Performance and quality assessment of salt curing of Taki (*Channa punctatus*), Shol (*Channa striatus*) and Tengra (*Mystus tengra*)

A

DISSERTATION SUBMITTED TO THE UNIVERSITY OF DHAKA FOR THE DEGREE OF DOCTOR OF PHILOSOPHY (Ph D) IN ZOOLOGY (FISHERIES)

BY

FARZANA BINTE FARID

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Department of Zoology
Faculty of Biological Sciences
University of Dhaka
Dhaka, Bangladesh
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DEDICATED
TO
MY BELOVED
PARENTS
My husband
and
My Children

CERTIFICATE

The undersigned certify that this is an original research work of Farzana Binte Farid entitled **"Performance and quality assessment of salt curing of Taki (*Channa punctatus*), Shol (*Channa striatus*) and Tengra (*Mystus tengra*)"** and the results which are embodied in this thesis are the investigation and works of her own for the degree of Doctor of Philosophy. This dissertation is suitable for submission.

Supervisor

Prof. Dr. Gulshan Ara Latifa
Department of Zoology
University of Dhaka
Dhaka-1000
Bangladesh

Co-Supervisor

Prof. Dr. Subhash Chandra Chakraborty
Department of Fisheries Technology
and Dean, Faculty of Fisheries
Bangladesh Agricultural University
Mymensingh-2202
Bangladesh

Declaration

I am submitting my dissertation to the University of Dhaka for the fulfillment of the degree of Doctor of Philosophy (Ph D). I do hereby declare that the research work contained in this dissertation entitled **"Performance and quality assessment of salt curing of Taki (*Channa punctatus*), Shol (*Channa striatus*) and Tengra (*Mystus tengra*)"** is my own investigation, original and belief it is not written by another person which has not been submitted for the award of any other degree.

Farzana Binte Farid

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ABSTRACT

Salting of fish is widely used as one of the most important low-cost traditional method for fish-preservation and considered as a concentrated source of nutrients to all categories of people throughout the world including Bangladesh. Salt-cured fishes are highly appreciated because of their characteristic taste, texture and storage stability. So, attempts have been made in the present study to preserve three freshwater fish-species of Bangladesh viz., large size fish Shol (*Channa striatus*, Bloch; 1801), medium size fish Taki (*Channa punctatus*, Bloch; 1793) and small size fish Tengra (*Mystus tengra*, Hamilton- Buchanan; 1822) with five different types of salt-curing method viz., Dry salting (DS), Pickle curing (PC), Brine salting (BS), Sun-dried salting (SDS) and Turmeric treated sun-dried salting (SDS+T). Studies were conducted on their sensory (characteristics & scores), proximate (moisture, protein, fat, ash) chemical (salt, TVB-N, FFA, pH) and bacteriological (SPC & HBC count) quality assessment during storage period both at room (26-32^{0}C) and refrigeration (4^{0}C) temperature using standard methods of analyses. Important mineral composition e.g. Ca, Mg, Fe, Cu Zn, and Mn were also determined in fresh fish and freshly processed salted fish-products.

The present study showed that, the content of moisture, protein, fat, ash, TVB-N (Total Volatile base-Nitrogen), FFA (Free Fatty Acid), pH and SPC (Standard Plate Count) of freshly collected shol fish was 77.03±0.12%, 19.52±0.07%, 1.93±0.07%, 1.44±0.11%, 4.41±0.01 mg/100g, 0.6±0.06%, 6.9±0.06, and 2×10^5 cfu/g; taki fish was 78.65±0.07%, 16.89±0.10%, 2.50±0.06%, 1.36±0.11%, 3.43±0.02 mg/100g, 0.5±0.10%, 7.0±0.06 and 1.1×10^5 cfu/g and tengra fish was 74.27±0.07%, 16.96±0.07%, 6.04±0.06%, 2.67±0.08%, 4.27±0.02 mg/100g, 0.9±0.21%, 7.0±0.10 and 5×10^5 cfu/g respectively. Fresh shol, taki and tengra fish had Ca 11.2, 16.35 and 22.025; Mg 10.125, 9.425 and 11.4; Fe 1.475, 1.275 and 2.25; Cu 0.7, 0.65 and 0.55; Zn 0.25, 0.425 and 1.275; Mn 0.1, 0.05 and 0.125 mg/100g of fish respectively.

'Salt ripening' of five different types of salted shol, taki and tengra was obtained by assessing changes in salt penetration rate and its effect on moisture content, fluid loss pattern in fish muscles and percent weight loss of fishes during ripening period.

After salt-ripening moisture (%) content, pH value, SPC (cfu/g) and HBC (cfu/g) load decreased whereas protein (%) (except BS tengra), fat (%), ash (%), salt (%) and FFA (%) value increased in salted fish samples when compared to the fresh fish samples. Salt (NaCl) content in freshly prepared salted products ranged between 13.77% & 17.10%. Preferability study was also done with the taste testing of 'Fish-curry' prepared from 5 different types of freshly processed salted Shol, Taki and Tengra fish-products so that these kinds of new products could find good market. Taste testing study showed the greater acceptability of turmeric treated sun-dried salted (SDS+T) shol, taki and tengra fish over the others. It was also observed that salted fish products are a good source of minerals. Mineral elements detected were in the decreasing order of Ca>Mg>Fe>Zn>Mn>Cu in fresh processed five types of shol, taki and tengra fish-products respectively.

During storage period, protein, fat, ash, salt and sensory score were found to be decreased whereas moisture, TVB-N, FFA, pH, SPC and HBC value increased both room and refrigeration temperature, but the increase in room temperature was more prominent than the products stored in refrigeration temperature. However, there was a general decline in sensory characteristics and scores on flavor, color and texture of salted fish-products during storage (a total period of 36 months) and the acceptability score <4.5 (in 1-9 point hedonic score on quality) was considered as rejected or spoiled for all salted fish samples. There was also found direct relation between moisture & chemical composition (TVB-N, FFA, pH) and moisture & bacterial count (SPC, HBC) whereas inverse relation was revealed between sensory score & TVB-N, FFA, pH value during entire storage period. Visually there was found no fungal attack in all of 5 types of salted shol, taki and tengra fish products during the storage period.

The overall quality of all salted fish-products were excellent in fresh process condition and five types of salted products stored at refrigeration temperature were found in better condition as determined by sensory, biochemical and bacteriological analysis.

From the overall performance, it has been proved that these three fishes were highly acceptable in salted condition and turmeric treated sun-dried salting (SDS+T) method was found as the best. On the basis of quality-standard and shelf-life performance, the five salting methods can be arranged as follows: Turmeric treated sun dried salting (SDS+T)>Sun-dried salting (SDS)>Dry salting (DS)>Pickle curing (PC)>Brine salting (BS). Among the three species, five categories of salted tengra fish-samples showed better performance in respects of shelf life study. The highest shelf life was found to be 36 months in SDS+T tengra at refrigeration storage whereas lowest was found in 75 days in BS taki at room temperature.

Thus the present study provides information about the suitability of different salting methods of three commercially important fishes to produce a very stable and safe product with long storage life. By assessing the nutritional quality as well as the feasibility of the method, it can be recommended to incorporate the turmeric treated sun-dried salting (SDS+T) method in commercial scale which further may contribute in national economy of Bangladesh.

CHAPTER-1

INTRODUCTION

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INTRODUCTION

1.1. Geography of Bangladesh

Bangladesh is a South Asian sub-tropical country located between latitude $20^0$34′ and $26^0$39′ north and longitude $80^0$41′ and $92^0$41′ east. It is bordered by India to the west, north and the northeast; Myanmar to the southeast and the Bay of Bengal to the south. The country is crisscross with hundreds of rivers. It consists of the deltaic flood plains of the Ganges, the Brahmaputra and the Meghna with their tributaries and branches. Winter lasts only for two months in the country. Temperature and rainfall ranges from 07^0 to 40^0C and 1170 to 3400 mm respectively. The climate of Bangladesh is a unique for aquaculture and fisheries resource management.

Bangladesh is one of the most aquatic resource (marine and freshwater) diverse countries in the world. It has vast water resources in the form of rivers, canals, haors, baors, beels, floodplains, ponds, lakes, estuaries and vast fishing grounds in the Bay of Bengal. Due to the sub-tropical climatic condition and a large number of natural freshwater reservoirs, wetlands, man-made ponds and lakes and flood plains water bodies shows a promise of further expansion of fish production in Bangladesh.

There have about 46.99 hundred thousand hectare inland water resources in Bangladesh in which 39.1 hundred thousand hectare are open water and 7.89 hundred thousand hectare is closed water (DOF, 2015). Bangladesh has also an area of 166.07 hundred thousand hectares of marine waters, including territorial and economic zone.

1.2. Status and importance of fish and fisheries

Fish like many other forms of life, are of immense values to mankind. They have long been a staple item in the diet of many people. Today they form an important commodity in the economy of many nations, while giving incalculable recreational and physiological values to the naturalists, sports enthusiast and home aquarist. Fish is a key ingredient on the global menu, a vital factor in the global environment and an important basis for livelihood worldwide (Bene and Heck, 2005). The global contribution of fish as a source of protein is high, ranging from 10% to 15% of the human food basket across the world (Wilson *et al.*, 2007).

Fish is a non-tetra pod chordate, i.e., an animal with a backbone that has gills throughout its life and has limbs in the shape of fins. (Nelson, 2006). Fish belongs to paraphyletic group of organisms that consist of all gill-bearing aquatic vertebrate (or craniate) animals. Most fish are "cold-blooded", or ectothermic, allowing their body temperatures to vary as ambient temperatures change, though some of the large active swimmers like white shark and tuna can hold a higher core temperature (Goldman, 1997). At 32,000 species, fish exhibit greater species-diversity than any other class of vertebrates. Fish species diversity is roughly divided equally between marine (oceanic) and freshwater ecosystems. In terms of weight of food consumed, fish ranks third after rice and vegetables (Minkin *et al.*, 1997; Hels *et al.*, 2002).

In view of the present population boom scarcity of protein is increasing day by day. According to a statistical report the earth will have eight billion people by the end of 2020 and there will be needed double increase in plant proteins and four times increase in animal proteins to maintain the existing nutritional status of people in different parts of the world. Chukwu (2009) stated that the gap between the demand and supply of fish is widening due to increase in population, poor post-harvest handling, lack of processing and storage facilities and utilization of unconventional fish species.

It is estimated that 60% of people in developing countries obtain 40-100% of the animal protein in their diets from fish (Clucas and ward, 1996). The LDC (less developed countries) capture 50% of the word harvest and a large proportion of the catch in many Asian countries over 50% of the animal protein intakes comes from fish (William *et al*., 1988).

In spite of these resources, malnutrition is one of the major factors faced by the people of developing countries today (Hossain and Afroze, 1991). In Bangladesh this nutritional deficiency has become more acute because of tremendous rate of population growth without any significant increase in animal production. The population of Bangladesh is near about 16 crore (BBS, 2015). Millions of people are suffering from serious nutritional problem owing to acute shortage of animal protein in their diet. The current fish consumption rate is 17.52 kg/people/year whereas the demand is 20.44 kg/people/year and is 29.74 MT per year (Kabir *et al.,* 2012; Minar *et al.,* 2012).

Due to high population growth there is an ever ending gap between supply and demand of fish and fisheries products in Bangladesh. Narrowing the gap not only requires increasing production but also improvement in all aspects of marketing and distribution system is also important (ICLARM, 1991). For this, the harvested fish and fisheries products should be processed properly to reduce the gap between supply and demand.

Tropical regions of the world have a rich diversity of demersal and pelagic fish. About 28 percent of the world's fish catch, nearly 90 million tons, are found in tropical waters. Bangladesh belongs to the sub-tropical region of the world.

1.3. Role of fish and fisheries in national economy of Bangladesh

From time immemorial, 'Rice' and 'Fish' dominate the diet of Bangladeshi peoples. Fish (including shrimp and prawn) is the second most valuable agricultural crop in Bangladesh (WorldFish Center, 2011). From 2004 to 2014, Bangladesh's fish production increased by 53%. Both marine and fresh water fishes are natural resources in Bangladesh, which cater to the enormous need of protein and other nitrogenous constituents (Mahamud *et al.*, 1999).

In the 2013-14 fiscal year, the country produced approximately 35.48 lakh Metric Ton fish (BBS, 2015). Bangladesh earned 4776.92 crore taka by exporting fish and fishery products in 2013-2014 (DOF, 2015). Fisheries contributed 4% GDP and 2.09% foreign exchange earnings in 2013-2014 fiscal years (DOF, 2015). In Asia, Bangladesh stands fourth in fish production; China is first in fish farming and next India and Myanmar (FAO, 2015).

Bangladesh is a reverine country and fisheries sector plays an important role in national economy and also in the national dietics. Fish production in ponds, lakes, burrow-pits, floodplains, oxbow lakes and semi closed water bodies are increasing day by day through transfer of modern technology (Ali *et al.*, 2001). It provides not only nutrition, employment generation, poverty alleviation and foreign currency but also helps to build up the socioeconomic condition of Bangladesh (Ali, 1998).

In our country about 12 million people are directly or indirectly involved in this sector. Fish plays an important role in the Bangladeshi diet, it constituting only animal protein source among rural households (Ferdose and Hossain, 2011; Shamsuddin, *et al.*, 2012; Azim *et al.*, 2012). Fish alone contributes about 63% of animal protein to the diet of the people of this country (DOF, 2009). There are 260 indigenous, 24 exotic fish species and 24 species of prawn and also 475 species of marine fish available so far in the waters of Bangladesh (DOF, 2013).

Fish as a food in one of the most familiar, popular, tasty and nutritionally enriched item of food around the world including Bangladesh**.** As a result of joining and accepting the policy of global market economy along with so many food items, garments, pharmaceutical products, so on and so forth fish and fishery products also get the opportunity to enter in the global market. The processed fish products are exported to the European countries, U.S.A and Japan. About 95% of total fish products are exported to those countries. Remaining portion is exported in Southeast Asia and Middle East.

1.4. Nutritional value of fish

Fish represents an important source of food for mankind throughout the world (Ravichandran *et al*., 2010; Sutharshiny and Sivashanthini, 2011). It is a good source of protein and lipid for the development of our body (Siddique *et al.,* 2011). Fish also rich in vitamin and minerals for both young and old age consumers (Edem, 2009; Koffi-Nevry *et al.,* 2011). The important constituents of fish are water (70.0-85.0%), protein (15.0-20.0%), lipid (1.0-10.0%), ash (1.0-1.5%) and carbohydrate (0.3-1.0%) (Moghaddam *et al.,* 2007; Sutharshiny and Sivashanthini, 2011) which make it good and healthy diet and it is easily digested by human body (Marcu *et al.,* 2010).

During the last decades in this century, healthy eating habits have received increased attention and it is widely recognized that regular fish consumption is one possible health improving practice (Sidhu, 2003). It also contains some bioactive compounds with therapeutic properties that are beneficial to human health (Nnaji *et al.,* 2010; Lordan *et al*., 2011). Furthermore, some nutritional components such as, fish oil is one of the most important natural sources of polyunsaturated fatty acids having eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), which have been proven to have useful effects on human body (Saoud *et al.,* 2008). The protection conferred may be due to Eicosapentaenoic acid (EPA), the omega-3 fatty acid

typically found in fresh fish oil (Anonymous, 1986). These fats are also needed to absorb fat-soluble vitamins A, D, E and K from food; and it also regulate cholesterol metabolism in body (Jabeen and Chaudhry, 2011). Fish has more unsaturated fatty acid than animal fats. Muscle of fish is largely composed of proteins of the globulin and albumin classes. Fish protein is more beneficial for human body because it has amino acids which are good for human body and also have free amino acids (Buchtova *et al*., 2010). Compared to mammalian meat, fish meat has more water and less connective tissue, which contains very little elastin (Kołakowska and Kołakowski, 2001). Fish are extremely important aspects for health conscious people particularly in affluent countries, where cardiovascular disease mortality is high (Huss, 1994). When the whole fish is consumed, it serves as a good source of calcium, phosphorus and iron, trace elements like iodine (in marine fishes) as well as vitamins (A and D).

In recent times, fish has been reported as the cheapest source of protein used to correct protein deficiency in human diets in the tropic region (Akinwumi, 2011). Arannilewa *et al*., (2005) and Ojutiku *et al.*, (2009) also highlighted that fish is rich in protein with amino acid composition very well suited to human dietary requirements comparing favorably with egg, milk and meat in the nutritional value of its protein. Fish also contains absorbable dietary minerals (Bruhiyan *et al.*, 1993). Fish demand is increasing as a result of the increasing world population, higher living standards and the good overall image of fish among consumers (Cahu *et al.*, 2004).

In Bangladesh, very little work has been done on the presence of macro and micro elements in freshwater fish, despite such data are important to assess the quality and safety of fish and fishery products for domestic consumption as well as for export.

The biochemical composition is an important aspect of fish quality and it influences both the keeping quality and technological characteristics of fish. Currently, the demand for traceability of food quality and food safety for consumers is increasing (Ólafsdóttir, 2005). The chemical composition of food gives its potential nutritive values. Proximate analysis provides information on the uncertain nutritional

significance of a particular organism or feed stuff (Crampton and Harris, 1969). So, the nutritional quality of fish is so high that can help to overcome the severe malnutrition problem with sharp and sudden shortage of animal protein in the diet of the people of Bangladesh.

It is necessary to know the composition of fish and its seasonal variation which will help as in determining the correct preserving method and is important in several ways in industrial processing of fish (Stansby, 1962).

1.5. Spoilage of fish

Spoilage is a metabolic process that causes food to be undesirable or unacceptable for human consumption due to changes in sensory and nutritional characteristics (Doyle, 2007). According to Yohanna *et al.* (2011), spoilage of fish can be due to rapid autolysis by the fish enzymes, and because of less acid reaction of fish flesh that favors microbial growth. Fish and shellfish are highly perishable foods and susceptible to faster postmortem deterioration (Smith and Hui, 2004).

Fish is highly perishable of all stable commodities especially in tropical climate regions and even in temperate climates of the world because it provides favorable medium for the growth of microorganisms after death (Aliya *et al.,* 2012; Oparaku and Mgbenka, 2012).

In the tropics where temperature and humidity of the climate are very favorable for the growth of microorganisms, spoilage is rapid; fish will spoil within 12-20 h depending on species, method of capture (Igene, 1983). If they are not processed immediately after harvesting, certain irreversible spoilage and deterioration of meat quality begin to take place (Conne, 1995).

The microorganisms present on fish include those which are associated with the raw material and acquired during harvesting, handling and processing (Huss *et al.*, 2003; Abraham, 2011). Fishes are in constant interaction with microorganisms which cause spoilage and their metabolic activities result in the appearance of slime, gross discoloration and strong odors and which become offensive to customers (Gram and Dalgaard, 2002; Paulsen and Smulders, 2003). Fish spoilage occurs following growth and activity of special microorganisms and lipid oxidation which cause off-odor and off-taste by production of some metabolites changing sensory characteristics and customer acceptability (Moini *et al*., 2009; Rostamzad *et al.*, 2010). Rahman, *et al.* (2001) stated that immediately after the fish dies, changes begin to taka place, which cause loss of quality and value of the products viz: a) Autolytic or enzymatic spoilage b) Bacterial and c) Physical damage.

The annual fish harvest fluctuates seasonally, with periods of high and low supply. In Bangladesh each year a large quantity of fish and fish products are spoiled particularly during the peak season of harvesting. It is estimated that about 30% (about 3,07,500 mt) of the freshly harvested fish are spoiled every year due to lack of poor processing and preservation at artisanal fishermen level, while acute shortage and increase costs of fish are experienced in periods of low harvest (Balachandran, 2001; Ghaly *et al*., 2010). Moreover, as the transportation and the process of handling of fish and fish products in Bangladesh are poor, a huge quantity is spoiled during transportation from one end of the country to other. Therefore there is an obvious need for development of new technologies and efficient fish preservation methods which permit shelf-life extension of these products (Chouliara *et al.,* 2004). However while considering the consumer's risks of health hazards, the study of bacteriological susceptibility of this perishable fish possess immense nutritional importance in Bangladesh. Spoilage is associated with Physical, Chemical and Bacteriological changes of fish which with species, stage of maturity, age and sizes of fish (Rubbi *et al.,* 1987; Muslemuddin *et al.,* 1991). While working on spoilage Khuda (1960) indicated the higher spoilage rate in younger fishes. So an attempt was undertaken to information on the effect of sizes on the spoilage rates both at room and refrigeration temperature.

1.6. Importance of processing and preservation of fishes

The term 'fish processing' refers to the processes associated with fish and fish products between the time fish are caught or harvested, and the time the final product is delivered to the customer. Curing is a simple and cheap method of processing; requiring least technical expertise but it has a great significance and relevance in the socio-economic system of small-scale fisher folk (Patterson and Ranjitha, 2009). Fish curing is defined as the method of preserving fish by means of salting, drying, smoking and pickling.

If not consumed within one day of capture fish becomes unfit for human consumption, unless subjected to some form of processing (Abolagba *et al.,* 1996). In order to reduce the wastage and spoilage of fish during periods of oversupply and to enhance long storage, it is necessary to adopt appropriate as well as affordable processing and preservation techniques for fish and marketing and during this period maintain proper quality. It is imperative to process and preserve some of the fish caught in the period of abundance, so as to ensure an all year round supply. Application of proper processing and preservation methods help to retard the spoilage change, invariably reduce post harvest losses, increase the shelf-life of fish, and guarantee a sustainable supply of fish during off season with concomitant increase in the profit of the fishermen (Eyo, 1997). Proper handling of the raw material, improved hygiene and sanitation in the process and improved packaging and storage can significantly improve the keeping quality and shelf-life of the traditional products (Nair, 2002).

Acording to Oluborode *et al.* (2010), preservation process starts when it is harvested and become complete when reaches the consumer's table. Inadequate storage techniques would imply a sustainable shortfall in fish availability thereby affecting the animal protein intake of the people in the tropics whose protein intake from fish ranges between 17.5-50% (Arannilewa *et al.,* 2005). Therefore, in order to prevent

potential future storages in the food supply, the maximum utilization of the existing catch will have to be assured (ASEAN-SEAEDEC, 2001). According to Whittle (2002), storage time and temperature are the major factor affecting the rate of loss of quality and shelf life of fish. Few studies are available which have been carried out in room temperature but no comparative data are available on various fish of tropical region. So it is felt desirable to investigate the effect of different temperatures during storage of these three processed fishes.

In recent years high cost investments had been done in process sector so consumption of aquatic products are increased. Processing technology has gained importance because of the structure and being suitable for microbial augmentation of aquatic products (Oğuzhan and Angis, 2013). Good fish preservation techniques must prevent microbial spoilage of fish without affecting its quality and nutritional value (Ghaly *et al.*, 2010). In addition, preservation suppresses undesirable chemical and biochemical changes and helps to maintain the products desirable physical and sensory properties (Lado and Yousef, 2007).

Traditional fish processing is an important livelihood activity for large number of people in Bangladesh. The nutrient content of processed fish is very much higher than raw fish. Traditional preserving methods of fish products have still wide acceptance around the world due to their accustomed taste and aroma (Kose, 2010). Cured fish are consumed mainly in tropical countries (FAO, 1981).

With the over growing world population and need to store and transport food, fish preservation becomes necessary to supply the distant market, to produce a range of products with different flavors and textures and creation of conditions unfavorable to the growth or survival of spoilage organisms (Yohanna *et al*., 2011).

At present there are about 123 fish processing industries in Bangladesh, among which only 59 are approved by government. The EU countries had banned import of processed fish from Bangladesh in 1997 due to lack of proper implementation of HACCP. At the time Bangladesh government had distributed up to 40 lakh taka loan

to each of the processing industries. According to the EU regulations, each processing industry should have proper facilities to ensure the required quality of their processed fish. At present 17 fish processing industries have gained approval from EU for exporting their processed fish to EU countries.

1.7. Methods of preservation

The overall goal in food production is to produce safe and wholesome products. Different processing methods have different effects on the nutritional compositions of fish. This is because heating, freezing and exposure to high concentration of salt lead to chemical and physical changes and therefore digestibility is increased, due to protein denaturation, but the content of thermo labile compounds and polyunsaturated fatty acids is often reduced (Chukwu and Shaba, 2009). Preservation and Processing invariably bring about some change (most of them are irreversible) in the intrinsic characteristics of the fish. It is a fact that consumers always prefer fresh to preserved fish. The lack of preference for preserved or processed fish is attributed to some extent, due to the changes in the appearance, color, texture, taste and flavor accompanying the processing involved. The severity as well as the irreversibility of the changes will depend on the type of the preservation/processing technique the fish has been subjected to.

The developments in fish processing technology over the years have been oriented towards presentation of preserved/processed products with characteristics closely resembling to those of the fresh fish. There are various methods of fish preservation, many of which are only of local importance.

The methods which are widely applied include the following:
A) Dehydration: drying, salting, pickling, smoking.

B) Lowering the temperature: Chilling with ice, Freezing and refrigeration, Storing in cold storage, deep freezing and freeze drying.
C) Rising the temperature: Canning.

According to Alam (2007) the simplest methods being employed are drying, salting, and semi-fermentation.

1.8. Salting as processing and preservation method of fishes and its importance

Salting is one of the oldest and commonly used processing techniques for fish preservation all over the world because of simplicity of the process and low production cost (Martinez-Alvarez & Gomez-Guillen, 2013) and the ease of combining it to other preservation methods, such as marinades, drying or smoking (Fuselli *et al.,* 2003; Yeannes and Casales, 2008). The preservative effect of salt has been recognised according to a decrease in water activity, less availability to microbial attack, and enhancement of functional properties, leading to an increase of the shelf-life time (Aubourg and Ugliano, 2002). A concentration of 6-10% salt in fish tissue can prevent the action of most spoilage bacteria. Length of salting period as well as salt concentration depends on the expected final product (Bellagha *et al*., 2007).

Though the technology of food preservation and processing has undergone revolutionary changes over the years and several new products processed employing diverse techniques have made their firm presence on the market, salting still continues to be the most widely used method for preservation of several foods including fish.

The salting of herring was probably first practiced in Scotland in the eighth century. Afterwards in the fourteenth to sixteenth centuries this method was improved by the

Dutch and Scotch and also by the Russians (Jarvis, 1950; Cutting, 1955). Salting is a method that has been used for centuries and in many places around the world such as Asia, Europe, Africa and Latin America (Wang *et al.,* 2000; Doré, 2008).

Egbal *et al.* (2010a) opined that, salt is effective to reduce water activity (aw), to destroy the growth of microorganisms and to increase the shelf life. It also provides desirable sensorial changes and ensures the safety for human consumption. The current demand for salted fish is driven more by sensorial alteration purposes, rather than preservation (Mujaffar and Sankat, 2005). The high concentration of salt has been shown to prevent microbial spoilage in similar products (Andersen *et al*., 2007).

Being a safe, antimicrobial and incidental food additive, toxic for some microorganisms, depressor of water activity (aw) of the food, sodium chloride has been used as a seasoning and flavor enhancer as well as a preservative or curing agent even in developed countries (Leroi and Joffraud, 2000; Lakshmanan *et al.,* 2002; Basti *et al.,* 2006; Turan *et al*., 2007). Salt causes plasmolysis and alters protein and enzyme states in such a way that proteins become impervious to enzyme action and lose their efficacy. It also has bacteriostatic and bactericidal effects (Ismail and Wootton 1992).

In India, Bangladesh, Burma, Thailand and Sri Lanka salting of fish is popular. In some countries e.g. Malaysia, Indonesia and some parts of India salting is followed by drying. In Japan almost all of the salted fish are dry salted except brine salted salmon for subsequent later smoking (Tanikawa *et al.,* 1985).

Salting is usually done by one of the four methods, dry salting, pickle salting, mixed salting and brine salting. A number of variations in the method of salting have arisen, thus in some areas the fish are used whole or are eviscerated in different ways. The amount of salt used and method of application is varied according to where the fish are caught and to their maturity. The objective of salting is to ensure that the salt penetration is rapid enough to similarly lower the water activity in the deepest parts of

the flesh. On completion of the process, a saline equilibrium between the muscle and the surrounding salt solutions is achieved.

In Bangladesh, the quantity of exported salted, dried and dehydrated fish products was 2,895 metric tons in 2013-2014 fiscal year, which valued Taka 51.32 crores (DOF, 2015).

Year wise (2004-2014) annual export of salted, dried and dehydrated fish products are given in Figure 1.8.

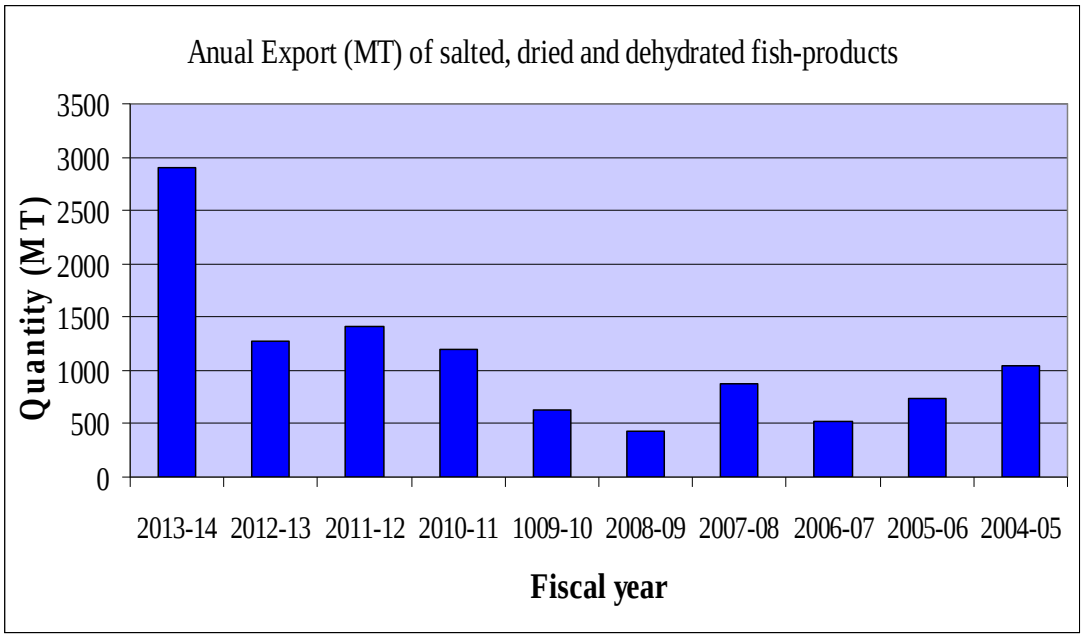


Figure. 1.8. Year wise annual export of salted, dried and dehydrated fish products

Although the method is well known to the fisherman, the effects of preservation on the quality of the products and scientific know-how are unknown to them. The quality of the traditionally salted fishes made by the fishermen is not quite good, due to their ignorance, unawareness and hence not practically suited for healthy reason. For the traditional long term preservation, salting is done without considering the quality of

the final product and the products that marketed are slimy and have unsavory appearance. It does not have universal acceptability. But the main importance is to preserve the fish well for a longer period of time, which can ensure the quality of the products acceptable and good for public health. This can be achieved by improving salting and storage techniques. The most important prerequisite to develop infrastructure for handling, storage, transport and marketing of a product is to know how long each particular product will remain in edible condition. Such information on salted fish products has not yet been accumulated over many years in the past by the technological research institute of Bangladesh. It is most urgent that this knowledge be acquired as much as possible in order that rational development of an industry can take place.

In our country, fish is the main source of dietary protein, and salted fish is an integral part of diet. With increasing demand in the most countries for high quality salted fish, a potential large commercial market is already established. To satisfy this market, practical ways to increase the production of salted fish are needed. Not only more efficient production technique necessary, but also these techniques should be adapted to a variety of fish species to broaden the source of raw materials.

1.9. Use of Turmeric as a newly introduced element in Salting

Turmeric (*Curcuma longa*) rhizome, an important tropical spice, which is a member of the ginger family Zingiberaceae (Aggarwal *et al.,* 2005). The rhizomes of turmeric provide a yellow, flavorful powder when dried and ground and have long been used in Chinese medicines (Joshi *et al.,* 2009).

Turmeric is used as a food additive, preservative and coloring agent in many other Asian countries, including China and South East Asia (Aggarwal *et al.,* 2003;

Chattopadhyay *et al.,* 2004). This spice commonly used to impart yellow color at household level, mostly for spicy preparation, is widely used due to its typical flavor and taste and its extract have various beneficial effects on human health (Nishiyama *et al*., 2005).

Now a days, many natural pigments are used not only as food coloring, but also as a substance that promotes health and well being by preventing or even healing diseases (Amin *et al.,* 2010). Turmeric is also known to have antibacterial properties and pharmacological activities (Sampathu *et al.,* 2000; Egan *et al.,* 2004; Haniffa *et al.,* 2006; Sasmal and Abraham, 2007). Curcumin, used in Indian cooking and credited with therapeutic properties (D'Souza and Prabhu, 2006).

In Bangladesh, turmeric is cheap, easily available and is considered as one of the important ingredient for cooking any kind of dish. Even in some parts of Bangladesh, rural people usually use turmeric for short time preservation of small sized fishes. But, there is very little scientific information about the use of turmeric in fish preservation. Due to the consumer awareness of chemical preservatives, extensive studies are being made on natural preservatives for preservation of meat and fish products.

The present research activities investigate on the effectiveness of the combination of salting and drying process which includes a newer dimension of salting by addition of turmeric with salt as preservative. This investigation on combination of salting and drying process had been termed as "turmeric treated sun-dried salting" (SDS+T).

For better understanding of the methodology of application of turmeric with salt and then drying of the salted fishes to preserve fishes, its performance had been compared with other general salt curing methods those were dry salting, pickle curing, brine salting and sun-dried salting methods as well as their quality assessment have been done at different storage conditions.

1.10. Selected fish species and cause of selection

Among the freshwater fish species, Shol (*Channa striatus*) and Taki (*Channa punctatus*) are snakehead fish belonging to the Channidae family. They are delicious, nutritious and popular to the consumers as well as bear high market price. They are indigenous to many tropical and sub-tropical countries and are fresh water, air breathing, carnivorous fish, which are a valuable source of protein throughout the Asia Pacific region (Mohsin and Ambak, 1983). These fishes widely consumed throughout South-East Asia, China and India for its biomedical properties (Mat Jais, 2007) and reputed positive effects on wound healing (Mat Jais *et al.,* 1997; Baie and Sheikh, 2000). *Channa spp.* fish possess anti-inflammatory (Somchit *et al.,* 2004) and pain reduction (Burstein *et al.*, 2000; Zakaria *et al.,* 2004) properties due to its high levels of arachidonic acid and key amino acids such as aspartic acid, glycine and glutamic acid that are also the key ingredient for polypeptide formation which is responsible for growth and wound healing (Zuraini *et al.*, 2006).

Another popular lean catfish *Mystus tengra* (Tengra) is selected for the present study which is one of the sole species of family Bagridae and a common catfish of the commercial catches of Bangladesh. These fishes have unique test and high demand from all corners of the country as these are economical in price and full of nutrients especially animal protein and fats. The small sized fishes (*e.g.* tengra) are eaten along with bones which are rich in calcium and eyes which are rich in vitamin A (Roos *et al.,* 2002). The calcium obtained by eating whole small fish is equivalent to the amount of calcium obtained from milk (24± 6% small fish, 22 ± 6% milk) (Larsen *et al.*, 2000).

These types of fish are acceptable to all sections of people because of its palatability especially during autumn and winter when they have high oil content.

In Bangladesh, the fatty fishes are generally used for salt-curing. But in the present research work, the above mentioned fishes are selected because these fishes considered widely accepted and preferred by the peoples and they need these fishes when these are not available. Moreover, these fishes are sufficiently fatty. To make these fishes available in off seasons, attempts have been made to cure them using commercial salt. Besides, these highly accepted fishes are not yet tried to preserve using salting or in any other forms.

1.11. Fish collection point (Meghna River)

The sampling sites (fish collection point) were selected at the Meghna River. The River Meghna is one of the major rivers of Bangladesh, especially famous for its great estuary that flows of the Ganges-Padma, the Brahmaputra-Jamuna and the Meghna itself. Spots were randomly selected from Bhairab branch of Meghna River and fish were collected from these sites. The sites were selected on both sides of the river just adjacent to the Bhairab Bridge.

The water of the selected zone was clear and not polluted and abundant fishes were available there.

Location of fish collection point is shown in map (Figure 1.11).

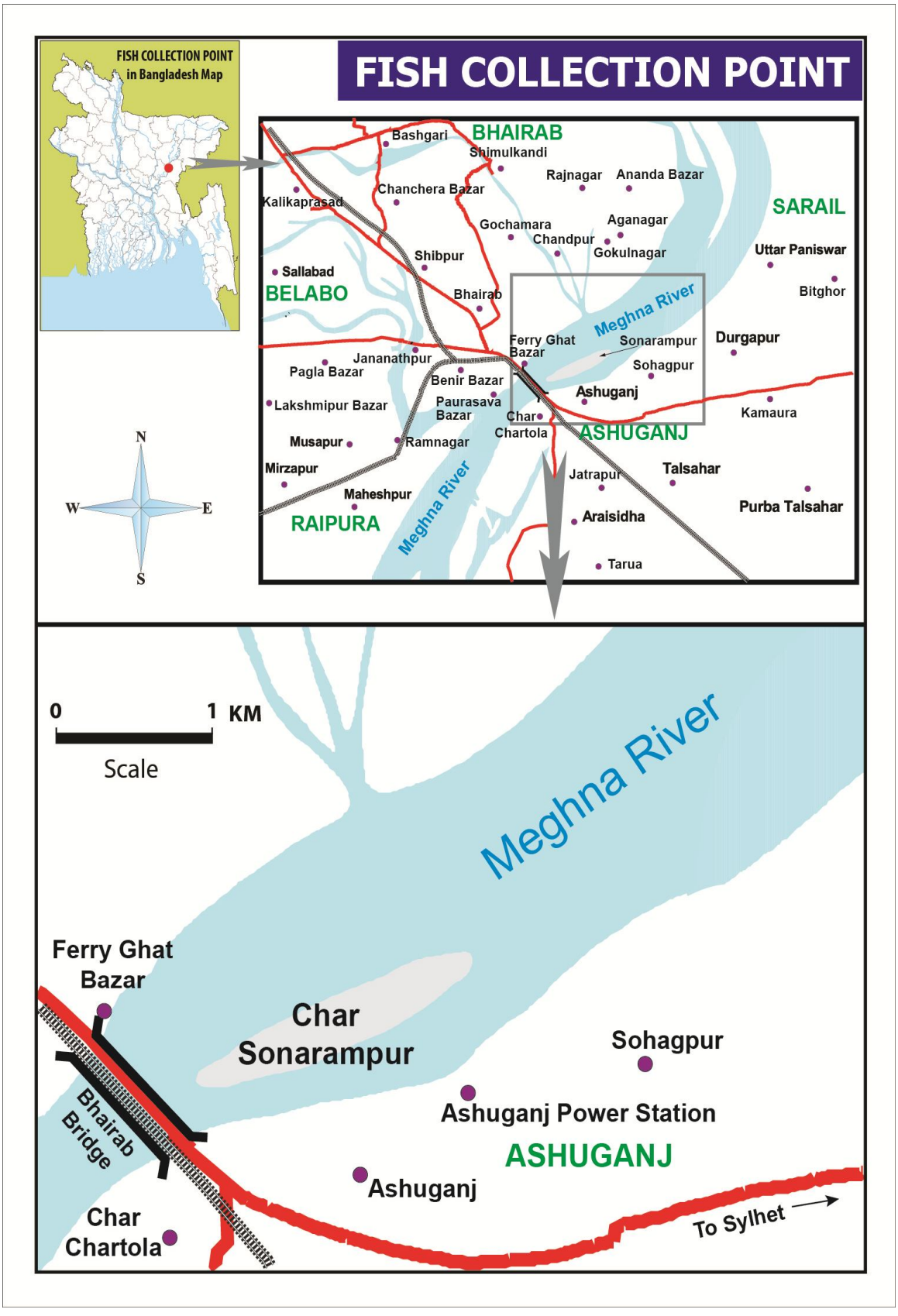


Figure.1.11. Location of fish collection point

1.12. Advantages

Regarding our existing resources, salting is an effective preservation method which can provide the following advantages**:**

1. Salted fish has a universal appeal to the consumer for its excellent flavor, lucrative color, and taste.

2. The method can be approached easily.

3. It is an effective and cost minimizing preservation technique.

4. It does not require any major equipment or machinery; it can be done almost anywhere throughout the year.

5. Moreover, the ingredients such as table salts are abundantly available in our country.

6. Salt hinders the growth and multiplication of spoilage microorganisms.

7. It keeps the fish in edible-form for a longer period of time compared to other preservation methods.

8. It has an additional advantage that refrigeration is not often necessary for the transportation and storage of salted fish.

9. In recent years a considerable amount of foreign currency is also earned through exporting salted and salted-dried fish-products.

1.13. Justification of the research work

1. Development of age-old salt curing techniques for its improvement to the processors.
2. Hazard-free and quality consumable products may be ensured to the consumers.
3. Nutritional aspects of the cured fish may properly be assessed.
4. To make the traditional products more attractive to the consumer and increase the shelf-life of these fish-products.
5. To provide high quality products to assure a good sell, more loyal to customer, increased income and wide marketing.
6. To fulfill nutritional and national goals of developments and decrease of unemployment and poverty.

1.14. Objectives of the research work

The main aims and objectives of the present study includes-

1. To explore the feasibility of preserving indigenous fishes like- shol, taki, tengra, their ability to be preserved in consuming state.
2. Study on the bio-chemical analysis, mineral contents and total bacterial load of fresh experimental fishes - Shol (*Channa striatus*), Taki (*Channa punctatus*) and Tengra (*Mystus tengra*) used in salting methods.
3. Preparation of different types of salt cured fish products.

4. Preparation and taste testing of 'Fish-curry' from different types of freshly processed salt cured fish products.

5. To study the effectiveness of different types of salt-curing methods by assessing nutritional quality, chemical analysis, minerals-composition, total and halophilic bacterial count of freshly prepared salted fish-products.

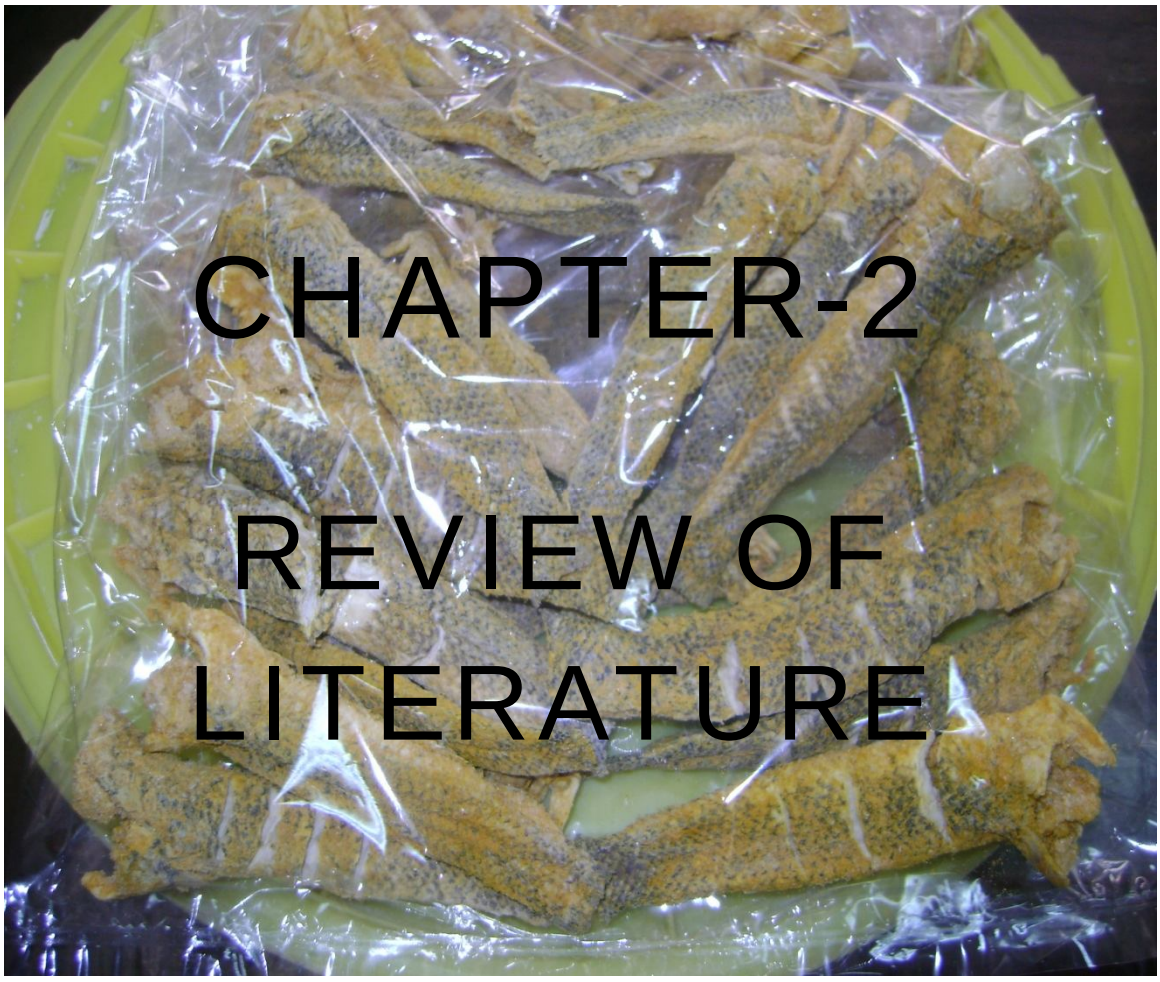
6. Shelf life study using sensory characteristics and sensory scores and quality assessment of the salted fish products stored at ambient (26-32^{0}C) and refrigeration (4^{0}C) temperature to determine the ideal storage condition of different salted products.

