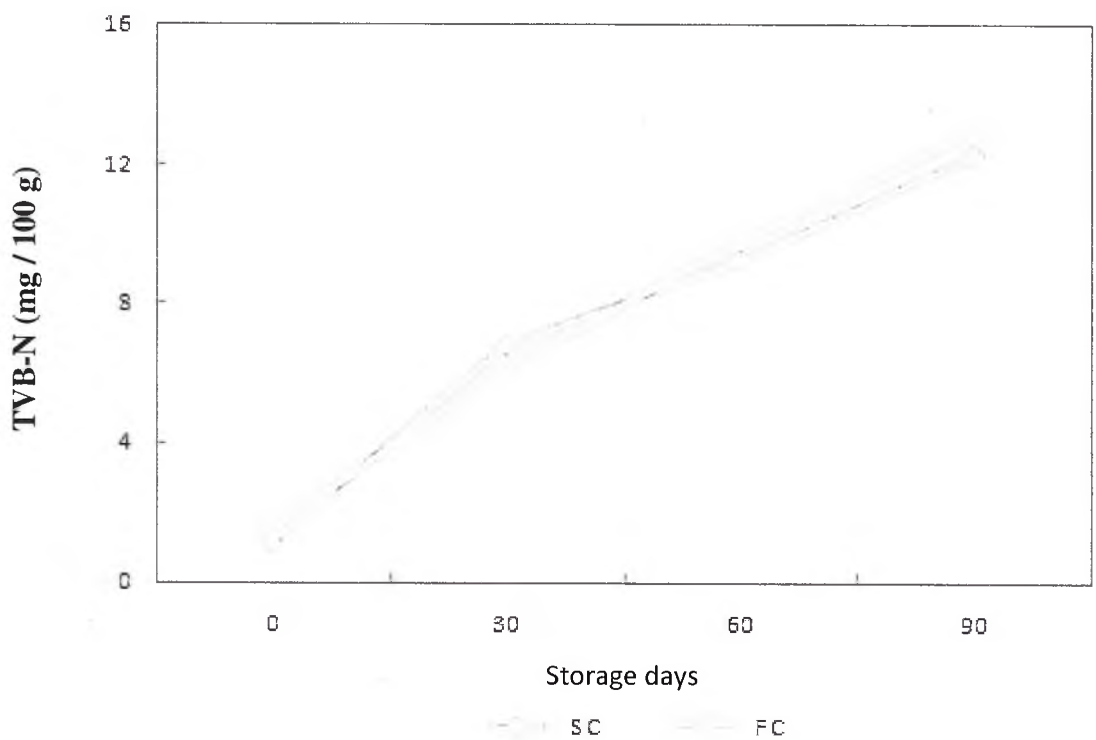


Fig : 21 Changes in Total volatile base nitrogen (TVB-N) content of fish ham stored at frozen temperature (- 18 ± 2 ^{0}C)

SC : Synthetic casing **FC :** Fibrous casing

Table 26 : Changes in Trimethylamine nitrogen (TMA-N) content of fish ham stored at frozen temperature (-18 ± 2 ^{0}C)

Storage days	**TMA-N (mg / 100 g)**	
	S C	**F C**
0	2.40 (± 0.34)	2.80 (± 0.36)
30	6.96 (± 1.20)	6.80 (± 0.08)
60	14.20 (± 0.60)	15.60 (± 1.02)
90	15.90 (± 0.11)	17.19 (± 1.06)

Note : * : Spoiled **SC** : Synthetic casing **FC** : Fibrous casing

( ) : Values in parenthesis indicate standard deviation, n = 3.

Table 27 : Changes in Peroxide value of fish ham stored at frozen temperature ($-18 \pm 2\ ^0C$)

Storage days	PV (milli eq O_2 / kg of fat)	
	S C	F C
0	1.38 (± 0.08)	2.45 (± 0.09)
30	8.64 (± 0.06)	6.23 (± 0.54)
60	12.38 (± 0.32)	12.76 (± 1.13)
90	15.16 (± 0.02)	16.57 (± 0.54)

Note : * : Spoiled **SC** : Synthetic casing **FC** : Fibrous casing

( ) : Values in parenthesis indicate standard deviation, n = 3.

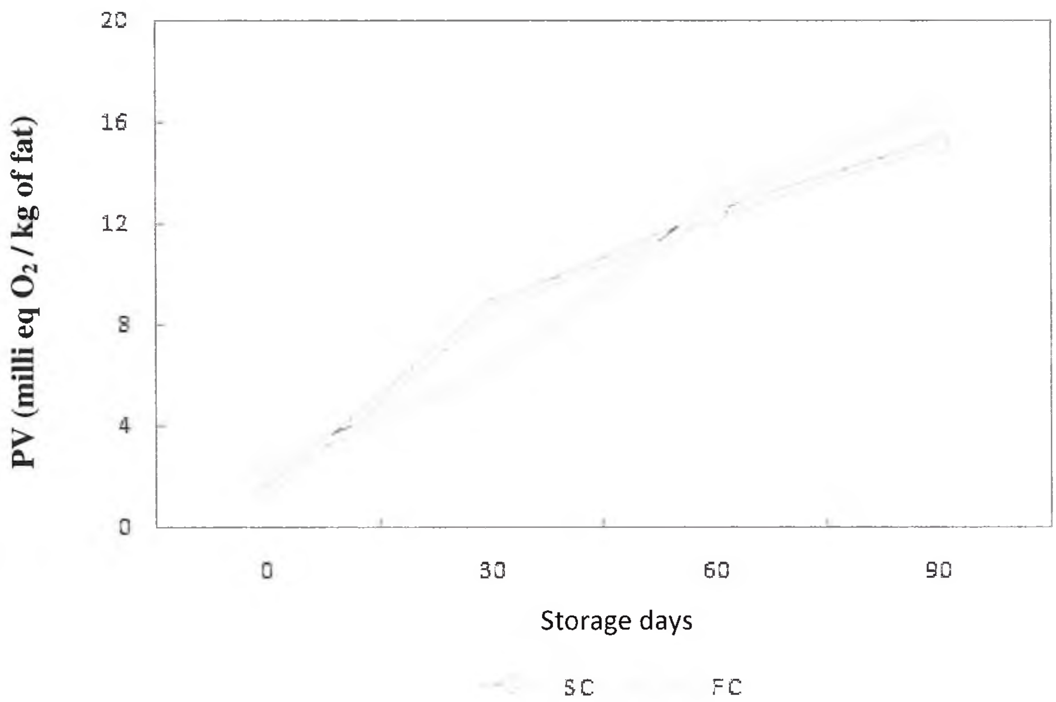


Fig : 23 Changes in Peroxide value of fish ham stored at frozen temperature (- 18 ± 2 ^{0}C)

SC : Synthetic casing **FC :** Fibrous casing

Table 28 : Changes in Free fatty acid of fish ham stored at frozen temperature ($-18 \pm 2\ ^0C$)

Storage days	FFA (% of Oleic acid)	
	S C	F C
0	2.39 (± 0.36)	2.90 (± 0.94)
30	3.78 (± 1.20)	4.88 (± 1.14)
60	4.91 (± 1.14)	5.62 (± 0.38)
90	6.02 (± 0.09)	6.29 (± 0.06)

Note : * : Spoiled **SC :** Synthetic casing **FC :** Fibrous casing

( ) : Values in parenthesis indicate standard deviation, n = 3.

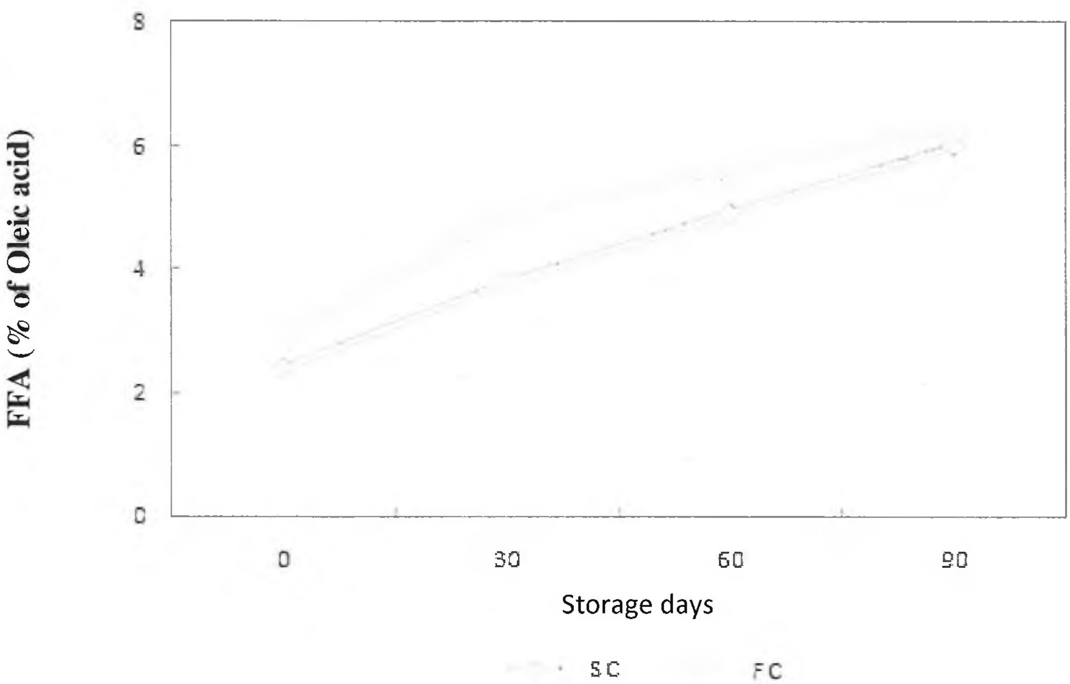


Fig : 24 Changes in Free fatty acid of fish ham stored at frozen temperature (- 18 ± 2 ^{0}C)

SC : Synthetic casing **FC :** Fibrous casing

Table 29 : Changes in TBA value of fish ham stored at frozen temperature (-18 ± 2 ^{0}C)

Storage days	TBA (mg malonaldehyde / kg meat)	
	S C	F C
0	0.11 (± 0.12)	0.21 (± 0.10)
30	1.15 (± 0.13)	1.60 (± 0.02)
60	1.55 (± 0.04)	2.54 (± 0.14)
90	2.29 (± 0.03)	2.86 (± 0.07)

Note : * : Spoiled **SC** : Synthetic casing **FC** : Fibrous casing

( ) : Values in parenthesis indicate standard deviation, n = 3.

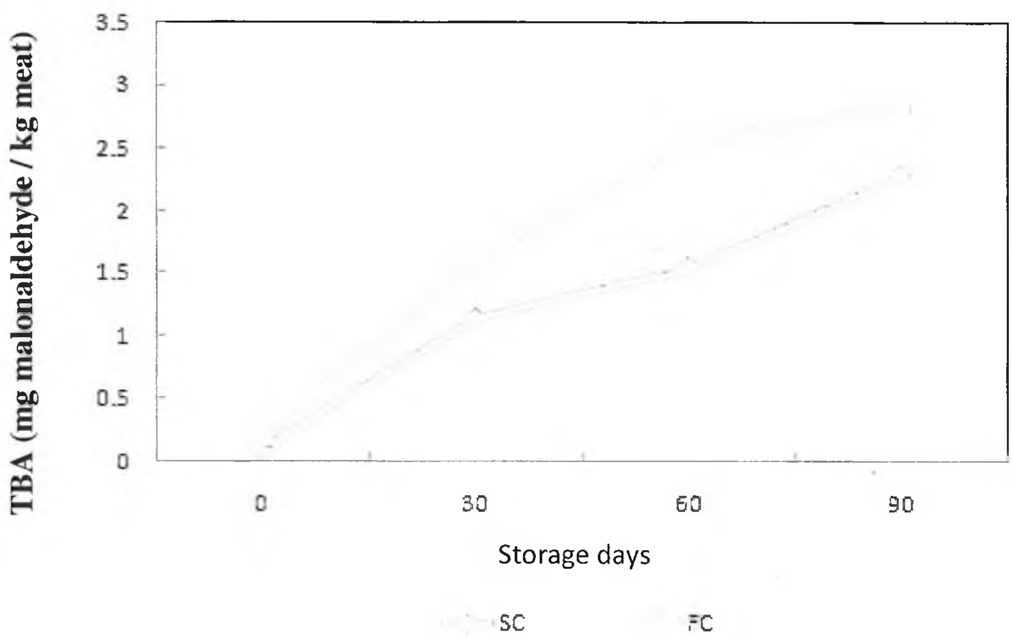


Fig : 25 Changes in TBA value of fish ham stored at frozen temperature (- 18 ± 2 ^{0}C)

SC : Synthetic casing **FC :** Fibrous casing

Table 30 : Changes in microbiological quality characteristics of fish ham stored at frozen temperature (-18 ± 2 ^{0}C)

Storage days	TPC (c f u /g)		Spores (No / g)	
	S C	F C	S C	F C
0	1.15×10^1 (1.24)	1.35×10^1 (1.48)	0.12×10^1 (0.28)	0.94×10^1 (0.40)
30	1.5×10^1 (2.54)	1.92×10^1 (3.10)	0.94×10^1 (1.14)	0.84×10^1 (2.10)
60	1.5×10^2 (2.82)	4×10^2 (3.10)	1.5×10^2 (2.40)	1.0×10^2 (2.10)
90	2.9×10^3 (2.94)	7.0×10^4 (0.34)	1.8×10^2 (4.70)	2.6×10^2 (2.94)

Note : ***** **:** Spoiled **SC :** Synthetic casing **FC :** Fibrous casing

( ) : Values in parenthesis indicate standard deviation, n = 3.

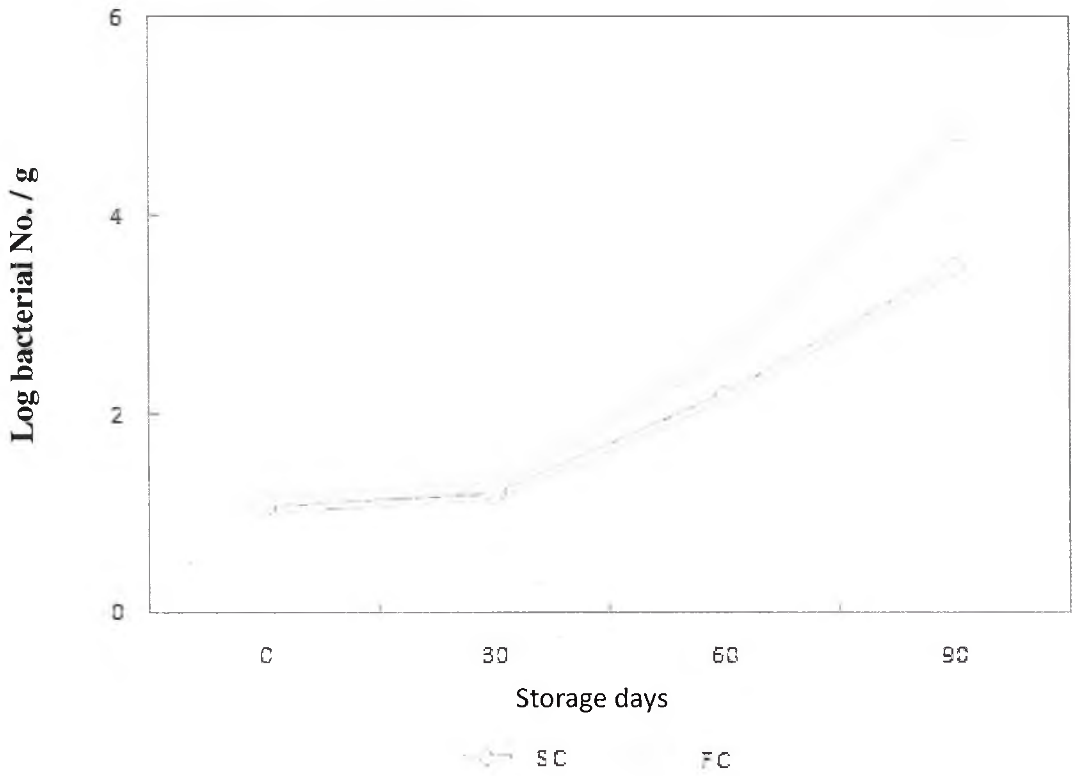


Fig : 26 Changes in microbiological quality characteristics of fish ham stored at frozen temperature (- 18 ± 2 ^{0}C)