7. To develop high quality salted fish-products and to explore the possibilities of the best method used for salt-curing in accordance with the observations and findings of the present study.

Besides above objectives, peer concentration should be made in order to know some different aspects related to this experiment.

Several works has been done in biochemical and microbial quality of different kinds of freshwater fishes only, but very few work have been done on the shelf life study of salted and salted dried fish products. Thus the present investigation was carried out to determine the overall shelf life quality of three freshwater fishes (taki, shol, tengra) treated with different salting process.

The purpose of the present study is to develop an efficient and effective model for curing of various fish species using cheapest ingredients (like salt, turmeric) for the production of high quality end products and transfer the technology to the rural small-scale fisher folks all over Bangladesh.



CHAPTER-2

REVIEW OF LITERATURE

CHAPTER-2
REVIEW OF LITERATURE

Salt curing method (salting) is one of the oldest and traditional methods of fish preservation and have been used since time immemorial throughout the world including Bangladesh. In our country, this easy and cheap method is used mainly for preservation of fatty fishes. Informations are available regarding the various aspects of salt preservation of Herring (*Clupea herengus*), Cod (*G. gadus*), Mackerel (*Scomber japonicus*), Hilsa (*Tanualosa ilisha*) that may forecast different information. Although much research has been conducted on the effect of salting on the quality parameters of these fish-species, very limited reference concerning the shelf life of salted and sun-dried salted fishes has been found in the literature. Therefore, the objective of this research work was to investigate the shelf life and changes to the sensory, proximate, chemical, mineral and bacteriological parameters of five types of salted Shol, Taki and Tengra fishes, as well as the quality characteristics of these salted fish-products during storage both at room and refrigeration temperature.

The literature reviewed here includes few works on salting and salted-products of those species, which are considered to have direct relationship with the present research work. The literature available also reveals that such information has not been accumulated much in the past in our research institutes in Bangladesh, although the shelf life of fishes is very important to develop marketing infrastructure and reduction of post-harvest losses of the commercially important fishes. Unfortunately precise information related to salted fish-products was scarce. The literature thus reviewed here necessarily includes studies conducted with salted and sun-dried salted fishery products prepared from other fishes, which are considered pertinent to the present study.

2.1. Spoilage of fish and its cause

After catching the fish from the water, a series of deteriorative changes starts which eventually render the fish unacceptable. These undesirable changes occurred initially by autolysis followed by microbial action. Due to the action of bacterial and chemical changes, the fish starts to spoil and finally become putrid.

- **Borgstrom (1965)** reported that as the fish spoils and finally becomes putrid, the surface loses its bright-color and becomes covered with a thicker slime, which grows increasingly turbid and lumpy. Finally the color of the slime becomes yellow or brown. The eyes gradually sink and shrink, the pupil becoming cloudy milky and the cornea opaque. The flesh gradually softens until it is easily stripped from the backbone and exudes juice under light pressure. If the fish are not gutted soon after catch the powerful digesting enzymes present in the gut attack the viscera and belly wall, causing discoloration, so called "Belly burn".
- **Daramola *et al.* (2007)** reported that chemical breakdown of protein, fat and water contents contribute to rapid spoilage of fish.
- **Frazier and Westhoff (1978)** reported that fish and other seafood may be spoiled by autolysis, oxidation or bacterial activity or most commonly by combination of these. Fish flesh is considered more perishable than meat because of the less acid reaction of fish flesh that favors microbial growth. The greater the load of bacteria on the fish, the more rapid the spoilage.
- **Huss (1994)** reported that most infections appear to be related to contamination of water or handling of food in unhygienic conditions.
- A large number of fishery resources are simply wasted due to the lack of post harvest technology **(James, 1984).**
- Fish are highly perishable, and they will spoil rapidly if improperly handled. Fresh iced fish generally are spoiled by bacteria, but dried fish are usually spoiled by fungi. In addition to microbial spoilage, fish that contains high levels of lipids (ex-salmon, herrings, and mackerel) are prone to oxidation and become rancid as microbial spoilage occurs **(Jay, 1992).**

- **Reay and Shewan (1949)** mentioned that freshly caught fish have a shining iridescent surface covered with nearly transparent, uniformly and thinly spread slime. The eyes are protruding, bright with a jet of black pupil and transparent cornea. The flesh is soft and flabby, tending to retain finger indentation. Soon after death when the body stiffens (rigor mortis) the flesh become hard and firm.
- According to **Regenstein and Regenstein (1997),** the greater the change in temperature, from the water to icing procedure, has a greater effect on the metabolism of the fish by slowing the postmortem biochemical changes. The rate of a fish's enzyme activities is optimized for its own normal ambient temperature; a marked decrease in temperature also slows this rate significantly. In absence of ice, tropical fish spoil much faster than temperate water fish because of the more rapid growth of non-psychotropic spoilage bacteria.
- The "Spoilage" potential is related to sensory impression and attributes of a product, particularly to the development of off-odor and off-flavor. The spoilage micro flora can be tested for their ability to produce compounds associated with spoilage and to cause rejectable off-odors. Spoilage potential of a bacterium native or invading a fish and other foods of aquatic origins after death depends on its specific nature i.e. on its genera and species **(Sen, 2005).** He also stated that different bacteria have different temperature optimal for survival and growth. Thus, the storage temperature also determines which cause spoilage.

2.2. Quality and quality changes of fish

- **Ababouch (1995)** reported quality changes in Sardines (*Sardinia pilchardus*) stored at ambient temperature.
- **Ahmed *et al.* (1981a)** found that the changes in quality were studied by chemical method and sensory evaluation, storage at higher temperatures was found to have a more deteriorating effect on quality than at lower temperature.

- **Ahmed *et al.* (1986b)** found that the quality of highly perishable flesh food like fish continuously changes during storage. Although, application of modern technology can reduce these deteriorative changes, it can not stop them altogether.
- Generally, the consumers will pay more for fish that they consider to be higher quality. **Connell (1975)** described the quality of food can be determined by the characteristics which make it acceptable to the consumers. The quality of fish is very important to the fish traders as well as the consumers.
- **Dyer (1986)** examined that water temperature, food supply, breeding cycle, size of the fish and the way in which they were killed affect the chemical-composition and quality of fish.
- **Fatima *et al.* (1988)** stated that, fish quality depends upon a number of factors including storage time and storage temperature. As the storage temperature of fishery products is lowered, the storage life is increased.
- **Joseph *et al.* (1986)** studied the chemical, bacteriological and organoleptic quality of cured fish of tamilnadu coast. They reported the main defect of the cured product, which were found to be unhygienic processing, inadequate salting, use of poor quality salt and incomplete drying.
- Quality has been defined as degree of excellence and includes such things as taste, appearance and nutritional content. It might also be said that quality is the composite characteristics that have significance and used for acceptability **(Joseph and Norman, 1996).**
- According to **Mansur (2012)**, fish quality is all those attributes which fish eater or buyer consciously or unconsciously consider or expect to be present in fish in terms of nutritional benefit, dietary satisfaction and that it does not contain any harmful bacteria or pathogen and that it is caught from unpolluted water.
- **Muslemuddin *et al.* (1983)** noted that quality changes were estimated by organoleptic method and also by objective parameters such as loss of water holding capacity and protein solubility.
- Once fatty compounds are oxidized, the breakdown products of lipid oxidation potentially can react with proteins and vitamins, leading to a loss of nutritional value and quality of the fish **(Pokorny, 1981).**

- **Shewan and Liston (1955)** stated that, the keeping quality of fish depends on the species of fish, fishing methods, size, seasons and fishing grounds.
- **Shewan (1961)** reported that the correlation, described above of the organoleptic with the bacterial and chemical data suggests the possibility of objectively assessing the quality of fish.
- Storage time and temperature are the major factors affecting the rate of loss of quality and shelf life of fish **(Srinivasan *et al*., 1997; Whittle, 1997).**
- Because of their unsaturated nature, fish body oils are susceptible to oxidation and also easily develop rancid and unacceptable odors and flavors during storage **(Waterman, 1976).**

2.3. Methods for estimating the quality of fish

- **Ahmed (1988)** observed that degree of spoilage or estimation of quality was determined by there parameters **(i)** Sensory scoring, **(ii)** Total volatile nitrogen (TVN) and (**iii)** Trimethylamine (TMA).
- **Joarder (1974)** studies the qualities of the preserved fillets by Chemical, Physical and sensory examination. The freshness of fish flesh was determined by simple organoleptic methods and by estimation of trimethylamine. The quantitative changes of the bacterial population of various stages of preservation were also determined.
- Methods for assessing the quality of fish are needed so that quality controls and as assurance systems can be properly regulated and monitored **(Wheatson and Lawson, 1985; Suwanrangsi, 1995; Clucas and Ward, 1996).**

There are various methods for assessing the quality of fish, but the followings are most widely used**:**

(i) Organoleptic or sensory evaluation:

- **Ahmed *et al.* (1986b)** found that organoleptic test suitable for the assessment of spoilage while chemical assessment did not give reliable indication.
- Another study by **Akter *et al.* (2013b)** reported that for fish ball prepared from frozen stored Thai pangas, produced moderate to low quality products where their quality parameters including texture, flavor and color of the fish flesh was affected by the quality of the raw material.
- According to **Al-Aswad (2000),** the sensory evaluation considered as one of the important tests used for general acceptance of meat, consumer desire; this includes flavor, tenderness, juiciness and general acceptance. Although the fish muscle have great nutritional value so the consumption can be increased by improving the palatability through flavor, smell, color, appearance, juiciness and tenderness.
- Off-odors and off-flavors, slime formation, gas production, discoloration and changes in texture are obvious signs of spoilage **(Huss, 1994).**
- **Nair *et al*. (1971)** studies that the quality characteristics considered for fish were odor, flavor, texture and color. The average score for each quality factor and the overall mean score for the samples were calculated.
- **Parisi *et al.* (2000)** have studied that, sensory analysis, is of vital importance to the fish industry, for assessment of products freshness. Fish consumers expect a product that is safe and has good appearance, odor, taste and texture and their decision to purchase a fish product is based first on appearance, followed by flavor and then texture.
- **Punna Reddy *et al.* (1991)** observed that the TVN, TMA and Amino Acid Nitrogen values along with the microbial counts significantly influenced the organoleptic quality.
- **Telesara and Kiran (1984)** noted that the texture of fish muscle undergoes changes at low temperature-which are mainly toughening and loss of gelatinousness.

(ii) Chemical Method:

a) TVB-N:

The study of many investigators about the test for volatile bases as a means of spoilage change came forward**:**

- **Botta *et al.* (1984)** examined six different methods for determination of TVB-N in relation to assessment of the quality of fresh cod.
- **Connell (1995)** observed that the upper limit of 30 mg TVB-N per 100 gm of fish muscle as acceptable limit for fresh water fish.
- **Egan *et al.* (1981)** reported that, a level of 35mg N/100g of fish is regarded as unfit for consumption.
- **El-Marrakchi *et al.* (1990)** stated that TVB-N value is affected by species, catching season and region, age and sex of fish.
- **Fatima *et al.* (1988)** stated that TVB-N found in small quantities in fresh fish flesh, and TVB-N during storage is consistent with the pattern of changes in bacterial counts. The levels of these compounds are primarily responsible for the fishy odors which increase as spoilage proceeds.
- **Gokoglu *et al.* (1998)** observed that during the frozen storage (4^0C) of Sardine (*Sardina pilclardus*) TVB-N and TMA-N increased.
- The quality classification of fish and fish products regarding TVB-N values would be ‘‘high quality” up to 25 mg/100 g, ‘‘good quality” up to 30 mg/100 g, ‘‘limit of acceptability” up to 35 mg/100 g, and ‘‘spoilt” above 35 mg/100 g **(Huss, 1995; EU/EC, 2008; Gulsun *et al.*, 2009; Amegovu *et al.*, 2012).**
- **Hye *et al.* (1990)** observed that total volatile base nitrogen (TVB-N) was found to increase with the increase of storage life of Hilsa fish.
- **Karara *et al.* (1984)** found the limit of acceptability of TVB-N value 40 mgN/100g for fresh water fish.
- The concentration of TVB-N in freshly caught fish is typically reported to vary between 5 and 20 mg/100 g **(Muhammet and Sevim, 2007).**

- According to **Orban *et al.* (2011),** TVB-N is considered as an indicator of spoilage of fish muscle tissues, and mentioned that has non-significant correlation between TVB-N and the storage period, and between TVB-N and the sensory quality of fish.
- According to **Rehbein and Oehlenschlaeger (1990),** TVB-N value the some of nitrogen bases (ammonia, di-methylamine, try-methylamine, di-ethylamine) is influenced by fish species, types of fishery products and storage condition.
- **Rubbi and Rahman (1986)** stated that, fish became unacceptable by the total sensory test when TVB-N values increased to about 40 mgN/100g of fresh fish (Immediately after death).
- The degree of spoilage in fish, as characterized by the production of TVB-N or other volatile bases, is different for each species and related to the type of processing and duration of storage. This is so because the composition of fish muscle varies from species to species, which is evidently reflected in differences in the course of decomposition **(Sadok *et al.*, 1996).**
- In case of TVB-N value, the upper limit for acceptability has been drawn at 30 mgN/100g of fish muscle above which fishery products are considered unfit for human consumption **(Sikorski *et al.*, 1989).**
- **Varma *et al.* (1983)** reported that the TVB-N values increased proportionately and value of TVB-N in iced Tilapia was less than the TVB-N value of Tilapia stored at room temperature.

b) Free fatty acid (FFA):

Fish tissues have lipases and phosphor-lipases that remain active both store in room and refrigeration temperature yielding free fatty acids (FFA) during enzymatic hydrolysis. The FFA formation itself does not lead to nutritional losses.

- **Han and Liston (1988)** reported that, FFA are known to cause texture deterioration by interacting with proteins and have been shown to be strongly interrelated with lipid oxidation.
- According to **Molla *et al.* (2007),** the percentages of free fatty acids (above 1.5%) are the determination or indication of unsuitability of the lipid for edible purpose.

(iii) Bacterial Method:

- **Mowlah and Mowlah (1990)** Observed that the bacterial counts were increased when fish kept at room temperature.
- **Shewan (1949)** mentioned that the estimation of the total bacterial load should be capable of correlation with the degree of spoilage.

2.4. Biochemical composition of fish

It is very important to know the biochemical composition of the flesh of fish in order to ascertain its nutritional quality and to determine the proper condition of its processing and preservation. The principal biochemical contents of fish flesh are water, protein, fat and ash. The nutritional status of fish varies due to different factors. There are many reports on the biochemical composition of fresh water and marine fish throughout the world. A brief description of the reports from several workers is given here**:**

- **Abolude and Abdullahi (2005)** reported that, the value of the ash content ranged between 3.85 to 7.97% when studied the proximate analysis and mineral contents in the components of *Clarias garipeneus*.
- **Adewumi *et al.* (2014)** studied comparative analysis of the proximate and elemental compositions of four species of fish; *C. gariepinus, P. obscura*, *T. zilli*, and *T. galilaeus*, they belong to the high-protein, high moisture and low-oil category.
- **Ahmed *et al*. (1981b)** found that *Tilapia nilotica* contained 22.10% protein, 1.32% fat, 1.72% ash, and 74.86% moisture. Female fish contained less protein, fat and ash than those of male fish.
- Food is the main source of energy for all living organisms. Fish and fishery products have got their supremacy to human consumption for their excellent protein source. Besides protein (15-24%), fish flesh also offers fat (0.1-22%),

minerals (1-2%), vitamins (A, D, E and B) and other essential nutrients. Freshwater fishes are important source of essential macro and micronutrients, which is found to be chronically scarce in Bangladesh **(Ahmed and Hasan, 1982).**

- **Al Habib (1990)** estimated the protein content of six fresh water fishes and these fishes contained 11-16.75% protein.
- **Begum and Minar (2012)** studied the body composition of different SIS commonly available in Bangladesh. In the study they found the moisture content was 76.01%, 77.91% and 75.46%; protein content was 16.78%, 17.31% and 18.17%; lipid content was 4.55%, 3.59% and 4.5% and ash content was 1.70%, 1.68% and 1.54% of *G. chapra, C. soborna* and *A. punctata* respectively.
- **Begum *et al.* (2012a)** reported moisture, protein, fat and ash content in *P. hypothalamus* was 78.29%, 12.78%, 16.55% and 1.78% respectively.
- **Beirao (2000)** reported that the chemical composition of the eatable parts of fish, crustaceans and mollusks found 25% ashes and these compositions varies with the species nutritional state, seasonality, age and gonadal condition.
- **Bijayalakshmi *et al.* (2014)** estimated the proximate composition of *A. mola* and moisture and lipid content was found to be 5.4% and 77.19% respectively.
- **Bogard *et al.* (2015)** reported the moisture, protein, fat and ash content was 81.3%, 16.5%, 1.3% and 1.1% in *C. batrachus*; 79.2%, 19.1%, 1.9% and 1.0% in *H. fossilis*; 65.5%, 16.0%, 17.7% and 0.9% in *P. sutchi*; 76.8%, 16.8%, 5.1% and 1.0% in *M. cavasius* respectively.
- **Chakraborty *et al.* (2003)** observed that, the proximate composition of fresh chapila 77.51% moisture, 15.59% protein, 3.58% fat and 2.75% ash and tengra 78.54% moisture, 17.01% protein, 2.15% fat, 2.06% ash content in edible portion. He concluded that the freshwater fishes of Bangladesh mostly belong to high protein-low oil and medium oil group.
- **Chandrashekhar and Deosthale (1993)** found that a wide variation existed between species in protein content (marine 8% - 21% and freshwater 13.5%-17.3%) and fat content (marine 0.7-14.7% and fresh water 0.6-1.3%).

- According to **Chukwu (2009),** mean moisture, protein, lipid, ash, fiber contents of raw tilapia fish (*Oreochromis nilotius*) were 70.15± 0.04, 23.06±0.04, 12.85+0.05, 28.16±0.02, 1.91±0.01 respectively.
- The crude protein of fresh fish ranges between 14-20% and higher levels are obtained during winter season **(Clucas and Ward, 1996).**
- **Dempson *et al.* (2004)** stated that the percentages of water are good indicator of its relative contents of energy, proteins and lipids. The lower the percentage of water, grater the lipids and protein contents and higher the energy density of the fish.
- **Devadasan *et al.* (1978)** determined the proximate composition of *Labeo rohita, Catla catla, Cirrhinus mrigala, Labeo calbasu, Mystus seenghala* and *Wallago attu.* They found moisture content in the above raw fishes was 77.71, 76.28, 77.47, 76.26, 80.83 and 77.11% respectively. The protein content was 19.60, 17.74, 18.72, 16.66 and 15.76% respectively. The protein content was 19.60, 17.74, 18.72, 16.66 and 15.76% respectively. The lipid content was 1.83, 1.32, 2.89, 1.19 and 3.16% whereas the ash content of the fishes was 1.31, 0.93, 1.40, 1.03, 0.91 and 0.72% respectively.
- **FAO (1991)** reported that normally fish contains 72% water, 19% protein, 8% fat, 0.5% calcium 0.25% phosphorus and 0.1% vitamin A, D, B and C etc.
- **Fawole *et al.* (2007)** studied the proximate composition of *O. niloticus, S. galilaeus, C. gariepinu and H. niloticus and* show encouragingly high crude protein contents of 38.40%, 41.28%, 44.28% and 46.28% respectively.
- **Gheyasuddin *et al.* (1979)** determined the proximate composition of some important fishes of the Bay of Bengal such as silver pomfret (*Stromateus cinereus*), ribbon fish (*Trichurus haumela*), Indian salmon (*Polynemus indicus*) and Bomby duck (*Harpodon nehareus*). The moisture content of the above fish species was 78.70, 77.34, 77.24 and 82.21% respectively. The protein content was 16.70, 16.60, 17.67 and 15.10%. The lipid content was 2.10, 2.52, 2.61 and 1.53% whereas the ash content was 1.95, 2.60, 2.40 and 1.20% respectively.
- **Gopalan *et al.* (2007)** determined the moisture content in *Mystus vitatus, Labeo rohita, Calta catla, Puntius* sp. and they obtained the result as 70%, 76.2%, 73.2%, 70% and 75% respectively. They also observed that *Mystus vitatus*

contained 6.4% fat and 18.85% protein, *Mystus bleckeri* contained 2.73% fat and 18.8% protein, *A. mola* contained 4.1% fat and 18% protein. *Catla catla* contained 2.4% fat and 19.5% protein and *Puntius sp*. contained 2.4% fat and 18.1% protein.

- **Govindan (1985)** analyzed the amount of protein content that was present in different fishes from both the fresh water and marine environment and he obtained that fish contained 9-25% of protein and in most cases the limit was 16 to 19%.
- **Hossain *et al.* (1999)** studied the maximum and minimum protein contents among the seven species and found that, higher protein contents were in *A. testudineus* (19.63%) and in *C. punctatus* (19.13%) whereas lower values were found in *C. batrachus* (14.87%) and in *M. vittatus* (15.62%).
- **Irianto and Irianto (1997)** reported that Nile Tilapia in Indonesia contains percentage of moisture, protein, fat and ash 76.8, 20.1, 2.2 and 1.0 respectively.
- **Islam (1977)** determined the proximate composition of fresh silver Jew fish (*Otolithes argenteus*), ribbon fish (*Trichiurus haumela*), Jwelled shad *(Ilisha fligera*) and dog fish (*Scoliodon sorrakowha*). Moisture content of these fishes was found to be 78.20, 79.29, 77.22 and 81.75% respectively. Protein contents were 16.31, 17.81, 16.26 and 20.93%. Lipid content was 2.47, 2.68, 3.10 and 0.92% and ash content was 3.20, 3.60, 3.30 and 2.40% respectively.
- **Islam *et al*. (2003)** experimented on the nutritional composition of a popular fresh water species *Cirrhinas reba* and found that the ash content of this species was 1.7%
- According to **Islam and Joadder (2005),** seasonal variation in the percentage of the proximate composition of male and female Gobi fish (*Glossogobius giuris*) was found in the range of moisture 78.05 to 80.26% and 79.08 to 81.41%; Protein 14.09 to 16.03% and 14.08 to 15.56%; fat 0.95 to 1.52% and 1.06 to 1.82%; ash 2.15 to 2.92% respectively. He concluded that, *G. giuris* is a 'low fat-high protein' fish.
- **Islam *et al.* (2010)** investigated the proximate composition of *Odontamblyopus rubicundus* commonly known as 'Lal Chewa' collected from the Meghna river estuary and Moisture (77.427 ± 0.18%), protein (15.135 ± 0.02 %), fat (5.273 ± 0.15%) and ash (2.035 ± 0.16%) contents of this fish were measured.

- **Islam *et al.* (2012)** found the percent of moisture content of whole *A. mola* was 75.79 ± 0.88 and protein, lipid and ash content was 15.40 ± 0.24%, 5.48 ± 0.02%, 1.60 ± 0.006% respectively.
- **Joseph *et al.* (1992)** reported that proximate composition of *Hirundichthys coramendelensis* was moisture 77.17%, protein 20.38%, fat 0.30-0.50% and ash 0.92-1.29% and *Cypselurus suttoni* contains moisture 76.29%, protein 20.88%, fat 0.55-1.27% and ash 0.84-1.18%.
- "Deshio Khadyadrobyer pustiman" published by Institute of Nutrition and Food Science **(Kamaluddin *et al.*, 1980)** reported that freshwater fishes contained 70-80% moisture, 15-18% protein, 0.1-8% fat.
- **Khuda *et al.* (1962)** estimated that the amount of moisture and ash content in *Puntius sophorae* and they observed that the fresh fish contained 77.14% moisture and 1.48% ash. They also estimated the amount of protein content on the two stages of growth of *Labeo rohita, Cirrhina mrigala and Labeo calbasu* and they found that in the cases of groups of fishes ranging from 55.5-78.5 gm, body weight and the crude protein level varied from 17.18% to 19.56% but, for higher body weight from 10.9-16.8 gm. They again reported that the decrease in protein and moisture content and increase in fat content were related with the increase of age, in case of carps.
- **Latifa *et al.* (1981)** reported protein 19.28% and 24.0%, Moisture 83.11% and 82.10%, fat 3.84% and 2.40%, ash 13.97% and 19.83%, in adults and juvenile of *Puntius stigma* respectively.
- **Martin *et al.* (1995)** studies on Europian catfish for proximate composition and reported that fat content varied with season and averaged 3.3% (range 0.6-8.6).
- **Mazumder *et al.* (2008)** analyzed the proximate composition of some small indigenous fish species. Protein was estimated in *A. mola* (18.46 %), *G. chapra* (15.23 %), *P. chola* (14.08%), *C. nama* (18.26%), *P. atherinoides* (15.84%) and in *A. coila* (16.99%) respectively. Fat content was recorded as 4.10%, 5.41%, 3.05%, 1.53%, 2.24% and 3.53% respectively in the six species of fish. The highest level of moisture content was found in *C. nama* (78.62%) and the lowest was in *A. coila* (65.88%). The percentage of ash content was highest in *C. nama* (3.92%) and lowest in *G. chapra* (1.55%).

- According to **Molla *et al.* (2007),** Bangladeshi fresh water fish *Mystus vittatus* was found to contain moisture 76.98%, protein 10.94%, lipid 1.89%, ash 1.01% and calcium 521.072 mg/100g respectively.
- **Mustafa *et al.* (2012)** stated that, the average moisture, protein, fat and ash contents of fresh hilsha and sarputi were 69.36±0.71% and 71.39±0.61%, 19.56±0.44% and 16.73±0.92%, 9.49±0.54% and 9.0±1.09%, 2.27±0.16% and 2.02±0.24% respectively.
- Proximate composition generally comprises the estimation of moisture, protein, fat and ash contents of the fresh fish body. The percentage composition of these constituents accounts for about 96%-98% of the total tissue constituents in fish **(Nowsad, 2007).**
- **Rahman (1989)** reported that the crude lipid in some Bangladeshi zeol fish ranged from 2.18% to 9.38%.
- **Rubbi *et al.* (1985)** observed that kachki, (*Corica soborna)* is rich in protein about 12.23% and fat contained about 3.6% as well as calcium and phosphorus.
- **Rubbi *et al.* (1987)** studied the proximate composition of 27 species of fresh water fish both scaly and non scaly fish and found moisture between 72.1-83.69, Protein; 11.9-21.9, Fat 0.8-15.0 and ash between 0.8-15.0 and ash between 0.8-5.11 g %.
- **Shahiduzzaman *et al.* (2004)** conducted an investigation on biochemical composition in batashi fish (*Clupisoma atherinoides).* He found fat content around 3%.
- According to **Viji *et al.* (2014)** proximate composition of the fresh cat fish (*Pangasianodon hypophhthalmus*) meat showed 77% moisture, 16.5% protein, 4% crude fat and 0.97% ash.
- Determination of some proximate profiles such as protein content, lipid, ash and other nutrients is often necessary to ensure that they are within the range of dietary requirement and commercial specifications **(Watchman, 2000).**

2.5. Processing and preservation of fish

Preservation is the methods by which fishes are kept in a “fresh state" as possible with minimum losses in flavor, texture, taste, odor, nutritive value, weight and digestibility of flesh. The principle of preservation based on such measures, which will keep a control on the factors that cause spoilage of fish. Some of the related reports are given as follows:

- **Bhattacharya and Chaudhuri (1990)** studied the quality change of *Clarias batrachus* while storing the fish at different storage temperature. As per organoleptic assessment, they found that the fish attained shelf life of 6^{th}, 8^{th} and 15 days at 37^0C, 22^0C and 0^0C respectively.
- **Bramsnaes (1965)** observed that during the summer season fish often have a shorter storage life than at other season.
- It has been observed that different processing methods have different effects on the nutritional compositions of fish. This is because heating, freezing and exposure to high concentration of salt lead to chemical and physical changes and therefore digestibility is increased **(Chukwu and Shaba, 2009).**
- According to **De Koning and Mol (1991),** controlling the temperature of fish is perhaps the most important element in the preservation of fresh fish.
- According to **Gopakumar (1997),** the pH level, temperature of storage, microbial load, salt content and aw are factors generally considered to influence storage life and safety of cured fish products.
- **Martenik and Jacobs (1963)** and **Doha (1964)** indicated that nutritionally the dehydrated products are very good and neither the nutritive value nor the digestibility of the protein is adversely affected due to dehydration process.
- The purpose of preservation is to reduce the moisture content of the fish because micro-organism that is responsible for spoilage and wastage cannot survive without moisture **(Ogunleye, 2006).**
- According to organoleptic evaluation Pabda fish was acceptable for 180, 210 and 240 in polythene pack, freeze pack and aluminum foil **(Rabbane *et al.*, 2007).**

- Preservation must be seen as a way of storing excess foods that are abundantly available at certain times of the year, so that they can be consumed in times when food is scarce and decreases the nutritional value **(Van Berkel *et al.*, 2004).**
- According to **Widjaja *et al.* (2009); Huda *et al.* (2010); Ahmed *et al.* (2010a); Kayim and Can (2010),** fish processing often results in the concentration of nutrients like crude protein and fat and for that, the quality of fish products is influenced by fish species, different method of processing, storage practices and duration of storage.

Salting/ Salt Curing

The ratio of salt to fish used in the west coast of India varies from place to place and also according to size of fish. But in Bangladesh, there is no recognized ratio of salt to fish. As a result no established ratio is maintained during salt processing of fish in any fish processing industry. They use the ratio of salt to fish by their assumption and the ratio is more or less 1:4.

- **Akter (2013)** prepared fish pickle using Thai pangas and reported that, the product prepared from dorsal muscle and whole fish muscle using both mustard oil and soybean oil was acceptable and their shelf life was 15 days at room temperature and more than 60 days under refrigeration condition.
- **Bahri *et al*. (2006)** observed that pH values in samples of salted trout, anchovy and mirror carp fish were found between 6.41-6.70 at the beginning and then changed depending on the storage period and varied between 5.34- 6.81.
- According to **Balachandran (2001)** in brine salt curing method, the amount of salt required to make a saturated brine solution is to use 26% salt by weight with pure clean drinking water.
- **Beatty and Fougere (1957)** demonstrated that thickness of flesh has a pronounced effect on salt uptake. Thicker fillets obtained lower amount of salt than that of thinner one when curing time in both the cases remained the same.
- **Berhimpon *et al.* (1990)** carried out a study on the salting and drying of low fat yellow tail (*Trychurus meculloch*i). They found that the rate of salt uptake

increased with brine concentration to reach constant values dependent upon brine concentration. In saturated brine the salt content reached 41% (dry basis) after 20 hrs and the salt uptake increased with time.

- The simplicity of the salting process, the low cost of production and the case with which it combines with other preservation methods, such as drying or smoking, has led to its popularity and extensive use **(Berhimpon *et al.*, 1991).**
- **Chakraborty *et al.* (1997)** reported that moisture content of dry salted, wet salted and sun dried salted fish showed significant decreases ($P<0.05$) from an initial 71.80% to 37.06%, 44.90% and 25.95% respectively.
- Salt retards the bacterial action and aids the removal of water by osmosis. When fishes are salted prior to drying, moisture content reaches between 35% and 45% in the flesh depending on the concentration of salt, which inhibit the bacterial growth **(Chinivasagm and Vidanapthirama, 1982).**
- According to **FAO (1992),** when water content falls below 25% of the wet weight, bacterial action stops, and if the water content is further reduced to below 15%, mold ceases to grow. When salt is added to the fish before drying, less water needs to be removed to achieve the same effect, and the product with a water content of 35% to 45%, depending on amount of salt present, is often dry enough to inhibit the growth of molds and bacteria under most climatic condition.
- **Gram (1991)** stated that, salt may have a significant inhibitory effect on mesophilic spoilage flora.
- **Islam (2007)** observed in dry salting, pickle salting, brine salting and experimental salting method moisture (%) content of chapila fish was 39.1%, 48.3%, 56.3 and 41.4% and kachki fish was 39.3%, 51.3%, 56.0% and 41.6% respectively.
- **Jittinandana *et al.* (2002)** have reported that brine concentration and time of salting affect the pH, protein solubility, water-holding capacity, water activity and textural properties of brine-salted rainbow- trout fillets.
- **Joseph *et al.* (1986)** have reported TVBN content up to 72.7% in salted anchovy and 105.0% in salted sole.
- **Josephson and Lindsay (1987)** studied the influence of processing on the volatile compounds characterizing the flavor of pickled fish (*Osmerus mordex*).

- **Kamruzzaman (1998)** reported the protein content of non-egg bearing salted Hilsa was 17.05% and it increases up to 21.96% after 16 days of ripening period.
- **Khan (1998)** found that the initial protein content of ordinary salt and pure salt processed Hilsa were 16.42% and 16.23% respectively and the final protein content were 26.58% and 26.87% respectively on 16^{th} day of observation at room temperature (28-32^0C).
- Dry salting is performed by distribution of solid salt over the fish surface, resulting in brine due to extraction of moisture from the fish muscle **(Lauritzsen, 2004).**
- **Mansur *et al.* (1997)** reported moisture, protein, lipid and ash contents of 46.87%, 18.75%, 19.93% and 1.30% respectively in salted mackerel (*Scomber scombrus*) fish.
- **Mansur *et al.* (1998)** found that the moisture content of dry-salted Hilsa ranged from 52.98% to 35.08% and protein and lipid content for dry salted Hilsa ranged 17.37% to 25.12% and 26.93% to 17.36% respectively on during 12 weeks of observation.
- **Muslemuddin *et al.* (1986)** described the effect of temperature and salt concentration of brine during short term preservation of Mola fish. They suggested that shelf-life of fish found to be increase with the increase of salt concentration in brine.
- **Mustafa *et al.* (2012)** stated that, moisture content of pickle salted, dry salted, mixed salted and brine salted hilsha and sarputi were 42.94% and 42.59%, 39.77% and 39.04%, 36.45% and 31.75%, 47.27% and 46.39% respectively.
- **Pillai *et al.* (1961)** determined that moisture content of the salted fish product varied from 20-50%, salt content between 7-21.5% and protein between 23-45%.
- **Rahman (1996)** stated that the ash content in raw (beheaded) fish was 1.68% which increased up to 18.92% for dry salted Hilsa on 14^{th} day of observation.
- **Rohomania *et al.* (2014)** shown the variation of proximate composition of dorsal and ventral parts of salted *T. ilisha*. The found that dorsal and ventral part was 45.13% and 40.20% moisture; 20.79% and 21.48% protein; 16.89% and 19.54% fat; 16..65% and 18.35% ash respectively.

- According to **Rubbi *et al.* (1981)** temperature was found to have little effect on salt up take and water loss, that's mean the greater the temperature the greater was the up take of salt and water loss, quality remained better at low temperature.
- It has been reported by **Sachithananthan *et al.* (1985)** that the composition of some salted dried marine fish products were 41.4% to 50.6% for protein, 1.0% to 6.5% for fat and 16.1% 30.6% ash.
- **Sayed (1997)** found that dry salted products, ash content slightly decreased after 15 days of observation.
- According to **Sharma *et al.* (2013),** dry salted and wet salted *G. chapra* had moisture content of 17.26% and 20.12%; crude protein content was 54.20% and 50.34%; total lipid content was 11.20% and 10.48%; ash content was 17.04% and 18.25% respectively.
- **Shewan (1961)** described that salting is in general variants of two fundamental methods, i.e., dry salting and wet salting or salt pickling. The former is used to cure the non-fatty species while the latter is appealed for curing fatty fishes such as herring, mackerel, river shad etc.
- **Siddiqui (1993)** studied on the shelf-life of salted Hilsa packed in polythene bag and kept at 4^0C had better quality than those kept in room temperature.
- **Siddiqui (1993)** and **Rahman (1996)** studied on the effect of salting on the chemical composition, protein solubility and essential amino acid composition of Hilsa fish showed that in addition to moisture, there was a considerable loss of lipid during salting.
- During dry salting, weight loss by the fish is the highest **(Voskresensky, 1958).**
- The fish kept at 0% salt concentration at 30^0C have shelf life of 6.25 hours while at 15% salt concentrations have shelf life of 28 hours at the same storage temperature **(Wahed *et al.*, 2005).**
- Use of crude NaCl which contains impurities such as chlorides, sulfates, calcium, and heavy metals accelerates lipid oxidation during fish processing and will adversely affect the overall quality of the finished product **(Yankah *et al.*, 1996).**

Turmeric as food additive

- The active ingredient of turmeric (*Curcuma longa*) having pesticidal action is curcumin (diferuloyl methane) **(Aggarwal *et al.*, 2007).**
- **Akter *et al.* (2007)** reported that a combination of turmeric and salt treated dried mola (*Amblypharyngodon mola*) contained 42.12% to 45.35% protein, 8.2 to 8.26% fat, 15.46 to 20.12% moisture; the shelf-life of those fishes was 4-5 months and the average sensory scores of these fishes was 4.87. The turmeric and salted treated dried fishes showed a prime level of effectiveness possessing a longer shelf-life, higher sensory scores, shorter curing period, better quality and more favorable nutrient properties for preservation than any other type of salt curing methods like dry salting, pickle salting, and brine salting.
- Turmeric is coded as E100 when used as a food additive and used in product systems that were packaged to protect them from sunlight. Investigations into the low incidence of colo-rectal cancer amongst ethnic groups with a large intake of curries compared with the indigenous population had discovered that some active ingredients of turmeric appear to have anti-cancer properties **(Cao *et al.*, 2007).**
- Turmeric is an orange yellow color derived from the root of the curcuma (turmeric) plant. Curcumin consists of three principal coloring components. It consists essentially of curcumins i.e. the coloring principle (1E, 6E)-1, 7-bis-(4-hydroxy-3- methoxyphenyl)-hepta-1, 6-diene-3, 5-dione and its desmethoxy- and bisdesmethoxy- derivatives **(ESFA, 2010).**
- **Shakila *et al.* (1996)** reported that turmeric exhibited the moderate effect on the formation of toxic histamine in mackerel.

Sun drying and sun-dried salting

Dry fish is the cheapest source of animal protein for the rural people of Bangladesh. As fresh fish is not available regularly throughout the year, dried fish is in demand during period of scarcity. In most of the studies the bio-chemical composition of sun

dried & salted dehydrated sun dried fisher products were found to have a wide range of variation.

- According to **Alam (2012)** salted-dehydrated fish is usually produced where a pre-treatment of salt is done in the fish during sundrying. Small amount of salt makes the texture compact, reduces the effects of contamination, destroys some of the bacteria and helps release water from the fish so that drying becomes easier and quick**.**
- **Azam *et al*. (2003b)** studied biochemical assessment of fourteen selected dried fish and observed that moisture, protein, fat and ash content ranging from 18.23-23.61%, 40.69-66.52%, 7.1-26.13% and 5.08-12.14% respectively.
- According to **Begum *et al.* (2011),** sun-dried tengra (*Mystus vittatus*) fish contained 16.01% moisture, 60.45% protein, 9.25% fat and 9.33% ash.
- According to **Bhattacharyya *et al.* (1985)** the markets samples of sun-dried *Gudusia chapra* had moisture ranging from 9.61% to 18.64%.
- **Bhuiyan *et al.* (1990)** reported that the sun dried fish should not contain more than 15% moisture to avoid the mould and bacterial growth.
- **Bhuiyan (1992)** observed 9.21% - 6.84% lipid in dried marine fishes.
- During the removal of moisture, drying rate was different at different stages. The drying rate depends on the temperature, internal diffusion, thickness and leanness of the fish (**Brennan *et al.*, 1969; Burgess *et al.*, 1965; Waterman, 1976).**
- **Chakraborty *et al.* (1997)** reported that in sun dried salted products, protein content changed from 17.06% to 35%.
- In Bangladesh, fishes are usually dried without preliminary salt treatment. Salt retards the bacterial action and aids the removal of water by osmosis. When fishes are salted prior to drying, moisture content reaches between 35% and 45% in the flesh depending on the concentration of salt, which inhibit the bacterial growth **(Curran and Trim, 1982).**
- **Faturoti (1985)** made a study on biological utilization of sun-dried African catfish (*Clarias nigrodigitus*). He showed that the gutted dried fish samples had a chemical composition of 6.27% to 10.92% moisture, 55.02% to 63.05% crude protein and 14.31% to 20.60% ash.

- **Flowra and Tumpa (2012)** determined the proximate composition of five sun-dried fish samples of *P.ticto, L.bata, W. attu, C. striatus* and *P. sp* and found that the moisture content ranged from 12.13% to 18.18%, protein varied between 28.20% to 51.19%, lipid ranged from 5.38% to 15.86% and ash content varied from 10.78% to 15.67% respectively.
- **Flowra *et al.* (2012)** studied the proximate composition of five dried fish samples of *Mystus vittatus*, *Channa punctatus*, *Chanda nama*, *Corica soborna* and *Trichuirus haumela* and found that the moisture content ranged from 14.06% to 24.58%, protein varied between 44.08% to 65.65% (moisture basis) and 53.45% to 76.39% (dry matter basis), lipid content of the selected dried fishes ranged from 1.91% to 17.76% (moisture basis) and 2.31% to 21.54% (dry matter basis). Ash content varied from 9.63% to 22.73% (moisture basis) and 11.21% to 28.15% (dry matter basis).
- Normally the sun-dried fishes contain an average of 10 to 20% of moisture 60 to 80% protein **(Haque, 2004).**
- **Humayun (1985)** stated that sun dried Rohu fish contained 10.30% moisture, 73.93% protein, 5.60% lipid and 9.31% ash.
- **Hussain *et al.* (1992)** found that the quality of the salted-dried headless and dressed Gonia fish products was better than the unsalted samples.
- **Islam (1982)** studied the proximate composition of traditionally dried Rohu fish and observed the moisture and protein content as 9.07% and 73.26%.
- **Islam (2001)** observed that total volatile base nitrogen (TVB-N) content of traditional dried ribbon fish, Bombay duck, Big-eye tuna, silver Jew fish and Chinese pomfret ranged from 16.56 to 44.83 mg/ 100gm.
- **Islam (2005a)** found that, in storage condition the salted sun dried *P. argenteus* has been found to have 16.6% to 18.9% moisture, 65.73% to 67.7% protein, 6.2% to 6.6% fat and 5.8% to 6.3% ash.
- **Islam (2005b)** in his project report on experimental sun dried *Amblypharyngodon mola* samples in fresh dried conditions contained moisture ranged from 15% to 20%, protein from 59% to 70%, fat from 6.1% to 8.8% and ash from 6% to 11%. He also reported that the experimental open sun dried *Gudusia chapra* contained

moisture content ranged from 15.5 to 20%, protein from 55.1% to 60.1%, fat from 15% to 19%, ash from 5.9% to 9.6%.

- A survey was conducted by the Indian Central Institute of Fisheries Technology (at four fish drying yards on the species used, drying practices and the quality of the dried products).The moisture content of the samples varied over a large range, from 12.3% to 54%. Likewise, protein 17.2% to 78%, fat 3.7% to 17.8% and ash 1.4% to 21.6% content varied widely over the 23 species analyzed **(Kalaimani and Kamasastri, 1998)**.
- **Kamruzzaman (1992)** suggested that when water content of fish falls below 25% of the wet weight, bacterial action stop. When the water content is further reduced to bellow 15%, mould ceases to grow. When salt is added to the fish before drying, less water needs to be removed to achieve the same effect, and the product with a water content of 35-45%, depending on amount of salt present, is often dry enough to inhibit the growth of molds and bacteria under most-climatic condition.
- **Keshvani (1964)** observed that well dried fish could be stored for up to one year in sealed polythene bags without serious loss of quality.
- **Kiaye (2004)** stated that brining reduces the microorganisms count on dry fish.
- For the protection of human health and production of safe dried fish, alternative additives such as salt, different herbal products such as chili, turmeric, neem powder have been suggested by **Lithi *et al.* (2012)** and **Roy *et al.* (2014).** The suitability of herbal pesticides including turmeric and neem in repelling dry fish insect.
- **Mansur *et al.* (1993)** conducted an investigation on the quality of dried fish where they found that the composition of traditional sun dried *Labeo rohita* contained 16.20% moisture, 59.85% protein, 10.80% lipid and 12.79% ash. For *Barbus sarana, Channa striatus* and *Gudusia chapra* that values of moisture, protein, lipid and ash were 16.71, 59.29, 13.26 and 10.54; 10.77, 70.29, 2.87 and 12.97; 18.32, 51.52, 11.76 and 16.73% respectively.
- **Mehbub (2004)** observed that, Moisture, protein, lipid and ash content of the dried product of Silver Jew fish (*Johnius argentatus*), Ribbon fish (*Trichiurus baumela*), Bombay duck (*Harpodon nehereus*) varied in the range of 11.48 to 16.83%, 58.51 to 68.49%, 6.08 to 8.62% and 13.05 to 14.88% respectively.

- **Morshed *et al.* (2004)** observed raw Bombay duck fishes contained 81% moisture, 15.3% protein, 1.30% fat and 1.90% ash and experimentally sun dried Bombay duck contained 21.8% moisture, 60.5% protein, 10.6% ash and 8% fat. They also observed the dried fishes when kept in polythene bags at low temperature of 4^0C, kept the fish in good condition for a period of 8 months.
- **Nabi *et al.* (2007)** studied the effects of hot water, turmeric solution and both hot water with turmeric solution on the shelf-life on sundried *Puntius ticto* and reported that the organoleptic scores revealed that the sample treated with turmeric solution had maximum shelf-life.
- **Nurullah (2005)** reported that the proximate composition of traditional dried SIS namely Mola, Dhela, Puni, Batashi, Tengra, Chapila were ranged from moisture content 23.26% to 26.42%, protein content range from 48.60 to 52.20%, lipid content range from 8.40% to 11.52% and ash content range from 12.20% to 19.32%. The author also reported the TVB-N value of the above mentioned species ranged from 20.30-28.40mg/100g.
- **Qudrat-E- Khuda *et al.* (1962)** estimated the protein content of sun dried 'shutki' of both marine and fresh water fishes varied from 55.50 to 74.18% in *Lates surinamensis* (Katkoi) and *Channa marulius* (Gazar) respectively.
- According to **Rahman *et al.* (1978),** the moisture, protein, lipid, and ash content of sun-dried rohu fish were 9.8%, 80.9%, 4.8%, 4.4% respectively
- **Rahman *et al.* (1982)** studied the proximate composition of marine dried fishes and found moisture between 15 and 29.5%, protein between 48 and 68.9% and fat between 7.2 and 12.3%.
- **Reza (2002)** observed that the moisture content of the dried product of Silver Jew fish, Ribbon fish, Bombay duck, Chinese Pomfret and Big-eye tuna fish-products ranged from 18.0 to 29.8% with the highest value in Big-eye tuna collected from retail market. The range of protein contents was 42.51 to 56.58%. Lipid and ash content were in the range of 1.97 to 8.08% and 15.5 to 26.84% respectively.
- **Roy *et al.* (2014)** stated that, dried fish treated with salt and herbal treatment (turmeric powder and chili powder) found organoleptically excellent quality.
- **Rubbi *et al.* (1982)** reported that dried fish wrapped in polythene bag were found to have better self-life than fish kept in gunny bags.

- **Saha and Choudhury (1994)** made nutritional investigations on the sun dried fish of West Bengal and reported that the sun-dried fish are nutritionally enriched and major source of concentrated form of nutrient for the all people. Their moisture content ranged from 12.6% to 17.5%, body fat from 2.7 to 14% and ash 8.7% to 27.5%.
- **Saha (1999)** observed 36.50% to 82.80% of moisture and 34.90% to 46.70% of protein in thirteen sun dried fishes which are of known as small indigenous species (SIS).
- **Siddique and Akter (2011)** studed the charges of nutritional value of sea dry fishes (*Harpodon nehereus, Johnius dussumieri and Lepturacanthus savala*) during 2 years of storage period. The findings of the study showed that nutritional value of dry fishes deteriorate with the increasing of storage period.
- **Siddiky (2015)** observed percentage of moisture, protein, fat and ash of five dried Sea fish. The value was 22.62, 60.61, 7.35 and 8.56 for Chinese pomfret; 17.93, 61.53, 7.44 and 12.78 for Bombay duck; 18.66, 58.36, 11.24 and 11.22 for Ribbon fish; 22.09, 50.16, 9.17 and 17.57 for Banded needle fish and 22.13, 52.82, 10.40 and 15.17 for Phasa fish respectively.
- According to **Siringan *et al.* (2006),** high salt environment decrease both an endogenous proteinase activities in raw materials and proteinase produced by certain bacteria.
- **Watanabe (1971)** and **Toots (1972)** recommended sealing dried fish in the bags, the later emphasizing the need to cool the fish after thorough drying and to store the product in a cool shady place to avoid sweating.
- Recommended intakes of proteins for adults is 0+75 g per kg of healthy body weight per day. For example, a 70 kg male needs 53 g of proteins per day. To fulfill this need he has to consume 66 g of dried fish. A 55 kg female needs 41 g of proteins per day. Or 51 g of dried fish **(Whitney *et al.*, 2002).**
- **Yu (1985)** observed that drying time up to 40 hours for sun drying and 8 hours for 20% salted oven-dried fish sample was judged the best.

2.6. Minerals and its role on fish

Considerable information is available in the literature about mineral contents of fishes elsewhere in the world, but systematically little work has been done about mineral contents of salted and sun-dried salted fish products.

- WHO (µg/g) suggested the maximum limits of Mn, Cu, Zn, Pb, and Cd for fish are 1 µg/g, 30 µg/g, 100 µg/g, 2 µg/g, 1 µg/g and FAO suggests the maximum limits of Cu, Zn, Cd for fish are 10 µg/g, 100 µg/g and 0.2 µg/g **(Adedeji and Okocha, 2011).**
- Acording to **Adeniyi *et al.* (2012)** minerals included potassium (91.51-102.86 mg/kg), calcium (16.32-24.53 mg/kg), sodium (59.21-75.12 mg/kg) and magnesium (29.61-41.44 mg/kg) in three different fishes, *Clarias gariepinus, Malapterurus electricus* and *Tilapia guineensis* while iron and zinc were present in trace amounts.
- **Adewoye and Omotosho (1997)** stated that, the variations in the concentration of minerals in fish muscles could have been as a result of the rate in which they are available in the water body and the ability of the fish to absorb these inorganic elements from their diets and the water bodies where they live.
- **Adewoye *et al.* (2003)** stated that the high calcium content recorded in *H. niloticus* and *C. angullaris* could probably be due to preferential accumulation and calcification of scales and hard tissues.
- **Adewumi *et al.* (2014)** studied comparative analysis of elemental compositions of four species of fish; *C. gariepinus*, *P. obscura*, *T. zilli*, and *T. galilaeus*, the pattern of elemental concentration in the fillets of each of the species, is in the order; Na>K>Ca>Mg>Fe>Mn>Cu>Zn.
- In a heavily polluted river of Bangladesh, Buriganga River, highest Cu was found in *C. punctatus* (5.27 mg/kg) and lowest in *G. chapra* (4.25 mg/kg) in pre-monsoon fish samples **(Ahmed *et al.*, 2010b).**

- **Akinneye *et al.* (2010)** studied mineral composition of sun-dried fish and found that, sun-dried *Bonga spp.* , *Sardinella spp*. and *Heterotis niloticus* have Ca content of 50.31, 80.34 and 33.21 mg/100g; Mg content of 134.40, 132.20 and 111.32 mg/100g; Fe content of 3.64, 8.81, 3.44 mg/100 g; Cu content of 0.34, 0.49 and 0.53 mg/100 g; Mn content of 3.07, 1.58 and 0.96 mg/100g; Zn content of 7.67, 6.44 and 5.68 mg/100g of fish respectively.
- Fish mineral and metal contents may vary according to the surrounding environment **(Ambedkar and Muniyan, 2011; Sen *et al.*, 2011).**
- **Banu and Salamatullah *(*1991)** compared the mineral contents of edible protein of 19 species of fresh water fishes of Bangladesh.
- According to **Basri *et al.* (2015),** nutritional trace elements in sun dried salted striped snakehead was Fe-130 ppb, Mn-4.729 ppb, Mg-3231 ppb, Cu-6.173 ppb, Zn-174.753 ppb respectively.
- According to **Committee on Animal Nutrition (1993),** minerals activate complex biochemical mechanisms, control, and regulate the uptake, storage and excretion of various inorganic elements allowing fish to live in a dynamic equilibrium with their aquatic medium. Minerals are important for vital body functions such as acid, base and water balance.
- **Dural *et al.* (2007)** and **Ploetz *et al.* (2007)** reported highest levels of Copper (Cu), Zinc (Zn), and Iron (Fe) in the liver and gills of fish species.
- **Fawole *et al.* (2007)** studied comparative analysis of elemental compositions of four species of dry fish; *O. niloticus*, *S. galilaeus*, *C. gariepinus*, and *H. niloticus*, the pattern of elemental concentration in the fillets of each of the species, is in the order; Zn>Ni>As>Cu>Pb>Cd.
- **Hadson (1998)** observed that the trace elements can be accumulated by fish both the food chain and water.
- All forms of aquatic animals require inorganic elements, or minerals, for their normal life processes. Unlike most terrestrial animals, fish have the ability to absorb some inorganic elements not only from their diets but also from their external environment in both fresh water and sea water. Many essential elements are required in such small quantities that it is difficult to formulate diets and maintain environments which are low in minerals to demonstrate a mineral

deficiency. Calcium and phosphorus are required for the formation of the skeletal structure of the body. Sodium, potassium and chloride, along with phosphates and bicarbonates, maintain homeostasis and the acid-base balance. Certain minerals, such as calcium, magnesium and manganese are of particular significance as enzyme activators **(Halver and Hardy, 2002)**.

- **Haque *et al.* (2006)** studied the seasonal variation of heavy metal concentrations in *Gudusia chapra* inhabiting the Sundarban mangrove forest and found the concentration of Cu, Zn, Fe, Pb, Cd, Cr and Ni seasonally varied from 0.527 to 3.99, 5.34 to 25.9, 0.038 to 0.221, 0 to 3.396 and 0.176 to 89.5 µg/g dry weight basis, respectively.
- **Hogstrand and Wood (1996)** mentioned that fifteen trace elements are considered to be essential in animals. Among these the physiological role of a deficiency of chromium, cobalt, copper, fluorine, iodine, iron, manganese, molybdenum, selenium, and zinc is well recognized. Most of these trace laments have been detected in fish tissues, including shellfish, however, the essentiality of only a few of these elements has been demonstrated.
- The Cu concentration in *M. armatus* was 0.86 mg kg-1 **(Javed and Usmani, 2012).**
- **Javed and Usmani (2014)** showed that the average accumulation of Cu, Fe, Mn and Zn in the edible part of *C. punctatus* was 13.65, 161.16, 21.20 and 124.05 mg kg-1 dry weight respectively.
- **Krishna *et al.* (2014)** observed that, the order of heavy metal concentration in *M. cephalus* was Zn>Pb>Mn>Cu>Cr>Hg and average concentrations in liver and muscle was 32.4, 10.8, 8.9, 6.4, 2.3 and 2.2 mg/kg respectively.
- **Kumar *et al.* (2012)** were determined the concentrations of Cu, Zn, Mn, Fe, Cd, Hg and As in muscle tissue of fish species collected from north East coast of India. The concentration range of Cu, Zn, Mn, Fe, Cd, Hg and As in fishes was 0.5-28.2, 3.0-99.1, 0.5-12.0, 10.4-249.7, 0.01-1.10, 0.05-1.60 and 0.02-2.37 µg g-1 dry wt. respectively.
- **Love (1980)** mentioned that the quantities of manganese, iron, copper, and zinc in the muscle and liver of fish reflect the agrochemical characteristics of the earth surrounding the aquatic body. The concentration of minerals in the body of

aquatic organisms depends on food source, environments, species, stage of development, and physiological status of the animal. Most organisms accumulate and retain minerals from the environment; however, their incorporation is highly selective.

- **Mansur and Sidky (2002)** stated that fish are often at the top of aquatic food chain and may concentrate large amount of some metals from the water.
- **Muslemuddin *et al.* (1991)** observed the effect of size, anatomical portion and habitat of major mineral content in muscle of mrigal fish (*Cirrhinus marigala*). Total mineral contents were lower in smaller fish than the lager ones.
- **Mustafa *et al.* (2012)** stated that, Ca, P and Fe content of fresh hilsha and sarputi fish was 56.49 and 222.68, 195.46 and 121.81, 0.40 and 0.42 mg/100g fish respectively.
- **Onyia *et al.* (2010)** determined mineral composition of some fresh water fishes of upper Benue River. They found that, *B. niloticus, M. rume, B. filamelosus, T. niloticus, C. gariepinus* has Fe content of 0.086, 0.074, 0.0009, 0.14 and 0.07 mg/g; Cu content of 0.042, 0.034, 0.038, 0.036 and 0.04 mg/g; Zn content of 0.082, 0.07, 0.074, 0.080 and 0.073 mg/g respectively.
- **Sharif *et al.* (1993)** reported that the calcium (Ca) content in *Mastacembelus armatus* (Baim) was 5232 mg kg^{-1}.
- **Taweel *et al.* (2013)** showed that Cu levels in liver, gills and muscles of tilapia fish were 491.30, 3.70 and 1.82 µg/g dry weights (dw) respectively.

2.7. Bacteriological study of fresh, salted and dried fishes

In Bangladesh, only some information is available on the occurrence and distribution of pathogenic bacteria in freshwater fish. There is no information about the microorganisms of salted and sun-dried salted stored snakehead fish (like-Taki, Shol) and small cat fish (like-deshi tengra), but there is available information on the microbial changes in other preserved fish.

- **Ahmed *et al.* (2010a)** studied the chemical and microbiological quality changes in salted (25% of the fish weight) *Hydrocynus forskalii* during storage at $+37\pm1^0$C and found that, salted techniques reduced the microbial counts of the salted fish.
- According to **Al-Basuni (1993),** determination of the microbial load of fishes and their products considered as the most important tests to determine the flesh quality and storage period.
- **Ali *et al.* (2007)** observed that organoleptic and microbiological assessment of ten commercially important dried and fresh fishes. They showed SPC (Standard Plate Count) of these fishes in fresh condition ranged from 8.2×10^4 to 3.1×10^5 cfu/g while the dried samples of these fishes ranged from 7.8×10^3 to 2.77×10^5 cfu/g respectively.
- **Balachandran (2001)** noted that most bacteria require a low concentration of salt for their growth. Many bacteria do not grow when the salt concentration is in the range 10-15%. However, haemophilic bacteria can developed even in concentrated solution of sodium chloride.
- **Brain *et al.* (1958)** described that, halophilic bacteria are frequently derived from contaminated solar salt. These bacteria are most troublesome at the wet stake stage in the salting and drying processes. They may be growing on fully dried salted fish and cause a pink or red discoloration.
- **Chaity (1992)** reported that the total bacterial count of market samples of traditional dried fish ranged from 1.0×10^5 to 1.63×10^6cfu/g.
- **Cheesbrough (2000)** stated that, fish with microbial load $>10^6$cfu/g is likely to be at the stage of being unacceptable from the microbiological point of view and unit for consumption.
- **Clucas (1984)** reported that halo tolerant microbes which can grow in a greater concentration of salt even unto saturation and salt loving microbes which include bacteria that require the obligatory presence of salt and can grow on media saturated with salt.
- The total bacterial count (TBC) of whole fish immediately after catch was found to range between 3.05×10^5 to 3.75×10^6cfu/g **(Devaraju and Setty, 1985).**
- **Enamul *et al.* (1987)** studied the total plate count at 30^0C for dried *Mugil sp.* and *Stromateus sp.* which was found to be 1.84×10^5/g and 2.82×10^5/g respectively.

- **Fatema (2005)** assessed the bacteriological and heavy metals concentration of Kacki (*Corica soborna*) and Mola (*Amblypharyngodon mola*) fishes. The results revealed that aerobic plate counts (APC) in local market raw fishes were quite high whereas in Kachki fish the highest APC was 8.5×10^7cfu/g and in mola fish, 2.4×10^7cfu/g.
- **Figueroa *et al.* (1990)** found that the mesophilic bacterial populations increased with storage time.
- According to **Flannery (1956),** the word halophile derives from the two Greek words, "halos" and "philus", meaning "salt" and "loving" respectively. Halophiles are named salt loving bacteria and they thrive in relatively high concentrations of salt. Both facultative and obligate halophiles will grow in media containing more than 2 % NaCl.
- **Frazier (1958)** recorded that solar salt contained a wide number of halophiles which are responsible for discoloration of fish flesh and it is assumed that fishes become contaminated with the bacteria from solar salt during salting process in addition to their normal flora.
- **Frazier and Westhoff (1978)** reported that no microorganism (yeast, mold, bacteria) can grow in fish product containing moisture less than 14%.
- Low temperatures are used to retard chemical reactions and the action of food enzymes and to slow or stop growth and activity of microorganisms in food **(Frazier and Westhoff, 1998).**
- **Freeman (1979)** stated that a variety of environmental factors influence the bacterial populations.
- According to **Gram and Huss (2000)** the microbial-infestation deteriorates the quality of the fish-product and thus human health is risked by infections and toxication. For this reason, information about the microorganisms (number of microorganisms) in the muscles of the fish is quite significant in terms of human health and preservation of the products.
- **Hasan (2006)** investigated that Aerobic Plate Count (APC) of solar tunnel dried Mola 2.16×10^8cfu/g; Tengra 2.59×10^8cfu/g and Kachki fish 2.08×10^8cfu/g.
- **Hasan *et al.* (2008)** found that, the microbial load in *Heteropneustes fossilis* and *Clarias batrachus* was 6.7×10^6cfu/g and 6.7×10^5cfu/g of fish sample.

- **Ichine *et al*. (1977)** described the micro flora of commercial slice dried fishes inclouding bonito and observed that the total plate count of dried fish samples averaged 1.2×10^6 cfu/g which ranged from 2.0×10^2 cfu/g to 9.5×10^6 cfu/g.
- According to **Kamruzzaman (1992),** total bacterial count ranged from 1.84×10^4 to 5.3×10^6cfu/g of commercially dried fresh water fish product samples.
- According to the definition of **Larsen (1986),** there are different categories of halo-tolerant microbes: non-tolerant, those which tolerate only a small concentrations of salt about 1% (%w/v); slightly tolerant, tolerating up to 6-8%; moderately tolerant, up to 18-22%; and extremely tolerant, those microbes that grow over the whole range of salt concentrations (0-32%, w/v).
- **Mansur *et al*. (1993)** investigated that total bacterial count of market sample soft dried fish ranged from 1.0×10^5 to1.63×10^6cfu/g. They also reported that total bacterial count of traditionally dried fishes were 1.5×10^6; 1.0×10^5; 1.8×10^5 and 1.6×10^6cfu/g in *Labeo rohita*; *Barbus sarana*; *Channa striatus* and *Gudusia chapra* respectively.
- **Mansur *et al.* (1998)** studied on the bacterial load of dry salted pre-spawning and post-spawning of hilsa. They found initial bacterial load was 0.78×10^6 calls/g and 0.97×10^6cell/g in pre-spawning and post-spawning hilsa which increased after dry salting to 0.01×10^6 and 1.14×10^6 cells/g of fish flesh respectively at the 18^{th} day of the study period.
- **Nabi *et al.* (1996)** mentioned that dry *Pampus chinensis* contained highest and lowest trend of bacteria were 5.23×10^7cfu/g and 3.20×10^6cfu/g in summer, 1.96×10^7cfu/g and 4.06×10^5cfu/g in monsoon, 1.79×10^5cfu/g and 4.05×10^4cfu/g in winter.
- Bacteriological quality is of public health importance as it directly relates to spoilage of fish and becomes the cause of outbreak of food poisoning **(Nilla *et al.*, 2012).**
- **Nurullah (2005)** mentioned that Aerobic Plate Count (APC) of some solar tunnel dried SIS products was found in the range of 1.45×10^5 to 2.52×10^6 cfu/g.
- **Poulter (1978)** mentioned that the number of bacterial growth better in 4^0C to 27^0C temperature.

- **Reza (2002)** studied the production of safe and high quality dried fish products and found the SPC were 1.8×10^3 to 2.6×10^4, 2.6×10^3 to 2.6×10^4, 5.4×10^4 to 6.0×10^5, 8×10^2 to 3×10^5 and 5×10^3 to 1×10^5 CFU/g respectively for silver jew fish, Bombay duck, big-eye tuna, Chinese pomfret and ribbon fish.
- **Shaheen *et al.* (1992)** reported that TBC in dried Punti averaged 6.0×10^5 cfu/g with a range between 1.30×10^4 cfu/g and 2.03×10^6 cfu/g. In dried Mola this count averaged 3.9×10^5 cfu/g which ranged between 1.2×10^4 cfu/g to 2.03×10^6 cfu/g.
- **Shewan (1977)** reported that, the fish from warm water frequently carry greater number of bacteria than cold temperate water fish.
- **Sufi *et al.* (1996)** showed that the annual average monthly total bacterial count TBC of dry samples of *Gudusia chapra* was 5.76×10^6 cfu/g. The seasonal bacteriological studies revealed that the highest TBC was 3.13×10^7 cfu/g in summer, 1.92×10^7 cfu/g in monsoon and 2.34×10^5 cfu/g in winter.
- **Sultana and Hossain (2010)** found among the dried fishes of three areas of Chittagong, the highest mean TVC was found 90.00±1.00 in Churi. On the other hand, the lowest TVC was 30.67±0.58 in Chingry respectively. Similarly highest TVC was 113.00±2 in Chapa and lowest TVC was 94.67± 0.58 in Challa of two areas of Mymensingh district.
- **Troller and Christian (1978)** observed that effect of various environmental factors such as pH, 0_2 and CO_2 concentrations besides temperature and water-activity for microbial growth.
- **Truelstrup *et al.* (1996)** reported that the salting and drying process often reduce the numbers of microorganisms and cause a change in the spoilage micro flora.
- According to **Valsan *et al.* (1985)** the cured fish product through preserved and protected with high level of salt and low content of moisture, the main spoilage factors associated with the cured fish were fungal and “Red” attack caused by certain halophilic or halo-tolerant microorganisms.

CHAPTER-3

MATERIALS AND METHODS

CHAPTER-3

MATERIALS AND METHODS

3.1. Experimental protocol

To carry out this research work, always easy and less expensive technology was practiced.

3.1.1. Selection of fish species

In this research work, three (3) varieties of fresh water fish species were used for this comparative study of different salting process. The size of fishes considered for the experiment was due to customary demand for consumers and some significance on the shelf life and quality of the processed product.

The fish varieties were:

1. Large size snake-headed fish, shol (*Channa striatus***;** Bloch, 1801) [Plate 3.1.1(a)]
2. Medium size snake-headed fish, taki (*Channa punctatus***;** Bloch, 1793) [Plate 3.1.1(b)] and
3. Small size bagrid catfish, tengra (*Mystus tengra***;** Hamilton-Buchanan, 1822) [Plate 3.1.1(c)].

Plate 3.1.1(a). Shol (*Channa striatus*; Bloch, 1801)

Plate 3.1.1(b). Taki (*Channa punctatus*; Bloch, 1793)

Plate 3.1.1(c). Tengra (*Mystus tengra*; Hamilton- Buchanan, 1822)

3.1.2. Collection of experimental fishes

Fresh mature fish samples were collected from fishermen of Meghna River in the early hours of the day. Mean length variation of the experimental shol, taki and tengra fish were 40±10, 22±8 and 10±2 cm respectively. Whereas, mean weight of the experimental shol, taki and tengra fish were 561.5 (±20), 128.5 (±6) and 9.1(±1.2) g respectively.

3.1.3. Handling of experimental fish in laboratory

Being air breathing fish, shol and taki fish were transported to the research laboratory in dram full with water. In case of tengra fish they were carried in clean, good quality sterile polythene bag with ice in order to keep the fish fresh and avoid any type of microbial contamination according to ICMSF, 1996.

3.1.4. Place/ location of the experiment

Biochemical analysis and Bacteriological study were carried out at the 'Fish Technology Section' and 'Food Microbiology Section' of the Institute of Food Science and Technology (IFST) of Bangladesh Council of Scientific and Industrial Research (BCSIR), Dhanmondi, Dhaka. Mineral analysis was done in Center for Advanced Research in Science (CARS).

3.1.5. Period of experiment

The whole experimental period was 43 months, started from June 2011 and continued for up to the period of December 2014.

3.1.6. Preparation of the experimental fishes

In the laboratory, the experimental fishes were carefully washed with normal cool tap water. The weight of whole fishes was measured by a sensitive balance and was recorded. The fishes were then dressed by removing scales, fins, gills and viscera and again washed with tap water to remove blood, slime and unnecessary adherents.
Cleaning depends on the fish species, size, and the organs accumulating the nutrients. So, the non-edible waste portion is not always related with the size of the fish. Normally, the operculum, jaws, fins, viscera (partial/full) and the scales are included among the waste. However, according to Roos *et al.* (2006) cleaning practices depend on the fish species and size of the fish.
Tengra is a small sized fish, so eaten along with bones whereas, because of hard and large shield-like bones of head, the bones and head of Taki and Shol are included as the waste.

After that, total cleaned fishes were then grouped into 5 batches. It should be mentioned here that to determine freshness of the raw fish, the nutrient contents of its body muscles was determined by applying the most acceptable Standard methodology applicable for fish analysis.

3.2. Materials used in salting procedure

3.2.1. Common Salt

For salting, the pure market table salt (salt brand-Super salt) with the fine grain was used. The finer the salt, the more rapidly the brine forms and thus the more rapidly the flesh is penetrated with salt. Salt for curing was collected from the local market. Ingredients of Super Salt are presented in Appendix [Appendix-A].

3.2.2. Turmeric

Fresh dried and powdered form of turmeric is used as a newly introduced element in salt curing method with a hypothesis that it would work as-

- A protection against insect, pest, fungus and other pathogens
- Cheap and easily available
- A good preservative
- Taste enhancer element.

3.2.3. Instruments / equipments

A. <u>For Taxonomic Studies</u>

1. Measuring scale

2. Books

B. <u>For Bio-chemical composition</u>

1. Analytical (Electronic) balance (Shimadzu Corporation, Type-AY, Max 220g, d=0.1 mg)

2. Drying oven (Gallen kamp Hotbox Oven with fan, Size=2)

3. Petri dishes

4. Porcelain basins

5. Desiccators

6. Laboratory grinder (Mortar and pestle)

7. Blender machine (Jaipan, Made in India)

8. Digestion Chamber (Buchi 426 Digestion Unit)

9. Distillation Machine (Gerhardt)

10. Filter paper (Whatman)

11. Exhausting chamber

12. Digestion tube

13. Burette

14. Conical flask

15. Volumetric flask

16. Measuring cylinder

17. Buchner Funnel

18. Water bath (HH-$S_{21.4}$ Thermostatic water bath, made in China)

19. Magnetic stirrer (Stuart Scientific, Hotplate Made in U.K.)

20. Porcelain crucible

21. Gas burner

22. Muffle furnace (NABER- L51/S, Germany)

23. Conway dish

24. Stopper glass bottle

25. Glass rod

26. Electrical sealing machine (PFS-300)

C. <u>For Bacteriological study</u>

1. Stomacher 400 (homogenizer) bag

2. Stomacher 400 (homogenizer) machine (ANNO 2010, Serial No. 030163)

3. Laminar flow

4. Magnetic stirrer (Stuart Scientific, Hotplate Made in U.K.)

5. Vortex Mixer (VM-1000, DIGISYSTEM LABORATORY INSTRUMENTS INC.)

6. Autoclave Machine (TOMY, SX-500)

7. Volumetric Flux

8. Micropipette

9. Petri dishes

10. Test tube

11. Colony counters (Digital Colony Counter, Ri)

D. <u>For Mineral Study</u>

1. Perkin-Elmer atomic absorption Spectrophotometer (Model A Analyst 200; Illinois, USA)

2. CEM Microwave digester (MARS, XP 1500 Plus)

3.2.4. Chemicals

A. For Bio-chemical composition

1. Concentrated sulfuric acid (Merck, Germany)

2. Digestion mixture (98 parts of anhydrous K_2SO_4 is mixed with 2 parts of $CuSO_4$),

3. Sodium hydroxide

4. 2% boric acid (2 g boric acid in 100 ml distilled water)

5. Mixed indicator (0.1% bromocresol green and 0.1% of methyl red indicator dissolved in 95% alcohol separately. Then 100 ml bromocresol green is added with 2 ml of methyl red solution)

6. N/70 sulfuric acid

7. Folch's reagent (Chloroform: Methanol = 2:1)

8. 10% Trichloro Acetic Acid (TCA) (Merck, Germany)

9. Potassium Carbonate (K_2CO_3)

10. 0.25 N Sodium hydroxide (NaOH) (Merck, Germany)

11. Ethanol (Merck, Germany)

12. Phenolphthalein solution

13. Chloroform (Merck, Germany)

14. Methanol (Merck, Germany)

B. For Bacteriological study

1. Plate count agar (Hi- media)

2. Ringers Stock

3. NaCl

4. Distilled water

C. For Mineral Study

1. Nitric acid (HNO_3), (65% Merck, Germany)

2. Deionized water

3.3. Working principle

3.3.1. Methods used in different kinds of salting

For the experiments, five different types of salting methods were used. The salting methods more or less relate with those described by Horner (1992). Salting methods are-

I. Dry salting (DS)

During this experiment the fresh fishes were mixed with dry commercial salt (NaCl) of about 30% by weight of the dressed fish stacked in containers and stored for a salting or curing period, at room temperature. In this method, the extracted water of the fish due to salt-in was removed from the container through a sieved basket so that the product remained nearly in dry condition.

II. Pickle curing (PC)

In this method, the fresh experimental fishes were salted by dry commercial salt (NaCl) of about 30% by weight of the dressed fish stacked in containers and stored at room temperature. The salt entered the fish and water extracted out from the fish-body and a salt-solution is formed. Thus in this method, the fishes are always, allowed to remain in such solution for the production of pickle-cured fish.

III. Brine salting (BS)

During this experiment about 30% salt solution was prepared which is called brine. The fresh fishes are kept at this super saturated brine solution stacked in containers and stored for a salt curing period, at room temperature (26^0C-32^0C) for the

production of brine salted fish. The brine solution was changed with new solution once every week for keeping nearly constant saturation outside the fish. The experimental fishes in brine were kept immersed by putting a glass weight on it.

IV. Sun-dried salting (SDS)

During this experiment the fresh fishes were well rubbed with dry common table salt (NaCl) of about 30% by weight of the dressed fish. They were then kept on a plastic basket in the sun. They were kept in sun regularly during day time (9:00 a.m. to 3 p.m.) for 2-7 days as sometimes the sky was cloudy and until the required drying period was over. During sun-drying, they were kept covered by small meshed nylon or mosquito net to avoid external contamination and to prevent bird attack and fly infestation.

The process and principle of sun dried salting can be expressed by the following model adapted from Nowsad, (2005)-

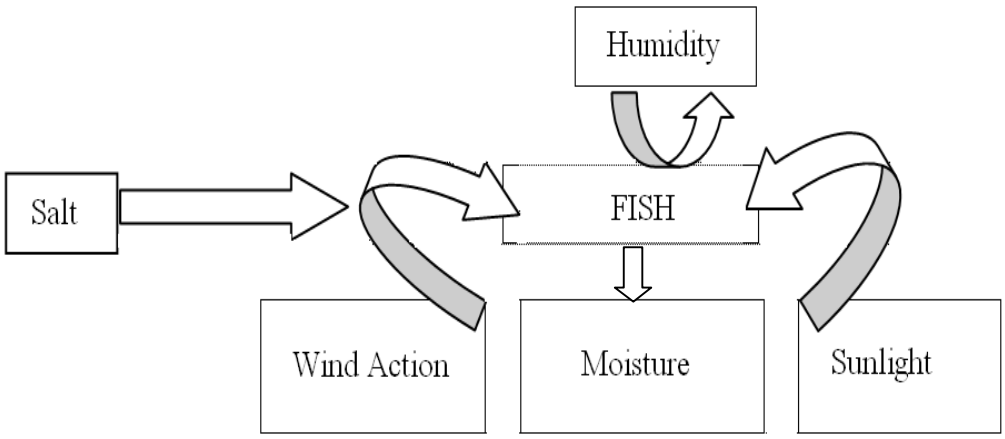


Figure. 3.3.1(IV). Process of sun dried salting (SDS) of fish

V. Turmeric treated sun-dried salting (SDS+T)

During this experiment the fresh fishes were treated with dry salt (NaCl) of about 30% mixed with 1% turmeric powder by weight of the dressed fish. They were kept on a plastic basket in the sun. They were kept in sun regularly during day time (12 a.m. to 3 p.m.) for 2-7 days until the ripening period was over. During sun-drying, they were kept covered by dense meshed nylon or mosquito net to avoid outside contamination and prevent bird attack and fly infestation.

The process and principle of sun dried salting can be expressed by the following model-

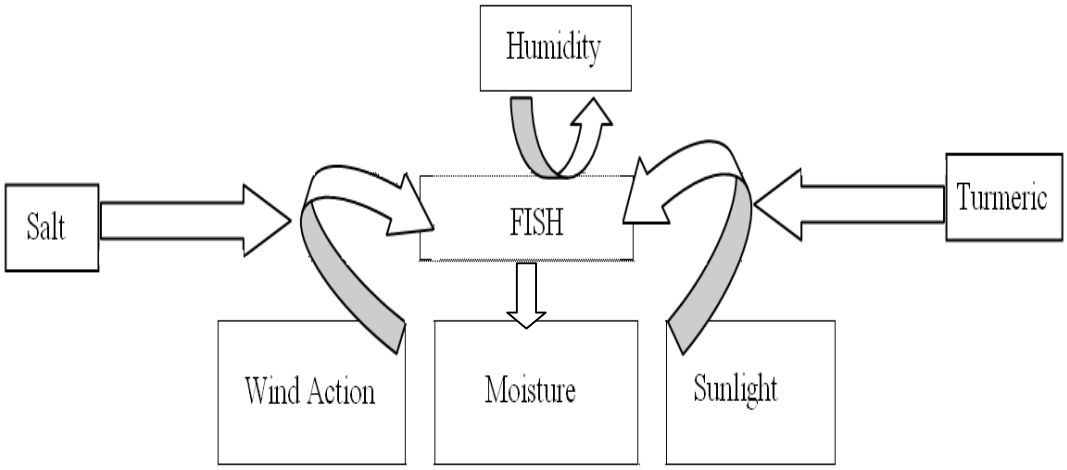


Figure. 3.3.1(V). Process of turmeric treated sun dried salting (SDS+T) of fish

3.3.2. Ripening period after salting process

The ripening of the salted product was determined by observing the changes in organoleptic characteristics such as color, texture, flavor etc. and changes in weight, moisture content and salt penetration rate. The completion of ripening of the salted fishes at different treatments was revealed by no further loss of moisture from the products. The loss of weight during different days of first, second and third stages of ripening of post-salted product was recorded.

The ripening of the products was confirmed by its salty taste, color and texture.

Various steps of ripening period of different treatments of shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish-products are shown in Plate-3.3.2 (a), 3.3.2 (b) and 3.3.2 (c).

Plate 3.3.2(a). Showing various steps of ripening period of different treatments of shol (*C. striatus*)

Plate 3.3.2(b). Showing various steps of ripening period of different treatments of taki (*C. punctatus*)

Plate 3.3.2(c). Showing various steps of ripening period of different treatments of tengra (*M. tengra*)

3.3.3. Storage of the salted fish-products for shelf life study

The processed shol, taki and tengra fish-products after completion of different salting process (ripening period) have been found to attain a level of quality. But storage of the salted products are very important to extend its shelf-life, because in a high-humid and high-temperature weather of Bangladesh, the salted and salted-dried fish-products losses its quality by absorbing more moisture from the surrounding air and thus facilitating the growth of microbes and spoilage became faster.

In the present study, two different storage condition were used to prolonged the shelf-life of the salted and salted-dried fish-products which are mentioned as follows-

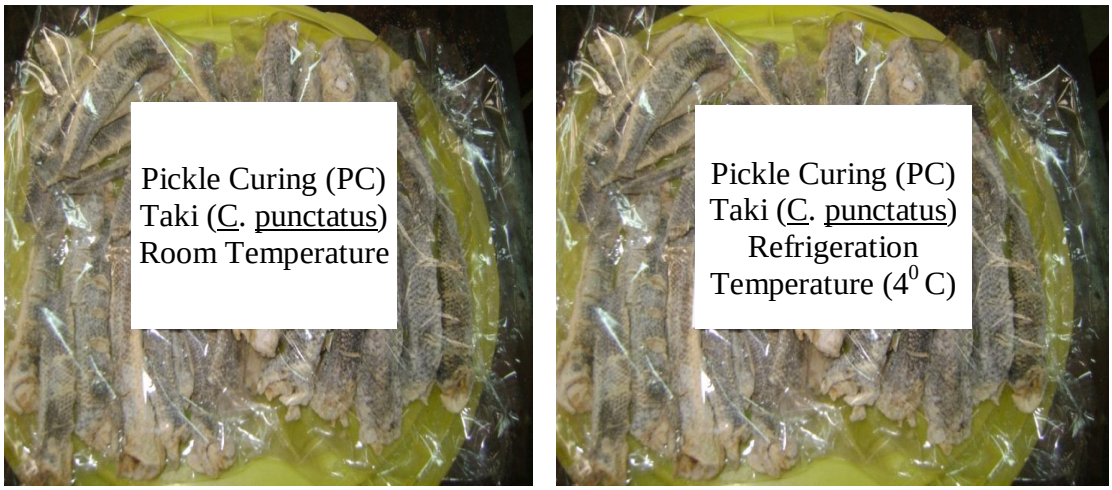
A) Room or Ambient Temperature (26^0C-32^0C)
B) Refrigeration Temperature (4^0C)

Procedure

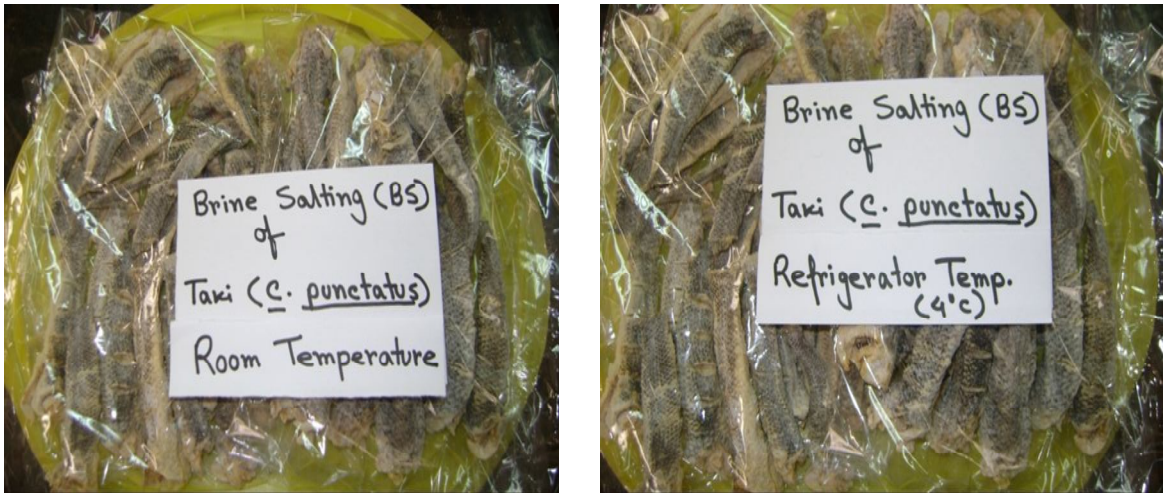
At the end of the ripening period, salted fish in each treatment was divided into two batches. Each batch are then weighed and packaged in plastic bag maintaining aseptic condition as far as possible. The bags were sealed by using an electrical sealing machine (PFS-300). After that, one batch of salted fish was stored at refrigeration temperature (4^0C) and the other batch at ambient or room temperature (26^0C-32^0C). The preservation period of products was found linked to the amount of salt added; therefore a straight proportion is present between the amount of salt used and the preservation period (Bahri *et al*., 2006).

To illustrated the storage conditions of different types of salted shol, taki and tengra fish products of the present study, the photographs are referred those are mentioned in the Plate 3.3.3.

(a) Dry salted (DS) products



(b) Pickle cured (PC) fish-products



(c) Brine salted (BS) fish-products

(d) Sun-dried salted (SDS) fish-products

(e) Turmeric treated sun-dried salted (SDS+T) fish-products

Plate 3.3.3. Showing DS, PC, BS, SDS and SDS+T shol, taki and tengra fish-products stored both at room temperature (26-32^{o}C) and refrigeration temperature (4^{0}C)

The salting procedure followed in this experiment is presented in a flow diagram given bellow-

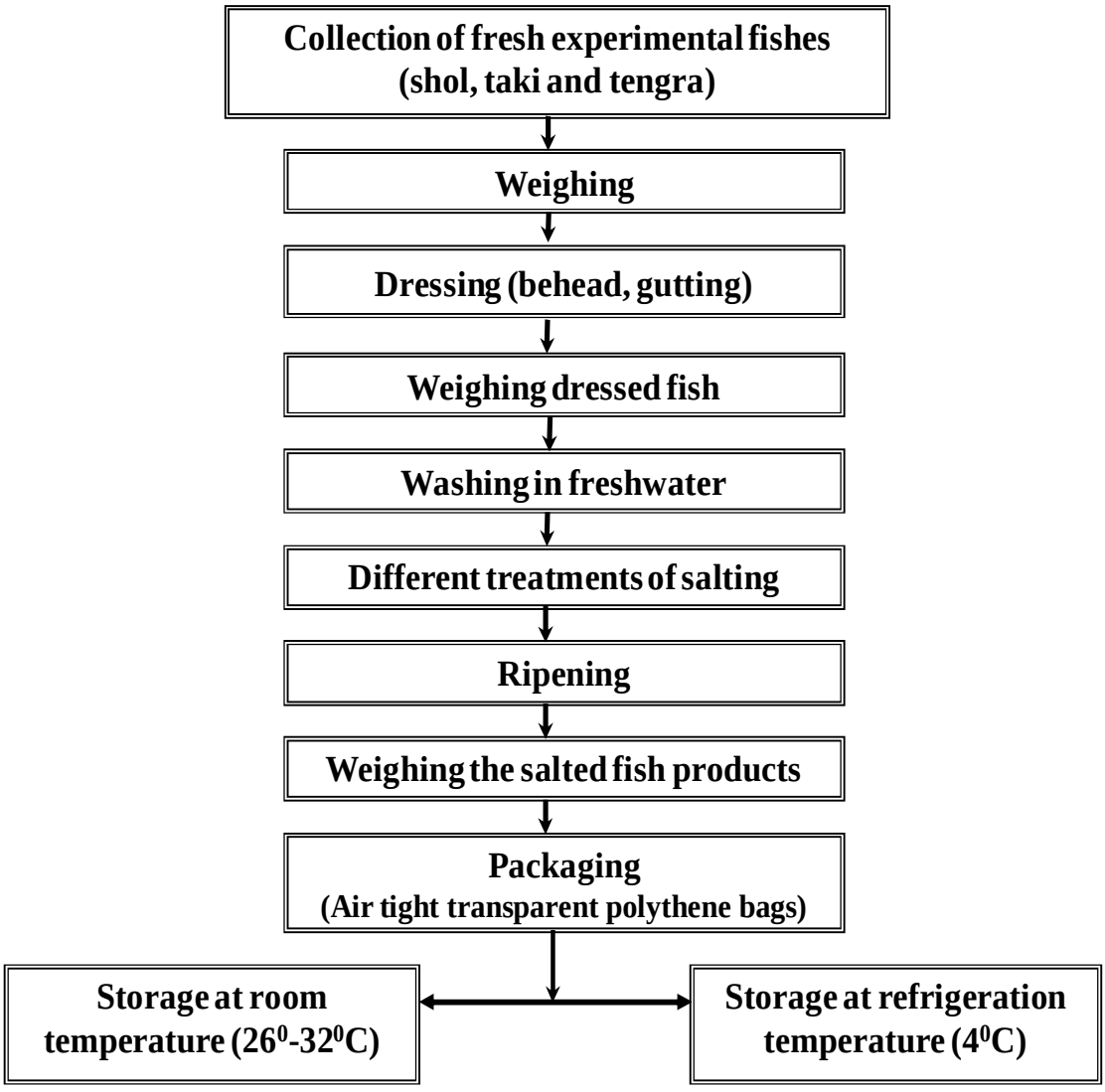


Figure. 3.3.3. Flow chart of salting procedure of shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*)

3.3.4. Sampling procedures

For quality analysis, sensory evaluation, proximate and chemical composition of 5 different treatments (DS, PC, BS, SDS, SDS+T) of the salted fishes were carried out at 15 days of interval and bacteriological study at 1 month of interval, for the fish kept at room temperature (26^0C-32^0C) and 2 months interval for the fish kept at refrigeration temperature (4^0C), until the fish became inedible. Important mineral composition i.e. Ca, Mg, Cu Zn, Fe and Mn were determined in fresh fish and freshly processed salted fish products.

For analysis, six or seven slices were taken randomly from different parts of the whole body of the fish. For sampling, adhered salt crystals (if any) were removed from the dry salted, sun dried salted and turmeric treated sun dried salted fish-products by using tissue paper. In case of pickle and brine salted fish-products, external water was absorbed with tissue paper putting a mild pressure on the slices from both the sides of fish by hand. Then the slices were chopped and finally ground by an electric blender machine (Jaipan, Made in India) to make a homogenous sample before being sampled for analysis.

3.3.5. Parameters of quality assessment

Food quality aspects represent the organoleptic evaluation, physical observation and biochemical parameters characteristics of food. These parameters served as important basis to evaluate the food quality of products under investigations. The salt cured fish products were assessed for their quality standard during different time interval from fishes at different storage conditions and the findings were recorded.