SC : Synthetic casing **FC :** Fibrous casing

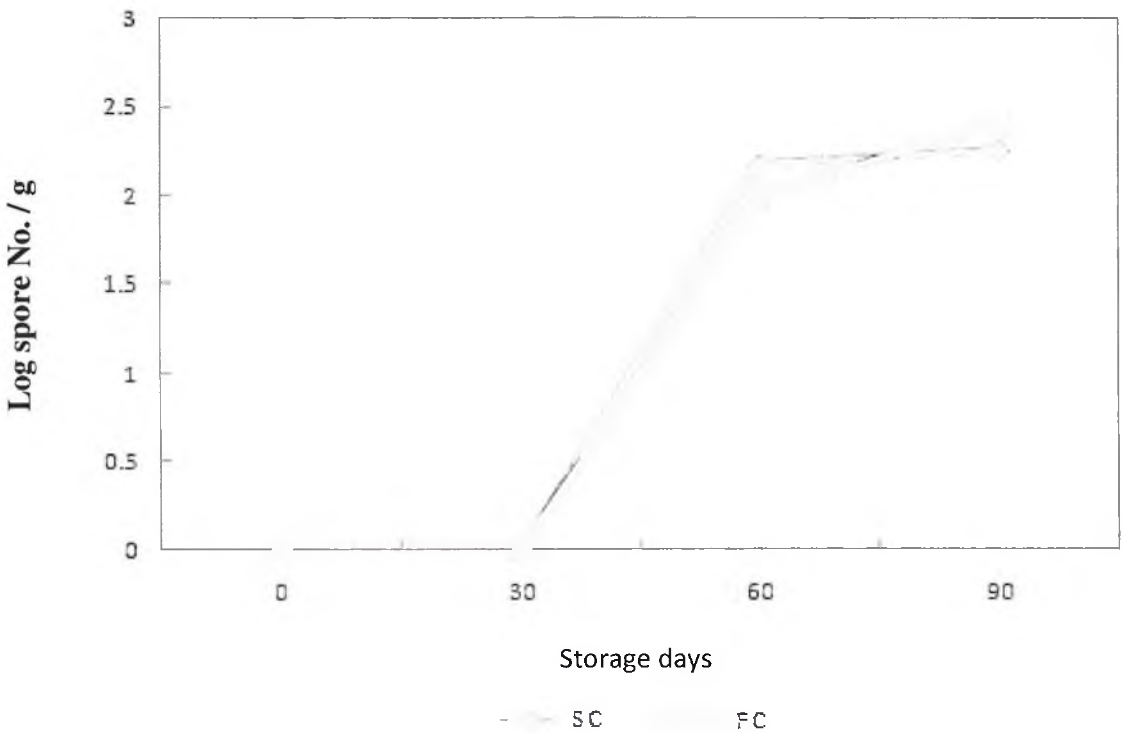


Fig : 27 Changes in microbiological quality characteristics of fish ham stored at frozen temperature (- 18 ± 2 ^{0}C)

SC : Synthetic casing **FC :** Fibrous casing

Table 31 : Changes in Organoleptic quality characteristics of fish ham stored at frozen temperature (-18 ± 2 °C

Attributes / Storage days	Appearance		Colour		Odour		Texture		Taste		Fl
	S C	F C	S C	F C	S C	F C	S C	F C	S C	F C	S C
0	8.00	7.70	8.00	7.90	8.13	8.13	8.06	8.06	7.80	7.94	8.00
30	8.80	8.30	8.40	8.30	8.30	8.10	8.20	7.70	8.40	8.10	8.00
60	7.50	6.70	7.00	7.20	7.30	6.90	7.50	7.00	7.50	6.60	7.60
90	5.90	4.00	4.50	4.00	5.00	4.00	4.00	3.50	4.50	3.40	4.00

Note : **SC** : Synthetic casing **FC** : Fibrous casing

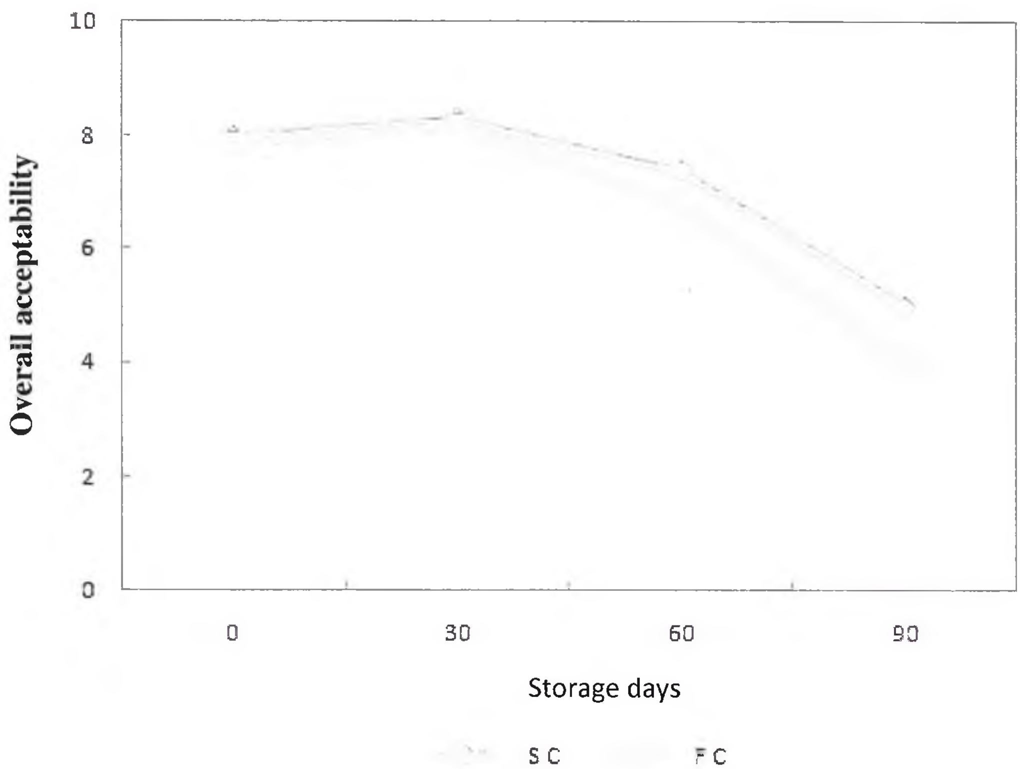


Fig : 28 Changes in Organoleptic quality characteristics of fish ham stored at frozen temperature (- 18 ± 2 ^{0}C)

SC : Synthetic casing **FC :** Fibrous casing

Discussion

V. DISCUSSION

Fish ham is one of the important popular paste products that is produced on commercial scale in Japan for the past several years (Raju, 1999). The key to the success of this processed product was due to the development of technology of fish ham from tuna meat by According to Tanikawa (1971), fish ham and sausages are popularly produced not only in Japan, but also in other countries because of their special taste and flavour.

5.1 Raw material characteristics

The Bull's eye is a deep sea fish locally called as "Disco fish" is an unfamiliar variety and hence, it is unfamiliar and underutilised. Among the fresh resources in deep water off the west coast *Priacanthus hamrur* forms about 40 % of harvested catch. At present this fish fetch very low value in the fresh fish market. The quality of the final product, its quality during storage and storage stability depends on mainly raw material (Bull's eye) and shrimp characteristics. It is very much essential to define the raw material characteristics used in the present study. A brief discussion on the important physical, chemical, microbial and sensory characteristics of Bull's eye (*Priacanthus hamrur*) used in the current investigation has been presented in the following sections.

5.1.1 Physical characteristics

The Bull's eye is characterised by its thick skin, big eyes with high flesh content, hence, used for the preparation of minced meat and surimi. Physical characteristics namely total length and standard length in centimetres, total weight in grams, dressing yield, picked meat yield and minced meat yield as percentage of Bull's eye (*Priacanthus hamrur*) are presented in Table 2.

The fish used in the present study has an average total length of 18.96 cm and an average weight of 84.35 g. Standard length of the fish was found to be 15.59 cm.