The quality assessment of fresh and salted products was performed by determination of 5 major analyses. viz,

1) Sensory characters and sensory score value evaluation
2) Proximate composition study
3) Determination of changes in quality parameters (Chemical changes)
4) Study of mineral composition
5) Bacteriological study

3.3.5.1. Sensory characteristics and sensory score evaluation

Principle

Various types of analyses have been developed to measure the loss of fish freshness and to detect spoilage. The deteriorative changes occurring in fish products were quantified by any standard techniques (Connell *et al.,* 1976). Sensory methods offer rapid ways of assessing acceptability of fish products and detecting spoilage.

The sensory evaluation of the salted products made from five different salting methods was analyzed for shelf life study by using a freshness-scoring sheet (Table-3.3.5.1) in a 9 point hedonic scale (Peryam and Pilgrim, 1957).

Table: 3.3.5.1. Freshness score sheet (Peryam and Pilgrim, 1957)

Overall acceptability score (9- point hedonic scale)	
Scores	**Description**
9	Like extremely/ Highly acceptable
8	Like very much/ Acceptable
7	Like moderately /Moderately acceptable
6	Like slightly/ Just acceptable
5	Neither like or dislike / Just Unacceptable
4	Dislike slightly / Unacceptable
3	Dislike moderately / Moderately Unacceptable
2	Dislike very much / very much Unacceptable
1	Dislike extremely / Extremely Unacceptable

Sensory characters considered in sensory score

a) Flavor or smell

When the salt curing process failed to preserve the fish in consumable condition, then the fish product released off-odor with time period lapsed. A good salt cured product showed a characteristic attractive smell created by the ripening reaction.

b) Color

The color reflects the fish condition. In case of general methods of salt curing, the fish skin remained shiny in good condition and became brown or reddish when crossed the border of acceptability.

c) Texture (External appearance)

The texture (external appearance) also reflects the condition of fish product. Closer the appearance to the original first conditions of the fish, fresher the fish product. But, if the appearance differs much from the original condition when it was stared for salt curing or during storage period, the assumption determines a lower rate of hedonic scale. In this experiment, it showed that with time lapsed the fish skin showed some signs of changes created by salt-loving bacteria or other pathogens or bio-chemical reactions indicating unacceptable to the consumers. At good preservation, the muscle texture of fish remains elastic, flexible and fresh.

There is a linear correlation between the sensory quality expressed as a demerit score and storage life on salt, which makes it possible to predict remaining storage life on salt (Luten and Martinsdóttir, 1998).

d) Taste testing of fish-curry

At good processing, the tastes of the fish products have to be delicious and highly acceptable which inspires the salting method to be applied on that kind of fish-product. When the taste is somewhat not satisfactory to the consumer it may mean that the preservation process was not suitable to that kind of fish.

For this reason, consumer preferability study was done after preparing five different types of freshly processed salted shol, taki and tengra fish curry with traditional south Asian recipe. These fish-curries were subjected to panel tests with 5 panel members. Cooked fish were evaluated with its taste. Panelists tasted the cooked samples and recorded their evaluations using a freshness-scoring sheet in a 9- point hedonic scale described by Peryam and Pilgrim, 1957 [Appendix-D].

3.3.5.2. Proximate composition study

This analysis was developed in Germany during the 1870 to evaluate feed ingredients and finished feeds (Halver and Hardy, 1992). Proximate analysis involved the partitioning of compounds in a sample into major four categories based upon the Chemical properties of the various constituents of feeds. Four categories are-

I) Moisture (Water)

II) Crude protein

III) Lipid or fat

IV) Ash

3.3.5.2(I). Estimation of moisture content

Principle

Determination of moisture content of the fresh as well as salt cured fishes was conducted by AOAC method (AOAC, 1990).

Procedure

Some washed and dried crucibles of known weight were taken. About 5 gram of previously prepared fairly minced fresh raw and salted fish samples were taken into each crucible and weighed in an electronic balance. The samples were allowed to dry into the oven at 105^0C for 24 hours in order to remove the moisture until constant weight. After that, the crucibles were taken out of the oven, cooled in desiccators and weighed in the digital balance. Moisture content was then calculated as shown below:

Calculation:

$$\% \text{ of Moisture} = \frac{\text{Weight Loss}}{\text{Original Weight of Sample Taken}} \times 100$$

Various steps of moisture content determination of the fish samples by AOAC method are shown in Plate 3.3.5.2(I).

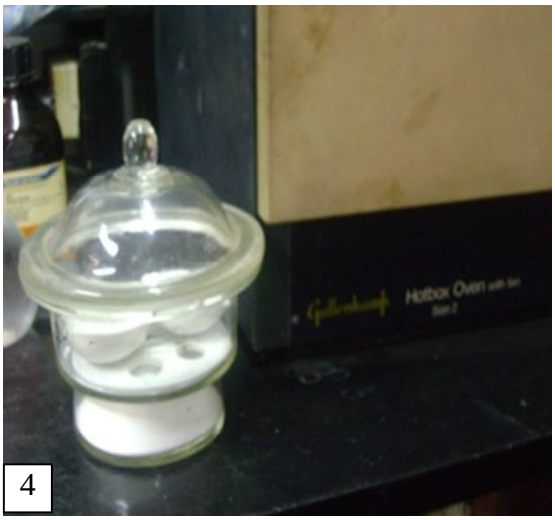


Plate 3.3.5.2(I). Showing various steps of moisture content determination of the fish samples by AOAC method

3.3.5.2(II). Estimation of protein content

Reagents required

1. Concentrated sulfuric acid
2. Digestion mixture**:** 98 parts of anhydrous K_2SO_4 is mixed with 2 parts of $CuSO_4$
3. Catalysts for digestion 40% sodium hydroxide (40 g NaOH in 100 ml Distilled water)
4. 2% boric acid (2 g boric acid in 100 ml distilled water)
5. Mixed indicator (0.1% bromocresol green and 0.1% of methyl red indicator was dissolved in 95% alcohol separately. Then 100 ml bromocresol green was added with 2 ml of methyl red solution)
6. N/70 sulfuric acid

Principle

The universally accepted method for determining total nitrogen of crude protein of the fish is "Micro-Kjeldahl" method (Pearson, 1999). The basic principle of this method involves the conversion of the nitrogenous protein into $(NH_4)_2SO_4$ when boiled with $H_2SO_4(NH_4)_2SO_4$, and distillation with access of sodium hydroxide (NaOH), gives ammonia (NH_3) which is absorbed in boric acid solution containing methyl red. The amount of Nitrogen (N_2) absorbed in boric acid is determined by titration with N/70 H_2SO_4.

Procedure: The process involves the following two steps-

i) Preparation of digestion solution

Some pieces of ash less filter paper were taken and weighed in an electronic balance. About 0.5-0.9 gm macerated experimental fish samples were taken in each piece of filter paper and they were also weighed. A record was kept for the identification of the

various types of fish samples. The samples with the ash less filter paper were taken into the washed and dried 50 ml Kjeldahl flasks. A mixture of Buchi digestion unit was prepared by adding 20 ml concentrated H_2SO_4 (20%) with the traditional digestion mixture (a white power). These were kept in that digestion unit until the mixture became clear. Thus a water color digestion solution was prepared.

ii) Preparation of sample solution

The digestion solution the made into 100 ml in a 100 ml volumetric flask with distilled water. 5 ml of the sample was transferred in a Micro- Kjeldahl distillation apparatus followed by 10 ml of 40% NaOH and 150 ml distilled water. 5 ml of 2% boric acid was taken into a conical flask and placed in the lower chamber of the Micro- Kjeldahl distillation unit. The prepared sample solution was kept into Micro Kjeldahl distillation unit for 50 minutes. Distillate was collect in access of 2% boric acid solution with indicator and was titrated by N/70 H_2SO_4. After titration the initial green color was changed into pink color.

Calculation

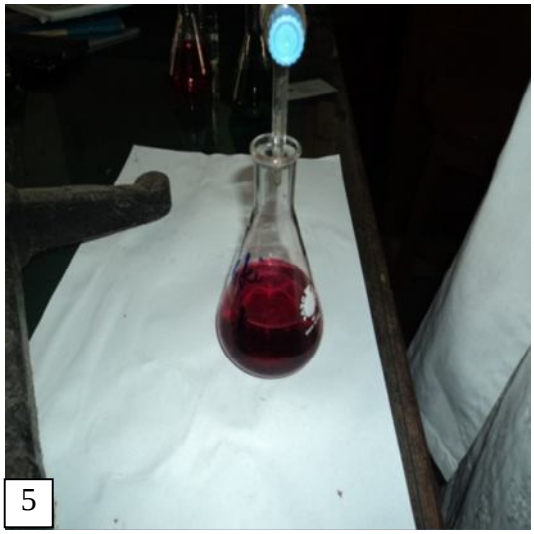
The percentage of nitrogen in the sample was calculated by the following equation:

$$\% \text{ of Protein} = \frac{\% \text{ of } N_2(\text{titration reading - blank reading}) \times \text{strength of acid} \times 0.002 \times \frac{100}{5} \times 100}{\text{weight of sample taken}}$$

For most routine purposes the percent of protein in the sample is then calculated by multiplying the % of N_2 with the protein conversion factor of 6.25 for fish.

% of protein = % of total $N_2 \times 6.25$

Various steps of protein content determination of the fish samples by Micro Kjeldahl method are shown in Plate 3.3.5.2(II).



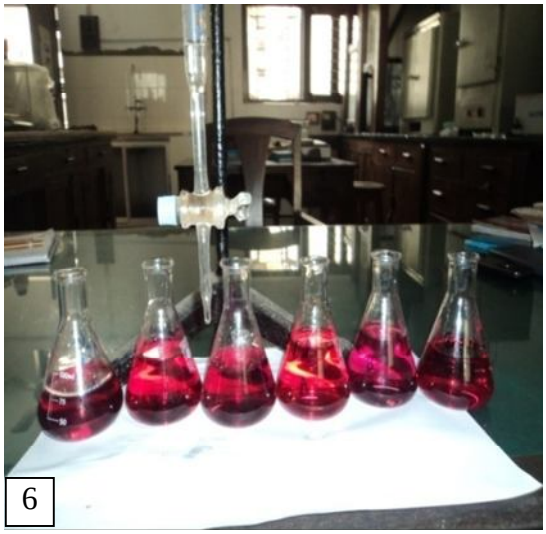


Plate 3.3.5.2(II). Showing various steps of protein content determination of the fish samples by Micro Kjeldahl method

3.3.5.2(III). Estimation of fat content

Reagents required

1. Folch reagent (Chloroform: Methanol = 2:1)

Principle

The estimation of fat content of experimental fresh raw fish and salted fish samples had been accomplished by following AOAC, 1990 (Folch *et al.*, 1957).

Procedure

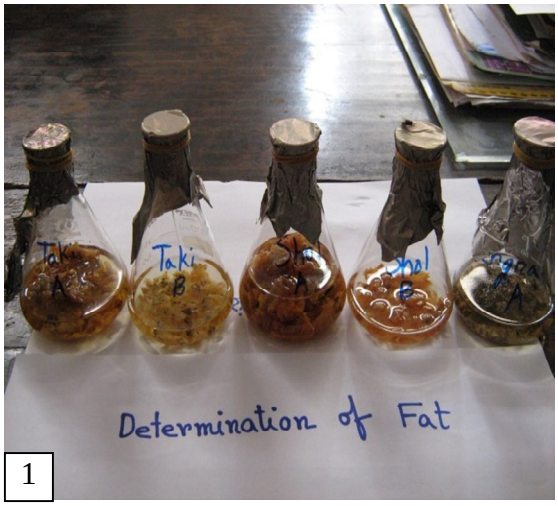
At first fresh and salted fish samples was taken and muscles were cut into pieces and minced with mortar and pestle for making homogenous one. Then about 5 g of the homogenous sample was taken into conical flasks. About 10 ml of folch reagent (Chloroform: Methanol = 2:1) was added into the sample and homogenized properly and kept in air-tight condition for 24 hours. Fat contents of the fish muscle react with that solvent and remains in the solution.

After 24 hours the solution of the flask was filtered in another weighed conical flask through a filter paper. Then these flasks were given in a hot water bath to dry up and removed the solvent. After that the flasks were kept into an oven for an hour to get the actual fat content. Then the flasks were weighed in an electronic balance to get the amount of fat content.

Calculation

$$\% \text{ of Fat} = \frac{\text{Weight of the residue}}{\text{Weight of sample taken}} \times 100$$

Various steps of fat content determination of the fish samples by AOAC method are shown in Plate 3.3.5.2(III).



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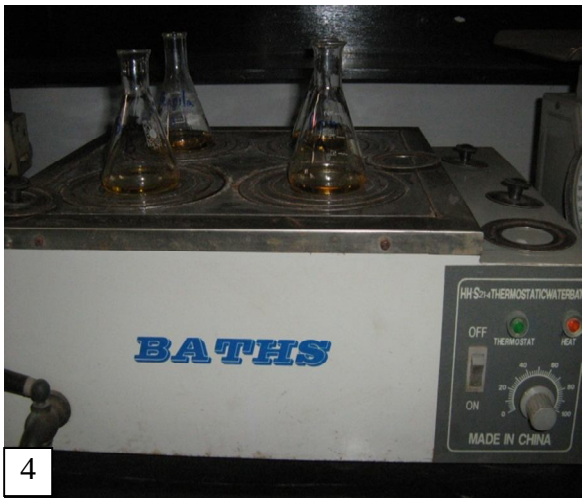

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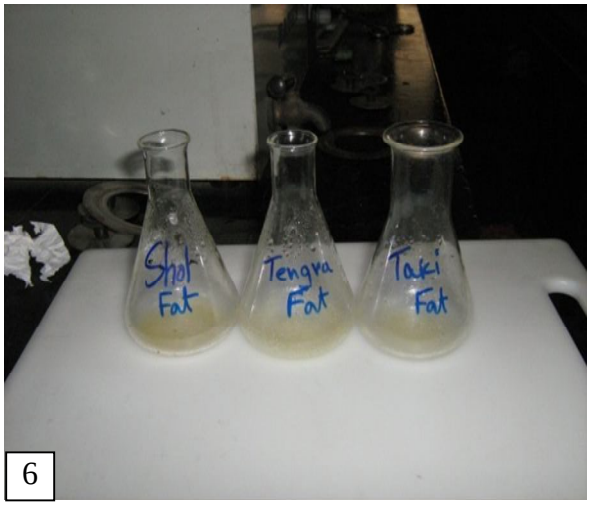

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Plate 3.3.5.2(III). Showing various steps of fat content determination of the fish samples by AOAC method

3.3.5.2(IV). Estimation of ash content

Principle

Determination of ash content of the fresh as well as salt cured fishes was conducted by AOAC method (AOAC, 1990).

Procedure

About 4-5 g fresh raw and salted macerated fish sample was weighed into a pre-weighed crucible. The crucible with the contents was heated first over a long flame till all the material was completely churned. Then it was transferred in the Muffle Furnace (NABER- L51/S, Germany) held at 600^0C for 5 hours until the residue become white. The crucible with ash were cooled in desiccators and weighed. Finally the % of ash content was calculated.

Calculation

$$\% \text{ of ash} = \frac{\text{Weight of fish}}{\text{Weight of sample taken}} \times 100$$

Various steps of ash content determination of the fish samples by AOAC method are shown in Plate 3.3.5.2(IV).

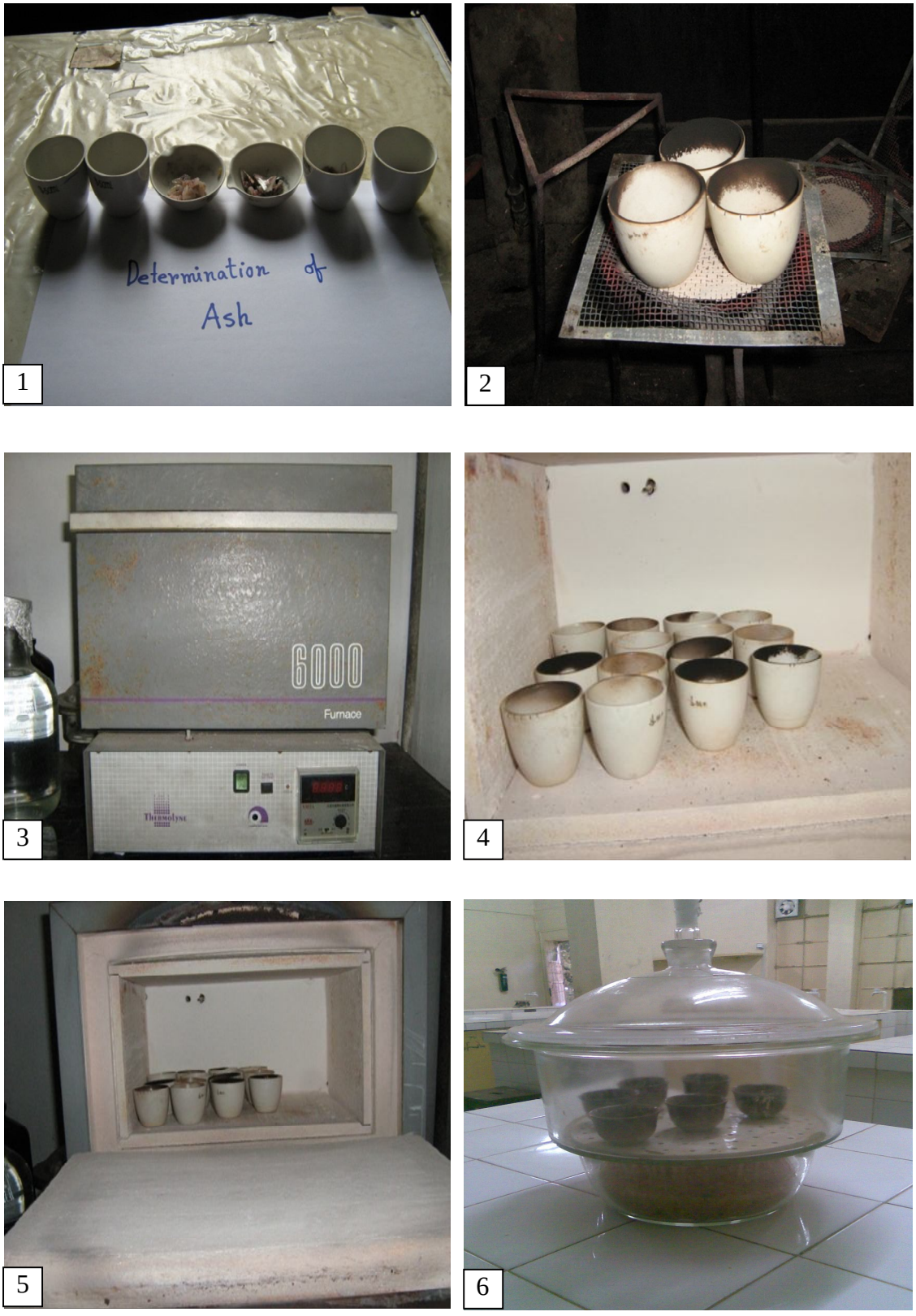


Plate 3.3.5.2(IV). Showing various steps of ash content determination of the fish samples by AOAC method

3.3.5.3. Determination of changes in quality parameters (Chemical changes)

Certain chemical changes in spoiling fish appear to run parallel within change in odor, texture, appearance, etc. Various attempts have been made to measure freshness by establishing the quantities of some of the end product that were the result of both the extra cellular and intracellular enzymatic actions.

In present research work, 4 chemical analysis were assessed to find out the shelf life of five types of salted shol, taki and tengra fish-products. These are-

I) Estimation of Salt (NaCl) content
II) Estimation of Total Volatile base Nitrogen (TVB-N)
III) Estimation of Free fatty acid (FFA) value
IV) Estimation of pH value

3.3.5.3(I). Estimation of Salt (NaCl) content

Reagents required

1. 0.05 N Silver Nitrate ($AgNO_3$)
2. 5% Potassium Chromate solution

Principle

Estimation of salt content of salt cured fish-products was conducted by AOAC method (AOAC, 1990).

Procedure

1.0g fish sample was taken from ground fish sample in a conical flask and was mixed with 10 ml distilled water, was stirring and mixed for half an hour so that all the salt in the muscle becomes soluble in water. Then the solution was filtered with filter paper and an aliquot of 0.2 ml from the solution (filtered) was taken in another conical flask to which 10 ml of distilled water was added following by an addition of 2 drops of 5% Potassium Chromate and mixed properly. Titration was done with 0.05 N $AgNO_3$ solutions up to the end point which was indicated by the brick-red color.

Calculation

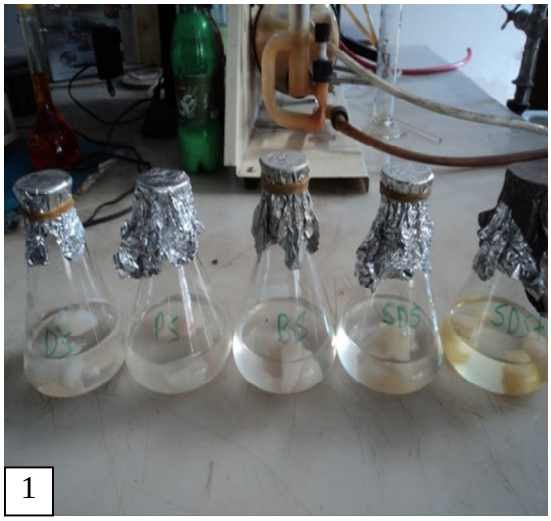
Salt content was determined by the following formula:

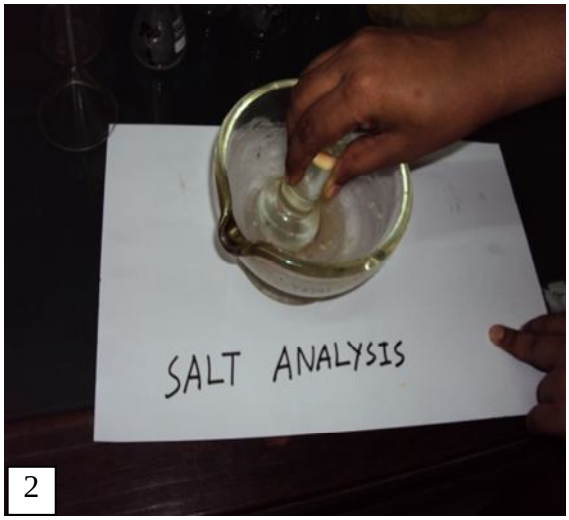
$$S_1 \times V_1 = S_2 \times V_2$$

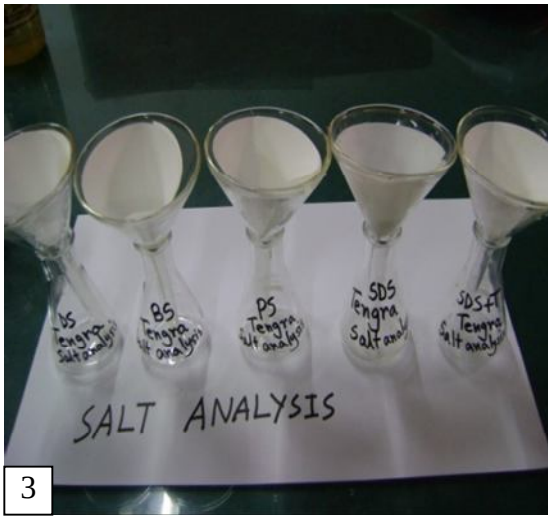
% of NaCl = $S_1 \times$ 58.5 (molecular wt. of NaCl)

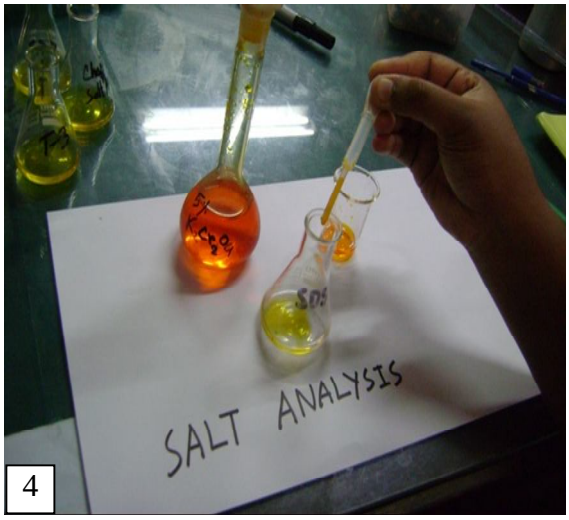
Where,
V_1 = Volume of sample
V_2 = Volume of titrant
S_1 = Strength of sample
S_2 = Strength of titrant ($AgNO_3$)

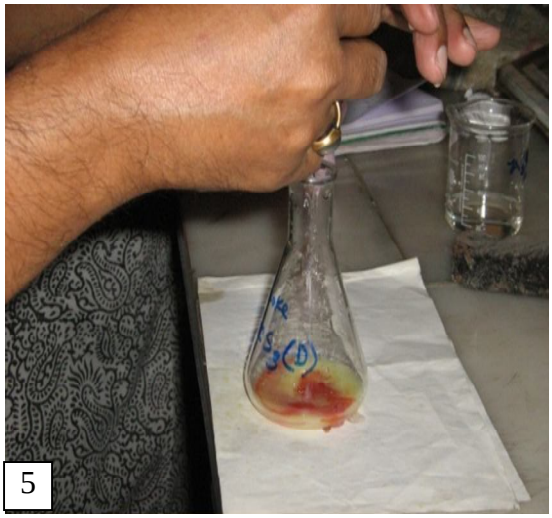
Various steps of salt content determination of the fish samples are shown in Plate 3.3.5.3(I).











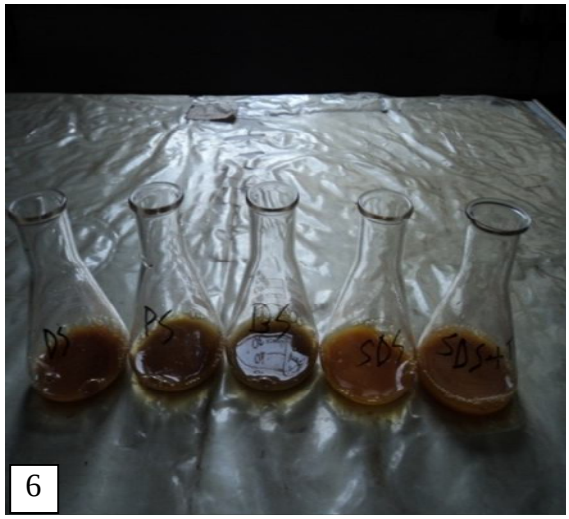


Plate 3.3.5.3(I). Showing various steps of salt content determination of the fish samples

3.3.5.3(II). Estimation of Total Volatile base Nitrogen (TVB-N)

Reagents required

1. 10% Trichloro Acetic Acid (TCA)
2. 2% boric acid
3. Potassium Carbonate (K_2CO_3)

Principle

TVB-N was determined by Conway modified micro-diffusion technique as described by Conway and Byrne (1933).

Procedure

Samples that were in the different levels of acceptability from highly acceptable condition to unacceptable condition had been selected for TVB-N analysis. 25 ml of 10% Trichloro-Acetic Acid (TCA) was added to 1-2 gm of ground fish sample and kept overnight and then filtrated with known volume. 2% boric acid, TCA, K_2CO_3 and the solutions made from the fish samples were taken into the Conway dishes in the following way (Table-2).

Table: 3.3.5.3(II). Showing the amount of chemicals taken in the inner and outer chamber of the Conway dishes

Conway dishes	Inner Chamber		Outer chamber	
	Chemical	Amount	Chemical	Amount
Blank	2% Boric acid	1 ml	K_2CO_3	1 ml
With Sample	2% Boric acid	1 ml	Sample Solution	1 ml
			K_2CO_3	1 ml

After the addition of Potassium Carbonate (K_2CO_3), each dish was covered by a piece of glass that was stacked with glue (Paraffin soft white) initially. Then it was kept for 24 hours. The samples and Potassium Carbonate (K_2CO_3) reacts to form NH_3 which was absorbed by the boric acid and then the solution of each Conway dish had been titrated by N/70 H_2SO_4 with the help of a micro-burette. Finally TVB-N was calculated.

Calculation

$$\text{TVB-N} = (\text{titration reading} - \text{blank reading}) \times \text{Strength of acid} \times 0.2 \times \frac{\text{Volume of extract}}{\text{Volume of extract taken}} \times \frac{100}{\text{Weight of sample taken}}$$

Flow chart of TVB-N Determination is given below:

Weiging 1-2 gm of prepared sample (homogenized)

↓

mixing with 25 ml of 10% TCA

↓

Grinding with mortal pastel

↓

Filtration the mixture

↓

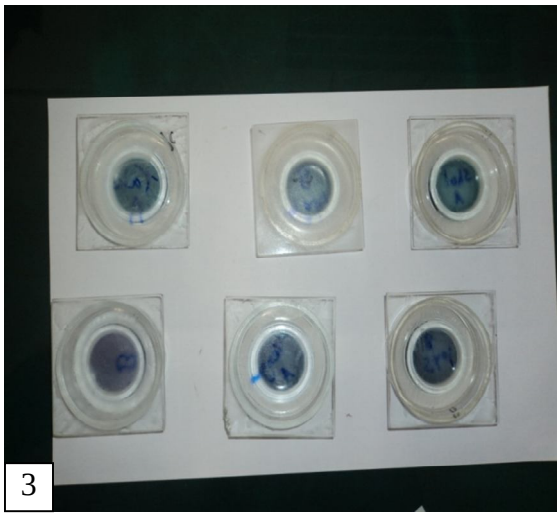
Filtration extract

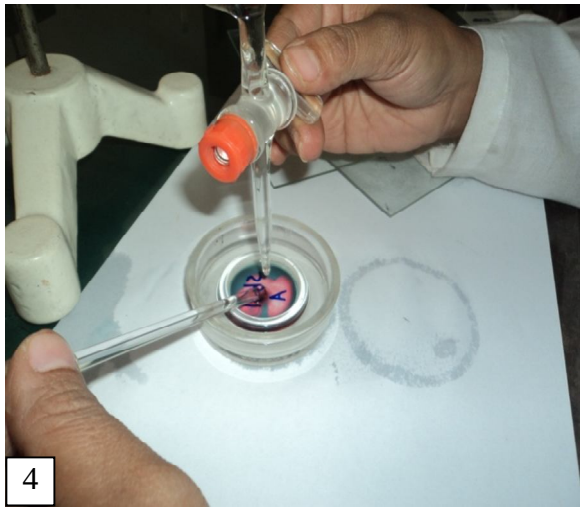
↓

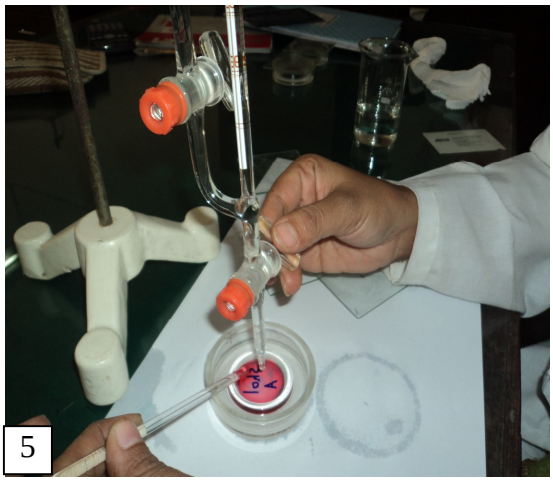
1 ml of 2% Boric acid in Inner Chamber and 1 ml of sample solution and K_2CO_3 in outer Chamber for over night in a dark place

Figure. 3.3.5.3(II). Flow chart for determination of TVB-N

Various steps of TVB-N determination of the fish samples are shown in Plate 3.3.5.3(II).







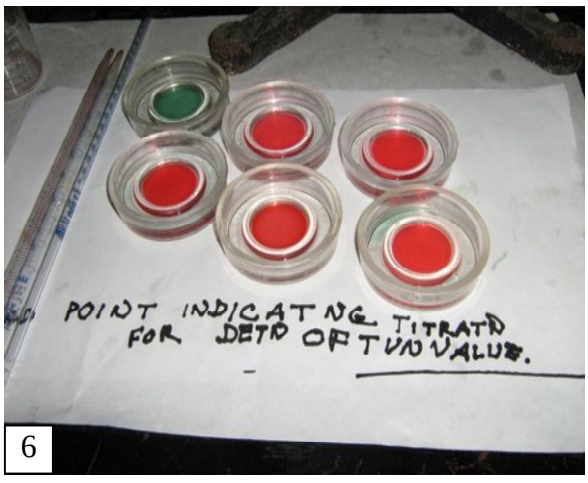


Plate 3.3.5.3(II). Showing various steps of TVB-N determination of the fish samples

3.3.5.3(III). Estimation of Free fatty acid (FFA) value

Reagents required

1. Folch reagent (chloroform and methanol in the ratio of 2:1 v/v)
2. 0.25 N sodium hydroxide (NaOH)
3. Ethanol
4. Phenolphthalein indicator

Principle

Among the various parameters to assess the extent of deterioration in fish, determination of free fatty acid (FFA) content has been widely used. Free fatty acid (FFA), a tertiary product of rancidity, increased during storage. The FFA is a measure of hydrolytic rancidity-the extent of lipid hydrolysis by lipase action. It produced as a result of fat oxidation (rancidity). FFA of the fish was determined by AOAC method (AOAC, 2000).

Extraction of fish oil from salted fish sample

Oil sample used throughout the work was prepared by extracting the salted fish by Folch reagent (chloroform and methanol in the ratio of 2:1 v/v).The salted fish was first cut into small pieces and then ground. The ground material was then mixed with folch reagent in a large wide mouthed stopper glass bottle for 24 hours at room temperature after 15 minutes stirring with glass rod. Extraction was facilitated by occasional stiring. The mixture was filtered through a Buchner funnel and the filtrate was evaporated in batches under heater or oven at 60^0 C.

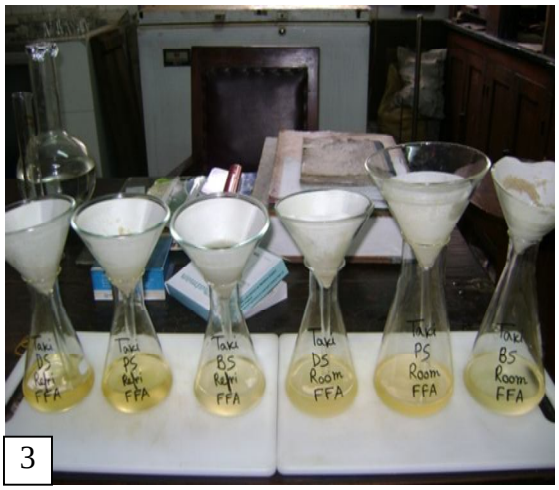
Procedure

Seven gram of well-mixed oil was taken into 250 ml flask and 50ml ethanol was added, previously neutralized by adding 2 ml phenolphthalein solution. Titration was done by 0.25 N sodium hydroxide with vigorous shaking until permanent final faint pink color appeared and persisted for at least one minute. The value was reported as percentage (%) of free fatty acid expressed as oleic acid. Milliliter of 0.25 N NaOH required for titration corresponds to the percentage of free fatty acid.

Various steps of FFA determination of the fish samples are shown in Plate 3.3.5.3(III).

1


2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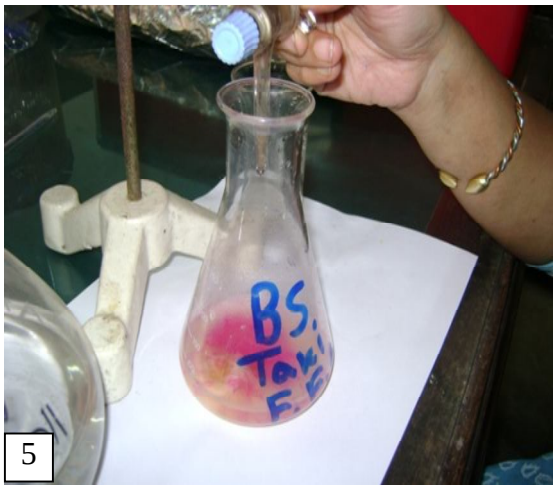

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Plate 3.3.5.3(III). Showing various steps of FFA determination of the fish samples

3.3.5.3(IV). Estimation of pH value

Principle

Eyo (1993) stated that pH is an indicator of the extent of microbial spoilage in fish and some proteolytic microbes produce acid after decomposition of carbohydrate, thereby increasing the acid level of the medium. pH was measured according to the method of Vyncke (1981).

Procedure

A 1 g sample of the fish flesh was homogenized in 10 ml of distilled water and the mixture was filtered. The pH of the filtrate was measured using a pH meter.

Various steps of pH determination of the fish samples are shown in Plate 3.3.5.3(IV).

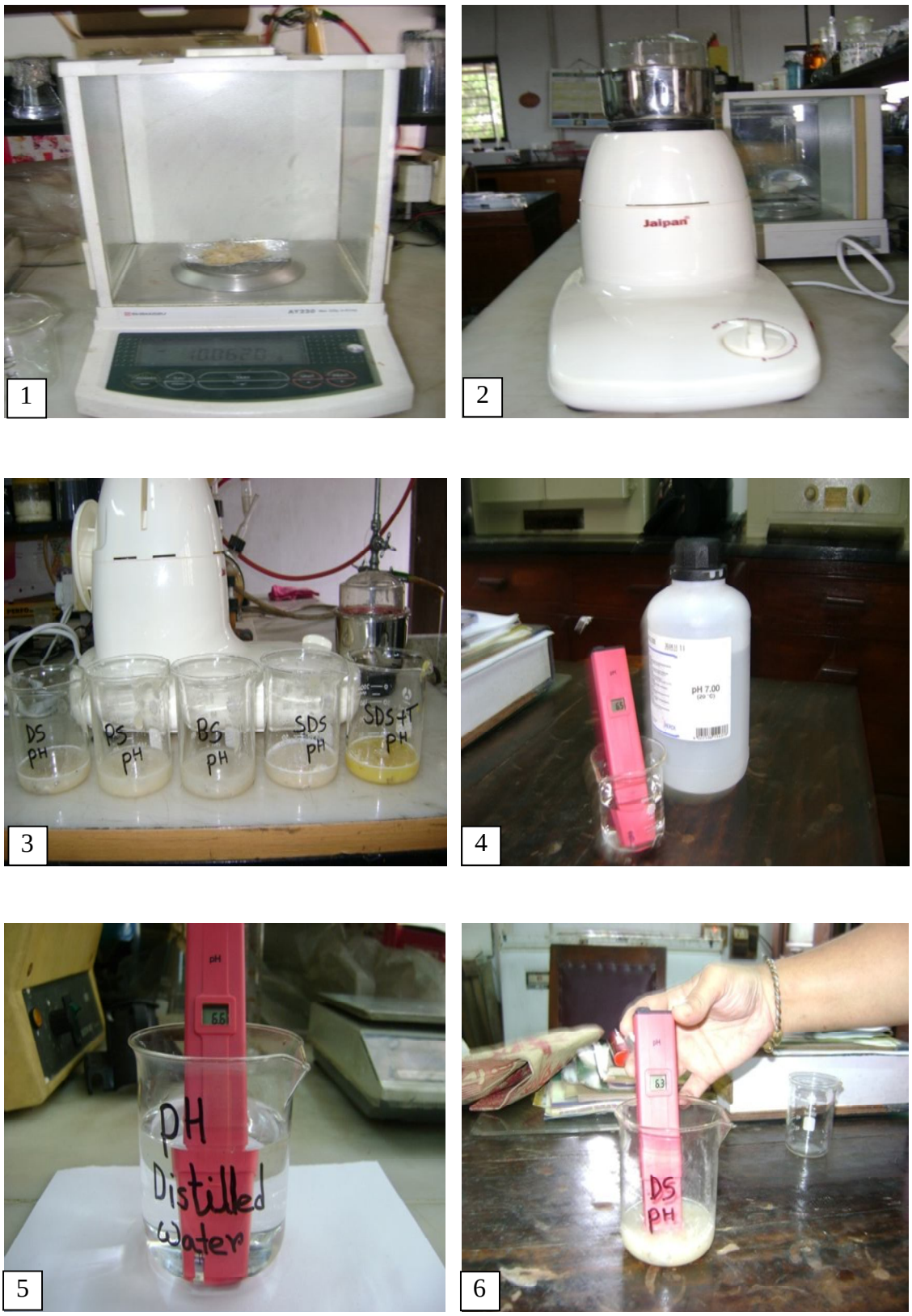


Plate 3.3.5.3(IV). Showing various steps of pH determination of the fish samples

3.3.5.4. Study of mineral composition

Principle

Samples for mineral analysis were prepared according to recommendations of Perkin-Elmer's procedures of Atomic Absorption Spectrometer (1996).

Procedure

(i) Sample preparation

Fish muscle was removed aseptically from the fish with sterile knife and was chopped in order to make smaller pieces and the sample was homogenized. Accurately 1 g of chopped sample was weighed and placed into a Teflon vessel.

(ii) Digestion

The digestion was done using CEM Microwave digester and the digested samples were serially diluted and the mineral content was analyzed by a Perkin-Elmer atomic absorption Spectrophotometer. All the samples were analyzed in the same procedure.

(iii) Mineral Analysis

1 gm sample was put into a Teflon vessel. 10 ml of concentrated nitric acid (HNO_3) was added and digested in a microwave oven. The samples were allowed to predigest by standing open for a minimum of 15 minutes before sealing the vessels and proceeding to heating program (CEM system).

The condition of microwave digestion was given in Table 3.3.5.4(a).

Table: 3.3.5.4(a). The condition of microwave digestion

Stage	Power (W)		Ramp Time (min)	Temperature	Hold time
	Level	%			
1	1600	100	10	180	15
2	1600	100	10	120	15

After digestion the content in Teflon vessel was dissolved in de-ionized water and filtered into 25 ml volumetric flask quantitatively and brought up to the mark with deionized water. The digested sample solutions were subsequently analyzed for the minerals Ca, Mg, Fe, Cu, Zn and Mn by an automatic sampler and analyzed using an air acetylene flame in combination with single element hollow cathode lamps into an atomic absorption spectrometer, having a detection limit as described in Table 3.3.5.4 (b).

Table: 3.3.5.4(b). List of Wavelength and detection limit for mineral estimation

Mineral	Hollow cathode lamps	Wavelength (nm)	Minimum detection limit
Calcium (Ca)	Calcium hollow cathode lamps	422.7	0.06 mg/L
Magnesium (Mg)	Magnesium hollow cathode lamps	285.2	0.01 mg/L
Iron (Fe)	Iron hollow cathode lamps	248.3	0.04 mg/L
Copper (Cu)	Copper hollow cathode lamps	324.8	0.03 mg/L
Zinc (Zn)	Zinc hollow cathode lamps	213.9	0.01 mg/L
Manganese	Manganese hollow cathode lamps	279.5	0.02 mg/L

The wavelength correlation coefficient and detection limit of each mineral in listed in Table 3.3.5.4(c).

Table: 3.3.5.4(c). Correlation of mineral in standard solution for calibration curve

Mineral	**Mg/L**	**Correlation coefficient**
Calcium (Ca)	0.5, 1.0, 3.0,5.0	0.997687
Magnesium (Mg)	0.5, 1.0, 3.0, 5.0	0.999787
Iron (Fe)	0.5, 1.0, 3.0	0.996787
Copper (Cu)	0.5, 1.0, 2.0, 3.0	0.999717
Zinc (Zn)	0.5, 1.0, 3.0, 5.0	0.998787
Manganese	0.5, 1.0, 3.0,5.0	0.997687

(iv) Mineral estimation

The sample had to be diluted many folds to keep the result in the analytical range. The mineral content of the sample was calculated by multiplication by the appropriate dilution factor. The relationship between spectrophotometer readings and mineral concentration was linear over a range of concentration represented in each working standard.

(v) Calculation of mineral

The amounts of minerals (Ca, Cu, Fe, Mn and Zn) in samples per 100g edible portion were calculated from the readings of AAS at specific wavelength using specific hallow cathode lamp by using the following formula:

$$\text{Mineral} = \frac{\text{Conc. from AAS (Mg/L) for sample} \times \text{Volume of original digest} \times \text{Dilution factor} \times 100}{\text{Weight of sample (g)} \times 1000}$$

Various steps of minerals determination of the fish samples are shown in Plate 3.3.5.4.

Plate 3.3.5.4. Showing various steps of minerals determination of the fish samples

3.3.5.5. Bacteriological study

In this study, different bacteriological analyses [Standard Plate Count (SPC) or total aerobic plate count (APC) and total halophilic bacteria count (HBC)] were carried out to determine the safety of 3 fish species, in fresh condition and 5 types of salted condition. Strict aseptic procedures were followed in every step of the study.

Reagents

1. Plate count Agar (Hi-media) - The composition of the media was used for culturing bacteria to perform Aerobic Plate Count (APC) is as follows:

Ingredients	**Quantity**
Tryptone	5.0g/l
Yeast extract	2.5g/l
Dextrose	1.0g/l
Agar	15.0g/l

2. Ringers Stock-

Ingredients	**Amount(gm/L)**
Sodium Chloride (NaCl)	9.0
Potassium Chloride (KCl)	0.42
Calcium Chloride (CaCl)	0.48
Sodium- bi-carbonate (Na_2CO_3)	0.20
Distilled water	1000 ml

3. NaCl for halophilic (Salt tolerance) Bacteria count

4. Distilled water

Principle

SPC and HBC were done according to the Standard methods of AOAC (AOAC, 1995) and FDA BAM (2001).

Procedure

(i) Preparation of Media

23.5 g of ager media were weighed by electric balance and were dissolved in 1 liter of distilled water with the help of magnetic stirrer. The mixture was then boiled to mix all the ingredients thoroughly.
For halophilic bacteria count 3g NaCl was added with 23.5 g of ager media and dissolved in 1 liter of distilled water (FDA BAM, 2001).

(ii) Preparation of Ringers Solution

100 ml ringer stock was mixed with 800 ml distilled water. So, 900 ml ringer solution was prepared.

(iii) Sterilization

All of the media and ringers solution were sterilized before using them in order to kill all bacteria, fungus or their spores present in the media or in the glassware if remain. Sterilization was performed by placing them into a horizontal autoclave for 30 minutes at a temperature of 121^0C under 15 lb per square inch pressure.

(iv) Sample preparation

Fish muscle was removed aseptically from the fish with sterile knife and was chopped in order to make smaller pieces and the sample was homogenized. Accurately 10 g of chopped sample was weighed and placed in a special bag of a homogenizer

(Stomacher 400). 90 ml of previously sterile ringer solution was added on this and it was blended for 30 - 60 seconds in the Stomacher machine for homogenized it. Thus 1: 10 dilution of the sample was obtained.

(v) Dilution

Serial dilutions of the samples (10^{-2} -10^{-7}) were prepared before subsequent culturing with standard methods (Harrigan, 1998). 1 ml of the diluted fish-sample was taken out from prepared stock solution and then was transferred with a sterile pipette to a test tube labeled as 10^{-1}, containing 9 ml of previously sterile ringer solution and the test tube was shaken thoroughly for mixing with the help of vortex machine. Again about 1 ml diluted solution from the test tube labeled as 10^{-2} was transferred to a test tube containing 9 ml sterile diluents and labeled as 10^{-3}. Using a similar process several ten-fold dilutions (decimal dilution) were made up to desired level.

Serial dilution chart of homogenized fish samples are shown in Figure 3.3.5.5.

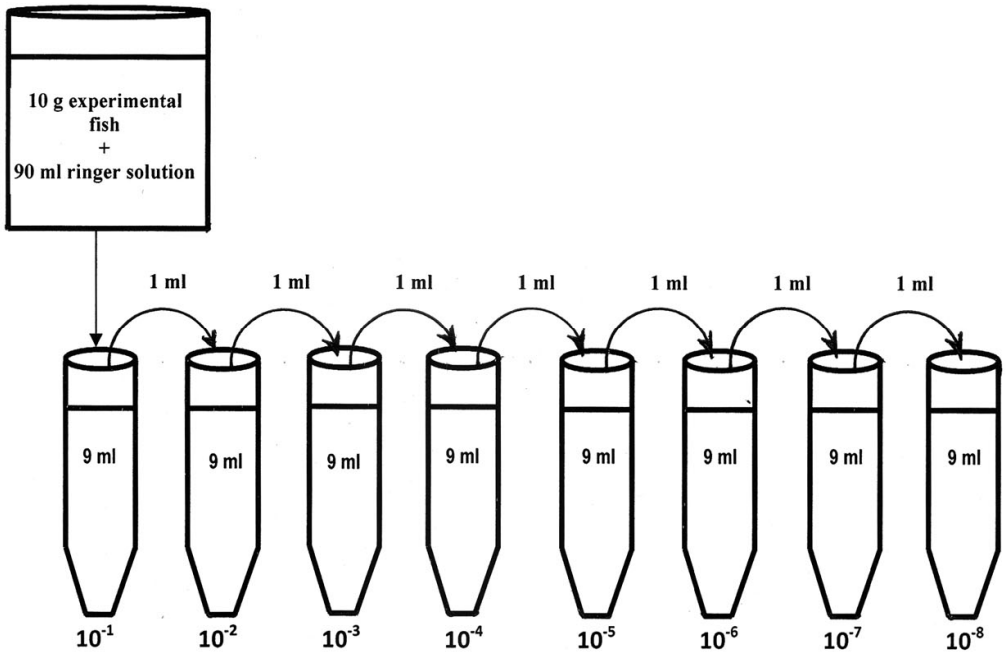


Figure. 3.3.5.5. Serial dilution chart of homogenized fish samples

(vi) Quantitative Study

Aerobic plate count (APC) or standard plate count (SPC) and halophilic bacteria count expressed as colony forming units per gram (cfu/g) of fish samples on different days of storage were determined by consecutive dilution technique.

(a) Enumeration of Standard Plate Count/Total Viable Count (SPC)

The samples pippeted were performed in pour plate techniques. 1 ml of the diluted solution was then transferred to Petri dish labeled as 10^{-1}, 10^{-2} -10^{-7} from the corresponding test tubes. Then the melted, cooled plate count agar was poured into the petri dishes containing different dilutions of sample. The plates were rotated by hand at least 5 times in the clockwise direction and 5 times in anticlockwise direction. Finally several times crosswise rotation was made for equal distribution of the media.

(b) Determination of Halophilic Bacteria

Sterilized agar media where 3% NaCl (salt) was added was poured on previously sterilized Petri dish just to cover its surface. It was done at a temperature of about 440C. Then it was allowed to cool down and solidify the media.
0.1 ml prepared, well shacked sample from a desired dilution was pipetted out using micropipette and transferred aseptically to the agar plates by raising the upper lids sufficient enough to admit the tip of the pipette. The pipetted samples were spread by L-shaped glass rods throughout the surface of the media until the samples were evenly distributed and dried out.

(vii) Incubation

The plates were incubated at 37˚C in an incubator. After 24 hours of incubation, colonies developed were counted using a colony counter for calculation.

(viii) Counting of plates

Only plates with 30-300 colonies were counted, and the average was multiplied by the relevant dilution factor to give the number of colony-forming units (cfu/g) of original sample (FDA BAM, 2001).

(ix) Calculation

The total Aerobic plate was calculated by following formula recommended by International Standard Organization (ISO).

$$\text{SPC in cfu/g of fish} = \frac{C \times D \times 10 \times V}{S}$$

Where,

C= Number of colonies found
D= Dilution factor
V= Volume of original sample
S= Weight of sample in grams
cfu= Colony Forming Unit

Various steps of bacteriological study of the fish samples are shown in Plate 3.3.5.5.

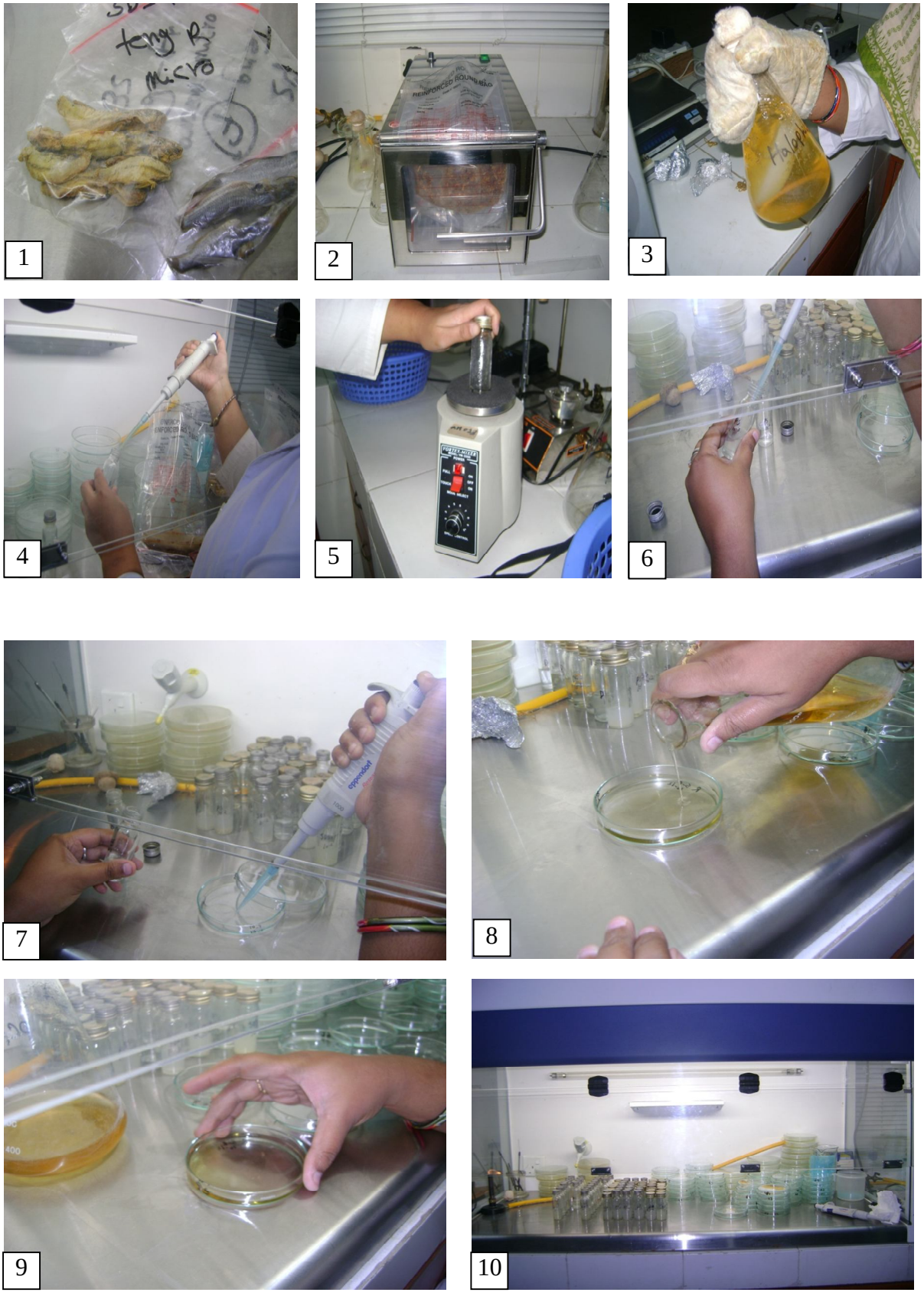


Plate 3.3.5.5. Showing various steps of bacteriological study of the fish samples

3.3.6. Statistical analysis

Biochemical composition of fresh fish and fresh process dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) shol, taki and tengra fish-products analysis was done with triplicates sample. Results presented are mean values of each determination ± standard deviation (SD). Analysis of variance was performed by one-way ANOVA procedures (SPSS, 2013). Differences between the mean values of fish and treatments were determined by the LSD (least significant difference) test and the significance was defined at $p<0.05$ (Sokal and Rohlf, 1987).

To see the degree and nature of relationship, between moisture & protein, moisture & fat, protein & fat and moisture & TVB-N of room and refrigeration stored 5 different types (DS, PC, BS, SDS, SDS+T) of salted shol, taki and tengra fish, coefficient of correlation ‘r’ and paired ‘t’ test was done with the help of SPSS 20 data analysis program.

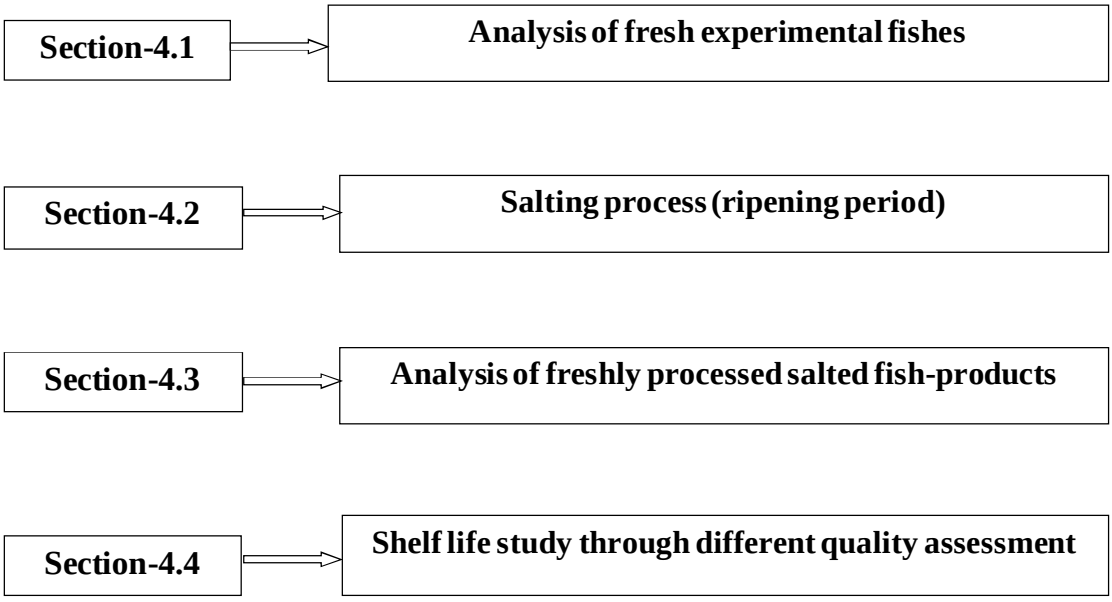
CHAPTER-4

RESULTS AND DISCUSSION

CHAPTER-4

RESULTS AND DISCUSSION

The results of the study in different experiments are arranged systematically in 4 sections, as cited bellow



Section-4.1: Analysis of fresh experimental fishes

It is well accepted fact that fish are a good source of animal protein throughout the world. Fish flesh generally contains up to 80% moisture, 15-25% protein, 1-2% mineral matter (CSIR, 1962). The composition of a particular species often appears to vary from one habitat to another, and season to season, but the basic causes of change in composition are usually variation in the amount and quality of food it eats and in the amount of movement it makes (Murray and Burt, 2009).

In this research work it was considered three freshwater fish species viz;

1) Large size shol (*Channa striatus*) fish,
2) Medium size taki (*Channa punctatus*) fish, and
3) Small tengra (*Mystus tengra*) fish- for salt curing.

The size variation of selected fish species for present experiments bears significant consideration. Rahman (2006) stated that soft textured fish tend to absorb salt faster than tough or firm-textured fish. He also stated that high fat content fish absorb salt slower than low-fat fish. The small indigenous species of fishes in Bangladesh are generally considered to be those which grow to a length of approximately 5-15 cm at maturity (Felts *et al.* 1996). It is assumed that the size of fishes considered to be selected for preparing any processed product has got some significance on the shelf life and quality of the product. This type of finding was revealed in the experiment of Hoffmen *et al.* (1974). They mentioned that size of the fish is an important factor of spoilage. Similarly, Ahmed *et al.* (1986a) showed in case of fresh water fish, that the small fish spoiled earlier than the larger ones at the same storage conditions at room temperature. Muslemuddin *et al.* (1991) had the same opinion that spoilage rate was found higher in small fish than the larger ones kept in same condition. Therefore, consideration of different fat content and different sizes of experimental fish species in this research work may be considered as justifiable. The initial quality of raw fish material strongly influences subsequent performance in processing and storage.

4.1.1. Physical features of the fresh experimental fish, shol (*Channa striatus*), taki (*Channa punctatus*) and tengra (*Mystus tengra*)

Physical features of three experimental fresh fish species, shol (*Channa striatus*), taki (*Channa punctatus*) and tengra (*Mystus tengra*) are shown in the Table 4.1.1.

Table: 4.1.1. Physical features of the fresh experimental fish, shol (*Channa striatus*), taki (*Channa punctatus*) and tengra (*Mystus tengra*)

Parameters	Shol (*C. striatus*)	Taki (*C. punctatus*)	Tengra (*M. tengra*)
Mean length (cm)	40.4	22.0	9.7
Mean weight (g)	561.5 (±20)	128.5 (±6)	9.1(±1.2)
Color	Grayish, abdomen whitish	Blackish	Blackish with a dark shoulder spot and with 4-5 longitudinal bands
Flesh / Texture	Firm and elastic	Firm and elastic	Firm and elastic

Mean length of shol, taki and tengra fish was 40.4, 22.0 and 9.7 cm and mean weight was 561.5 (±20), 128.5 (±6) and 9.1(±1.2) g respectively. The color of shol fish was grayish, abdomen whitish; taki fish was blackish and tengra fish was blackish with a dark shoulder spot and with 4-5 longitudinal bands whereas flesh or texture of three fishes was firm and elastic.

4.1.2. Proximate and chemical (bio-chemical) composition of the fresh experimental fish, shol (*Channa striatus*), taki (*Channa punctatus*) and tengra (*Mystus tengra*)

Proximate and biochemical analysis provides information on the nutritional value of a particular organism used as a source of food (Zafar *et al.*, 2004). It has been established that the proximate composition of fish may vary in different species and even within the same species from one individual to another is mainly due to age, sex, season, size, species, starvation of the day, environmental factors, nutritional status, energy spending procedure and so on (Sankar and Ramachandran, 2001).

Mean initial proximate composition and some chemical composition of three experimental fresh fish species, shol (*Channa striatus*), taki (*Channa punctatus*) and tengra (*Mystus tengra*) are shown in the Table 4.1.2.

Table: 4.1.2. Mean initial proximate and chemical composition of the fresh experimental fish, shol (*Channa striatus*), taki (*Channa punctatus*) and tengra (*Mystus tengra*)

Parameters	Shol (*Channa striatus*)	Taki (*Channa punctatus*)	Tengra (*Mystus tengra*)
Moisture (%)	77.03±0.12^{a}	78.65±0.07^{b}	74.27±0.07^{c}
Protein (%)	19.52±0.07^{a}	16.89±0.10^{b}	16.96±0.07^{b}
Fat (%)	1.93±0.07^{a}	2.50±0.06^{b}	6.04±0.06^{c}
Ash (%)	1.44±0.11^{a}	1.36±0.11^{a}	2.67±0.08^{c}
TVB-N (mg/100g)	4.41±0.01^{a}	3.43±0.02^{b}	4.27±0.02^{c}
FFA (%) (Free fatty acid)	0.6±0.06^{a}	0.5±0.10^{ba}	0.9±0.21^{ca}
pH	6.9±0.06^{a}	7.0±0.06^{a}	7.0±0.10^{a}

Values are shown as mean ± standard deviation of triplicate measurements**;** **a, b, c:** Means significant differences ($p<0.05$) between groups. Common superscript letter **(Blue color)** within a row are not significantly different ($p>0.05$) from each other using LSD test.

From the table it is clear that fresh fish samples have high moisture and low protein content, similar to previously reported by Eyo (1998) and Davies and Davies (2009).

The major component of fish muscle was found to be moisture. The moisture contents of shol, taki and tengra fish in fresh condition was found 77.03±0.12, 78.65±0.07 and 74.27±0.07% respectively which resembles with the findings of Clucas and Ward (1996) who observed the moisture content of fresh water fish ranged from 70-80%. It also indicates that the percentage moisture in fish muscles was within the acceptable level (60-80%) in all the samples. The high moisture content in all three species is within the previously reported range in other fishes (Gallagher *et al*., 1991). This result also coincides with the findings of Nabi and Hossain (1989) in *M. aculeatus,* Salam *et al.* (1995) in *P. gonionotus* and of Mazumder *et al.* (2008) in *Amblypharyngodon mola, Panchax chola, Gudusia chapra* and in *P. atherinoides.* This finding also agreed with observation of Marais and Erasmus (1977) in several freshwater fish species.

The protein (%) of shol, taki and tengra fish in fresh condition was 19.52±0.07, 16.89±0.10 and 16.96±0.07% which are in accordance with the findings of FAO (2015) for freshwater fish. The relatively high to moderate percentage of crude protein may be attributed to the fact that these fishes are good source of protein (Burgess, 1975). But the differences observed among the selected species could be as a result of fish consumption or absorption capability and conversion potentials of essential nutrients from their diets or their local environment. Similar findings were revealed by Onyia *et al.* (2010); Jabeen and Chaudhry (2011) and Fawole *et al.* (2007). Present findings also have got more or less similarities with the findings of Nabi and Hossain (1989) in *Mastacembelus aculeatus* and of Mazumder *et al.* (2008) in *G. chapra and P. chola*. However, Salam (2002) reported that the protein in *Heteropneustes fossilis* was 18.25% which was higher compared with the present findings of taki and tengra and this might be to species variation. The high protein content of shol fish may results from the equally high amount of live feed in the regular diet (fish items, crustaceans, algae and diatoms). However, the average protein content may vary from the seasonal physiological variation.

All the fish species examined belonged to high-protein group of fish which resembles with the findings of Govindan (1985) who observed the protein content of such fresh water fish ranged from 13-20%. Adewumi *et al.* (2014) observed that fresh water fish contains protein ranged from 18-23%. Indeed the protein content of the fish species is higher than that of the egg yolk (15%) as reported by CFCD (2002).

It is also shown in Table 4.1.2 that fat content of fresh shol, taki and tengra fish, was 1.93±0.07, 2.50±0.06 and 6.04±0.06% respectively which was more or less similar to the findings of Habashi (1972) in *Cyprinus carpio*, Nabi and Hossain (1989) in *P. gonionotus* and of Mazumder *et al.* (2008) in *G. chapra, P. chola, A. coila* and *A. mola.* Salam (2002) estimated the highest fat content as 3.25% in *H. fossilis* which was less than that of tengra. These differences might be due to the reason of species variation. According to Bandarra *et al.* (1997) and Osman *et al.* (2001), fat content varies within species and is affected by the catching season whereas Agren *et al.* (1991) stated that, fat content is found to be influenced by geographic location.

Protein and fat are the major nutrient in fish and their level help to define the nutritional status of a particular organism (Aberoumad and Pourshafi, 2010). According to Ackman (1989), fish can be grouped into four categories according to their fat content: lean fish (< 2 %), low fat (2 to 4 %), medium fat (4 to 8%), and high fat (> 8%). According to Stansby (1962), low oil-high protein fish contained <5% oil and 15-20% protein; medium oil-high protein fish contained 5-15% oil and 15-20% protein and low oil-very high protein fish contained <5% oil and >20% protein.

From above point of view it can be said that, shol fish can be grouped as low oil-very high protein fish, taki can be grouped as low oil-high protein fish whereas tengra can be treated as medium oil-high protein fish. Mohamed (2013) stated that fish species with low levels of fat are suitable to be processed.

The value of ash in fresh shol, taki and tengra fish was 1.44±0.11, 1.36±0.11and 2.67±0.08% respectively which was nearer to the values obtained by Mazumder *et al.*

(2008) in *Ailia coilia* and in *Amblypharyngodon mola*; by Salam (2002) in *Heteropneustes fossilis* and by Abimbola *et al*. (2010) in *T. guineensis* and *T. melanotheron. However,* comparatively higher ash value of 3.06% in *Clarias gariepinus* reported by Chukwu and Shaba (2009), which was higher than that of shol, taki and tengra.

The ash content gives a measure of the total mineral content in the tissue (Nair and Mathew, 2001). From this point of view, the present findings state that the ash contents of these three freshwater fishes might be a good source of minerals such as calcium, potassium, zinc, iron and magnesium which is in harmony with the findings of Oniya *et al.* (2010).

Findings of this experiment in respect of fat and ash content of fresh fish have got similarities with the findings of Srivastava (1985) and Balachandran (2001) who has got percent of fat and ash ranged from 1.0-7.0% and 0.4-3.0% respectively.

According to Chakraborty *et al.* (2003), Moisture (%), Protein (%), Fat (%) and Ash (%) content of edible portion of shol (*C. striatus*) was 75.48 (%), 20.10 (%), 1.66 (%) and 2.63 (%); taki (*C. punctatus*) was 78.85 (%), 16.81 (%), 2.40 (%) and 1.94 (%); tengra (*M. tengra*) was 74.12 (%), 18.38 (%), 5.62 (%) and 1.77 (%) respectively which was more or less similar with present findings.

The Table 4.1.2 also shows that, TVB-N contents of fresh shol, taki and tengra fish was 4.41±0.01, 3.43±0.02 and 4.27±0.02 mg/100g respectively. The level of TVB-N in freshly caught fish is generally between 5 to 20 mg/100 g muscles (Egan *et al*., 1981). According to EEC (1995), the TVB-N value of this three fresh fish was much lower than the acceptable upper limits of 25-35 mg/100 g for some fresh fish species. The TVB-N of fish is an indicator of the freshness of the raw material (Zhou *et al.*, 2011). According to Pons-Sanchez-Cascado *et al.* (2006), TVB-N levels of 10 mg/100g or less for fresh fish.

FFA (%) value of fresh shol, taki and tengra fish was 0.6±0.06%, 0.5±0.10% and 0.9±0.21% respectively. According to Huss (1988) and Eyo (1993) the acceptable limit of FFA in fresh fish is about 0.5 to 1.5%. The FFA (%) value in this experiment with three fish species showed the values within the range of acceptable limit. Free fatty acids are not only important from the point of view of oxidation products, but they have also been reported to have a direct sensory impact (Refsgaard *et al.* 1998).

According to Paul *et al.* (2013), FFA (%) value of fresh *C. striatus* and *C. marulius* were found in the ranges of 0.88 ± 0.02% and 0.92 ± 0.04%. According to Molla *et al.* (2007), FFA (%) value of fresh *M. vittatus* was 0.96%. The above study was just similar of present findings.

pH value of fresh shol, taki and tengra fish was 6.9±0.06, 7.0±0.06 and 7.0±0.10 respectively. The pH in fresh condition fresh-water fish flesh is almost neutral (Virta, 2009; Osibona *et al.*, 2010). This is in agreement with the initial pH values of these three fishes.

It is also shown in Table 4.1.2 that, moisture, fat and TVB-N content of shol, taki and tengra fish are significantly different ($p<0.05$). But, in case of protein, no significant difference ($p>0.05$) was shown between taki and tengra fish. No significant difference ($p>0.05$) between shol and taki in case of ash content whereas in case of FFA value, Shol is insignificantly different with both taki and tengra but between FFA value of taki and tengra they are significantly different ($p<0.05$). No significant different ($p>0.05$) were present in shol, taki and tengra in case of pH.

The variation in proximate composition of experimental fish, shol (*Channa striatus*), taki (*Channa punctatus*) and tengra (*Mystus tengra*) in fresh condition are shown in Figure 4.1.2.

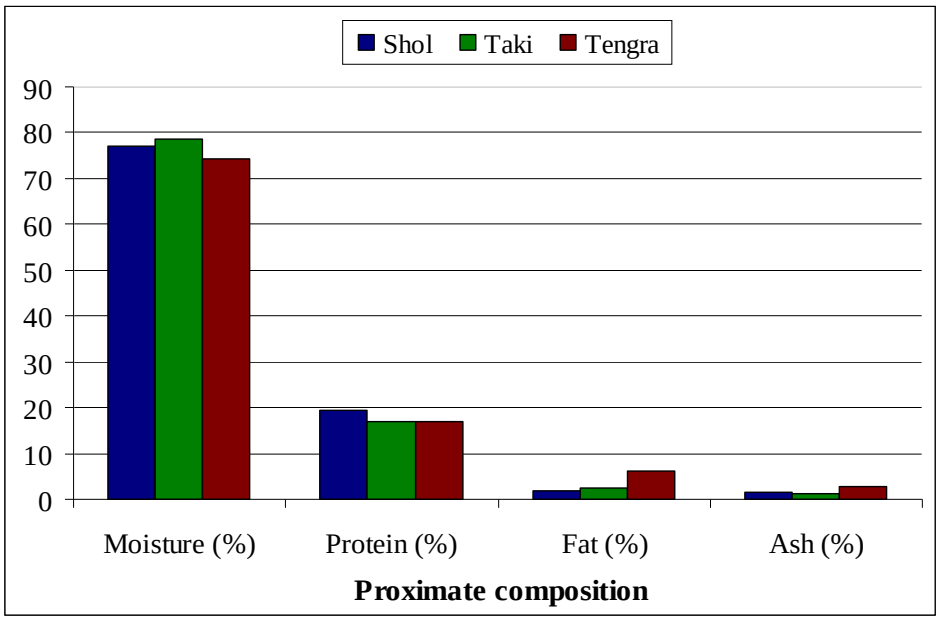


Figure. 4.1.2. Showing variation in proximate composition of experimental fish, shol (*Channa striatus*), taki (*Channa punctatus*) and tengra (*Mystus tengra*) in fresh condition

In this figure, it is clear that the highest moisture content was in taki fish whereas lowest was recorded in tengra fish. The variation in the protein content of shol was found to be little higher than taki fish. It is in likely with the findings of previous workers and is within the range found for other members of the family Channidae (Fapohunda and Ogunkoya, 2006; Baliu *et al.*, 2007; Narhasan, 2008; Ama-Abasi and Ogar, 2013). The fat content recorded for shol and taki was on the low range as against tengra having high fat. Ash is a measure of the mineral content of food item. In the Figure 4.1.2 the highest ash content was in tengra whereas the lowest was in taki. The highest ash content of tengra fish could be examined by the presence of the bones in the samples. It is in agreement with Daramola *et al.* (2007) who stated that; smaller sized fish species has higher ash content due to the higher bone of flesh ratio.

4.1.3 Important mineral (mg/100g of fish) composition in fresh shol (*Channa striatus*), taki (*Channa punctatus*) and tengra (*Mystus tengra*) fish

The study of micro-nutrients present in living organisms is of biological importance because many of such micro-nutrients take part in some metabolic processes and are known to be indispensable to all living things (Shul'man, 1974). Fishes contain small amount of these micro-nutrients, some of which are essential nutrients, being components of many enzymes system and metabolic mechanisms that contribute to the growth of the fish.

Important mineral (mg/100g of fish) composition of macro and micro elements in three experimental fish in fresh condition is reported in Table 4.1.3.

Table: 4.1.3 Important mineral (mg/100g of fish) composition of macro [Calcium (Ca) and Magnesium (Mg)] and micro elements [Iron (Fe), Copper (Cu), Zinc (Zn) and Manganese (Mn)] in experimental fresh fish, shol (*Channa striatus*), taki (*Channa punctatus*) and tengra (*Mystus tengra*)

Experimental fishes	Macro elements		Micro / Trace elements			
	Ca mg/100g	Mg mg/100g	Fe mg/100g	Cu mg/100g	Zn mg/100g	Mn mg/100g
Shol (*C. striatus*)	11.2	10.125	1.475	0.7	0.25	0.1
Taki (*C. punctatus*)	16.35	9.425	1.275	0.65	0.425	0.05
Tengra (*M. tengra*)	22.025	11.4	2.25	0.55	1.275	0.125

From this table it can seen that, the macro elements Ca, Mg were abundant in all the fishes examined while micro elements Fe, Cu, Zn, Mn were present in trace amounts.

Fresh shol, taki and tengra fish had calcium (Ca) content of 11.2, 16.35 and 22.025 mg/100g and magnesium (Mg) content of 10.125, 9.425 and 11.4 mg/100g of fish respectively. Small fishes are mostly eaten with bones, so the amount of available Ca and Mg is high in tengra fish than in other two fishes. Ca is essential in human body for the formation of bones, muscle tone and nervous impulse (Mollah *et al.,* 2000) and it has been reported that, fractional Ca absorption in human body is 24% from small fish (Larsen *et al.,* 2000). Ca may also protect against cardiovascular diseases by lowering blood pressure (Appel *et al.,* 1997; NNR, 2004). In that case, tengra fish as a whole one may be considered as one of the good source of Ca in the diet. Fishes generally have higher Ca content than the meat alone. Rose (1933) reported Ca contents of 22 mg/100 g in the meat of lean fish (edible parts) and 19 mg/100 g in fat fish. The Ca content of the flesh of some Mediterranean fish ranged from 15.4 mg/l00 g in plaice and 80.1 mg/100 g in sole (Martinez, 1995); 4.7-51.4 mg/l00 g in Indian fish (Chandrashekar and Deosthale, 1993) and 3.0-12.7 mg/ l00 g in Canadian fish (Belinsky *et al.,* 1996). On the other hand, Mg is an essential co-factor for multiple enzymes involved in the glucose metabolism (Murray *et al.,* 2003).

The trace element contents in the fishes examined recorded very low in trace amounts. Some trace elements were present below the level of detection in some fish samples. Fresh shol, taki and tengra fish had Fe content of 1.475, 1.275 and 2.25 mg/100g; Cu content of 0.7, 0.65 and 0.55 mg/100g; Zn content of 0.25, 0.425 and 1.275; Mn content of 0.1, 0.05 and 0.125 mg/100g of fish respectively which is more or less similar with the findings of Achionye-Nzeh *et al.* (2011) who observed that Fe, Cu, Zn and Mn of fresh water fish (*Channa obscura*) was 10.3, 0.25, 2.05 and 4.8 mg/kg of fish.

Among the trace elements, Fe content was dominant in these three fishes. This trend agrees with other studies where elevated amount of Fe was found in fish tissue (Asuquo *et al.,* 2004; Dural *et al.,* 2006; Yilmaz *et al*., 2007; Uysal *et al.,* 2009; El-Naggar *et al.,* 2009; Dahunsi *et al.,* 2012; Ogundiran *et al*., 2014). Report has shown that Fe is one of the most abundant metals in the earth crust (Ibrahim and Tayel, 2005). The high value of Fe observed in this study compared to other minerals may be

due to increase in total dissolved iron in the Meghna River. Fe content of three fishes was found near to Rohu fish (9.9 mg/kg) as reported by Jyothirmayi *et al.* (2009). Some other marine-fish like bass, cod, salmon and halibut are good source of Fe containing 4.2, 9.4, 8.6 and 9.5 mg/100g of fish respectively (Gehring *et al.*, 2011) which were much higher from our results. The Fe levels reported in this study fall within the range of Fe concentration reported by FAO (2015) for fish muscles.

Zinc (Zn) is known to be involved in most metabolic path-ways in humans (Malakootian *et al.,* 2011). The maximum zinc level permitted for fish is 50 mg/kg according to Food Codex. The values of Zn obtained from this study were lower from that finding and more or less similar with the findings of freshwater fishes in China (Du *et al.,* 2012). But, United States environmental protection agency and the European Commission (US-EPA and EC) have not considered any standards or limits for the zinc concentrations in fish (WHO, 2008).

Khan *et al*. (2014) observed Ca, Fe, Zn, Mn contents of farmed *L. rohita* was 0.142, 0.414, 0.049 and 0.052 mg/kg; farmed *C. catla* was 0.137, 0.376, 0.047 and 0.054 mg/kg; farmed *C. cirrhosus* was 0.124, 0.387, 0.039 and 0.043 mg/kg; farmed *H. molitrix* was 0.101, 0.246, 0.031 and 0.034 and farmed *P. hypophthalmus* was 0.096, 0.119, 0.035 and 0.047 mg/kg respectively. According to Belinsky *et al.* (1996), Fe contents ranged between 0.2 and 0.4 mg/l00g and Cu contents ranged between 0.1 and 0.5 mg/l00g in whitefish *(Coregonus clupeaformis),* cisco *(Coregonusartedii)* and leak trout *(Salvelinus namaycush)* raw roe which is more or less similar with this 3 experimental fishes.

Cu concentration in three experimental fishes of this study varied from 0.55-0.7 mg/100g of fish (Table 4.1.2), which were lower than the maximum recommended standards of 30 mg/kg in food fish (FAO/WHO, 1983; WHO, 1985; FEPA, 2003). According to TFC (2002), standard level of 20 mg/kg dw (dry weight) has been reported for Cu. However, after new regulation in 2008, no maximum level was specified for Cu in the fish species and sea foods (TFC, 2008). Also, there is no

guideline on acceptable levels of Cu in fish suggested by European Economic Community or FAO/WHO (Mol *et al.*, 2010).

The Zn contents in three fishes of this study ranged from 0.25 to 1.275 mg/100g which were much lower than the recommended maximum limits of 50 mg/kg in fish (FAO/WHO, 1983; WHO, 1985).

According to Chandrashekar and Deosthale (1993) Mn concentration ranged from 0.009 to 0.07 mg/l00g in edible muscle of 20 Indian fish species which is lower from this 3 experimental fishes. Mn contents of shol (0.1), taki (0.05) and tengra (0.125) fish were in the acceptable level when compared to 0.01-0.05 mg/kg (WHO, 1985; FEPA, 2003; FAO, 2015) standards.

Variations in the concentration of minerals in fish muscles could be due to their concentration in the water bodies where they live (Yeannes and Almandos, 2003; Ali *et al*., 2001), the fish physiological state (Ako and Salihu, 2004) and the ability of the fish to absorb the elements from their diets and the water bodies (Adewoye and Omotosho, 1997). This is supported by the findings of Ricardo *et al*. (2002), Adewoye *et al*. (2003) and Fawole *et al*. (2007).

The variation in important mineral composition of macro and micro elements analyzed in experimental fish, shol (*Channa striatus*), taki (*Channa punctatus*) and tengra (*Mystus tengra*) in fresh condition is shown in Figure 4.1.3.

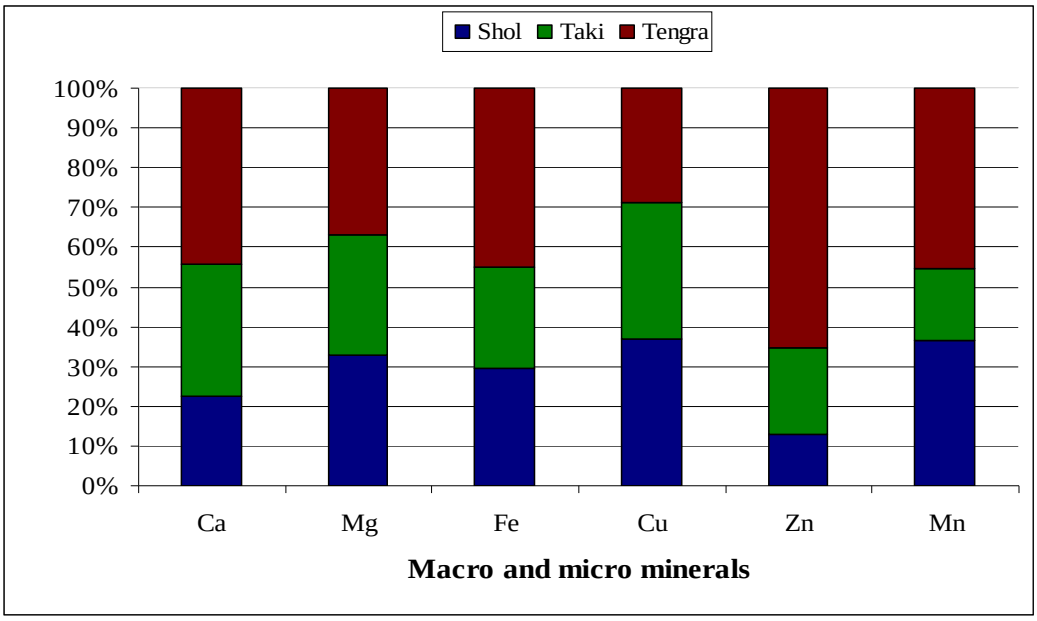


Figure. 4.1.3. Showing variation in important mineral (mg/100g of Fish) composition of macro and micro elements in experimental fish, shol (*Channa striatus*), taki (*Channa punctatus*) and tengra (*Mystus tengra*) in fresh condition

The highest amount of Calcium (Ca) was found in tengra and the lowest was found in shol. The highest calcium found in tengra fish is expected because of its deposition in the bone. Comparatively higher concentration of magnesium (Mg) was observed in tengra fish than other fish, which could be linked to the magnesium deposit in the fish bone.

Among the major elements (Ca and Mg), no well-defined order of magnitude within the same specie of fish was evident. However, the order of magnitude of these elements reported by Kirchgessner and Schwarz (1986) was Ca>K>Na>Mg which is similar with present findings. But this is contrary to the report of Oladimeji and Sadiku (1991) where the decreasing order K>Na>Mg>Ca was found in the edible muscle tissues of *S. galilaues*, *Lates niloticus* and *Synodontis schall*. Teeny *et al.* (1984) reported a similar decreasing order for minerals in several other species of fish.

Mineral (macro and micro) elements detected in this study were in the decreasing order Ca>Mg>Fe>Cu>Zn>Mn for shol and taki and Ca>Mg>Fe>Zn>Cu>Mn for tengra.

Jabeen *et al.* (2015) studied mineral composition of 3 freshwater fishes (*C. carpio, L. rohita and W. attu*) and found that these fishes have mineral composition in the decreasing order of Ca>Mg>Fe>Zn>Mn which is similar with our findings. Likewise, Gopakumar (2000) reported the values of major elements of Indian fishes in the above same order in most fishes.

The order of magnitude of the three trace elements of tengra fish are Fe>Zn>Cu and the same order of magnitude was reported by Kinsella *et al*. (1997); Ako and Salihu (2004); Onyia *et al.* (2010) in fillets of several species of fresh water fishes.

Fawole *et al.* (2007) reported decreasing order of Zn>Fe>Ni>Cu>As in the studies of some fresh water species. Other researchers have similar observation with regards to lack of agreement of different reports on the order of magnitudes of mineral contents of a given species of fishes (Akinneye *et al.*, 2007).

Mazumder *et al*. (2008) defined the decreasing order of magnitude (Zn>Fe>Mn>Cu) which was dissimilar from present findings.

4.1.4. Standard plate count (SPC) or total bacterial count (TBC) (cfu/g) of shol (*Channa striatus*), taki (*Channa punctatus*) and tengra (*Mystus tengra*) fish in fresh condition

The standard plate count (SPC) of three experimental fish shol (*Channa striatus*), taki (*Channa punctatus*) and tengra (*Mystus tengra*) in fresh condition is reported in Table 4.1.4.

Table: 4.1.4. Standard plate count (SPC) (cfu/g) of fresh experimental fish, shol (*Channa striatus*), taki (*Channa punctatus*) and tengra (*Mystus tengra*)

Shol (*C. striatus*) (cfu/g)	**Taki (*C. punctatus*) (cfu/g)**	**Tengra (*M. tengra*) (cfu/g)**
2×10^5	1.1×10^5	5×10^5

The standard plate count (SPC) in fresh three experimental fish shol, taki and tengra was recorded in 2×10^5, 1.1×10^5 and 5×10^5 cfu/g. From the table it was found that the highest bacterial load was in the tengra fish, whereas lowest was in taki fish.

Although it is widely accepted that the initial microbial load of freshwater fish varies depending on water conditions and temperature, most of the available literature on different freshwater and marine water species (Tilapia, Striped bass, Rainbow trout, Silver perch and Sea bream) reports bacterial counts of 2 to 7 log cfu/g (Moini *et al.*, 2009).

According to Vishwanath *et al.* (1998), total plate counts (SPC) in fresh *Monopterus albus* about 1.2×10^{-6} to 1.0×10^{-7}.

According to Das *et al.* (2007) 12 different species of fish were analyzed and highest standard plate count (5.1×10^6 cfu/g) was observed in Batashi (*Clupisoma atherinoides*) and lowest (2.3×10^5 cfu/g) in Rui (*Labeo rohita*).

Begum *et al.* (2010) observed that total bacterial count (SPC) was 2.64×10^6, 6.60×10^6, 1.64×10^6, 4.08×10^6, and 7.95×10^5 cfu/g in *Barbodes sarana*, *Labeo rohita*, *Oreocromis niloticus, Sperata seenghala* and *Corica soborna* fish respectively which was brought from local market.

The above results are in accordance with present study.

International Commission on Microbiological Specifications for Foods (ICMSF, 1996) indicated a limit of acceptability as less than 1.0×10^6 cfu/g for the total viable count in any food to be safe for consumption.

According to Surendran *et al.* (2006) the acceptable limit for bacterial count is 5×10^5/g for fresh fish. Yousef *et al.* (2007) stated that, if SPC counts 6 log cfu/g or higher, fresh fish begins to spoil.

The bacterial count in this experiment with three fish species showed the values within the range of acceptable limit.

Section-4.2: Salting process (ripening period)

Salting process is considered as one of the oldest methods of fish preservation and this process is still been used in several places around the world. Salting is generally aimed at reducing water activity (a^w) to reduce, obstruct or destroy the growth of the microorganism as well as inactive autolytic enzymes, where at the end, the fish meat goes its way to durability (Ashie *et al.*, 1996; Horner, 1997). The aim of salting is not only to prolong the shell life of fresh fish (Ahmed *et al.*, 1981a) but also to provide desirable sensorial changes (Andres *et al.*, 2005). The role of salt is highly significant to guarantee the quality and stability of the finished products. The preservative action of salt lies in the reduction of water activity of a system thus renders a condition less favorable for the microbial life (Barbut *et al.*, 1986; Luck and Jager, 1997).

Salting process starts when the surface of fish goes in contact with salt and is completed when all the fish reach the appropriate salinity, taste, consistency and odor. Performance of a salt curing method is judged by the capacity of the salting process to keep the fish fresh with a longer duration. Longer the existence of fish freshness, more acceptable the salt curing method for that kind of fish, as it helps to preserve the fish to be marketed in the off-seasons.

The concentration of salt has a great influence on the rate of surface evaporation (Lee *et al.*, 1994). In addition, depending on the salt concentration and relative humidity, the salted fish may reabsorb moisture from the environment during storage (Nketsia-Tabiri and Sefa-Dedeh, 1995). Shewan (1937) noted that, using too little salt in fish the normal microbes active. A salt solution of approximately 15.5% is required to inhibit most bacteria (Davidson, 1997). The ratio of salt used varies from one production site to another and from one processor to another.

In this experiment 30% salt was used so that microbes do not spoil the muscle of fish and for a better shelf life.