According to the experiment conducted by Siddappa (2002), an average total length of 18.6 cm, average weight of 83.4 g and standard length of fish was found to be 15.54 cm. Bull's eye (*P. hamrur*) used by Dhananjaya *et al.* (1984), Murthy (1999) and (Morey, 2003) have used the fish having a mean total length of 18.2 cm, 22.02 cm and 18.19 cm and mean weight of 76.9, 132.6 g and 87.9 g respectively. The fish used by Bhatta (2001), was bigger in size. The mean total length of 23.3 cm, standard length of 18.15 cm and average weight of 164.46 g has been reported by Bhatta (2001) for *P. hamrur.* Mahesh (2002) had also used *P hamrur* of 18.03 cm mean total length, 14.42 cm standard length and 84.35 g mean weight in his studies. Based on growth, mortality and stock assessment studies, Chakraborty and Vidyasagar (1996) have reported that *P. hamrur* attained the length of 171 mm, 260 mm, 308 mm and 334 mm at the end of 1st, 2nd, 3rd and 4th year respectively. Chakraborti (2002) has reported that the weight range of the same species was about 100 - 250 g. Hence, it can be concluded that the fish used in the present experiment belong to within second year of its growth.

The dressing yield percentage, after beheading, eviscerating and descaling was found to be 55.85 %, picked meat yield was found to be 32.42 % and minced meat yield percentage 30.84 % was reported (Table 2). Murthy (1999), Siddappa (2002) and Morey (2003) have recorded 57 %, 67 % and 68.94 % for dressing yield, 34 % , 40.30 % and 42.43 % for picked meat yield and about 29 %, 32.50 % and 27.83 % for minced meat yield. The difference between picked meat to minced meat yield may be attributed to small bones and small amount of meat remaining in shrimp commonly called red rings (*Aristeus alcocki*) as 37.3 % and 19.5 % respectively. Pandian (1994) has reported that the peeled and deveined yield percentage of 47.62 % for marine species (*Parapenaeopsis stylifera*). About 5 to 13 % higher yield in the present study may be due to small scale of processing and variation in size grades of shrimp.

5.2 Chemical characteristics

5.2.1 Proximate composition

The proximate composition of Bulls eye (*P. hamrur*) used in the present study is presented in Table 2. The fish used contained 77.80 % moisture, 18.53 % protein, 1.45 % ash and 1.38 % of total lipid. The proximate composition of shrimp (Table 3) indicate that the protein, ash, fat and moisture content of 16.40 %, 1.56 %, 1.12 % and 81.40 % respectively. Morey (2003) has reported the mean values for moisture, protein, fat and ash are 78.30 %, 19.32 %, 1.34 % and 1.44 % respectively. The proximate composition Table indicate that both fish and shrimp have low fat content, hence belong to lean variety of fish.

Dhananjaya *et al.* (1984), Murthy (1999), Bhatta (2001) and Siddappa (2002) have reported a protein content of 17.54 %, 18.60 % 19.30 % and 18.87 % respectively. The fat content of *P. hamrur* was found to be 5.08 % (Dhananjaya *et al.*, 1984), 1.38 % (Murthy, 1999) 1.28 % (Bhatta, 2001) and 1.40 % (Siddappa, 2002). The ash content of 1.20 and 1.50 % have been reported by Murthy (1999) and Bhatta (2001) respectively. According to Mahesh (2002) *P hamrur* contain 76.66 % moisture, 19.78 % protein, 1.38 % fat, and 1.44 % ash content. Moisture and fat content of the same species was found to be 78 .3 % and 1.4 % respectively (Chakraborti, 2002). Zaitsev *et al.*(1969) and Connell (1975) have reports that the proximate composition of fish depends on several factors like season, sex, habitat, environmental condition food and feeding habit. The proximate composition of shrimp is found similar to the values reported by Krishnamurthy (1983a). Pandian (1994) and Thimmappa (2002) have reported that the fresh *A. alcocki* and *P stylifera* contained 81.18 and 87.45 % moisture, 17.14 and 11.68 % protein, 0.77 and 0.46 % total lipids and 0.90 and 0.30 % ash respectively. The proximate composition of shrimp used in the study generally compares well with the range reported by Govindan (1974), Wheaton and Lawson (1985), Pandian (1994) and Thimmappa (2002). The results obtained in the present study with regard to proximate composition is in close agreement with the results of Bhatta (2001) and Siddappa (2002).

5.2.1.1 Other chemical characteristics.

The results of the biochemical parameters such as pH, total volatile base nitrogen, (TVB-N), trimethylamine nitrogen (TMA-N), peroxide value (PV), Free fatty acid (FFA) and thiobarbituric acid (TBA) of the lipid fraction of the fish shrimp and minced meat are presented in Table 2,

The Bull's eye and shrimp used in the present experiment was found to have a pH value of 6.84, and 7.18 respectively. Siddappa (2002), have reported pH of Bull's eye has 6.80 and for fresh Bull's eye. Krishnamurthy (1983a), has reported 6.4 to 6.5 for fresh shrimp which increased during ice storage to 7.5. If the pH of shrimp rises to above 7.8 to 8.0, it indicate the spoilage (Krishnamurthy, 1983a).Volatile base nitrogen of 5.60 mg %, trimethylamine nitrogen of 3.36 mg %, peroxide value of 1.11 milli eq of O_2 per kg of fat, free fatty acid content of 0.48 % of oleic acid, thiobarbituric acid content of 1.98 mg of malonaldehyde / kg in total lipid was detected in minced meat. TVB - N and TMA - N were not detected in these values indicate that the quality of fish used in the present study was extremely high quality. In fresh disco fish Murthy (1999), Bhatta (2001), Siddappa (2002) and Mahesh (2002) have reported 7.40 mg %, 5.88 mg % 7.38 mg % and 7.46 mg % for TVB-N in fresh Bull's eye respectively. The same authors have also reported TMA-N content of 3.73 mg %, 2.15 mg %, 3.77 mg % and 2.80 mg % respectively in fresh Bull's eye used in their studies. Chakraborti (2002) reported a TVB-N content of 7.2 mg % in Bull's eye. According to Morey (2003) fresh *P. hamrur* had 12.30 mg % of TVB-N, 5.04 mg % TMA-N, 3.64 % of oleic acid for FFA, 12.2 milli eq per kg of fat for PV and pH value of 6.40. In spite of high values reported for TVB-N and TMA-N, they have considered the fish was of high quality. Connell (1975) has stated that TVB-N content of 30 - 40 mg % and TMA-N of 10 - 15 mg % in fresh fish can be considered as the limit for borderline for acceptance. According to Nair *et al.*(1971), the determination of TVB-N for assessment of freshness of fish was widely used, as there is good relation with sensory quality changes in fish. The TVB-N, TMA-N, PV, FFA and TBA content of 4.48 mg %, 2.80 mg %, 2.30 milli eq of O_2 / kg of fat, 1.05 % as oleic acid and 0.44 mg of melonaldehyde per kg of fat has been reported for the shrimp used for the preparation of fish ham. Pandian (1994) and Thimmappa (2002) have reported 9.86 and 12.60 mg % for TVB-N 0.23 and 3.50 mg % for TMA-N 3.14 and 1.24 % of oleic acid for FFA and 1.01 and 2.20 milli eq of O_2 / kg of fat for fresh shrimp respectively. TBA value of 0.023 and 0.003 mg. of malonaldehyde / kg of sample has been reported for *Metapenaeus monoceros* and *P stylifera* respectively by Pandian (1994).

The chemical quality characteristics as indicated by TVB-N and TMA-N content revealed that the shrimps were of prime quality. The higher values reported in the present study are well below the acceptable limit as suggested by many workers for TVB-N values of 30 - 40 mg /100 g and TMA-N content of 5 mg / 100g (Pillai, *et al.,* 1961, Connell, 1980 and Pandian 1994). The pH of fish meat and shrimp was found to be 6.84 and 7.18 respectively. According to Siddappa (2002) and Thimmappa (2002), the pH of fresh Bull's eye and shrimp was 6.80 and 7.40 respectively, Murthy (1999), Bhatta (2001) Mahesh (2002) and Siddappa (2002) have reported PV, content of 16.13, 15.21 17.38 and 15.87 milli eq of O_2 / kg of fat and free fatty acid content of 3.40 %, 3.51 % 2.93 % and 3.37 % of oleic acid. TBA value of 0.54 mg of malonaldehyde / kg of fat has been reported by Mahesh (2002) for fresh Bull's eye.