4.2.1. Changes in salt penetration rate and its effect on moisture content during ripening period in experimental fishes

Moisture content and salt percentage play an important role in the shelf life and keeping quality of salt-cured fish products (El-Sharnouby, 1989). During salting, the mass transfer occurs basically between salt and water: the fish muscle takes up salt and loses water (Chaijan, 2011; Oliveira *et al.*, 2012).

Many workers agree that maximum salt uptake takes place within 6-7 days of salting without further uptake during subsequent storage (Narayanaswamy *et al.*, 1980; Leitao *et al.*, 1983; Simmonds, 1984). Similar results also obtained in the present study where the fish contained maximum salt content in 7 days of salting.

During salting process, the changes in chemical and physico-chemical characteristics takes place and in certain stage, the original characteristics of the raw fish is found virtually absent. This stage is regarded as **'salt ripening of fish'.** According to Voskresensky (1965) these changes are induced by enzyme which breakdown both proteins and fats. A series of complex biochemical processes including proteolysis, lipolysis and lipid oxidation takes place during ripening stage. The ripening stage renders a salted product with a firm consistency having characteristics pleasant aroma and taste. The physical and chemical changes that occur during ripening, determine the overall sensory qualities of salted fish-products (Sikorski *et al.*, 1995).

Itou and Akahane (2000) stated that, the decrease in moisture is due to osmotic migration of salt into and water out of the fish. Borgstrom (1965) reported that salt

ripening is the autolytic phenomena caused by the enzyme of the muscle or gastrointestinal tract. During ripening changes induced by enzyme led to breakdown of proteins in tissue structures of muscle and the body organs of the fish. As a result some of the nitrogenous substances chiefly of low molecular weight diffuse from the fish into the salt brine.

According to the method of Ikeme, (1993) saturated brine was made by dissolving 36g sodium chloride in 100ml water. In brine-salting, the higher concentration of brine solution in which the fish was immersed results an osmotic change in concentration affecting a loss of moisture from the fish tissue. In present research work, 30% salt was used in five types of salting method. The properties of fish muscle vary due to changes in water and salt content: the muscle gains salt, whereas water is lost or gained depending on the salting procedure (Thorarinsdottir *et al.,* 2002, 2004).

It is generally accepted that salt migration by diffusion plays an important role in the salting of fish. Salt uptake depends on many factors including species, muscle type, fish size, fillet thickness, weight, composition (lipid content and distribution), physiological state, fish-quality, salting method, salt-concentration, duration of salting step, fish-to-salt ratio, ambient temperature, freezing and thawing and the concentration gradients within the muscle (Hilderbrand, 1992; Wang *et al.*, 2000; Jittinandana *et al.*, 2002; Barat *et al.*, 2003; Erikson *et al.*, 2004).

The rates of water diffusion are positively correlated with increasing of salt concentration (Bellagha *et al.*, 2007; Boudhrioua *et al.,* 2009). The rate of salt uptake is very important with regard to weight change and quality of the final product. The fast rate of salt uptake may lead to inactivation of enzymes and bacteria. The rate of salt uptake depends on the type of process and product. Similar results were reported by Thorarinsdottir *et al*. (2004) on the brine curing of cod and by Sveinung *et al*. (2005) on brine salting of herring fillets.

The changes in salt content in shol, taki and tengra fish tissues in 5 different types of salting process (DS- dry salting, PC- pickle curing, BS- brine salting, SDS- sun-dried

salting, SDS+T- turmeric treated sun-dried salting) and their relationship with moisture content during salting process are presented in Table 4.2.1 (a) - 4.2.1(c) and Figure 4.2.1(a) - 4.2.1(e).

Table: 4.2.1(a). Changes in salt penetration rate and its effect on moisture content in different treatments (DS- dry salting, PC- pickle curing, BS- brine salting, SDS- sun-dried salting, SDS+T- turmeric treated sun-dried salting) of experimental fish Shol (*Channa striatus*), during 7 days of ripening period

Days of ripening period	DS		PC		BS		SDS		SDS+T	
	Moisture (%)	Salt (%)	Moisture (%)	Salt (%)	Moisture (%)	Salt (%)	Moisture (%)	Salt (%)	Moisture (%)	Salt (%)
0*	77.03	0	77.03	0	77.03	0	77.03	0	77.03	0
1	64.57	4.6	64.52	5.22	70.42	4.9	57.31	6.9	50.88	6.1
2	54.15	8.4	59.46	10.3	67.01	7.78	39.09	9.7	39.28	8.9
3	52.01	11.1	55.44	12.5	65.33	9.46	35.87	10.1	34.98	9.2
4	51.40	13.6	53.25	13.5	63.84	11.24	33.12	11.3	33.27	10.95
5	50.15	14.5	52.00	14.3	62.54	12.9	32.55	12.8	32.52	12.1
6	49.00	15.11	51.28	15.50	61.93	14. 52	30.48	13.2	31.25	13.0
7	48.84	16.00	50.29	16.08	60.17	15. 2	29.77	14.9	30.92	14.3

*= Initial (Fresh fish)

Table: 4.2.1(b). Changes in salt penetration rate and its effect on moisture content in different treatments (DS- dry salting, PC- pickle curing, BS- brine salting, SDS- sun-dried salting, SDS+T- turmeric treated sun-dried salting) of experimental fish Taki (*Channa punctatus*), during 7 days of ripening period

Days of ripening period	DS		PC		BS		SDS		SDS+T	
	Moisture (%)	Salt (%)	Moisture (%)	Salt (%)	Moisture (%)	Salt (%)	Moisture (%)	Salt (%)	Moisture (%)	Salt (%)
0*	78.65	0	78.65	0	78.65	0	78.65	0	78.65	0
1	68.48	6.1	67.62	6.0	73.68	4.4	56.08	6.8	59.63	5.7
2	56.39	10.3	62.16	10.9	70.03	7.3	43.84	10.22	45.27	10.04
3	52.38	12.52	59.00	12.91	67.98	9.31	38.07	11.30	42.76	10.98
4	50.00	13.16	56.77	14.08	65.45	11.96	31.42	12.57	36.93	11.44
5	48.43	14.47	54.24	15.12	63.98	13.11	26.57	13.25	28.94	12.30
6	47.90	15.35	53.84	15.95	62.82	14.65	19.05	14.16	23.55	13.08
7	46.21	16.06	52.71	16.22	62.28	15.50	9.77	14.72	12.72	13.77

*= Initial (Fresh fish)

Table: 4.2.1(c). Changes in salt penetration rate and its effect on moisture content in different treatments (DS- dry salting, PC- pickle curing, BS- brine salting, SDS- sun-dried salting, SDS+T- turmeric treated sun-dried salting) of experimental fish Tengra (*Mystus tengra*), during 7 days of ripening period

Days of ripening period	DS		PC		BS		SDS		SDS+T	
	Moisture (%)	Salt (%)	Moisture (%)	Salt (%)	Moisture (%)	Salt (%)	Moisture (%)	Salt (%)	Moisture (%)	Salt (%)
0*	74.27	0	74.27	0	74.27	0	74.27	0	74.27	0
1	60.73	7.2	63.23	7.01	69.95	6.9	55.99	7.5	53.32	8.09
2	54.23	10.9	57.41	10.31	66.12	9.96	37.02	10.56	36.76	12.44
3	49.70	12.5	53.44	12.6	64.01	12.94	34.56	11.08	35.42	12.69
4	45.68	14.08	50.21	14.14	62.20	14.6	8.66	13.92	10.64	13.78
5	42.22	15.33	47.18	15.5	60.05	15.43	8.16	14.19	10.53	13.90
6	41.81	16.60	46.17	16.98	58.44	16.2	5.28	14.87	6.97	14.65
7	41.41	16.80	45.88	17.10	57.35	16.5	4.9	15.2	6.08	14.80

*= Initial (Fresh fish)

The moisture content of DS, PC, BS, SDS and SDS+T shol was observed in 48.84%, 50.29%, 60.17%, 29.77% and 30.92% respectively during ripening period of 7 days [Table.4.2.1 (a)].

On the other hand, DS, PC, BS, SDS and SDS+T taki had moisture content of 46.21%, 52.71%, 62.28%, 9.77% and 12.72% respectively during 7 days of ripening period [Table 4.2.1 (b)].

A similar trend of reducing moisture content of 41.41%, 45.88%, 57.35%, 4.9% and 6.08% was observed during 7 days of ripening period in DS, PC, BS, SDS, SDS+T tengra fish [Table 4.2.1(c)].

The moisture content of salted fish was significantly reduced which indicates a probable longer shelf life and limited chance of deterioration which is in aggrement with Kingley- Ekow (1999) and Omafra (2011). Comparatively higher decrease in moisture content become emphasize during the sun drying in case of SDS and SDS+T. This step which is normally combined with salting and drying, have duel

effects such as the lowering of the water activity (aw) level and a specific inhibitory effect on the growth of some species of microorganisms through the Na+ ion. So the two steps (Salting and Drying) are interrelated to reduce the moisture sufficiently. In this aspect, Nazir and Magar (1983) reported that fish dried after salting with 15 to 30% salt or brining in 9.5 to 15% brine solution for 18 hours showed higher degree of retention of all the materials than plain dried fish.

The decrease in moisture content may be attributed to the loss in the water holding capacity of fish protein as a result of proteolysis (Hernandaz-Herroro, 1999). Zaitsev *et al.* (1969) reported that the loss of water from fish depends mainly on the strength of salt, the temperature and salting time. Present results are in a good agreement with above mentioned research.

In case of shol fish, the highest salt content of 16.08% was observed in PC and the lowest (14.30%) salt content was observed in SDS+T whereas DS, BS, SDS, had salt concentration of 16.00%, 15.2% and 14.9% respectively after 7 days of ripening period [Table 4.2.1(a)].

In case of taki fish, comparatively highest (16.22%) salt content was observed in PC and lowest (13.77%) salt content was observed in SDS+T whereas DS, BS, SDS, had salt concentration of 16.06%, 15.50% and 14.72% respectively after 7 days of ripening period [Table 4.2.1(b)].

Comparatively higher (17.10%) salt content was found in PC tengra than that in BS tengra (16.5%) whereas DS, SDS, SDS+T had salt concentration of 16.80%, 15.2% and 14.8% respectively after 7 days of ripening period in tengra fish [Table 4.2.1(c)].

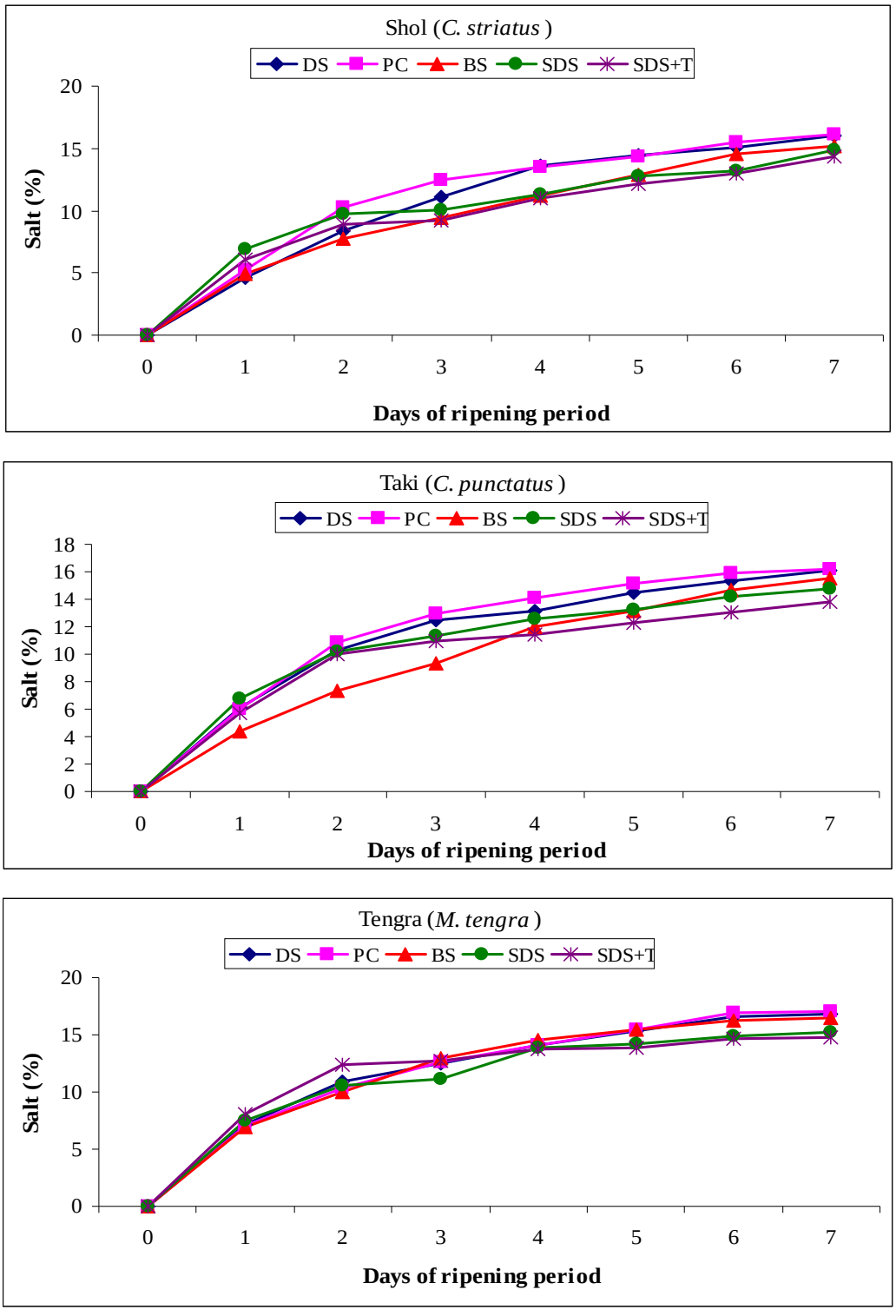


Figure. 4.2.1(a). Changes in salt penetration rate during 7days of ripening period in 5 treatments (DS- dry salting, PC- pickle curing, BS- brine salting, SDS- sun-dried salting, SDS+T- turmeric treated sun-dried salting) of shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish products

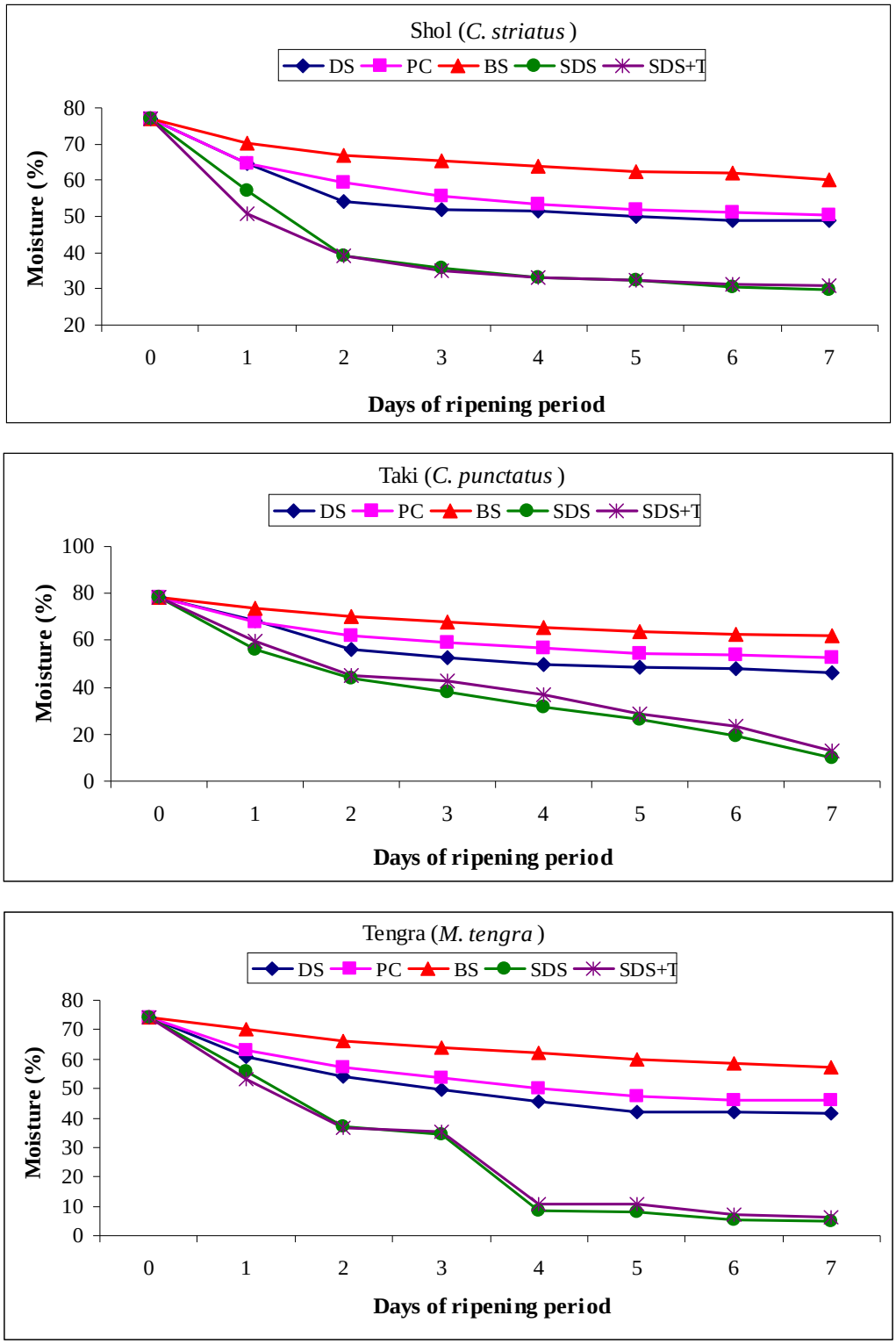


Figure. 4.2.1(b). Changes in moisture (%) content during 7days of ripening period in 5 treatments (DS- dry salting, PC- pickle curing, BS- brine salting, SDS- sun-dried salting, SDS+T- turmeric treated sun-dried salting) of shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish products

In Figure 4.2.1(a) comparatively highest salt content was observed in PC shol, taki and tengra fish and lowest salt content was observed in SDS+T shol, taki and tengra fish. Whereas DS, BS, SDS, had lower salt concentration at the end of ripening period.

On the other hand at the end of ripening period, comparatively highest moisture was found in BS shol, taki and tengra fish and comparatively lowest moisture was found in SDS shol, taki and tengra fish [Figure 4.2.1(b)].

In case of sun- dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) shol, taki and tengra fish products [Figure 4.2.1(b)], significant reduction of moisture content was assumed to be increased due to action of salting and sunlight, because the salt uptake and consequently moisture loss is temperature dependent (Duerr and Dyer, 1952). Similar opinion about the influence of air temperature in drying was described by Van klaveren and Legendre (1965). They opined that, even a small increase of only a few degrees in the intensity of sun-light may appreciably improve the over-all efficiency of the operation. This is certainly a surprising distinctive feature in this study.

It is also shown in Figure 4.2.1(b) that, during ripening period no significant effect on moisture loss in brine salted shol, taki and tengra fish. Similar results were reported by Osibona *et al*. (2010). The differences in water content between five treatments were due to the different salting processes (Andrés *et al.* 2005).

The significant differences noted in the yields of the different salting methods particularly the SDS and SDS+T could be associated with loss in the water content. This suggests that as continuation of salting, the fish muscle loses its water holding capacity, thus drying the muscle. The relatively high increase in salt content could be linked to the equivalent loss in moisture during this period. The salt concentrations, osmotic pressures and the salting medium results in salt-water exchange (salt-uptake by the fish muscle).

An inverse relationship was found between salt penetration rate and the moisture content of fish during 7 days of ripening period. According to Beraquet and Barrera (1983), moisture content was inversely proportional to NaCl content of the fish. As the salt concentration increased, the amount of moisture content decreased almost linearly. This can be explained by the fact that since salt is hygroscopic; increase in salt concentration increases the amount of salt particles for absorbing water molecules from the fish samples (Graivier *et al.* 2006).

It is clear from Figure 4.2.1(a) - 4.2.1(e) that, during the first two days the salt penetration rate into the fish flesh was very rapid. In the first stage of salting the fish was exposed to high osmotic pressure. At that time, the active movement of salt towards inside the fish flesh accompanied by rapid diffusion of water from the flesh to outside of the fish body which resulted a considerable decline in moisture content of the fish. This phenomenon was markedly evident in the first few days of salting. On the second stage, osmotic pressure was believed to still exert influence although on a reduced scale and there was no great difference in the rate of salt moving into the fish or water leaving the fish. This process continued about 7 days of salting. Then, penetration of salt in the salted product practically stopped. Rapid penetration in the initial stage of salting method and slow penetration at the latter stages indicated that an exchange of water was largely responsible for the movement of salt. Cutting and Waterman (1965) stated that, in dry salting, though the fish were completely buried in salt they could not take more salt because water exchange was then practically absent.

The study on the changing pattern of moisture (%) and salt (%) content in 5 treatments (DS=dry salting, PC=pickle curing, BS=brine salting, SDS=sun-dried salting, SDS+T=turmeric treated sun-dried salting) of shol, taki and tengra during 7 days of ripening period are presented in Figure 4.2.1(c)-(e).

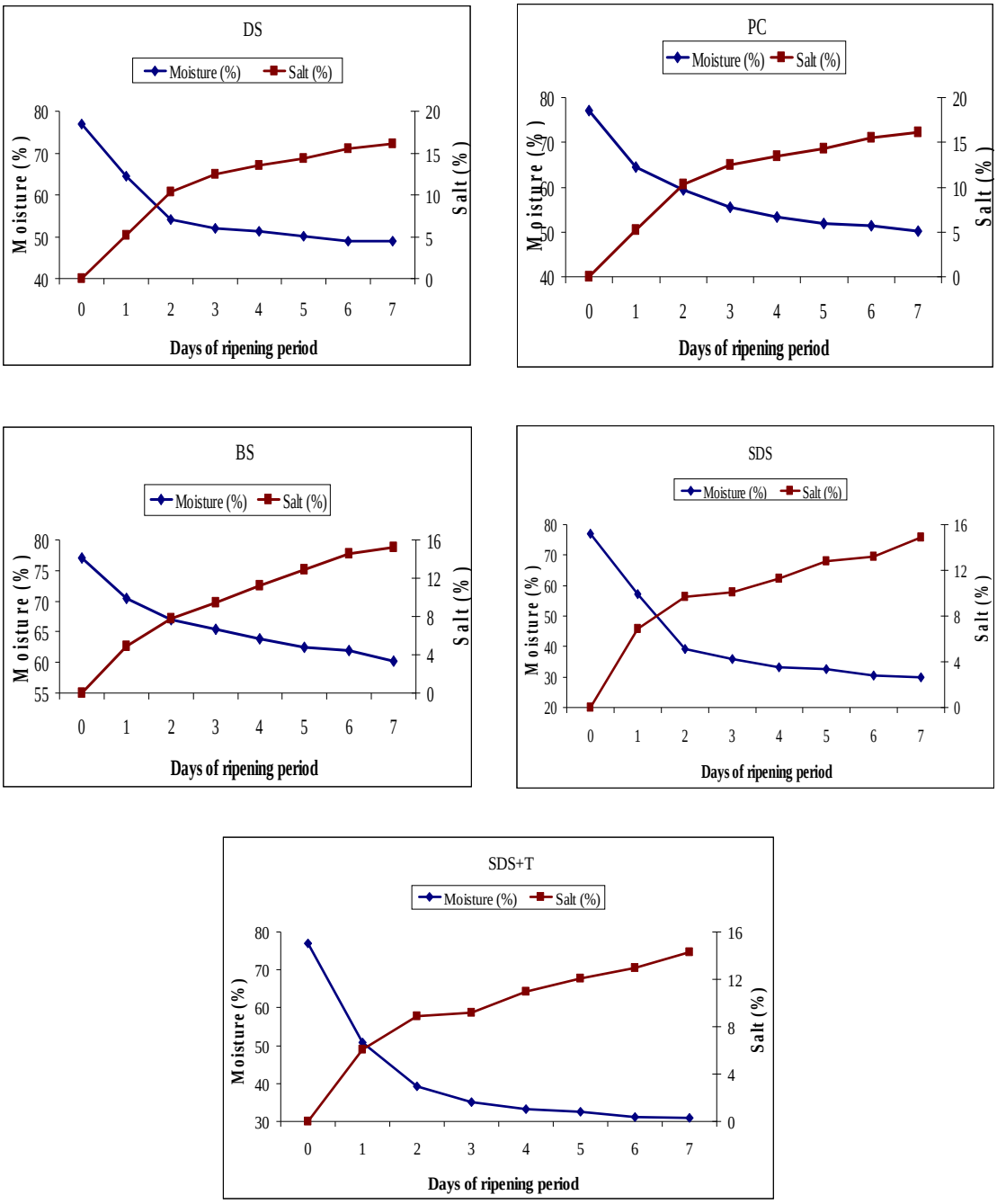


Figure. 4.2.1(c). Relationship of changing pattern of moisture (%) and salt (%) content during 7 days of ripening period in 5 treatments (DS, PC, BS, SDS and SDS+T) of shol (*C. striatus*) fish products [DS=dry salting, PC=pickle curing, BS=brine salting, SDS=sun-dried salting, SDS+T=turmeric treated sun-dried salting]

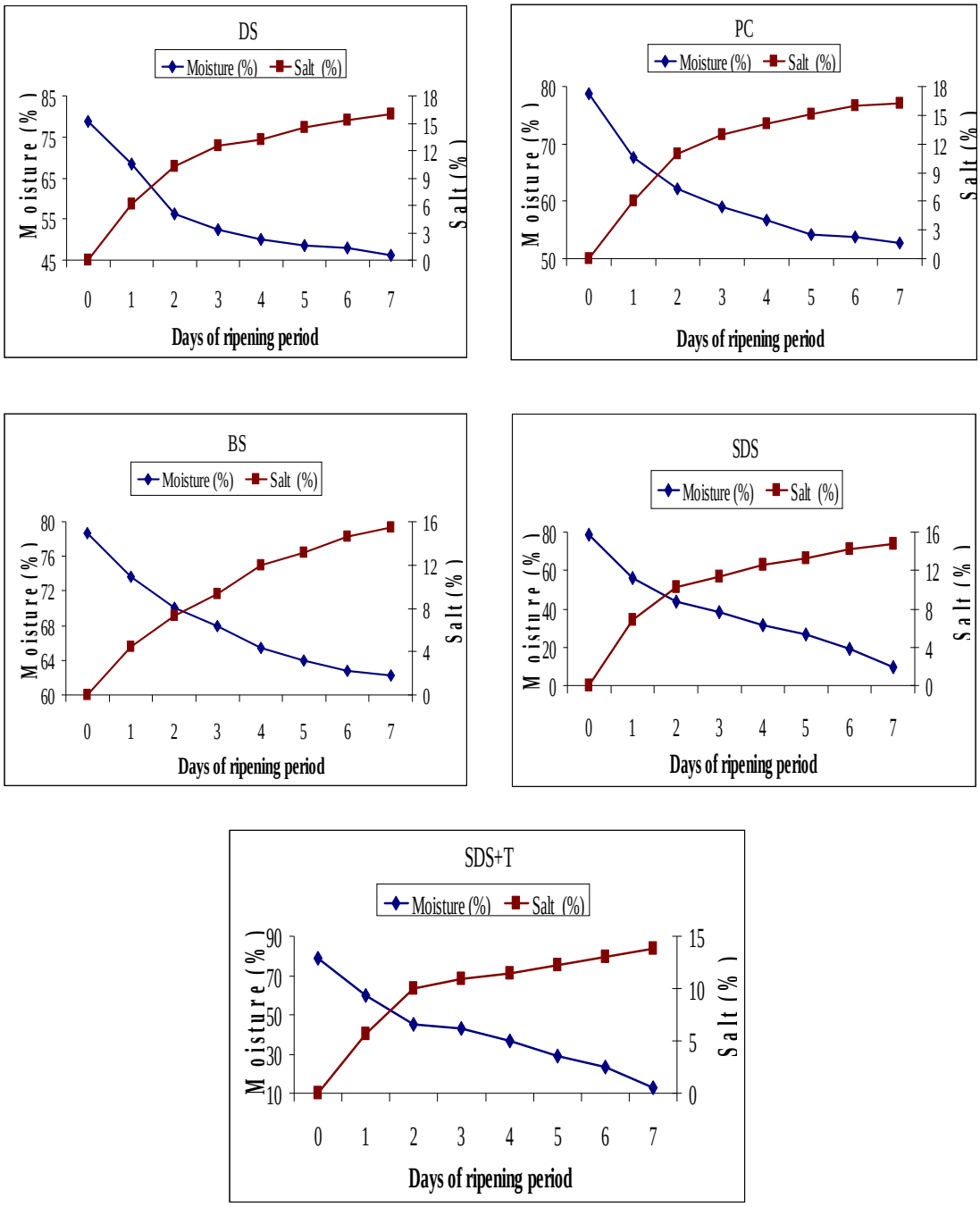


Figure. 4.2.1(d). Relationship of changing pattern of moisture (%) and salt (%) content during 7 days of ripening period in 5 treatments (DS, PC, BS, SDS and SDS+T) of taki (*C. punctatus*) fish products [DS=dry salting, PC=pickle curing, BS=brine salting, SDS=sun-dried salting, SDS+T=turmeric treated sun-dried salting]

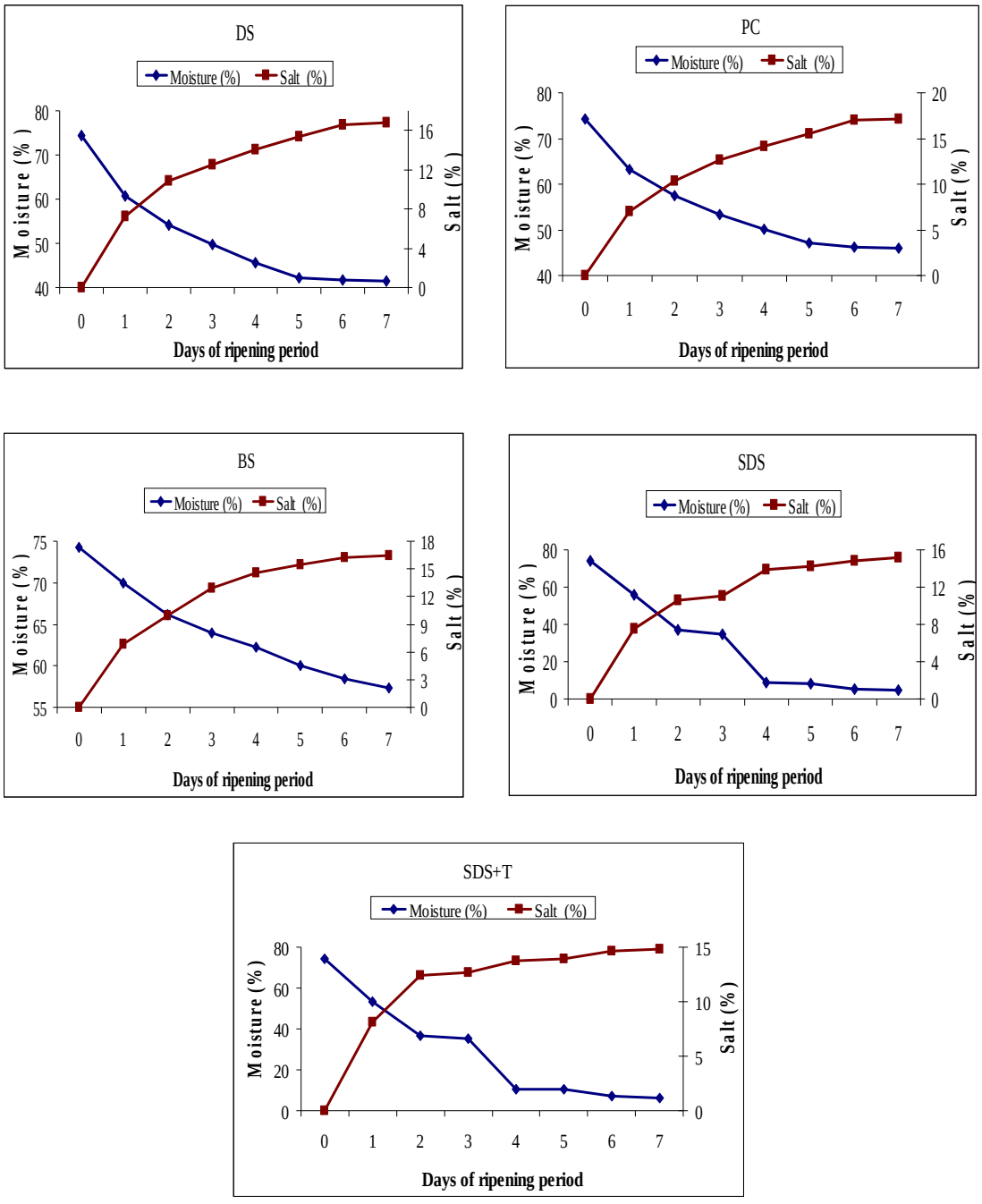


Figure. 4.2.1(e). Relationship of changing pattern of moisture (%) and salt (%) content during 7 days of ripening period in 5 treatments (DS, PC, BS, SDS and SDS+T) of tengra (*M. tengra)* fish products [DS=dry salting, PC=pickle curing, BS=brine salting, SDS=sun-dried salting, SDS+T=turmeric treated sun-dried salting]

4.2.2. Fluid loss pattern in fish muscles during ripening period

From first day of salting considerable amount of fluids started to be diffused from the fish flesh. From second day, the quantity of fluid loss decreased gradually until the loss of fluids ceased in 6-7 days. Ripening period ends when the salt concentration of fish tissue under osmotic diffusion becomes equal to the concentration of salt in the surrounding solution. After the salt concentration in the cellular fluid of various parts of the fish body reaches to 15-20%, the bound water (30-35%) is converted to 'Free State' (Akiba, 1955, 1961; Voskresensky, 1958). Martínez-Alvarez and Gomez-guillén (2006) stated that, the dry salting method produced considerable loss of water due to heavy uptake of salt. Present result is more or less in agreement with those suggests that maximum water loss take place when salt concentration in the fish body approaches to about 15%. This study clearly indicates that salting process is accompanied by fluid losses because fluids in this experiment in all the treatments showed a loss of moisture content with other nutrients. The decrease of moisture or loss of fluid in dry-salted (DS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) products are simply a result of curing due to salt penetration in the fish tissue and in all the 3 cases salting resulting in the loss of moisture due to osmosis diffusion, the moment the fish surface came into contact with salt. Here the condition of the salt whether crystals or in solution was not the matter (Voskresensky, 1965).

Attempts were also made to quantify the amount of fluid that lost during ripening period. However, pickle cured (PC) and brine salted (BS) were not considered in the figure because of its nature ie. liquid media. The fluid loss pattern showed a sharp decrease on first three days followed by a slow decrease of fluid up to 7 days of ripening period in shol and in taki fish whereas tengra showed a sharp fall of fluid on the first two days followed by slower decreased up to ripening becomes completed [Figure 4.2.2].

Figure 4.2.2 shows the comparative feature on Fluid loss pattern during 7days of ripening period in 3 treatments (DS, SDS & SDS+T) in shol, taki and tengra fish products. [Values are presented in Appendix B (Table-1, 2, 3)].

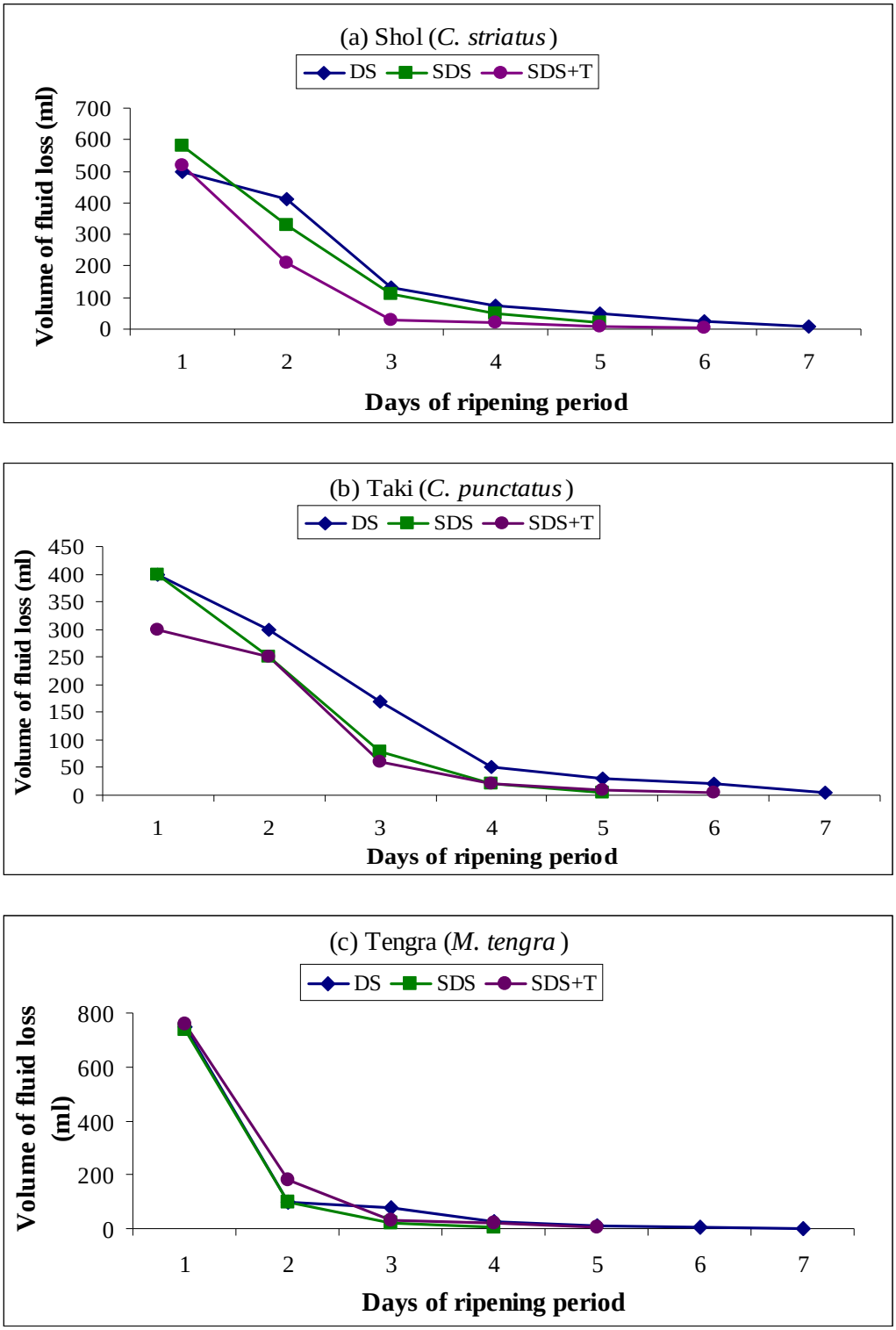


Figure. 4.2.2. Fluid loss pattern during 7days of ripening period in 3 treatments (DS=Dry salting, SDS= Sun-dried salting & SDS+T= Turmeric treated sun-dried salting) of shol (*C. striatus*), taki (*C. punctatus)* and tengra (*M. tengra*) fish products

4.2.3. Percent weight loss of fishes during ripening period

Percent weight loss of 5 different types of salted [DS=dry salted, PC=pickle cured, BS=brine salted, SDS=sun-dried salted, SDS+T=turmeric treated sun-dried salted] Shol, Taki and Tengra fish through 7 days of ripening period are shown in Table 4.2.3(a)-4.2.3(c).

The weight of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) shol fish product, immediately after salting (day-1) was 2798g, 2812g, 2920g, 3012g and 3242g which decreased to 2073g, 2286g, 2520g, 1849g and 1980g respectively on termination of the study at 7 days of ripening period. At the end of ripening period, percent weight loss was found of 35.62%, 26.57%, 20.75%, 52.47% and 50.03% respectively in DS, PC, BS, SDS and SDS+T shol fish products [Table 4.2.3(a)].

On the other hand, the weight of DS, PC, BS, SDS and SDS+T Taki fish product, immediately after salting (day-1) was 2011g, 2115g, 2000g, 2420g and 2500g which decreased to 1495g, 1600g, 1730g, 1019g and 1077g respectively on termination of the study at 7 days of ripening period. At the end of ripening period, percent weight loss was found of 38.98%, 34.43%, 17.62%, 63.21% and 61.41% respectively in DS, PC, BS, SDS and SDS+T taki fish products [Table 4.2.3(b)].

Similarly, the weight of DS, PC, BS, SDS and SDS+T Tengra fish product, immediately after salting (day-1) was 2460g, 2490g, 1975g, 1932g and 2600g which decreased to 1959g, 1980g, 1820g, 1170g and 1450g respectively on termination of the study at 7 days of ripening period [Table 4.2.3(c)].

It is also shown in Table 4.2.3(c) that, at the end of ripening period, percent weight loss of DS, PC, BS, SDS and SDS+T tengra fish products was 25.29%, 24.49%, 9.0%, 61.0% and 57.35% respectively.

Table: 4.2.3(a). Changes in weight and percent weight loss of salted shol (*C. striatus*) fish in treatments DS, PC, BS, SDS and SDS+T during ripening period [DS=dry salting, PC=pickle curing, BS=brine salting, SDS=sun-dried salting, SDS+T=turmeric treated sun-dried salting]

Ripening period	DS		PC		BS		SDS		SDS+T	
	Average wt. of fish after salting	Wt. loss (%)	Average wt. of fish after salting	Wt. loss (%)	Average wt. of fish after salting	Wt. loss (%)	Average wt. of fish after salting	Wt. loss (%)	Average wt. of fish after salting	Wt. loss (%)
0*	3220	0	3113	0	3180	0	3890	0	3962	0
1	2798	13.11	2812	9.67	2920	8.18	3012	22.57	3242	18.17
2	2450	23.91	2601	16.45	2730	14.15	2646	31.98	2828	28.62
3	2281	29.16	2471	20.62	2620	17.61	2337	39.92	2506	36.75
4	2213	31.27	2409	22.61	2580	18.87	2077	46.61	2233	43.64
5	2178	32.36	2369	23.9	2550	19.81	1980	49.1	2119	46.52
6	2137	33.63	2319	25.51	2530	20.44	1909	50.93	2040	48.51
7	2073	35.62	2286	26.57	2520	20.75	1849	52.47	1980	50.03

*= Initial (Fresh fish)

Table: 4.2.3(b). Changes in weight and percent weight loss of salted taki (*C. punctatus*) fish in treatments DS, PC, BS, SDS and SDS+T during ripening period [DS=dry salting, PC=pickle curing, BS=brine salting, SDS=sun-dried salting, SDS+T=turmeric treated sun-dried salting]

Ripening period	DS		PC		BS		SDS		SDS+T	
	Average wt. of fish after salting	Wt. loss (%)	Average wt. of fish after salting	Wt. loss (%)	Average wt. of fish after salting	Wt. loss (%)	Average wt. of fish after salting	Wt. loss (%)	Average wt. of fish after salting	Wt. loss (%)
0*	2450	00	2440	00	2100	00	2770	00	2790	00
1	2011	17.92	2115	13.32	2000	4.76	2420	12.64	2500	10.39
2	1819	25.76	1847	24.30	1870	10.95	1842	33.50	1970	29.39
3	1630	33.47	1720	29.51	1820	13.33	1577	43.07	1671	40.11
4	1615	34.08	1690	30.74	1800	14.29	1466	47.08	1528	45.23
5	1538	37.22	1630	33.20	1775	15.48	1340	51.62	1430	48.75
6	1500	38.78	1620	33.61	1748	16.76	1225	55.78	1200	56.99
7	1495	38.98	1600	34.43	1730	17.62	1019	63.21	1077	61.41

*= Initial (Fresh fish)

Table: 4.2.3(c). Changes in weight and percent weight loss of saled tengra (*M. tengra*) fish in treatments DS, PC, BS, SDS and SDS+T during ripening period [DS=dry salting, PC=pickle curing, BS=brine salting, SDS=sun-dried salting, SDS+T=turmeric treated sun-dried salting]

Ripening period	DS		PC		BS		SDS		SDS+T	
	Average wt. of fish after salting	Wt. loss (%)	Average wt. of fish after salting	Wt. loss (%)	Average wt. of fish after salting	Wt. loss (%)	Average wt. of fish after salting	Wt. loss (%)	Average wt. of fish after salting	Wt. loss (%)
0*	2622	00	2622	00	2000	00	3000	00	3400	00
1	2460	6.18	2490	5.03	1975	1.25	1932	35.6	2600	23.53
2	2175	17.05	2207	15.83	1920	4.00	1770	41.0	2311	32.03
3	2110	19.53	2190	16.48	1900	5.00	1463	51.23	2130	37.35
4	2014	23.19	2130	18.76	1870	6.5	1292	56.93	1805	46.91
5	1980	24.48	2099	19.95	1850	7.5	1223	59.23	1611	52.62
6	1969	24.90	2045	22.01	1830	8.5	1199	60.03	1530	55.00
7	1959	25.29	1980	24.49	1820	9.0	1170	61.0	1450	57.35

*= Initial (Fresh fish)

It is clear here that the weight of salted fish gradually decreased throughout the ripening period and this may occur due to maximum water was coming out from the fish flesh because of osmosis and diffusion. This type of weight loss was observed as reported by several researchers during salting of fish (Voskresensky, 1952, 1953, 1958; Minder, 1952; Semenov and Makarova, 1953). The relationship between changes in weight, salt and water contents is almost linear.

The comparative features of changing pattern of weight loss (%) of shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish during ripening period are shown in Figure 4.2.3.

From Figure 4.2.3 it is clear that, significantly higher amount of weight loss observed in sun-dried salted (SDS) shol, taki and tengra fish whereas, the least percent weight loss was found in brine salted (BS) shol, taki and tengra fish-products.

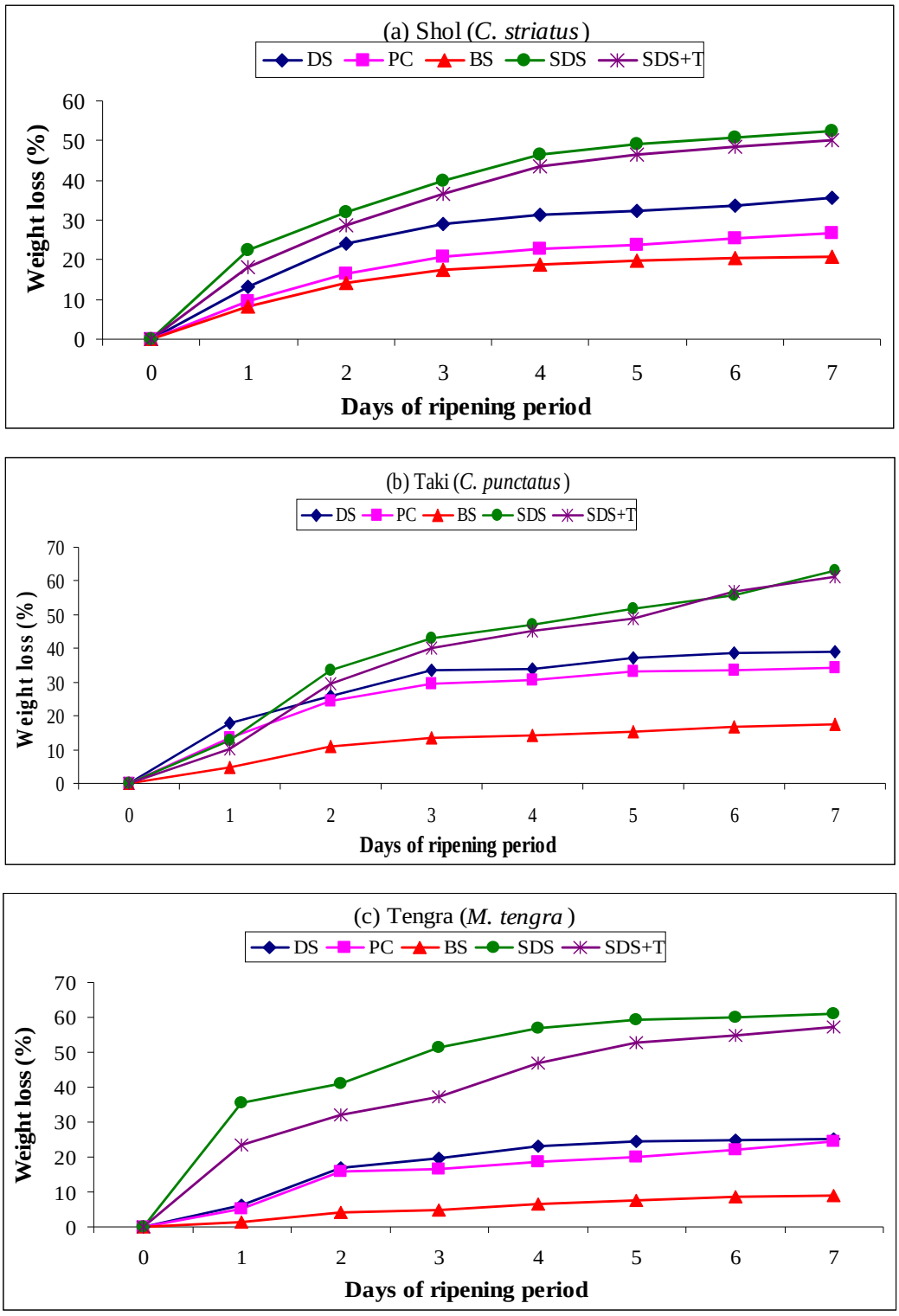


Figure. 4.2.3. Weight loss (%) of DS, PC, BS, SDS and SDS+T shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish during ripening period (DS=Dry salted, PC=Pickle cured, BS=Brine salted, SDS=Sun-dried salted and SDS+T=Turmeric treated sun-dried salted)

Section-4.3: Analysis of freshly processed salted fish-products

4.3.1. Biochemical composition of freshly processed Dry salted (DS), Pickle cured (PC), Brine salted (BS), Sun-dried salted (SDS) and Turmeric treated Sun-dried salted (SDS+T) shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish-products

Bio-chemical composition of freshly processed Dry salted (DS), Pickle cured (PC), Brine salted (BS), Sun-dried salted (SDS) and Turmeric treated Sun-dried salted (SDS+T) shol, taki and tengra fish-products are shown in Table 4.3.1.

The biochemical composition of cured fish-muscle describes the final product quality. In present study, freshly processed (just after ripening) dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) **shol** fish-products had moisture (%) content of 48.84±0.06%, 50.29±0.05%, 60.17±0.06%, 29.77±0.04% and 30.92±0.05%; protein (%) content of 28.21±0.01%, 27.64±0.02%, 20.72±0.02%, 41.48±0.01% and 41.00±0.03%; fat (%) content of 3.99±0.01%, 3.79±0.03%, 3.20±0.03%, 5.10±0.02% and 4.79±0.01%; ash (%) content of 18.98±0.01%, 18.69±0.02%, 16.06±0.01%, 22.80±0.01% and 22.41±0.01%; salt (%) content of 16.00±0.05%, 16.08±0.04%, 15.20±0.05%, 14.90±0.04% and 14.30±0.06%; TVB-N content of 5.25±0.01, 5.28±0.01, 3.64±0.01, 4.89±0.01 and 5.17±0.02 mg/100g of fish; FFA (%) value of 1.5±0.15%, 1.3±0.10%, 1.8±0.15%, 2.3±0.06% and 2.4±0.10%; pH value of 6.3±0.10, 6.4±0.15, 6.5±0.25, 6.2±0.10 and 6.3±0.06 respectively.

In case of freshly processed dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) **taki** fish-products, moisture (%) content were 46.21±0.04%, 52.71±0.06%, 62.28±0.02%, 9.77±0.02% and 12.72±0.04%; protein (%) content were 23.58±0.01%, 21.39±0.02%, 18.02±0.01%, 47.69±0.01% and 46.06±0.01%; fat (%) content were 3.93±0.01%, 3.40±0.01%, 2.76±0.01%, 7.47±0.01% and 7.09±0.02%; ash (%) content were 26.37±0.02%, 22.96±0.01%, 17.24±0.01%, 35.16±0.01% and 34.29±0.02%; salt (%) content were 16.06±0.06%, 16.22±0.04%, 15.50±0.04%, 14.72±0.06% and 13.77±0.05%; TVB-N content were 4.18±0.00, 3.79±0.02, 5.70±0.01, 2.27±0.01 and 3.58±0.01 mg/100g of fish; FFA (%) value were 1.9±0.15%, 2.4±0.21%, 2.6±0.15%, 1.8±0.10% and 2.3±0.06%; pH value were 6.5±0.06, 6.6±0.15, 6.8±0.10, 5.9±0.15 and 5.9±0.06 respectively.

It is also shown in Table 4.3.1 that, freshly processed dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) **tengra** fish-products had moisture (%) content of 41.41±0.04%, 45.88±0.03%, 57.35±0.05%, 4.9±0.03% and 6.08±0.02%; protein (%) content of 22.05±0.01%, 20.43±0.01%, 15.30±0.02%, 43.00±0.01% and 42.60±0.02%; fat (%) content of 10.65±0.03%, 9.40±0.02%, 6.84±0.02%, 15.99±0.01% and 15.58±0.02%; ash (%) content of 26.15±0.04%, 24.62±0.01%, 20.80±0.03%, 36.20±0.02% and 35.60±0.01%; salt (%) content of 16.80±0.03%, 17.10±0.02%, 16.50±0.03%, 15.20±0.04% and 14.80±0.04%; TVB-N value of 3.90±0.02, 4.92±0.03, 2.59±0.04, 1.9±0.07 and 2.52±0.08 mg/100g of fish; FFA (%) value of 2.8±0.21%, 3.2±0.11%, 1.0±0.15%, 1.8±0.15% and 1.6±0.10%; pH value of 6.0±0.11, 6.0±0.06, 5.9±0.10, 6.3±0.15 and 6.4±0.06 respectively.

Moisture determination is one of the most important and most widely used measurements in the processing and testing of foods (Kiin-Kabari *et al.,* 2011). The moisture content plays an important role in the spoilage of fish. Stansby and Olcott (1963) reported that reduction of moisture content retards the spoilage of fish. It was observed that during salting, the moisture content of salted fish varied considerably according to the type of fish (lean or fatty fish), the duration of ripening period, the

quality of salt used and the amount of salt as well. Santhnaraj *et al.* (1973) has recorded a moisture content of 9.2 to 11.6% after sun drying anchovies before storing. Mansur *et al.* (1993) showed that, moisture content of traditionally dried fishes ranged from 10.77 to 18.32%. Moisture content of 25% salted-dried *Clarias sp*. was found to be a range of 4.55-5.95% (Adam Sulieman and Sidahmed, 2012). This result also coincides with present findings of moisture content of SDS and SDS+T tengra. According to Islam (2005a), sun-dried salted shol fish have 32.0% moisture which is similar with present findings of SDS shol. According to Al-Reza *et al.* (2015), sun-dried salted Chela fish contain 5.88% moisture which is similar with present findings of SDS Tengra.

The salting process and salting along with sun-drying, resulted in a significant decrease ($P < 0.05$) in moisture content and a significant increase ($P<0.05$) in protein (except-BS tengra), fat and ash content of 5 types of salted shol, taki and tengra fish samples (Table 4.3.1). This is consistent with the observations by Vasiliadou *et al.* (2005); Mujaffar and Sankat (2006), Sereno *et al*. (2006); Bouriga *et al.* (2008); Kituu *et al*. (2008) and Abraham-Olukayode *et al.* (2012). Similar results for the proximate composition of salted fish were reported in previous studies (Bilgin *et al.*, 2008; Goulas and Kontominas, 2005; Unlusayın *et al.*, 2001). Nutritional components, such as protein, lipid, and ash, were increased due to the loss of water in fish muscle in the salting process (Bras & Costa, 2010; Chaijan, 2011).

Significant increase in protein levels ($p<0.05$) in all salted fish-products (except-BS tengra), when compared with the fresh fish, suggested that protein nitrogen was not lost during salting (Puwastien *et al.*, 1999; Tao and Linchun, 2008; Nahid *et al.*, 2014). The increase in protein contents may be due to product dehydration which concentrated proteins, thus increasing the nutritional value of freshly processed salted and sun-dried salted fish products. Similar results were obtained by Mohamed (2008); Ojutiku *et al.* (2009); Ahmed *et al.* (2011) and Immaculate *et al.* (2012). Chukwu and Shaba (2009) reported that protein nitrogen was not lost during drying, and it increased with the reduced moisture content in cat fish (*Clarias gariepinus)*. Pigott and Tucker (1990) stated that, as the moisture content reduces the concentration of

other nutrient components increases. In this experiment, it was found that among the five types of salting methods, brine salted (BS) shol, taki and tengra has lowest protein content. Due to diffusion and osmotic process in brine salted products, reduced the water soluble and salt soluble proteins from the fish muscles. That is why brine salted products showed a lower decreased value of protein content after ripening period. According to Clucas (1981), brine-salting of fish was usually accompanied by protein losses, as water is drawn out and meal brine is formed, some protein is dissolved into the brine. This is in accordance with present findings. Increase of protein was due to dehydration of water molecule present between the proteins causing aggregation of protein and this result in the increase in protein content of dried fishes (Ninawe and Rathnakumar, 2008).

The findings in the protein and fat contents were also in agreement with related studies by (Thorarinsdottir *et al.* 2001, 2004) where quite related trends were observed with indications that the increase amount of protein and fat was a function of salt concentration and water content. The increase in fat content of all salted products during freshly processed condition '0 days' could be attributed to a reduction in water content. This observation could also be related to similar findings by Olsen and Skara (1997) and Alvarez and Guillen (2005). Dried *Rita rita* contains 13.92% fat (Mollah *et al.*, 1998) and *M. vittatus* contains 17.76% fat (Flowra *et al.*, 2012) which is more or less similar with present findings of fat content of SDS and SDS+T tengra.

The higher value of total ash content in freshly processed 5 types of salted shol, taki and tengra fish than fresh fish was attributed to high salt content which added more ash components to the products. These results were in accordance with Beauchamp and Engelman (1991); Srikar *et al.* (1993); Ahmed (2006); Kiin-Kabari *et al.* (2011); Bakhiet and Khogalie (2012); El-Bassir *et al.* (2015a). It is also shown in table 4.3.1 that, ash content was significantly high in SDS tengra fish than other fish products which indicate an increased inorganic content (Oloyede, 2005). In a similar study, Rahman (1976) recorded 20.1% and 19.4% ash content in traditional market dry salted hilsa and laboratory dry salted hilsa respectively.

Table: 4.3.1. Bio-chemical composition of freshly processed Dry salted (DS), Pickle cured (PC), Brine salted (BS), Sun-dried salted (SDS) and turmeric treated Sun-dried salted (SDS+T) shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish

Fish sample	Salting Treatments	Moisture (%)	Protein (%)	Fat (%)	Ash (%)	Salt (%)	TVB-N (mg/100g)	FFA (%)	pH
Shol (*C. striatus*)	**DS**	48.84±0.06^{aA}	28.21±0.01^{aA}	3.99±0.01^{aA}	18.98±0.01^{aA}	16.00±0.05^{aA}	5.25±0.01^{aA}	1.5±0.15^{aA}	6.3±0.10^{aA}
	PC	50.29±0.05^{bB}	27.64±0.02^{bB}	3.79±0.03^{bB}	18.69±0.02^{bB}	16.08±0.04^{aB}	5.28±0.01^{bB}	1.3±0.10^{bB}	6.4±0.15^{aB}
	BS	60.17±0.06^{cC}	20.72±0.02^{cC}	3.20±0.03^{cC}	16.06±0.01^{cC}	15.20±0.05^{cC}	3.64±0.01^{cC}	1.8±0.15^{cC}	6.5±0.25^{aC}
	SDS	29.77±0.04^{dD}	41.48±0.01^{dD}	5.10±0.02^{dD}	22.80±0.01^{dD}	14.90±0.04^{dD}	4.89±0.01^{dD}	2.3±0.06^{dD}	6.2±0.10^{aD}
	SDS+T	30.92±0.05^{eE}	41.0±0.03^{eE}	4.79±0.01^{eE}	22.41±0.01^{eE}	14.30±0.06^{eE}	5.17±0.02^{eE}	2.4±0.10^{dE}	6.3±0.06^{aE}
Taki (*C. punctatus*)	**DS**	46.21±0.04^{fF}	23.58±0.01^{fF}	3.93±0.01^{fF}	26.37±0.02^{fF}	16.06±0.06^{fA}	4.18±0.00^{fF}	1.9±0.15^{fF}	6.5±0.06^{fA}
	PC	52.71±0.06^{gG}	21.39±0.02^{gG}	3.40±0.01^{gG}	22.96±0.01^{gG}	16.22±0.04^{gG}	3.79±0.02^{gG}	2.4±0.21^{gG}	6.6±0.15^{gfB}
	BS	62.28±0.02^{hH}	18.02±0.01^{hH}	2.76±0.01^{hH}	17.24±0.01^{hH}	15.50±0.04^{hH}	5.70±0.01^{hH}	2.6±0.15^{hgH}	6.8±0.10^{gH}
	SDS	9.77±0.02^{iI}	47.69±0.01^{iI}	7.47±0.01^{iI}	35.16±0.01^{iI}	14.72±0.06^{iI}	2.27±0.01^{iI}	1.8±0.10^{fE}	5.9±0.15^{iI}
	SDS+T	12.72±0.04^{jJ}	46.06±0.01^{jJ}	7.09±0.02^{jJ}	34.29±0.02^{jJ}	13.77±0.05^{jJ}	3.58±0.01^{jJ}	2.3±0.06^{jgE}	5.9±0.06^{iJ}
Tengra (*M. tengra*)	**DS**	41.41±0.04^{kK}	22.05±0.01^{kK}	10.65±0.03^{kK}	26.15±0.04^{kK}	16.80±0.03^{kK}	3.90±0.02^{kK}	2.8±0.21^{kK}	6.0±0.11^{kK}
	PC	45.88±0.03^{lL}	20.43±0.01^{lL}	9.40±0.02^{lL}	24.62±0.01^{lL}	17.10±0.02^{lL}	4.92±0.03^{lL}	3.2±0.11^{lL}	6.0±0.06^{kL}
	BS	57.35±0.05^{mM}	15.30±0.02^{mM}	6.84±0.02^{mM}	20.80±0.03^{mM}	16.50±0.03^{mM}	2.59±0.04^{mM}	1.0±0.15^{mM}	5.9±0.10^{kM}
	SDS	4.9±0.03^{nN}	43.00±0.01^{nN}	15.99±0.01^{nN}	36.20±0.02^{nN}	15.20±0.04^{nN}	1.9±0.07^{nN}	1.8±0.15^{nE}	6.3±0.15^{nD}
	SDS+T	6.08±0.02^{oO}	42.60±0.02^{oO}	15.58±0.02^{oO}	35.60±0.01^{oO}	14.80±0.04^{oO}	2.52±0.08^{mO}	1.6±0.10^{nO}	6.4±0.06^{nO}

Values are shown as mean ± standard deviation of triplicate measurements; $^{a\text{-}o:}$ Means significant differences within fishes. $^{A\text{-}O:}$ Means significant differences within treatments. Means followed by the same letter on the vertical column $^{(Blue\ color)}$ are not significantly different ($p > 0.05$) from each other.

According to Hernandaz-Herrero *et al.* (1999), after salt ripening of Anchovies, salt content increased from 0.32 to 19.3%, ash content increased from 1.6% to 21% and pH decreased from 6.13 to 5.72 which are in harmony with present findings. Monsur (2007) observed that, 30% dry salted, pickle salted and brine salted punti fish had salt content of 24.21%, 24.68% and 17.28% respectively and chapila fish had salt content of 25.11%, 21.43% and 15.94% respectively which is comparatively higher from the present study.

According to Begum *et al.* (2012b), freshly processed sun-dried salted punti fish (*Puntius sophore*) have moisture, protein, fat and ash in the range of 18.1-19.8%, 55.9-58.1%, 11.8-14.1% and 7.1-9.0% respectively in laboratory basis.

According to the research of Imtiaz (2013), freshly processed sun-dried salted ribbon fish (*Trichiurus haumela*) have moisture, protein, fat and ash content of 24.78%, 51.05%, 5.17% and 12.91% and turmeric treated sun-dried ribbon fish (*Trichiurus haumela*) have moisture, protein, fat and ash content of 19.52%, 54.96%, 6.97% and 16.95% respectively. According to the research of Patterson and Ranjitha (2009), freshly processed sun-dried salted *Somberoides tol* was found to be 17% protein, 1.2% fat and .01% FFA.

Present findings in respect of moisture, protein, fat and salt content of sun-dried salted fish-products is similar with the findings of Kumar *et al.* (2013) who observed that, 30% (1:3) salted sun-drying reduced moisture content from 77.46% to 7.14% and increased protein and fat content from 17.94% and 2.42% to 51.32% and 9.48% and salt content is 17.5% in case of *Labeo gonius* fish.

According to the research of El-Bassir *et al.* (2015b) freshly processed 20% salted sun-dried *Bagrus bayad* have moisture, potein fat and ash content was 5.20%, 57.93%, 6.95% and 27.39% respectively. According to the research of Karrar *et al.* (2015) freshly processed 30% salted sun-dried *Oreochromis niloticus* have moisture, potein fat and ash content was 2.74%, 67.42%, 12.47% and 18.64% respectively. On the other hand, El-Bassir *et al.* (2015a) observed that, freshly processed 20% salted

sun-dried *Clarias lazera* have moisture, potein fat and ash content was 23.14%, 66.82%, 11.99% and 17.85% respectively. The above findings are in accordance with the present findings of SDS and SDS+T shol fish.

Reza *et al*. (2008) observed that the Total Volatile Base Nitrogen (TVB-N) content were 5.3 to 19.0 mg/100g for traditionally sun dried Ribbon fish. Sun dried salted and turmeric treated sun-dried ribbon fish had TVB-N value of 18.78 and 17.46 mg/100g (Imtiaz, 2013) which was higher from present result. According to Hernandaz-Herrero *et al.* (1999), TVB-N in salted Anchovies fish muscle appeared to decrease from 24 (fresh) to 19.9 mg/100g during the first 7 days of ripening which is similar with BS shol, SDS taki and DS, BS, SDS, SDS+T tengra. But in case of DS, PC, SDS, SDS+T shol; DS, PC, BS, SDS+T taki and PC tengra fish-products, there is an increase of TVB-N value than fresh one. The increase in TVB-N values may be due to bacterial and enzymatic action, particularly the growth of halophilic bacteria.

From the fresh fishes, there was found an increase in FFA% value in all five types of salted shol, taki and tengra fish and it was in accordance with the findings of Bouriga *et al.* (2008) who found an increase in FFA% value from 1.8% to 3.67% in fresh to sun-dried Atherina fish. The FFA (%) value recorded in this work is low and hence there is no fear of rancidity. This agrees with the work of Frazier and Westhoff (1998). Azad Shah *et al.* (2009) observed that, FFA (%) value increased from 4.55-5.12% within 4 days of drying of herring fillet and then gradually increased up to 10 days of drying (6.86%).

The salting process caused a decrease in pH values in all salted fish samples (Table 4.3.1) which is similar with the findings of Shiau *et al*. (1998) for commercially salted mackerel. According to Lauritzsen *et al.* (1999) and Thorarinsdottir *et al.* (2001), salt-curing induces conformational changes of the proteins and normally a reduction in the muscle pH was observed and lowering of the muscle pH increased the water loss from the muscle during the process. Goulas and Kontominas (2005) observed that pH values of mackerel (*Scomber japonicus*) decreased after salting. The pH value decrease in freshly processed fish products from fresh fish is explained by the

increase of the ionic strength of the solution inside of the cells (Goulas & Kontominas, 2005; Leroi and Joffraud, 2000). Present result is in the normal ranges reported by other researchers (Agab and Shafie, 1989; Eltom, 1989; Ahmed, 2006; Al-Reza et al., 2015). The report of Eyo (1993) indicating that a decrease in pH during ripening period is due to the fact that carbohydrate of the fish was fermented to acids. Pacheco-Aguilar *et al.* (2000) stated that, the pH of fish flesh has an important influence on its freshness because of its influence on bacterial growth; the lower the pH of a fish muscle, the slower the bacterial growth, and vice versa.

From the Table 4.3.1 it is clearly shown that, moisture content of DS shol fish was $48.84\pm.06^{\mathbf{aA}}$**.** Here $^{\mathbf{a}}$-means significant difference ($p<0.05$) present within five treatments (DS, PC, BS, SDS, SDS+T) of shol fish, whereas $^{\mathbf{A}}$-means significant difference ($p<0.05$) within 3 fishes in case of treatment DS (DS shol, DS taki and DS tengra).

It is also shown in this table that, salt content of DS shol fish was $16.00\pm.05^{\mathbf{aA}}$ and DS taki fish was $16.06\pm.06^{\mathbf{fA}}$**.** Here $^{\mathbf{A}}$-means there was no significant difference ($p>0.05$) present between salt content of DS shol and salt content DS taki fish.

In same way, TVB-N content of BS tengra fish was $2.59\pm.04^{\mathbf{mM}}$ and SDS+T tengra fish was $2.52\pm.08^{\mathbf{mO}}$. Here, $^{\mathbf{m}}$-means, there was no significant difference present ($p>0.05$) in TVB-N content of this 2 treatments (BS & SDS+T) in tengra fish.

pH value of PC Taki was $6.6\pm.15^{\mathbf{gfB}}$. Here $^{\mathbf{g}}$-means there was no significant difference ($p>0.05$) present in pH value between PC Taki and BS Taki ($6.8\pm.10^{\mathbf{gH}}$) whereas $^{\mathbf{f}}$-means there was no significant difference ($p>0.05$) present in pH value between PC Taki and DS Taki ($6.5\pm.06^{\mathbf{fA}}$). On the other hand, $^{\mathbf{B}}$-means there was no significant difference ($p>0.05$) present in pH value between PC Taki and PC Shol ($6.4\pm.15^{\mathbf{aB}}$).

Other results from this Table can be interpreted in the same way.

4.3.2. Mineral composition of freshly processed Dry salted (DS), Pickle cured (PC), Brine salted (BS), Sun-dried salted (SDS) and turmeric treated Sun-dried salted (SDS+T) shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish-products

The effect of processing methods on the fish samples recorded variation in the concentrations of minerals.

The mineral composition of freshly processed salted and salted-dried shol, taki and tengra fish products are shown in Table 4.3.2.

Table: 4.3.2. Important mineral (mg/100g of fish) composition of macro [Calcium (Ca) and Magnesium (Mg)] and micro elements [Iron (Fe), Copper (Cu), Zinc (Zn) and Manganese (Mn)] of freshly processed Dry salted (DS), Pickle cured (PC), Brine salted (BS), Sun-dried salted (SDS) and turmeric treated Sun-dried salted (SDS+T) shol (*Channa striatus*), taki (*Channa punctatus*) and tengra (*Mystus tengra*) fish-products

Fish sample	Salting Treatments	Macro elements		Micro / Trace elements			
		Ca mg/100g	Mg mg/100g	Fe mg/100g	Cu mg/100g	Zn mg/100g	Mn mg/100g
Shol (*C. striatus*)	**DS**	438.75	83.69	2.65	0.275	2.125	0.45
	PC	201.5	32.19	1.82	0.225	1.725	0.31
	BS	196.25	14.75	1.5	0.15	1.45	0.225
	SDS	468.75	85.69	3.32	0.375	2.675	0.775
	SDS+T	219.5	43.687	2.8	0.30	2.275	0.525
Taki (*C. punctatus*)	**DS**	600	147.75	3.75	0.425	1.525	0.625
	PC	497.5	65.5	2.95	0.25	1.43	0.60
	BS	123	52.5	2.07	0.2	1.175	0.55
	SDS	700.75	157.5	5.35	0.84	3.50	0.925
	SDS+T	578.25	126.5	4.99	0.62	3.0	0.775
Tengra (*M. tengra*)	**DS**	1250	520.5	4.3	0.435	2.175	0.65
	PC	910	77.375	3.75	0.40	1.625	0.45
	BS	310.75	52.5	2.425	0.3	1.275	0.375
	SDS	1405	795.5	6.95	0.625	3.6	1.02
	SDS+T	976.25	331	5.025	0.475	3.575	0.722

In present study, freshly processed (just after ripening) dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) **shol** fish-products had Ca content of 438.75, 201.5, 196.25, 468.75 and 219.5 mg/100g of fish; Mg content of 83.69, 32.19, 14.75, 85.69 and 43.687 mg/100g of fish; Fe content of 2.65, 1.82, 1.5, 3.32 and 2.8 mg/100g of fish; Cu content of 0.275, 0.225, 0.15, 0.375 and 0.30 mg/100g of fish; Zn content of 2.125, 1.725, 1.45, 2.675 and 2.275 mg/100g of fish; Mn content of 0.45, 0.31, 0.225, 0.775 and 0.525 mg/100g of fish respectively.

In case of freshly processed dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) **taki** fish-products, Ca content were 600, 497.5, 123, 700.75 and 578.25 mg/100g of fish; Mg content of 147.75, 65.5, 52.5, 157.5 and 126.5 mg/100g of fish; Fe content of 3.75, 2.95, 2.07, 5.35 and 4.99 mg/100g of fish; Cu content of 0.425, 0.25, 0.2, 0.84 and 0.62 mg/100g of fish; Zn content of 1.525, 1.43, 1.175, 3.50 and 3.0 mg/100g of fish; Mn content of 0.625, 0.60, 0.55, 0.925 and 0.775 mg/100g of fish respectively.

It is also shown in table 4.3.2 that, freshly processed dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) **tengra** fish products had Ca content of 1250, 910, 310.75, 1405 and 976.25 mg/100g of fish; Mg content of 520.5, 77.375, 52.5, 795.5 and 331 mg/100g of fish; Fe content of 4.3, 3.75, 2.425, 6.95 and 5.025 mg/100g of fish; Cu content of 0.435, 0.40, 0.3, 0.625 and 0.475 mg/100g of fish; Zn content of 2.175, 1.625, 1.275, 3.6 and 3.575 mg/100g of fish; Mn content of 0.65, 0.45, 0.375, 1.02 and 0.722 mg/100g of fish respectively.

From this table it is clear that all five types of salting method raised the mineral composition (except Cu) of shol, taki and tengra fish products. It has been reported that sun-dried *Channa punctatus* contain Ca of 1093 mg/100g, respectively of raw edible parts (Roos *et al*., 2003). The results of the table provided the information that the salted fishes are more nutritive than they are in fresh condition.

According to Rodrigo *et al.* (1997), hake and ling salted-dried roes have Ca content of 16.7 mg/100g and 21.6 mg/100g**;** the Mg ranged from 34.0 mg/100g and 36.8 mg/l00g**;** Fe concentrations 2.13 mg/l00g and 4.46 mg/l00g; Cu concentrations of 0.25 mg/100g and 0.66 mg/l00g; Zn concentration of 4.10 mg/100g and 10.94 mg/l00g and Mn concentration of 0.23 mg/100g and 0.25 mg/l00g of fish respectively. According to Begum *et al.* (2012b), freshly processed salted-dried punti (*Puintius sophore*) fish have calcium (Ca) and Iron (Fe) was in a range of 320-330 mg/100g and 3.1-3.9 mg/100g of fish respectively.The above findings got more or less similarities with present research.

Mormede and Davies, (2001) found that the copper concentrations of fish muscle between 0.10 and 0.83 mg/kg for fish from the North East Atlantic which is similar with present findings. Salting treatments did not influence the copper content of these three fishes. Moreover, the concentrations of these macro and micro minerals were within the FAO/WHO (1983); FAO/WHO (1989) recommended levels of daily intake of minerals [Appendix-C].

Among the macro and micro elements, five types of salted and salted-dried shol, taki and tengra fish-products showed decreasing order of magnitude; Ca> Mg> Fe> Zn> Mn> Cu which is in agreement with the findings of Ako and Salihu (2004). Akiinneye *et al.* (2010) studied micro elements in sun-dried *Sardinella* spp., where Fe was found to be highest in concentration (Fe > Zn > Mn > Cu > Pb). In another report, salted *Clarias gariepinus* showed mineral components in a decreasing order of magnitude Ca> Mg> Fe> Zn (Ekpenyong and Ibok, 2012) which is in aggrement with the present findings. However, Khan *et al.* (1987) and some other researchers did not observe any definite order in the magnitude of the elements. Burgess *et al.* (1965) opained that, nutritional composition of fish muscles varies both from species to species and within species from one season to another. This view is supported by the findings of Window *et al.* (1987) and Khan *et al.* (1987), which showed that variation in the concentrations of elements from one sample of fish to another was due to the chemical forms of the element and their concentrations in the local environment.

4.3.3. Bacteriological study of freshly processed Dry salted (DS), Pickle cured (PC), Brine salted (BS), Sun-dried salted (SDS) and turmeric treated Sun-dried salted (SDS+T) shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish-products

Bacterial growth is the main cause of fish spoilage; therefore it is logical to use bacterial count as an index of fish quality.

4.3.3.1. Standard plate count (SPC) or total bacterial count (TBC)

Standard plate count (SPC) (cfu/g) of freshly processed five types of salted shol, taki and tengra fish-products is given in Table 4.3.3.1.

After salting (end of ripening period) standard plate count (SPC) of freshly processed dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) **shol** fish products were 2.7×10^3, 3.6×10^3, 2.1×10^4, 2.4×10^3 and 2.1×10^3 cfu/g**; taki** fish products were 4.0×10^3, 3.0×10^4, 4.3×10^4, 3.9×10^3 and 2.8×10^3 cfu/g whereas **tengra** fish products were 3.8×10^3, 1.2×10^4, 1.5×10^4, 2.8×10^3 and 2.5×10^3 cfu/g respectively.

Table: 4.3.3.1. Standard plate count (SPC) (cfu/g) of freshly processed salted shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish-products in different treatments (DS = Dry salted, PC = Pickle cured, BS = Brine salted, SDS = Sun-dried salted, SDS+T = Turmeric treated sun-dried salted)

Storage period (Month)	DS	PC	BS	SDS	SDS+T
Shol (*C. striatus*)	2.7×10^3	3.6×10^3	2.1×10^4	2.4×10^3	2.1×10^3
Taki (*C. punctatus*)	4.0×10^3	3.0×10^4	4.3×10^4	3.9×10^3	2.8×10^3
Tengra (*M. tengra*)	3.8×10^3	1.2×10^4	1.5×10^4	2.8×10^3	2.5×10^3

From this table it was observed that in comparison with the fresh fish, there was a decrease in total bacterial count (SPC) which may be due to the presence of high salt concentration, so the growth of pathogenic microorganism controlled. This result is in agreement with the findings of El-Tom (1989); Abu Gideiri (2001); Ahmed (2006); Yanar *et al.* (2006).

The result shows that freshly processed five types of salted shol, taki and tengra fish-products indicated an acceptable microbial load ($<10^5$ cfu/g^{-1}) (ICMSF, 1998).

Kilinc and Cakli (2005) found that, SPC count of raw sardine fillets was 4.5×10^4 cfu g^{-1}, after the sardine fillets were put into barrels, all these microorganisms were inhibited. After marination, the same negative results for microbiological counts were also found in other studies (Aksu *et al*., 1997; Fuselli *et al*., 2003).

According to Abbas Bakhiet and Khogalie (2011), salting process reduces total bacterial count of *Hydrocynus spp.* fish and found that SPC of fresh fish was 58.1×10^3 cfu/g and it reduces in freshly processed 15%, 20% and 25% salted fish-products in 10.0×10^3, 7.8×10^3 and 4×10^3 cfu/g respectively which are in agreement with the present study.

Comparison of total bacterial count (log cfu/g) of freshly processed dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish-products is given in Figure 4.3.3.1.

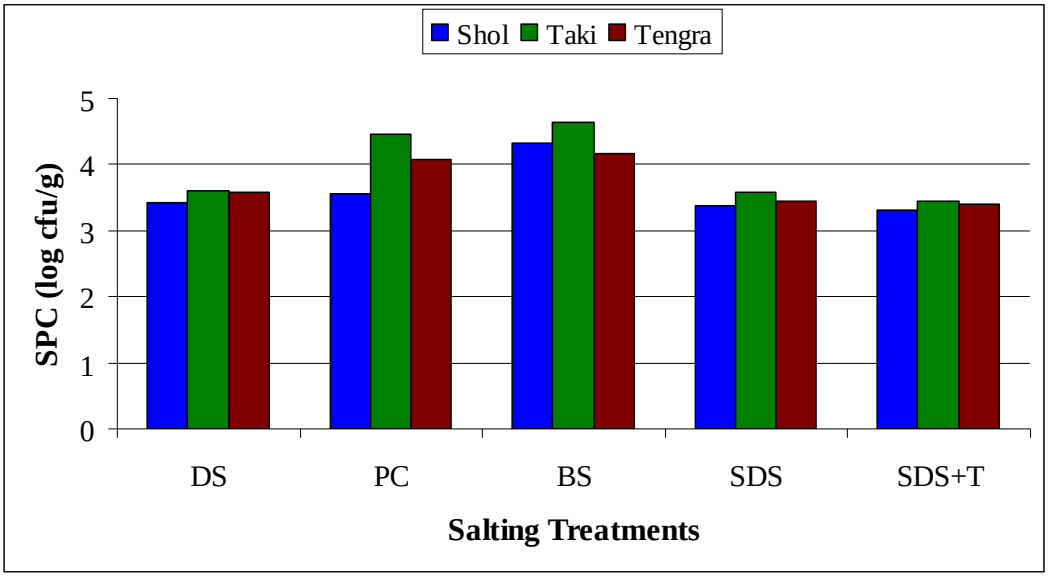


Figure. 4.3.3.1. Comparison of standard plate count (SPC) (log cfu/g) of freshly processed salted shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) in different treatments (DS = Dry salted, PC = Pickle cured, BS = Brine salted, SDS = Sun-dried salted, SDS+T = Turmeric treated sun-dried salted)

From this Figure it is shown that, in comparison of SPC count of 5 types of salted shol, taki and tengra fish products, SPC count was highest in taki fish whereas lowest count was in shol fish products (except BS).

SDS and SDS+T had the lowest SPC count, out of the processing methods studied. The introduction of sun-drying during the salting methods employed would not only kill microorganisms but will also reduce the moisture content of the fish muscles making the environment less favorable for microbial growth whereas BS samples has comparatively highest SPC count. In comparison to all salting treatments, salt and turmeric treated sun-drying reduces bacterial growth more efficiently. The study by Karthikeyan *et al.* (2007) showed that freshwater fish dried to a moisture content of 6.8 to 19.9% had microbiological and sensory factors within acceptable limit.

Sankat and Mujaffar (2004) stated that, adequate salt absorption helps to stop normal bacterial spoilage. It was also observed that, sun-drying along with salting and use of turmeric, might reduced microbial load in fish flesh but did not eliminate completely contaminants in salted fish-samples. The present investigation of salting and sun-dried salting is in agreement with the findings of Salan *et al.* (2006); Kumolu-Johnson and Ndimele (2001) that spoilage of fish can be slowed down by the addition of salt and salt+sun-drying as well as reduction of moisture.

According to Voskresensky (1965) and Filsinger (1987), decrease in total bacterial count was due to the bacteriostatic or bactericidal effects of salt. These findings are in accordance with those of some investigators (Patir *et al*., 2001, 2009; Gurel and Patir, 2004). Whereas, Shewan (1952) reported that the effect of salt was due to the restriction of microbiological activity as a result of the decrease in water content of the tissues and to the direct action of NaCl on the putrefactive micro-organisms. This result agree with the direct relationship between the microbial count and moisture content of the sample (Lilabati *et al.,* 1999) because when moisture content decreased in freshly processed fish, bacterial load also decreased.

The preferable medium for bacteria is neutral and semi acidic (Eltom, 1989). The relative importance of the fish and microbial enzymes in salted fish probably depends on the procedure followed in the preparation of salted fish-products. These results were in agreement with Knøchel and Huss (1984), who studied the microbiology of barrel salted herrings, revealed that both aerobic and anaerobic viable counts (in media containing 15 percent sodium chloride) were low, i.e. not more than 3×10^5 cfu/g of fish.

4.3.3.2. Halophilic bacterial count (HBC)

The halophilic bacterial count (cfu/g) of freshly processed dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish-products are given in Table 4.3.3.2.

At the end of ripening period, halophilic bacterial count (HBC) of freshly processed dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) **shol** fish products were 3.0×10^2, 4.1×10^2, 4.0×10^3, 2.5×10^2 and 2.0×10^2 cfu/g**; taki** fish products were 3.0×10^2, 3.3×10^3, 4.1×10^3, 2.4×10^2 and 2.1×10^2 cfu/g whereas **tengra** fish products were 3.2×10^2, 1.4×10^3, 1.8×10^3, 1.5×10^2 and 1.2×10^2 cfu/g respectively.

Table: 4.3.3.2. Halophilic bacterial count (cfu/g) of freshly processed salted shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish-products in different treatments (DS = Dry salted, PC = Pickle cured, BS = Brine salted, SDS = Sun-dried salted, SDS+T = Turmeric treated sun-dried salted)

Storage period (Month)	DS	PC	BS	SDS	SDS+T
Shol (*C. striatus*)	3.0×10^2	4.1×10^2	4.0×10^3	2.5×10^2	2.0×10^2
Taki (*C. punctatus*)	3.0×10^2	3.3×10^3	4.1×10^3	2.4×10^2	2.1×10^2
Tengra (*M. tengra*)	3.2×10^2	1.4×10^3	1.8×10^3	1.5×10^2	1.2×10^2

The high salt concentration leaves only salt tolerant microorganisms to survive (Horner, 1997). Salted-dried fish have low water activity resulting from rapid dehydration and only halophilic microorganisms are able to develop in them (Rodrigues *et al.*, 2003; Bras and Costa, 2010).

Ames *et al.* (1991) reported that when the water activity is considerably reduced, most microorganisms become inactive but haploidic microorganisms become the major causes of microbial spoilage.

According to the literature (Horner 1992, Kolodziejska *et al.* 2002) most microorganisms normally associated with fish spoilage are halophobic and will not grow in salt concentration exceeding 5%. Salt also forms a membranous surface which could prevent the growth of both spoilage and pathogenic bacteria (Leroi and Joffraud, 2000) and chloride ions in salt are toxic for some microorganisms (Leroi *et al.*, 2000). This kind of bacteria is very important in salted foods (Felix, 2004, 2007).

The presence of extremely halophilic bacteria in foods gives them a reddish coloration; sometimes causing odors and textural changes that promotes rejection by consumers (Hernandez-Herrero, 1999; Doré, 2008).

Halophilic bacteria are natural contaminants in salt used in preserving fish (Felix, 2007). The good shelf stability of salted- fish can be affected to the quality of the salt used. These bacteria can have proteolytic and lipolytic characteristics, their growth can produce unwished changes; therefore, the determination of halophilic bacteria content is an important matter in salted products (Yeannes and Casales, 2008).

Comparison of halophilic bacterial count (log cfu/g) of freshly processed dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish-products is given in Figure 4.3.3.2.

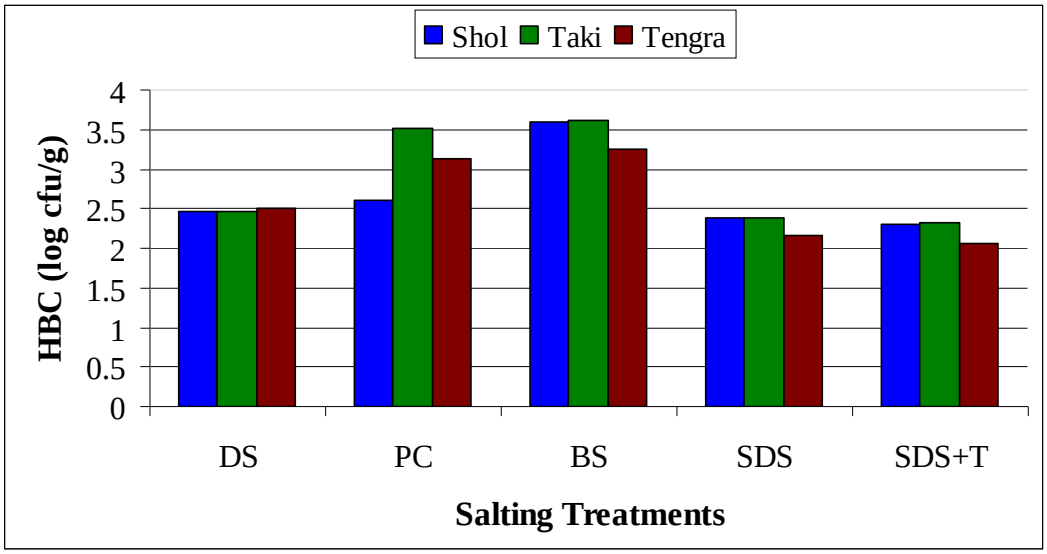


Figure. 4.3.3.2. Comparison of halophilic bacterial count (log cfu/g) of freshly processed salted shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish-products in different treatments (DS = Dry salted, PC = Pickle cured, BS = Brine salted, SDS = Sun-dried salted, SDS+T = Turmeric treated sun-dried salted)

In case of HBC count, the results showed a significant difference between DS, PC, BS, SDS and SDS+T fish samples. In case of dry-salted (DS) shol, taki and tengra, HBC count is more or less similar but in case of PC, BS, SDS and SDS+T, highest HBC count was in taki and lowest was in tengra fish (except PC).

4.3.4. Taste of 'Fish-curry' prepared from freshly processed Dry salted (DS), Pickle cured (PC), Brine salted (BS), Sun-dried salted (SDS) and Turmeric treated Sun-dried salted (SDS+T) shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish-products

In Bangladesh, shol, taki and tengra is mainly consumed as fresh and there was found very limited valid reports on using these fishes for producing salted products. It is therefore, of particular importance, whether these types of products will be acceptable to its consumer or not. Studies on eating quality and consumer preferability of freshly processed Dry salted (DS), Pickle cured (PC), Brine salted (BS), Sun-dried salted (SDS) and turmeric treated Sun-dried salted (SDS+T) shol, taki and tengra fish-products are necessary so that these kinds of new products could find good market.

The taste of the cooked fish were applied for taste-testing panel for different treatments were performed for curry samples prepared from five types of salted shol, taki and tengra fish-products and results are shown in Appendix [Appendix-D].