5.2.1.1.2 Microbiological characteristics of fish and shrimp

Fresh fish and shell fish naturally contain microorganisms in gills, gut, skin and through incidental contamination during post harvest handling and processing. As the fish and shrimp are stored, microbial load increases as the natural flora of fish and shrimp enters into the tissue and degrade it (Wheaton and Lawson,1985). Since microbes are most important agents of food spoilage, keeping the activities of tissue enzymes and the initial microbial load as low as possible are of primary importance. In the present study, total plate count was found to be 8.0×10^3 and 6.4×10^3 c f u / g and spore were not detectable. Bhatta (2001), Mahesh (2002) and Siddappa (2002) have reported 5.65×10^4, 2.48×10^4 and 3.3×10^4 c f u /g respectively in disco fish. According to Morey (2003), fresh fish had shown 4.0×10^4 c f u /g. The initial total plate counts in deep sea shrimp and *P. stylifera* were found to be 6.5×10^3 and 5.1×10^5 c f u /g of meat have been reported by Thimmappa (2002) and Pandian (1994). Almost similar TPC counts were reported by Pillai *et al.* (1961)

(10^4 to 10^7 / g); Khan, 1986 (9.8×10^5 to 7.9×10^6 /g); Bhobe and Pai, 1986 (2.6×10^4 /g).

The bacterial flora of shrimp are influenced by environmental factors and differ widely with species and season (Surendran, *et al.,* 1985). According to Wheaton and Lawson, (1985), Nirmala and Gopakumar (1991) are of the opinion that tropical shrimps carry gm $^{+ve}$ bacteria such as *micrococcus, Bacillus* etc. Where as in cold water species the *psychrophiles* predominate gm $^{-ve}$ such as *pseudomonas, flavobacterium* etc. In the present study initial low total plate count of fresh shrimp (peeled and deveined) may be attributed to better handling and hygienic practices and reduction of temperature by good iced condition.

5.2.1.1.3 Sensory characteristics of fish and shrimp

The organoleptic characteristic of raw materials decides the ultimate acceptability of the final product by the consumers. The mean organoleptic scores for fresh fish, and shrimp (uncooked and cooked) are presented in Table. 2. Based on the scores obtained for various attributes namely appearance, colour, texture, taste, odour, flavour and overall acceptability were found to vary between 8 (like very much) and 9 (like extremely), Bhatta, (2001) and Siddappa (2002) and Patel, (2003) have also reported similar quality attributes of Bull's eye used in their studies. Based on all the above experimental results, it can be confidently deduced that the fish and shrimp used in the fish ham preparation was of extremely good quality and highly acceptable in raw and cooked form by the panellists and scores correlates well with chemical and microbiological characteristics. Similar, organoleptic quality evaluation of raw and cooked shrimp was reported by Pandian (1994) and Thimmappa (2002).

5.3 Biochemical characteristics of fish ham

5.3.1 Changes in pH

pH is one of the important parameter used to assess the quality of fish and fishery products. In the present experiments changes in pH during storage at ambient, refrigerated and frozen storage these are presented in Table 6, 15, 24 and Fig 2, 11 and 20.

The initial pH of fish ham products was 6.82 which gradually decreased during storage at three different temperature. The pH of fish ham in synthetic casing decreased to 6.63 after 24 hrs and finally to 6.25 after 96 hrs (4 days) where as fish ham in fibrous casing the decrease in pH was faster and shown a pH of 6.10 after 72 hrs (on 3rd day). Many workers have reported a steady decrease in pH of fish sausage with increase in storage period (Hegde, 1989 and Srinivas,1998). A pH of 6.8 has been reported by Chandrashekhar *et al.*(1988) in corn flour incorporated fish sausage without any preservative. According to Okada and Takesu (1965), mild acidity would be favourable fish sausage from the point of prolonging the shelf life. The results of the present study compares well with reported value of pH 6.0 for fish ham containing shrimp chunks at room temperature.

It has been observed that the changes in pH in both type of products stored at refrigerated temperature were very marginal, pH increased from its initial value of 6.82 to 7.00 and 7.30 at the end of 14 and 21 days in fibrous casing ham, on the other hand pH has slowly raised to 7.00 after 28 days of storage in synthetic casing fish ham. Ueno (1968) and Desai *et al.*(1984) have reported gradual increase in pH during refrigerated storage. Similar results have also been reported by Cross,1984 and Chandrasekhar (1986). The increase of pH value might be attributed to the increase in volatile bases (e.g., ammonia and trimethylamine) produced by either endogenous or microbial enzymes (Manat *et al.*, 2005). Siddappa (2002) has reported an increase in pH of frozen chikuwa during cold storage and also in chikuwa made from frozen fish paste.

Sliced fish ham stored in cold storage at -18 ± 2 ^{0}C has also recorded a gradual increase in pH from 6.82 to 6.90 and 6.98 in synthetic casing and fibrous casing products respectively, over a storage period of 90 days. Srinivasa (1998), has reported gradual increase in pH of fish sausage during frozen storage. pH is suggested to be a good index of freshness. Generally crustacean muscle show a tendency to become alkaline rapidly during storage at lower temperature. Average slow drop in pH values in all the products at refrigerated and cold storage may be due to the production of nitrogenous compounds by microbial enzymes.

The results of the present study compare well with the result of other scientists report on different type of Japanese paste products.

5.3.2 Changes in total volatile base nitrogen (TVB-N)

The changes in TVB-N values of fish ham during ambient, refrigerated and frozen storage are presented in Table 7, 16, 25 and Fig 3, 12, 21.Total volatile base nitrogen (TVB-N), is mainly composed of ammonia and primary, secondary and tertiary amines (Beatty, 1938) resulted from degradation of proteins and non-protein nitrogenous compounds, which is chiefly caused by microbial activity (Ruiz - Capillas and Moral, 2005). TVB-N is widely used as an indicator for quality deterioration of meat (Olafsdottir, *et al.,* 1997). The results of change in volatile base nitrogen content in fish ham packed in fibrous casing during storage at different temperatures showed faster increase compared to fish ham in synthetic casing. The initial value of TVB - N in fibrous casing fish ham stored at room temperature was recorded as 1.50 mg %

which increased sharply to 41.84 mg % at the end of 3 days, whereas it was 41.40 mg % for synthetic casing fish ham at the end of 4 days.

The TVB-N values of fish ham in fibrous casing, during its storage at refrigerated temperature has gradually increased to 28.64 mg % after 21 days, from 16.60 mg % recorded on 7th days but on 28th day the product was spoiled completely. Although a TVB-N content of 26.44 mg % was reached on 28th day, the product was still in acceptable condition in synthetic casing fish ham. Kose *et al.* (2009) found that, TVB-N value for whiting burgers during refrigerated storage increased from the initial value of 2.02 mg % to maximum value of 42.03 mg %. A gradual increase was observed for both fibrous and synthetic casing fish ham from 1.50 and 1.16 mg % to 12.94 mg % and 12.28 respectively at the end of 90 days of frozen condition. Ueno (1968) has reported a TVB - N value of 10.0 mg % for freshly prepared sausage on initial day. On storage at 40 °C the values gradually increased to 18 mg % on 6th day. Suzuki (1981) also reported a TVB - N value of more than 30 mg % in the product, which was found acceptable. Chandrasekhar *et al.* (1988) have reported that the initial TVB-N value of 19.6 mg % in fish sausage prepared by mixing dried spice powder and oleoresins. However, sausages containing spice powder were found spoiled after 24 hrs of storage at ambient temperature. Joshi (1990), Ravishankar (1990), Srinivasa (1998) and Siddappa (2002), have also reported TVB-N values less than 30 mg % before spoilage in different Japanese style paste products stored at ambient, refrigerated and cooler stage temperatures. A gradual increase in TVB-N content up to 24 to 26 mg % in chikuwa paste and chikuwa prepared from frozen stored chikuwa paste was observed by Siddappa (2002), during storage at -18 ± 2 °C.

In the present study fish ham slices were made and packed in polythene bags (*polyethalian*) has also recorded a maximum TVB-N content in fibrous casing and synthetic casing products. Siddappaji (1993) has suggested that the increase in TVB-N values may be due to the activity of spore formers which survive heat processing and generally these organisms are not involved in putrefactive reactions. In the present experiment fish ham was prepared with the ion of incorporated spice powder which (spores present) could be attributed to early spoilage. Gerrard (1969), Chandrasekhar *et al.*(1988) have suggested the use of oleoresins in place of spice powders to reduce bacterial load. According to Menabrito *et al.*(1988) have reported as much as 75 mg TVB-N per 100 g in sugar and salted herring products of acceptable sensory quality. The variability of TVB-N contents makes it useful as an index of freshness or acceptable quality only at the terminal storage life of fish not in additive added fishery products (Siddappaji. 1986, Venugopal 2006). Venugopal (2006) reported that there is a good correlation between TVB-N values and sensory data. Connell (1975) has recommended 100 - 200 mg % of TVB-N as acceptable limit for cured products.

5.3.3 Changes in trimethylamine nitrogen (TMA-N)

The changes in the TMA-N content of fish ham stored at ambient, refrigerated and frozen storage are presented in Table 8,17,26 and Fig 9, 13 and 22.The TMA-N production in fish tissue during refrigerated storage could be used as an indicator of bacterial activity and it is an accepted measure of deterioration. The pungent odour of spoiled fish has been often related to the presence of TMA-N, also with the number of spoilage organisms present in many fish species (El Marrakchi, 1990). The acceptable limit of TMA-N for fishery product is 10 to 15 mg / 100 g (Connell, 1975).

According to Easter, *et al.* (1983), at higher pH, production of ammonia from amino acid, inhibit TMAO reductase and reduce its contribution to spoilage. TMAO reductases of some spoilage flora can result in increased TMA-N production in micro-aerobic and anaerobic condition.

Since TMA-N is produced only during bacterial spoilage of fish, its content can only be used as an index of spoilage due to microbial activity in product and not as an index of freshness. TMA-N concentration at sensory rejection is dependent on the products and storage conditions (Venugopal 2006).

The initial TMA-N content of products in fibrous and synthetic casing was found to be 2.80 mg % and 2.40 mg % respectively. During ambient storage temperature TMA-N values for these products gradually increased to 18.67 mg % and 8.50 mg % on 3rd day whereas it was 16.25 mg % in synthetic casing ham on 4th day. The TMA-N content during refrigerated storage on 14th day for fibrous casing was 4.48 mg % and spoiled at the end of 28 days. While the it was only 6.01 mg % in synthetic casing product on 28th day but later on 35th day it was found spoiled. TMA-N is often used as an index in assessing the shelf-life and keeping quality of fishery products because of their rapid accumulation in the meat under refrigerated condition (Ocano-Higuera *et al.,* 2006). Chandrasekhar *et al.* (1988) Hegde (1989) and Siddappa (2002) have reported initial TMA-N value of 3.50 mg %, 3.42 mg % respectively. Siddappji (1986) has reported TMA-N value of 28.7 mg % from the initial value of 2.68 mg % at ambient temperature for fish ham prepared using cured tuna cubes. Joshi (1990) has reported 5.50 mg % of TMA-N content on initial day of storage in fermented sausage.

However Siddappji (1993) has reported a TMA-N value of 7.90 mg % and 9.50 mg % in fish sausage containing defatted soyabean meal (DFSM) and full fat soyaflour (FFSF) stored at cooler storage. Fish ham prepared using cured tuna meat stored at refrigerated temperature and cooler storage has shown 23.52 mg % and

19.6 mg % before spoilage (Siddappaji, 1986). Similarly the fish ham stored at - 18 ± 2 °C reported that the fish ham in fibrous and synthetic casing reached a value of 17.19 mg % and 15.90 mg % at the end of 90 days. According to Siddappa (2002) fish chikuwa stored at – 20 °C and chikuwa made from frozen fish meat paste has recorded TMA-N value of 11.97 mg % 5 mg % and 11.92 mg % before spoilage.

5.3.4 Changes in peroxide value (PV)

The changes in lipid during storage is conventionally used as an index of quality in fish and fishery products (Hardy, 1980). The peroxide value (PV) is an index of oxidative rancidity (Labuza, 1971) According to Venugopal (2006) lipoxygenase, peroxidise and microsomal enzymes can initiate lipid peroxidation producing hydroperoxides. Peroxide value has been used as an index of rancidity in lipids of fish and fishery products. In the early stage of lipid oxidation Lipids rich in omega -3-polyunsaturated fatty acid are very susceptible to oxidation and produced distinctive oxidative off flavours (Ackmen, 1992).

The changes in the peroxide values of the products stored at different temperature throughout the storage period are presented in Table 9, 18, 27 and Fig 5, 14, and 23 respectively. The initial peroxide of fish ham packed in synthetic casing and fibrous casing stored at different temperature were found to be 1.38 and 2.45 milli eq of O_2 / kg of fat respectively. The peroxide value content of both the products in synthetic casing and fibrous casing stored at ambient temperature steadily increased with the increase of storage period from 10.31 to 20.23 milli eq of O_2 / kg of fat on 1st day to 4th day in synthetic casing ham. Whereas peroxide value in fibrous casing ham increased at slightly faster rate from 11.01 to 25.58 milli eq of O_2 / kg of fat at salt and its impurities such as copper and iron, as well as high moisture content of the product (Burt,1989).

5.3.5 Changes in free fatty acids (FFA)

The FFA is an index of hydrolytic rancidity. The changes in the free fatty acid content of the different samples stored at different temperature throughout the storage period are presented in Table 10, 19, 28 and Fig 6, 15 and 24. In the present study the initial content of free fatty acids in lipids of ham in synthetic casing and fibrous casing stored at different temperature were found to be 2.39 and 2.90 % of oleic acid respectively.

The free fatty acid content of both the products stored at ambient temperature showed marked increased with increase of storage period. Free fatty acid of fish ham in synthetic casing from 5.29 to 8.50 % of oleic acid on 1st day and 4th day , where as free fatty acid in fibrous casing fish ham also increased from 5.29 after 24 hrs to 8.23 % as oleic acid at the 3rd day respectively. The FFA content in fibrous casing showed that the product was hydrolysed faster when compared to synthetic casing fish ham.

The free fatty acid content of fish ham in synthetic casing and fibrous casing stored at refrigerated temperature showed increasing trend with increase of storage period from 2.39 to 7.88 % of oleic acid on 28thday in fish ham in synthetic casing. The free fatty acid in fibrous casing fish ham also increased from 2.90 to 8.05 % of oleic acid on 28th day.

The fish ham in synthetic casing at frozen temperature showed marginally rise in free fatty acid over fibrous casing products during its storage at -18 ± 2 °C. The initial values of both the products were found to be 2.39 % and 2.90 % of oleic acid respectively, which has increased to 6.02 % and 6.29 % of oleic acid at the end of 90 days of cold storage. Increase in free fatty acid during stored at different temperature may be due to hydrolysis of triglycerides and phospholipids (Bilinski *et al.*, 1981). It is well known that FFA is a result of enzymatic decomposition of lipid in fish and fisheries products (Hardy, 1980). Free fatty acids have been shown to oxidize faster than their triacylglycerols (Labuza *et al.*,1969; Yanishlieva-Maslarova,1985) and faster than ethyl and methyl esters (Miyashita and Takagi, 1986). Thus the free fatty acids exert a pro-oxidative effect. This effect is due to complex formation between hydroperoxides and carboxyl groups through a hydrogen bond, which results in an accelerated decomposition of hydroperoxides into free radicals (Miyashita and Takagi, 1986).

Siddappa (2002) has reported initial free fatty acid content of 2.91 % for freshly prepared chikuwa which has steadily rose to 10.48 % of free fatty acid as oleic acid % during cooler storage. Similar trend has been observed in free fatty acid content of smoked (8.57 %) and unsmoked (9.84 %) sausages during cooler storage (Srinivasa,1998). Siddappa (2002) has recorded arise in free fatty acid content to 8.05 %, and 11.19 % in frozen chikuwa and frozen fish paste before the products were unacceptable. The lower values of free fatty acid recorded in frozen sliced ham products may be due to volatilisation during cold storage. According to Damodaran (1980) low concentration of salt in fish has no effect on FFA formation and with increase in concentration it inhibits FFA formation rancidity in products. These values do not indicate any change in the quality of fat oil present in fish ham products. Singhal *et al.* (1997) has suggested FFA as a criterion for assessing quality of seafood containing significant amounts of fat. Fish ham products were prepared with 2 % salt and 5 % refined oil.

5.3.6 Changes in Thiobarbituric acid value (TBA)

The thiobarbituric acid (TBA) test is the most commonly used method to determine rancidity in fishery products. Rancidity occur as a result of auto oxidation, which is measured in terms of the content of malonaldehyde, the principal compound in oxidized lipids (Tarladgis *et al.,*1960). The presence of TBA reactive substances is due to the second stage of auto-oxidation during which peroxides are oxidized to aldehydes and ketones (Lindsay, 1991).

The changes in the TBA value in fish ham stored at different storage temperature are presented in Table 11, 20, 29 and Fig 7, 16 and 25. The initial TBA values of fish ham in synthetic casing and fibrous casing were found to be 0.11 and 0.21 mg malonaldeyde / kg of fat respectively. These values gradually increased to 2.01 and 2.94 mg malonaldehyde / kg of fat on 4^{th} and 3^{rd} day respectively at ambient temperature. TBA value of 2.00 mg malonaldehyde / kg of fat is regarded as the limit, beyond which the fish will normally develop an objectionable odour and taste (Connell, 1990).

The TBA content of fish ham in synthetic casing stored at refrigerated condition showed a gradual increase in its content with increase of storage days from 1.18 on 7^{th} day to 1.97 mg malonaldehyde / kg of meat on 28^{th} day, where as TBA in fibrous casing fish ham also increased from 1.13 to 3.94 mg maloaldehyde / kg of meat at the end of 7^{th} and 28^{th} day respectively, indicating spoilage due to secondary stage of auto oxidation. This observation of the increase of the TBA value during refrigerated storage is in agreement with results reported by Yanar and Fenercioglu (1998) for fish balls made from carp and by Gelman and Benjamin (1988) for minced meat of silver carp. The changes in TBA during cold storage showed fluctuation with maximum value of 1.60 and 2.86 mg of malonaldehyde / kg of meat at the end of 90 days storage in fibrous casing fish ham, whereas in synthetic casing fish ham the changes were slightly lower with 2.09 mg of malonaldehyde / kg of meat at the end of 90 days. The results of TBA analysis indicate that there was very significant increase in its content at different storage temperatures.

5.4 Microbiological quality changes of fish ham

5.4.1 Changes in total plate count (TPC) and spore count (SPC)

According to Shewan (1961) bacterial number ranges from 10^2 to 10^6 cfu / sq. cm on the fish skin. The results of microbiological TPC and spore count of products stored at different temperature are presented in Table 12, 21, 30 and Fig 8, 17 and 26. The initial TPC of the ham in synthetic casing and fibrous casing stored at different temperature for microbial load were found to be in increasing order. During ambient storage, the fibrous casing ham showed an increasing range from initial counts of 1.35×10^{1} cfu / g to 3.18×10^{6} cfu / g on 3rd day whereas it was 3.40×10^{6} cfu / g on 4th day for synthetic casing. The spore count for the same products also increased. Microbiological counts of both the samples increased gradually during the storage period at different temperature. The fish ham in fibrous casing and synthetic casing stored at various temperature recorded almost same number of the spores after two days, after 14 days and two months in synthetic casing product stored at different temperature. Majority of the total plate counts were found to be spore formers.

The TPC and spore counts of all the products at ambient and cold storage were in the range of 1.35×10^{1} to 7.0×10^{4} cfu / g and 0.12×10^{1} and 1.8×10^{2} for spore counts in fibrous and synthetic casing products. The products stored at refrigerated temperature showed gradual increase in TPC and spore counts up to 28 days in synthetic casing and fibrous casing products. Raccach and Baker, (1978) have reported increased average plate count by approximately 1 log unit from initial levels of 10^4 to 10^5 during mechanical deboning of headed and gutted *Cod, Pollack* and *Whiting*.

According to Tanikawa (1971) spoilage occurring in fish paste products is attributed to microorganisms originally present in fish meat and other added materials. Similar observation were also by Ravishankar (1990) and Raju (1999) for fish sausage. Siddappaji (1986) has recorded 2.1×10^{1} cfu / g of sample and he has also reported that the spore counts was not detectable soon after processing in fish ham containing cured tuna meat. In the present study, the lower counts may be attributed to the use of blanched shrimp (salt in shrimp) and spice powders. The alkaline nature of shrimp meat, higher sodium chloride content in the fish ham products and the preservative effect of spice powder might have resulted in low counts. Heat processing of fish ham for 90 min at 88 - 90 °C. Apart from its storage at refrigerated and cold storage. At ambient temperature, because of higher temperature, microbes have grown faster and caused spoilage.

The muscle tissues of healthy fish is sterile, consequently pathogens and spoilage flora, are introduced to fresh fish during post harvest processing and handling. The rate of spoilage in fish ham was high because it is a protein - rich medium that encourages microbial growth. The increased microbial growth resulted in the undesirable odour, flavours and signs of decay on the surface of the product. Spoilage of fish ham due to changes in flavour and odour during storage is almost invariably a consequence of microbial activity. However, even oxidative rancidity also contributes to spoilage. Mahesh (2002) has reported 1.8×10^2 to 2.2×10^2 cfu / g.

According to Banwart (1999), the greatest reduction in microbial load occurs during or shortly after the freezing of food and the numbers reduce further during frozen storage. This may be the reason for very low counts in fish ham stored in cold storage.

5.5 Organoleptic evaluation of the products.

The results of the analysis of the mean panel scores conducted for assessing the organoleptic quality of non-measurable attributes of the fish ham such as appearance, texture, odour, colour, taste, flavour and overall acceptability are recorded and presented in table 13, 22, 31 and Fig 9, 19, 28.

Freshly prepared fish ham in synthetic casing scored 8.0 and above for all the attributes except for taste (7.8). The overall acceptability of fish ham in fibrous casing was 7.9. As the storage period increased, there was gradual reduction in mean organoleptic scores of all attributes. The score for taste, texture and flavour for the fish ham in synthetic casing stored at ambient temperature on 3rd day was 6.0 and 6.0 and 5.5 respectively, which were much higher than the scores of fish ham in fibrous casing on 2nd day, which scored 5.5 for texture, 5.2 for taste and 5.1 for flavour. Both products were found to be spoiled on 4th and 3rd day respectively. Similar result have been reported by Siddappaji (1986) for fish ham containing cured tuna meat. Maragal (1979), Joshi (1990) and Srinivasa (1998), have also made similar observation when fish sausage was stored at room temperature.

The results of the organoleptic scores recorded for fish ham in synthetic and fibrous casing samples stored at refrigerated temperature were almost similar up to 21 days, but the scores for products in synthetic casing was higher throughout the storage period of 28 days. The score for taste, texture and flavour of synthetic products on 28 days was 4.0, 5.0, 3.5 respectively. The corresponding scores for same attributes of fish ham in fibrous casing were 3.0, 3.5 and 3.0 respectively. But the overall acceptability on 21st day for fibrous casing product was 5.6 with score for texture was 5.1 and 5.2 for taste and texture. Hence the study showed that the fibrous casing fish ham has a shelf life of 21 days and fish ham in synthetic casing were highly acceptable for 28 days at refrigerated storage temperature.

In the present study, it was observed that the pH of all the products stored at different temperature was high (> 6), thus the spoilage becomes apparent in organoleptically detectable level. Hence low organoleptic score after 2nd and 3rd day for fibrous casing and synthetic casing products respectively stored at ambient temperature, Whereas the products at 6 ± 2 °C and at -18 ± 2 °C recorded low scores for all the attributes after 21 and 60 days respectively. Consumers acceptance of fish ham is dependent on the appearance of the products. The primary factor determining the texture of fish ham can be the final pH, which affect the water content of the product.

A food is said to be spoiled scientifically; when it is unacceptable to consumers due to its considerable sensory attributes or when it contain lot of chemicals at levels hazardous to health. The sensory scores decreased in all the products stored at different storage temperatures to 5.0 and below indicating that the products are not acceptable.

From the results it can be concluded that the shelf life of fish ham in fibrous casing spoiled little earlier when compared to fish ham in synthetic casing. It was observed that, the ham packed in fibrous casing was very well accepted for 2 days at ambient condition, 21 days at refrigerated temperature and nearly 90 days at cold storage. Hence, fibrous casing can be used as an alternative to synthetic casing.

Summary

VI. SUMMARY

Fish is high in nutritional and health benefit characteristics as well as taste. Dieticians now advice that fish be eaten at least twice a week, a factor that has heightened demand for its regular supply, it is suggested to effectively utilise by catch of trawl for human consumption by preparing ready to serve heat processed nutritious fish products (about 30 % of the catch constitute low value fishes). At present a wide variety of fish paste products including fish ham, are successfully processed and marketed using synthetic polyvinyledene chloride plastic casing. Now a days consumers are demanding for products packed in natural material, biodegradable, environment friendly packaging material. Hence, an attempt has been made to utilise deep sea trawl catches such as *Priacanthus hamrur* and small shrimps (instead of cured red meat of tuna) to prepare fish ham in fibrous casing as an alternative to synthetic plastic casing.

Disco fish meat was separated, minced and ground with various additives namely salt, sugar, spices powder, groundnut oil, corn flour with sufficient quantity of chilled water into fine paste. The shrimp was deveined and washed then blanched in 6 % brine solution for 6 min then cut into small pieces. Cooled and mixed with fine fish paste in the ratio of 60 : 40 (Fish paste : blanched shrimps) used to prepare fish ham. Fish ham was divided into 2 batches. One batch of fish ham was stuffed into synthetic casing (SC) and the another batch was stuffed into fibrous casing, (FC) after sealing the ends of the casings, the products were washed, then heat processed at 90 °C for 90 min. After cooling for 15 min. The heat processed products were stored at ambient (28 ± 2 °C) and refrigerated (6 ± 2 °C) temperature to assess the changes in quality and shelf life. One batch of ham product was stored at frozen storage at (-18 ± 2 °C).

- The physical characteristics of raw material indicate that the Bull's eye sho- wed an average total length of 18.96 cm. The average weight of Bull's eye used in the present experiment was found to be 84.35 g.
- The mechanical separation of meat from the dressed fish has given a good proportion of picked meat and minced meat yield percentage of 32.42 %.
- The physico - chemical tests that were carried out for ham in fibrous casing and synthetic casing include proximate composition, pH, TVB-N, TMA-N, PV, FFA, TBA. Microbiological tests include TPC and spore counts of raw materials and final products during storage studies at different temperature.
- The average values for moisture, protein and ash content of Bull's eye and shrimp were found to be 77.80 %, 18.53 %, and 1.45 %. The proximate composition values indicate that the raw materials belong to lean variety with an average fat content of 1.38 %. The percentage of fish paste and blanched shrimp mixed in the ratio of 60:40 was found optimum with respect to sensory characteristics such as taste, odour and flavour. A processing duration of 90 min was ideally suitable for fish ham products having 450 g, when heat processed at 90 ± 2 °C.
- After processing the proximate composition of fish ham packed in synthetic casing was (SC) found to be, 18.15 % protein, 5.66 % fat 71.39 % moisture and 2.50 % of ash and that of fish ham in fibrous casing (FC) was found to be 19.2 % protein, 6.33 % for fat, 2.21 % ash and 67.63 % moisture content.
- During storage at ambient temperature the initial values of pH, TVB-N and TMA-N were found to be 6.82, 1.5 mg % and 2.8 mg % in FC where as the same for SC were 6.82, 1.16 mg % and 2.4 mg % respectively. The initial values of PV, FFA and TBA of SC were 1.38 milli.eq. of O_2 / kg of fat, 2.39 % of oleic acid and 0.11 mg of malonaldehyde/ kg of meat. During storage at ambient temperature these

values gradually increased to 20.23 milli.eq. of O_2 / kg of fat, 8.5 % of oleic acid, 2.01 mg of malonaldehyde / kg of meat, where as the same in FC was 25.58 milli.eq. of O_2 / kg of fat, 8.23 % of oleic acid and 2.94 mg of malonaldehyde / kg of meat. TPC and Spore counts of fresh fish ham were in the order of 1.15×10^1 cfu/g and 0.12×10^1 spore No./g in SC and in FC it was observed to be 1.35×10^1 cfu/g and 0.94×10^1 spore No./g . During 4th day it was observed in SC that the TPC and spore counts were 3.4×10^6 cfu/g and 6.4×10^3 spore No./g respectively whereas in FC they were 3.18×10^6 cfu/g and 6.1×10^4 spore No./g respectively. The overall acceptability scores of SC and FC products was 8.0 and 7.9 which has been reduced to 6.35 and 5.86 on 3rd day and 2nd day respectively.

- During refrigerated storage the pH of SC and FC has gradually increased from 6.82 to 7.0 and 7.3 respectively, at the end of storage period. TVB-N and TMA-N values of SC were reported to be increased to 26.44 mg % and 6.01 mg % where as the values of FC were 28.64 and 9.8 mg % on 28th and 21st day of storage. There was a noticeable rise in the values of PV, FFA and TBA from 14 days and 21 days with the values 16.73 and 12.43 milli.eq. of O_2 / kg of fat , 5.33 % and 4.75 % of oleic acid and 2.14 and 1,60 mg of malonaldehyde/ kg of meat in FC and SC respectively. The rise in TPC (3.9×10^6 cfu / g) of FC was faster than SC and a marginal variation in spore counts were recorded in all the products at refrigerated storage temperature.
- The organoleptic quality characteristics of FC and SC were remained very well in terms of taste, texture and flavour for a period of 21 days and 28 days respectively with the score being 5.20, 5.10 and 5.20 for SC 5.70, 5.90 and 5.60 and 4.0, 5.00 and 3.5 respectively in FC.

- The frozen stored sliced ham at - 18 ± 2 °C showed a decreasing trend up to 30 days and 60 days in SC and FC products before increased to 6.9 and 6.98 on 90th of storage during storage at -18 ± 2 °C, the TVB-N and TMA-N values increased to 12.94 mg % and 17.19 mg % in FC respectively whereas the same for SC were found to be 12.28 mg % and 15.90 mg % on 90th days of storage. A slow and steady increase in PV, FFA and TBA values were observed with 15.16 and 16.57 milli.eq. of O_2 / kg of fat, and 6.02 and 6.29 % of oleic acid and 2.09 and 2.86 mg of malonaldehyde/ kg of meat in SC and FC respectively. The TPC and spore counts of FC and SC have slowly increased from 1.35×10^1 and 1.15×10^1 to 7.0×10^4 and 2.9×10^3 cfu / g. The average spore count of SC and FC on the 90th day of storage was found to be 2.2×10^2 No / g.
- It was observed that, the ham packed in fibrous casing was very well accepted for 2 days at ambient condition, 21 days at refrigerated temperature and nearly 90 days at cold storage. Even though the synthetic casing proved to be slightly better than fibrous casing, the fibrous casing can be used very well as an alternative to synthetic casing for the for the production of ham.

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Abstract

VIII. ABSTRACT

In the present study, ready-to-eat fish ham was prepared using *P. hamrur* and small shrimps. Fish ham was prepared using cured meat cubes of tuna where sodium nitrate were used generally for curing to retain the attractive red colour, but this chemical is known to cause cancer long run. Hence, uniform pieces of small size shrimp at a ratio of 60 : 40 were used in place of cured red meat cubes.

The fish paste containing shrimp was divided into 2 batches. One batch was stuffed into synthetic casing (SC) and another batch was stuffed into fibrous casing (FC), sealed and cotton twine was wound around the products and heat processed at standardised processing temperature and time of 88 ± 2 °C and 90 min. respectively, and cooled for 20 min. The prepared ham were divided in to three batches and stored at different conditions *viz.*, ambient (28 ± 2 °C), refrigerated (6 ± 2 °C) and at frozen (-18 ± 2 °C) temperature respectively.

The quality analyses of ham stored at ambient, refrigerated and frozen conditions were conducted at regular intervals of one day, week and one month respectively. The results showed that TVB-N and TMA-N increased significantly in FC ham at the end of 3 days, 21 days and 3 months respectively when compared to SC ham. The lipid quality such as PV, TBA, FFA also showed a similar trend in FC ham when compared to SC ham in both ambient and refrigerated temperature, which [illegible]tributed to the good barrier properties of SC, whereas during frozen storage [illegible] The initial TPC (1.15×10^1 and $1.35 \times$