Five experienced panelists (Teachers, Ph D researcher, Lab-technicians) recorded their scores in the Score sheet. Samples showing a good range of super quality on taste were scored between 7 and 9. Whereas score 5 indicated fish on the borderline of acceptable taste. Scores below 5 having slightly unpleasant taste (if any) were considered.

From the evaluation of the panelists it was found that taste of freshly processed turmeric treated sun-dried salted (SDS+T) shol, taki and tengra fish-curry was 'liked extremely' (sensory score-9) whereas freshly processed dry-salted (DS), pickle-cured (PC), sun-dried salted (SDS) shol, taki and tengra fish-curry were 'liked very much' (sensory score-8) by the panel members. On the other hand freshly processed BS shol and tengra fish-curry were also 'liked very much' (sensory score-8) by the panel

members but freshly processed brine salted (BS) taki fish-curry was 'liked moderately' (sensory score-7).

This is the first report on preferability study of five different types of salted shol, taki and tengra fish-products.

Large group of consumers have become more health conscious and interested in free from food health hazard materials. Different types of salt curing methods may be widely applied when it is largely accepted by consumers.

Section-4.4: Shelf life study through different quality assessment

Shelf-life

The shelf-life of a food is the period for which it remains safe and suitable for consumption. On the other hand shelf-life is the length of time that corresponds to a tolerable loss in quality of a processed food and other perishable items. This means that the food has not deteriorated in quality or spoiled in any way that the consumer would find unacceptable. There should be no formation of toxic products within the food and no loss of significant nutrients below the levels listed on the label. The food must stay safe to consume i.e. should not cause food-poisoning because of the growth of pathogens or the production of toxins in the food during storage (Manat, 2012). The actual length of the shelf life of any given product will depend on a number of factors such as processing method, packaging, and storage conditions (Joseph and Norman, 1996).

Salting is one of the most common pretreatments used for the fish products. Salting converts fresh fish into shelf-stable products by reducing the moisture content, and acting as a preservative. In combination with drying, these processes contribute to the development of characteristic sensory qualities in the products, which influence their utilization as food (Hardy, 1980). According to Andres *et al.* (2005), salted fish-products have a long shelf-life and are considered low risk foods because of their low moisture and low water activity levels.

Limited work is known on quantifiable information on shelf life of salted and sun-dried salted fish-products under various storage conditions. In order to protect the salted and salted-dried fish products from different kinds of spoilage (Enzymatic, bacteriological), it is important to investigate the storage life under different storage conditions (room and refrigeration storage).

Shelf life study of experimental fishes during different duration of storage at room (26-32^{0}C) and refrigeration (4^{0}C) temperature through quality assessment

Performance of a salt curing method is judged by the capacity of the salting process to keep the fish fresh with a longer duration. Longer the existence of fish freshness, more acceptable the salt curing method for that kind of fish, as it helps to preserve the fish to be marketed in the off-seasons.

Quality assessments are necessary to ensure the food safety of any processed product (Azam *et al.*, 2003b). During this study, high quality salted fish products with excellent sensory and physical properties were obtained through these five different types (dry salting, pickle curing, brine salting, sun-dried salting and turmeric treated sun-dried salting) of salting process. Now it is of interest to see how long these salted products could be kept in acceptable under different storage condition.

Spoilage of fish not only occurs at room temperature but also at the refrigeration temperature (Mahin *et al.*, 2011). Microbes are not killed in refrigeration at 5°C. Low temperatures are also used to retard chemical reactions and action of food enzymes and to slow down or stop growth and activity of microorganisms in fish. Low temperature slows the enzymatic action, chemical action and microbial growth (Bakermans and Skidmore, 2011). Ahmed *et al.* (1981a) noted that the lower temperature, the fish remained acceptable for the longest period. Telesara and Kiran (1984) suggested that lower temperature of fish muscle store at long storage period.

According to Clucas (1981), there are several ways of assessing shelf life quality of fish product and these are physical examination, biochemical, microbiological, entomological and sensory methods.

In this study, different types of salted fish products were kept both at room temperature (26-32^{0}C) and refrigeration temperature (4^{0}C) to assess the shelf life through determining the following quality parameters.

1. Sensory characteristics and sensory score evaluation
2. Proximate composition analysis
3. Chemical composition analysis for determining quality changes
4. Bacteriological study

The detail findings of these quality parameters are given under their respective headings-

4.4.1. Sensory characteristics and sensory score evaluation

The sensory evaluation of food products to any processing technology is very important in determining the consumer acceptability (Azam *et al.,* 2003a). Sensory tests have been conducted by human beings to evaluate the goodness and badness of food (Meilgaard *et al.,* 2006) via their senses (i.e. tasting, smelling, touching, etc.). The descriptive analysis involves the detection (discrimination) and the description of both the qualitative and quantitative sensory aspects of a product by trained panels. The sensory characteristics of fish are clearly visible to the consumer and are essential for consumer satisfaction (Reineccius, 1990).

The sensory characteristics and sensory score of Dry salted (DS), Pickle cured (PC), Brine salted (BS), Sun-dried salted (SDS) and turmeric treated Sun-dried salted (SDS+T) shol, taki and tengra fish-products stored at room and refrigeration temperature are presented in Table 4.4.1 (A)-(F).

The entire Table shows that with the lapse of storage time, all products produce a salty taste with different degree of smell, color and texture. Quality assessment of salted and sun-dried salted fish products while stored in sealed polythene bag at ambient or room temperature (26-32^{0}C) and refrigerated temperature (4^{0}C) have been found to have variation in their shelf life in accordance with storage condition and method of salting of fish.

Salted fishery products are characterized by their specific taste, color and flavor and sometimes by texture. Although, 5 types of salted fish-products prepared here were ripened and achieved characteristics ripened salted fish, turmeric treated sun-dried salted shol, taki and tengra fish products possess best organoleptic characteristics compared to remaining 4 types of salted products.

In this experiment, the changes in color appeared to be associated with the solar salts of the market which was not entirely pure. Yellow and brown discoloration of the salted fish indicated the presence of iron in the salt. Usually fish salted with only NaCl may be soft and yellow colored. As a result of impurities in salt particularly iron content in trace amount probably responsible for brownish color of the salted product (Dyer and Gunnarson, 1954; Arnesen, 1954). One group of halophiles (ex-*Halobacterium*), the red or pink bacteria may also cause a reddening of wet and dry salted fish (Clucas, 1984). According to Sefa-Dedeh (1995) and Anihouvi *et al.* (2005), these halophilic bacteria, which can grow at salt concentrations of 13% and above, confer a reddish color on salted and fermented fish. On the other hand, the changes of color in the products from whitish to brownish may be due to lipid oxidation during storage period. This is quite clear from the present study that lipid oxidation presence of oxygen was more prominent in products stored at room temperature than that of products stored in refrigeration temperature. Sen *et al.* (1961)

reported that the main deteriorative changes with salted mackerel samples were reddening, fungal attack, development of rancid and off flavor and browning, when stored at room temperature. Such product also showed higher bacterial count. In the present study there was found no fungal attack in 5 types of salted shol, taki and tengra fish products.

Acceptability test on the basis of flavor, color and texture of the fish-products prepared from 5 different types of salting methods was undertaken by a consumer taste panel. The products were found acceptable by the panel members due to the presence of nice, acceptable aroma and flavor of traditionally salted and salted-dried fishery-products [Table 4.4.1(A)-(F)].

In case of salted Shol products kept at room temperature, at the 135^{th} day, BS (brine salted) shol product showed faded odor with reddish color, soft and slimy and less acceptable product whereas the remaining four types of salted product were still are in good and acceptable condition [Table 4.4.1(A)]. On the other hand, BS shol product stored at refrigeration temperature remained good up to 18 month of storage [Table 4.4.1(B)]. Whereas, in room temperature stored Taki fish products, BS taki fish-product showed dominant fishy odor with faded color, soft and less acceptable at the 75^{th} day whereas another 4 types of salted products are still in good and acceptable condition [Table 4.4.1(C)]. On the other hand, BS taki product stored at refrigeration temperature remains good until 10 month of storage [Table 4.4.1(D)].

In case of salted Tengra products stored at room temperature, at the 120^{th} day, BS tengra product showed faded odor and color with comparatively soft texture and less acceptable product whereas another 4 types of salted products are still in good and acceptable condition [Table 4.4.1(E)]. On the other hand, BS tengra product kept at refrigeration temperature remains good till 24 month of storage [Table 4.4.1(F)].

In case of refrigerated stored fish samples, five types of salted shol and taki fish-samples were evaluated on their sensory quality by panel members at every two months. The overall acceptability showed no significant changes up to 2 months.

However, after 2 months of storage, all the sensory attributes of these fish samples more or less changed and slight signs of quality deterioration were observed during rest of the storage period [Table 4.4.1(B) and Table 4.4.1(D)]. Whereas five types of tengra fish-samples showed no significant changes up to 4 months and duration of shelf life of these fish products was too long. So after 1st four months of observation, the data was presented at following 4 months intervals in five treatments of tengra fish-products stored at refrigeration temperature [Table 4.4.1(F)].

According to the results of the sensory evaluation, changes in the parameters during storage were significant ($P < 0.05$) [Table 4.4.1(A) - (F)]. The remarks on these tables were made on the basis of the scores determined by the observation of panel members as stated in Table 3.3.5.1 (Peryam and Pilgrim, 1957) in materials and methods section. 9-point hedonic scale was used for sensory analysis in this experiment showed its acceptability. The sensory score of the fish products 9.0 indicates that the products were 'excellent' in over all quality whereas 4.5 refer as 'rejected'. Miyauchi *et al.* (1964) suggested that the average sensory score of 5 might be accepted in case of sensory test which is similar with present findings. The highest sensory score '9' observed in the fresh process condition (0 day) of all salted fish products.

Dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun dried salted (SDS+T) **shol** fish products turned to slightly acceptable to the panel members with sensory score of 5 in the end of 165 days, 150 days, 135 days, 165 days and 180 days respectively in room temperature stored products whereas 26 months, 22 months, 18 months, 24 months and 28 months in case of refrigeration storage products.

Similarly, room temperature stored dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun dried salted (SDS+T) **taki** fish products turned into slightly acceptable at the end of 150 days, 135 days, 75 days, 240 days and 300 days and at the end of 26 months, 24 months, 10 months, 26 months and 28 months in case of refrigeration storage products.

In room temperature, the highest shelf life of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun dried salted (SDS+T) **tengra** fish products was 210 days, 180 days, 120 days, 360 days and 420 days respectively whereas 32 months, 28 months, 24 months, 32 months and 36 months in case of refrigeration storage products with sensory score of 9 to 5 in entire storage period.

The reduction in the sensory qualities with increase in the storage period of processed fish could be attributed to higher activities of the spoilage agents. Similar trend was observed in variously processed catfish and Telapia stored at ambient temperature (Olatunde *et al.,* 2013); Hilsa at refrigerator temperature (Alam *et al.,* 2009) and crustaceans (Oyster and Shrimps) under storage (Llobreda *et al.,* 1986).

Based on sensory assessment, the processed fish samples stored at room temperature were decreased greatly in odor and flavor as the duration of storage increased compared with refrigerator temperature stored samples. Most of the samples stored at room temperature did not have the characteristics smell after 75 days of storage. On the other hand, the flavor, color, texture of the samples stored at refrigerator temperature had the characteristics smell of freshly prepared sample and acceptable condition even up to 10 months of storage.

According to Norhana *et al*. (2010) refrigeration and freezing lower the temperature of food to levels at which bacterial metabolic processes are stopped and the rates of chemical and biochemical reactions reduced and therefore, are well known techniques for extending the shelf-life of food products which is in agreement in present study.

According to Abbas *et al.* (2008) sensory assessment has always played a key role in quality and freshness evaluation in fish industry. Yu (1985) applied this hedonic rating scale to evaluate the acceptability of the sun dried fishes by their external morphological and quality changes. This hedonic rating scale was also applied by using 9- points for the sensory evaluation of the dried and dehydrated fish by Morshed *et al.* (2005).

Table: 4.4.1(A). Changes in sensory characteristics and scores of different salted shol (*C. striatus*) during different days of observation at room temperature (Peryam and Pilgrim, 1957) [DS = Dry salted, PC = Pickle cured, BS = Brine salted, SDS = Sun-dried salted, SDS+T = Turmeric treated sun-dried salted]

Days of storage	Product	Flavor (Smell / Odor)	Color	Texture	Remarks	Hedonic scale score (0-9)
0	DS	Attractive salty odor	Natural color	Slightly tough	Excellent	**9**
	PC	Attractive salty odor	Natural color	Elastic	Excellent	**9**
	BS	Attractive salty odor	Natural color	Elastic	Excellent	**9**
	SDS	Attractive salty odor	Natural color	Tough	Excellent	**9**
	SDS+T	Attractive turmeric odor	Deep Yellow	Tough	Excellent	**9**
15	DS	Attractive salty odor dominant	Natural color	Tough	Excellent	**8.8**
	PC	Attractive salty odor dominant	Natural color	Elastic	Excellent	**8.7**
	BS	Attractive salty odor dominant	Natural color	Elastic	Excellent	**8.6**
	SDS	Attractive salty odor dominant	Natural color	Tough	Excellent	**8.9**
	SDS+T	Attractive turmeric odor dominant	Deep Yellow	Tough	Excellent	**9**
30	DS	Attractive salty odor dominant	Slightly whitish	Tough & shrunken	Excellent	**8.7**
	PC	Attractive salty odor dominant	Slightly whitish	Semi-elastic	Excellent	**8.6**
	BS	Attractive salty odor dominant	Slightly whitish	Elastic	Excellent	**8.4**
	SDS	Attractive salty and slightly fishy odor	Whitish	Comparatively tougher& shrunken	Excellent	**8.8**
	SDS+T	Attractive turmeric odor dominant	Deep Yellow	Comparatively tougher& shrunken	Excellent	**8.9**
45	DS	Attractive salty odor dominant	Fade	Comparatively tougher& shrunken	Excellent	**8.5**
	PC	Attractive salty odor	Fade	Semi-elastic	Excellent	**8.4**
	BS	Characteristics salty odor	Grayish	Semi-elastic	Very much good	**8.2**
	SDS	Attractive salty and fishy odor dominant	Whitish	Comparatively tougher& shrunken	Excellent	**8.6**
	SDS+T	Attractive turmeric & slightly fishy odor dominant	Deep Yellow	Comparatively tougher &shrunken	Excellent	**8.8**

60	DS	Attractive salty odor	Fade	Comparatively tougher& shrunken	Excellent	**8.4**
	PC	Characteristics salty odor	Fade	Slightly elastic	Very much good	**8.2**
	BS	Salty & stimulating fishy odor	Brownish	Slightly elastic	Moderately Good	**8.0**
	SDS	Attractive salty and fishy odor dominant	Whitish	Comparatively tougher& shrunken	Excellent	**8.5**
	SDS+T	Attractive turmeric & fishy odor dominant	Deep Yellow	Comparatively tougher &shrunken	Excellent	**8.6**
75	DS	Characteristics salty odor	Fade brown.	Nearly firm	Very much good	**8.2**
	PC	Salty & stimulating fishy odor	Slightly brown	Nearly firm	Moderately Good	**8.0**
	BS	Stimulating fishy odor dominant	Fade brown	Slightly soft	Slightly Good	**7.5**
	SDS	Salty & stimulating fishy odor	Fade brown	Semi-Elastic	Excellent	**8.4**
	SDS+T	Attractive turmeric & fishy odor dominant	Yellowish	Semi-Elastic	Excellent	**8.5**
90	DS	Salty & stimulating fishy odor	Fade brown.	Firm	Moderately Good	**8.0**
	PC	Stimulating fishy odor dominant	Slightly brown.	Firm	Slightly Good	**7.5**
	BS	Characteristic fishy odor	Brownish red	Slightly soft	moderately accepted	**7.0**
	SDS	Slightly salty & characteristic fishy odor	Fade brown	Semi-Elastic	Very much good	**8.2**
	SDS+T	Attractive turmeric & stimulating fishy odor dominant	Yellowish	Semi-Elastic	Excellent	**8.4**
105	DS	Stimulating fishy odor dominant	Brownish	Firm	Slightly Good	**7.5**
	PC	Characteristic fishy odor	Slightly reddish	Slightly soft	Moderately accepted	**7.0**
	BS	Very low stimulating fishy odor	Brownish red	Comparatively soft & little slimy	Slightly accepted	**6.5**
	SDS	Stimulating fishy odor dominant	Slightly yellowish brown	Elastic	Moderately Good	**8.0**
	SDS+T	Slightly fishy and dominant turmeric odor	Yellowish	Elastic	Very much good	**8.2**

120	DS	Distinctive fishy & slightly salty odor	Brownish	Slightly soft	Moderately accepted	**7.0**
	PC	Very low stimulating fishy odor	Slightly reddish	Slightly soft	Slightly accepted	**6.5**
	BS	Slightly fishy odor	Reddish	Soft & slimy	Just accepted	**6**
	SDS	Slightly salty odor present	Yellowish brown	Elastic	Slightly Good	**7.5**
	SDS+T	Slightly turmeric & characteristic fishy odor	Yellowish	Elastic	Moderately Good	**8.0**
135	DS	Very low stimulating fishy odor	Brownish	Slightly soft	Slightly accepted	**6.5**
	PC	Slightly fishy odor present	Reddish brown	Soft	Just accepted	**6**
	BS	Faded odor	Reddish	Soft & slimy	Neither like or dislike	**5**
	SDS	Distinctive fishy odor But salty odor absent	Yellowish brown	Firm	Moderately accepted	**6.8**
	SDS+T	Stimulating fishy with turmeric odor	Yellowish	Firm	Slightly Good	**7.5**
150	DS	Slightly fishy odor present	Deep brown	Slightly soft	Just accepted	**6**
	PC	Faded fishy odor	Reddish brown	Comparatively Soft	Neither like or dislike	**5**
	BS	Putrid odor	Reddish	Soft & slimy	Rejected	**4.5**
	SDS	Very low stimulating fishy odor	Yellowish brown	Firm	Slightly accepted	**6.2**
	SDS+T	Stimulating fishy with little turmeric odor	Fade yellow	Firm	Moderately accepted	**7.0**
165	DS	Faded odor	Deep brown	Comparatively soft	Neither like or dislike	**5**
	PC	Slightly rancid odor	Reddish brown	soft	Rejected	**4.5**
	BS	*	*	*	*	*
	SDS	Slightly fishy odor present	Yellowish brown	Slightly soft	Neither like or dislike	**5**
	SDS+T	Stimulating fishy odor	Fade yellow	Slightly soft	Slightly accepted	**6.5**
180	DS	Slightly rancid odor	Deep brown	soft	Rejected	**4.5**
	PC	*	*	*	*	*
	BS	*	*	*	*	*
	SDS	Faded odor	Faded radish	Comparatively soft.	Rejected	**4.5**
	SDS+T	Very low stimulating fishy odor	Faded yellow	Slightly soft	Neither like or dislike	**5**
195	DS	*	*	*	*	*
	PC	*	*	*	*	*
	BS	*	*	*	*	*
	SDS	*	*	*	*	*
	SDS+T	Faded odor	Faded yellow	Comparatively soft.	Rejected	**4.5**

***= Rejected as per sensory evaluation, acceptability score, < 4.5 is considered as rejected or spoiled.**

Table: 4.4.1(B). Changes in sensory characteristics and scores of different salted shol (*C. striatus*) during observation at refrigeration (4^0C) storage (Peryam and Pilgrim, 1957) [DS = Dry salted, PC = Pickle cured, BS = Brine salted, SDS = Sun-dried salted, SDS+T = Turmeric treated sun-dried salted]

Months of Storage	Product	Flavor(Smell/ Odor)	Color	Texture	Remarks	Hedonic scale score (0-9)
0	DS	Attractive salty odor	Natural color	Slightly tough	Excellent	**9**
	PC	Attractive salty odor	Natural color	Elastic	Excellent	**9**
	BS	Attractive salty odor	Natural color	Elastic	Excellent	**9**
	SDS	Attractive salty odor	Natural color	Tough	Excellent	**9**
	SDS+T	Attractive turmeric odor	Deep yellow	Tough	Excellent	**9**
2	DS	Attractive salty odor dominant	Natural color	Slightly tough	Excellent	**8.9**
	PC	Attractive salty odor dominant	Natural color	Elastic	Excellent	**8.8**
	BS	Attractive salty odor	Natural color	Elastic	Excellent	**8.6**
	SDS	Attractive salty odor	Natural color	Tough	Excellent	**8.9**
	SDS+T	Attractive turmeric odor	Deep yellow	Tough	Excellent	**9**
4	DS	Attractive salty odor dominant	Natural color	Slightly tough	Excellent	**8.8**
	PC	Attractive salty odor dominant	Natural color	Elastic	Excellent	**8.7**
	BS	Characteristics salty odor	Natural color	Elastic	Excellent	**8.5**
	SDS	Attractive salty odor dominant	Natural color	Tough	Excellent	**8.8**
	SDS+T	Attractive turmeric odor	Deep yellow	Tough	Excellent	**8.9**
6	DS	Attractive salty odor	Slightly whitish	Slightly tough	Excellent	**8.8**
	PC	Characteristic salty odor	Slightly whitish	Elastic	Excellent	**8.6**
	BS	Salty & stimulating fishy odor dominant	Brownish	Elastic	Moderately good	**8**
	SDS	Attractive salty and fishy odor dominant	Whitish	Semi-elastic	Excellent	**8.7**
	SDS+T	Attractive turmeric & slightly fishy odor dominant	Deep yellow	Tough	Excellent	**8.9**
8	DS	Characteristic salty & slightly fishy odor	Fade brown	Nearly firm	Excellent	**8.7**
	PC	Characteristic salty odor	Whitish	Elastic	Excellent	**8.5**
	BS	Salty & stimulating fishy odor	Brownish	Slightly elastic	Slightly good	**7.5**
	SDS	Stimulating salty and fishy odor dominant	Fade brown	Semi-elastic	Excellent	**8.6**
	SDS+T	Attractive turmeric & slightly fishy odor	Deep yellow	Tough	Excellent	**8.8**

10	DS	Stimulating fishy odor	Fade brown	Firm	Excellent	**8.6**
	PC	Flavorful fishy & salty odor dominant	Slightly brown	Firm	Moderately good	**8**
	BS	Very low stimulating fishy odor	Fade brown	Slightly elastic	Moderately accepted	**7**
	SDS	Flavorful fishy odor dominant	Fade brown	Semi-elastic	Excellent	**8.5**
	SDS+T	Attractive turmeric & slightly fishy odor	Deep yellow	Tough	Excellent	**8.7**
12	DS	Flavorful fishy odor	Comparatively brownish	Firm	Excellent	**8.5**
	PC	Flavorful fishy & salty odor dominant	Slightly brown	Nearly firm	Slightly good	**7.5**
	BS	Slightly undesirable fishy odor	Fade brown	Slightly soft	Slightly accepted	**6.5**
	SDS	Slightly salty & characteristic fishy odor	Slightly yellowish brown	Elastic	Moderately Good	**8**
	SDS+T	Turmeric & fishy odor dominant	Yellowish	Semi-elastic	Excellent	**8.6**
14	DS	Distinctive fishy & slightly salty odor	Comparatively brownish	Firm	Moderately good	**8**
	PC	Stimulating fishy & slightly salty odor	Slightly brown	Firm	Moderately accepted	**7**
	BS	Fishy odor dominant	Brownish red	Slightly soft	Just accepted	**6**
	SDS	Slightly salty & characteristic fishy odor	Slightly yellowish brown	Elastic	Slightly good	**7.5**
	SDS+T	Distinctive turmeric & slightly fishy odor	Yellowish	Semi-elastic	Excellent	**8.5**
16	DS	Slightly salty & distinctive fishy odor	Brownish	Slightly soft	Slightly good	**7.5**
	PC	Stimulating fishy odor dominant	Slightly reddish	Slightly soft	Slightly accepted	**6.5**
	BS	Fishy odor absent	Brownish red	soft & little slimy	Not so good	**5.5**
	SDS	Slightly salty &dominant fishy odor	Yellowish brown	Firm	Moderately accepted	**7**
	SDS+T	Slightly fishy & characteristic turmeric odor	Yellowish	Elastic	Moderately good	**8**
18	DS	Slightly salty & fishy odor	Brownish	Slightly soft	Moderately accepted	**7**
	PC	Salty odor absent & slightly fishy odor	Slightly reddish	Slightly soft	Just accepted	**6**
	BS	Slightly rancid odor	Reddish	Soft & slimy	Neither like or dislike	**5**
	SDS	Slightly salty &dominant fishy odor	Yellowish brown	Firm	Slightly accepted	**6.5**
	SDS+T	Stimulating fishy & turmeric odor	Yellowish	Elastic	Slightly good	**7.5**

20	DS	Salty odor absent	Brownish	Slightly soft	Slightly accepted	**6.5**
	PC	Slightly fishy odor	Reddish brown	Soft	Not so good	**5.5**
	BS	Slightly rancid odor	Reddish	Soft & slimy	Rejected	**4.5**
	SDS	Salty odor absent	Yellowish brown	Slightly soft	Just accepted	**6**
	SDS+T	Stimulating fishy & slightly turmeric odor	Fade yellow	Firm	Moderately accepted	**7**
22	DS	Slightly fishy odor	Brownish	Slightly soft	Just accepted	**6**
	PC	Faded odor	Reddish brown	Soft	Neither like or dislike	**5**
	BS	*	*	*	*	*
	SDS	Slightly fishy odor	Yellowish brown	Slightly soft	Not so good	**5.5**
	SDS+T	Flavorful fishy odor	Fade yellow	Firm	Slightly accepted	**6.5**
24	DS	Slightly faded fishy odor	Deep brown	Slightly soft	Not so good	**5.5**
	PC	Slimy and semi decomposed odor	Reddish brown	Soft & slimy	Rejected	**4.5**
	BS	*	*	*	*	*
	SDS	Slightly rancid odour	Faded radish	Comparatively soft.	Neither like or dislike	**5**
	SDS+T	Slightly turmeric odor still present	Fade yellow	Slightly soft	Just accepted	**6**
26	DS	Faded odor	Deep brown	Comparatively soft	Neither like or dislike	**5**
	PC	*	*	*	*	*
	BS	*	*	*	*	*
	SDS	Slightly undesirable fishy odor	Faded radish	Comparatively soft	Rejected	**4.5**
	SDS+T	Slightly fishy odor	Faded yellow	Slightly soft	Not so good	**5.5**
28	DS	Slightly rancid odor	Deep brown	soft	Rejected	**4.5**
	PC	*	*	*	*	*
	BS	*	*	*	*	*
	SDS	*	*	*	*	*
	SDS+T	Faded odor	Faded yellow	Comparatively soft.	Neither like or dislike	**5**
30	DS	*	*	*	*	*
	PC	*	*	*	*	*
	BS	*	*	*	*	*
	SDS	*	*	*	*	*
	SDS+T	Slightly unpleasant odor	Faded yellow	Comparatively soft.	Rejected	**4.5**

***= Rejected as per sensory evaluation, acceptability score, < 4.5 is considered as rejected or spoiled.**

Table: 4.4.1(C). Changes in sensory characteristics and scores of different salted Taki (*C. punctatus*) during different days of observation at room temperature (Peryam and Pilgrim, 1957) [DS = Dry salted, PC = Pickle cured, BS = Brine salted, SDS = Sun-dried salted, SDS+T = Turmeric treated sun-dried salted]

Days of storage	Product	Flavor(Smell/ Odor)	Color	Texture	Remarks	Hedonic scale score (0-9)
0	DS	Attractive salty odor	Fade blackish	Tough & shrunken	Excellent	**9**
	PC	Attractive salty odor	Fade blackish	Slightly tough & shrunken	Excellent	**9**
	BS	Attractive salty odor	Fade blackish	Elastic	Excellent	**9**
	SDS	Attractive salty odor	Whitish	Comparatively Tougher & shrunken	Excellent	**9**
	SDS+T	Attractive turmeric odor	Deep yellow	Comparatively Tougher & shrunken	Excellent	**9**
15	DS	Attractive salty odor dominant	Fade blackish	Tough & shrunken	Excellent	**8.9**
	PC	Attractive salty odor dominant	Fade blackish	Slightly tough& shrunken	Excellent	**8.8**
	BS	Slightly salty and fishy odor dominant	Fade blackish	Semi-elastic	Excellent	**8.5**
	SDS	Attractive salty odor dominant	Whitish	Comparatively tougher & shrunken	Excellent	**9**
	SDS+T	Attractive turmeric odor	Deep yellow	Comparatively tougher & shrunken	Excellent	**9**
30	DS	Attractive salty odor dominant	Fade blackish	Comparatively tougher & shrunken	Excellent	**8.7**
	PC	Attractive salty & slightly fishy odor	Fade blackish	Tough & shrunken	Excellent	**8.6**
	BS	Stimulating salty & fishy odor	Fade blackish	Soft & elastic	Moderately good	**8**
	SDS	Attractive salty odor dominant	Whitish	Comparatively tougher & shrunken	Excellent	**8.9**
	SDS+T	Attractive turmeric odor dominant	Deep yellow	Comparatively tougher & shrunken	Excellent	**9**
45	DS	Attractive salty odor dominant	Fade blackish	Comparatively tougher	Excellent	**8.6**
	PC	Stimulating salty & fishy odor	Fade blackish	Tough	Excellent	**8.5**
	BS	Characteristic fishy & slightly salty odor	Fade blackish	Slightly soft	Slightly good	**7.5**
	SDS	Attractive salty odor dominant	Whitish	Comparatively tougher & shrunken	Excellent	**8.9**
	SDS+T	Attractive turmeric odor dominant	Deep yellow	Comparatively tougher & shrunken	Excellent	**8.9**

60	DS	Stimulating salty & slightly fishy odor	Fade blackish	Comparatively Tougher	Excellent	**8.5**
	PC	Characteristic salty & fishy odor	Fade blackish	Nearly firm	Moderately good	**8**
	BS	Salty odor absent	Fade	Comparatively soft	Moderately accepted	**7.0**
	SDS	Attractive salty odor dominant	Whitish	Comparatively tougher & shrunken	Excellent	**8.8**
	SDS+T	Attractive turmeric odor dominant	Deep yellow	Comparatively tougher & shrunken	Excellent	**8.9**
75	DS	Stimulating salty & fishy odor dominant	Fade blackish	Tough	Moderately good	**8**
	PC	Distinctive fishy & slightly salty odor	Fade blackish	Nearly firm	Slightly good	**7.5**
	BS	Fishy odor dominant	Fade	Soft	Not so good	**5.5**
	SDS	Stimulating salty & slightly fishy odor	Whitish	Comparatively tougher & shrunken	Excellent	**8.8**
	SDS+T	Attractive turmeric odor dominant	Deep yellow	Comparatively tougher & shrunken	Excellent	**8.9**
90	DS	Distinctive salty & fishy odor	Fade	Nearly-firm	Slightly good	**7.5**
	PC	Flavorful fishy odor dominant	Fade	Firm	Moderately accepted	**7.0**
	BS	Slightly putrid odor	Fade	Soft	Rejected	**4.5**
	SDS	Stimulating salty & fishy odor dominant	Whitish	Comparatively Tougher & shrunken	Excellent	**8.7**
	SDS+T	Attractive turmeric odor dominant	Deep yellow	Comparatively Tougher & shrunken	Excellent	**8.8**
105	DS	Flavorful fishy odor dominant	Fade	Firm	Moderately accepted	**7.0**
	PC	Characteristic fishy odor	Fade	Firm	Slightly accepted	**6.5**
	BS	*	*	*	*	*
	SDS	Flavorful salty & fishy odor	Whitish	Tough	Excellent	**8.6**
	SDS+T	Attractive turmeric odor dominant	Deep yellow	Comparatively Tougher & shrunken	Excellent	**8.8**
120	DS	Characteristic fishy and slightly salty odor	Fade	Firm	Slightly accepted	**6.5**
	PC	Salty odor absent	Fade	Slightly soft	Just accepted	**6**
	BS	*	*	*	*	*
	SDS	Flavorful salty & fishy odor	Whitish	Firm	Excellent	**8.6**
	SDS+T	Attractive turmeric odor	Deep yellow	Comparatively Tougher & shrunken	Excellent	**8.7**

135	DS	Salty odor absent	Fade	Slightly soft	Just accepted	**6**
	PC	Slightly Faded fishy odor	Fade	Slightly soft	Neither like or dislike	**5**
	BS	*	*	*	*	*
	SDS	Characteristic fishy odor	Whitish	Firm	Excellent	**8.5**
	SDS+T	Stimulating turmeric & slightly fishy odor	Deep yellow	Comparatively Tougher & shrunken	Excellent	**8.7**
150	DS	Faded fishy odor	Fade	Slightly soft	Neither like or dislike	**5**
	PC	Slightly rancid odor	Fade	Slightly soft	Rejected	**4.5**
	BS	*	*	*	*	*
	SDS	Characteristic fishy odor dominant	Whitish	Firm	Moderately good	**8.0**
	SDS+T	Stimulating turmeric & slightly fishy odor	Deep yellow	Comparatively Tougher & shrunken	Excellent	**8.6**
165	DS	Slightly Rancid odor	Fade	Slightly soft	Rejected	**4.5**
	PC	*	*	*	*	*
	BS	*	*	*	*	*
	SDS	Distinctive fishy & slightly salty odor	Whitish	Firm	Slightly good	**7.5**
	SDS+T	Flavorful turmeric with fishy odor	Yellowish	Comparatively Tougher & shrunken	Excellent	**8.6**
180	DS	*	*	*	*	*
	PC	*	*	*	*	*
	BS	*	*	*	*	*
	SDS	Distinctive fishy & slightly salty odor	Whitish	Nearly firm	Moderately accepted	**7.0**
	SDS+T	Flavorful turmeric with fishy odor	Yellowish	Tough	Excellent	**8.5**
195	DS	*	*	*	*	*
	PC	*	*	*	*	*
	BS	*	*	*	*	*
	SDS	Fishy odor dominant	Whitish	Nearly firm	Slightly accepted	**6.5**
	SDS+T	Flavorful turmeric with fishy odor	Yellowish	Firm	Very much good	**8.2**
210	DS	*	*	*	*	*
	PC	*	*	*	*	*
	BS	*	*	*	*	*
	SDS	Salty odor absent	Whitish	Nearly firm	Just accepted	**6**
	SDS+T	Characteristic fishy with turmeric odor	Yellowish	Firm	Moderately good	**8**
225	DS	*	*	*	*	*
	PC	*	*	*	*	*
	BS	*	*	*	*	*
	SDS	Slightly fishy odor	Whitish	Nearly firm	Not so good	**5.5**
	SDS+T	Characteristic fishy with turmeric odor	Yellowish	Firm	Slightly good	**7.5**

240	DS	*	*	*	*	*
	PC	*	*	*	*	*
	BS	*	*	*	*	*
	SDS	Slightly faded fishy odor	Fade	Comparatively soft	Neither like or dislike	**5**
	SDS+T	Distinctive fishy & slightly turmeric odor	Yellowish	Firm	Moderately accepted	**7.0**
255	DS	*	*	*	*	*
	PC	*	*	*	*	*
	BS	*	*	*	*	*
	SDS	Slightly Rancid odor	Fade	Comparatively soft	Slightly accepted	**4.5**
	SDS+T	Slightly turmeric & fishy odor dominant	Yellowish	Firm	Slightly accepted	**6.5**
270	DS	*	*	*	*	*
	PC	*	*	*	*	*
	BS	*	*	*	*	*
	SDS	*	*	*	*	*
	SDS+T	Slightly turmeric & fishy odor dominant	Fade yellow	Slightly soft	Just accepted	**6**
285	DS	*	*	*	*	*
	PC	*	*	*	*	*
	BS	*	*	*	*	*
	SDS	*	*	*	*	*
	SDS+T	Slightly turmeric odor present	Fade yellow	Slightly soft	Not so good	**5.5**
300	DS	*	*	*	*	*
	PC	*	*	*	*	*
	BS	*	*	*	*	*
	SDS	*	*	*	*	*
	SDS+T	Faded odor	Fade yellow	Comparatively soft	Neither like or dislike	**5**
315	DS	*	*	*	*	*
	PC	*	*	*	*	*
	BS	*	*	*	*	*
	SDS	*	*	*	*	*
	SDS+T	Slightly undesirable odor	Fade yellow	Comparatively soft	Rejected	**4.5**

***= Rejected as per sensory evaluation, acceptability score, < 4.5 is considered as rejected or spoiled.**

Table: 4.4.1(D). Changes in sensory characteristics and scores of different salted Taki (*C. punctatus*) during observation at refrigeration (4^0C) storage (Peryam and Pilgrim, 1957) [DS = Dry salted, PC = Pickle cured, BS = Brine salted, SDS = Sun-dried salted, SDS+T = Turmeric treated sun-dried salted]

Days of storage	Product	Flavor(Smell/ Odor)	Color	Texture	Remarks	Hedonic scale score (0-9)
0	DS	Attractive salty odor	Fade blackish	Tough & shrunken	Excellent	**9**
	PC	Attractive salty odor	Fade blackish	Slightly tough & shrunken	Excellent	**9**
	BS	Attractive salty odor	Fade blackish	Elastic	Excellent	**9**
	SDS	Attractive salty odor	Whitish	Comparatively Tougher & shrunken	Excellent	**9**
	SDS+T	Attractive turmeric odor	Deep yellow	Comparatively Tougher & shrunken	Excellent	**9**
2	DS	Attractive salty odor dominant	Fade blackish	Tough & shrunken	Excellent	**9**
	PC	Attractive salty odor dominant	Fade blackish	Slightly tough& shrunken	Excellent	**8.9**
	BS	Slightly salty and fishy odor dominant	Fade blackish	Semi-elastic	Excellent	**8.5**
	SDS	Attractive salty odor dominant	Whitish	Comparatively Tougher & shrunken	Excellent	**9**
	SDS+T	Attractive turmeric & slightly salty odor	Deep yellow	Comparatively Tougher & shrunken	Excellent	**9**
4	DS	Attractive salty odor dominant	Fade blackish	Comparatively Tougher &shrunken	Excellent	**8.9**
	PC	Attractive salty & slightly fishy odor	Fade blackish	Tough & shrunken	Excellent	**8.8**
	BS	Salty odor slightly present	Fade blackish	Soft & elastic	Moderatel ygood	**8.0**
	SDS	Attractive salty odor dominant	Whitish	Comparatively Tougher & shrunken	Excellent	**8.9**
	SDS+T	Attractive turmeric odor dominant	Deep yellow	Comparatively Tougher & shrunken	Excellent	**9**
6	DS	Stimulating salty & slightly fishy odor	Fade blackish	Tough	Excellent	**8.8**
	PC	Attractive salty & fishy odor dominant	Fade blackish	Tough & shrunken	Excellent	**8.7**
	BS	Salty odor completely absent	Fade blackish	Slightly soft	Slightly good	**7.5**
	SDS	Stimulating salty & slightly fishy odor	Whitish	Comparatively Tougher & shrunken	Excellent	**8.8**
	SDS+T	Stimulating fishy with turmeric odor	Yellowish	Firm	Excellent	**8.9**

8	DS	Stimulating fishy & salty odor dominant	Fade blackish	Tough	Excellent	**8.7**
	PC	Stimulating fishy & salty odor	Fade blackish	Comparatively Tougher	Excellent	**8.6**
	BS	Slightly faded fishy odor	Fade	Comparatively soft	Slightly accepted	**6.5**
	SDS	Stimulating fishy & salty odor dominant	Whitish	Firm	Excellent	**8.7**
	SDS+T	Stimulating fishy with turmeric odor	Yellowish	Firm	Excellent	**8.8**
10	DS	Stimulating fishy & salty odor	Fade blackish	Tough	Excellent	**8.6**
	PC	Characteristic fishy odor dominant	Fade blackish	Nearly firm	Excellent	**8.5**
	BS	Faded fishy odor	Fade	Comparatively soft	Not so good	**5.5**
	SDS	Stimulating fishy & salty odor	Whitish	Firm	Excellent	**8.6**
	SDS+T	Flavorful fishy with turmeric odor	Yellowish	Firm	Excellent	**8.7**
12	DS	Characteristic fishy odor dominant	Fade	Nearly-firm	Excellent	**8.5**
	PC	Characteristic fishy odor	Fade blackish	Nearly firm	Moderately good	**8**
	BS	Slightly rancid odor	Fade	Soft & slimy	Rejected	**4.5**
	SDS	Characteristic fishy odor dominant	Whitish	Firm	Excellent	**8.5**
	SDS+T	Flavorful fishy with turmeric odor	Yellowish	Nearly firm	Excellent	**8.6**
14	DS	Characteristic fishy odor	Fade	Nearly-firm	Moderately good	**8**
	PC	Stimulating fishy odor dominant	Fade	Firm	Slightly good	**7.5**
	BS	*	*	*	*	*
	SDS	Characteristic fishy odor dominant	Whitish	Nearly firm	Very much good	**8.2**
	SDS+T	Characteristic fishy with turmeric odor	Yellowish	Nearly firm	Excellent	**8.5**
16	DS	Characteristic fishy odor and slightly salty odor	Fade	Firm	Slightly good	**7.5**
	PC	Stimulating fishy odor	Fade	Firm	Moderately accepted	**7.0**
	BS	*	*	*	*	*
	SDS	Characteristic fishy odor and slightly salty odor	Whitish	Nearly firm	Slightly good	**7.6**
	SDS+T	Characteristic fishy with turmeric odor	Yellowish	Nearly firm	Moderately good	**8**

18	DS	Salty odor slightly present	Fade	Firm	Moderately accepted	**7**
	PC	Slightly fishy odor	Fade	Firm	Slightly accepted	**6.5**
	BS	*	*	*	*	*
	SDS	Salty odor slightly present	Whitish	Nearly firm	Moderately accepted	**7.2**
	SDS+T	Turmeric odor dominant	Yellowish	Nearly firm	Slightly good	**7.5**
20	DS	Salty odor absent	Fade	Firm	Slightly accepted	**6.5**
	PC	Salty odor slightly present	Fade	Slightly soft	Just accepted	**6**
	BS	*	*	*	*	*
	SDS	Salty odor absent	Fade white	Slightly soft	Slightly accepted	**6.7**
	SDS+T	Slightly turmeric odor	Yellowish	Nearly firm	Moderately accepted	**7**
22	DS	Slightly fishy odor	Fade	Firm	Just accepted	**6**
	PC	Salty odor absent	Fade	Slightly soft	Not so good	**5.5**
	BS	*	*	*	*	*
	SDS	Slightly fishy odor	Fade white	Slightly soft	Just accepted	**6.2**
	SDS+T	Slightly turmeric and distinctive fishy odor	Yellowish	Nearly firm	Slightly accepted	**6.5**
24	DS	Fishy odor dominent	Fade	Slightly soft	Not so good	**5.5**
	PC	Slightly faded fishy odor	Fade	Slightly soft	Neither like nor deslike	**5**
	BS	*	*	*	*	*
	SDS	Fishy odor dominent	Fade	Slightly soft	Not so good	**5.5**
	SDS+T	Slightly turmeric and fishy odor dominant	Fade yellow	Slightly soft	Just accepted	**6**
26	DS	Slightly faded fishy odor	Fade	Slightly soft	Neither like nor deslike	**5**
	PC	Slightly rancid odor	Fade	comparatively soft	Rejected	**4.5**
	BS	*	*	*	*	*
	SDS	Faded fishy odor	Fade	Comparatively soft	Neither like nor dislike	**5**
	SDS+T	Turmeric odor absent	Fade	Slightly soft	Not so good	**5.5**
28	DS	Slightly rancid odor	Fade	Slightly soft	Rejected	**4.5**
	PC	*	*	*	*	*
	BS	*	*	*	*	*
	SDS	Slightly rancid odor	Fade	Comparatively soft	Rejected	**4.5**
	SDS+T	Fishy odor dominant	Fade	Comparatively soft	Neither like nor dislike	**5**
30	DS	*	*	*	*	*
	PC	*	*	*	*	*
	BS	*	*	*	*	*
	SDS	*	*	*	*	*
	SDS+T	Slightly unpleasant odor	Fade	Comparatively soft	Rejected	**4.5**

***= Rejected as per sensory evaluation, acceptability score, < 4.5 is considered as rejected or spoiled.**

Table: 4.4.1(E). Changes in sensory characteristics and scores of different salted Tengra (*M. tengra*) during different days of observation at room temperature (Peryam and Pilgrim, 1957) [DS = Dry salted, PC = Pickle cured, BS = Brine salted, SDS = Sun-dried salted, SDS+T = Turmeric treated sun-dried salted]

Days of storage	Product	Flavor(Smell/ Odor)	Color	Texture	Remarks	Hedonic scale score (0-9)
0	DS	Attractive salty odor	Whitish	Slightly tough	Excellent	**9**
	PC	Attractive salty odor	Whitish	Elastic	Excellent	**9**
	BS	Attractive salty odor	Whitish	Elastic	Excellent	**9**
	SDS	Attractive salty odor	Whitish	Tough	Excellent	**9**
	SDS+T	Attractive turmeric odor	Deep yellow	Tough	Excellent	**9**
15	DS	Attractive salty odor dominant	Whitish	Tough & shrunken	Excellent	**9**
	PC	Attractive salty odor dominant	Whitish	Tough & shrunken	Excellent	**8.8**
	BS	Attractive salty odor dominant	Whitish	Elastic	Excellent	**8.7**
	SDS	Attractive salty odor dominant	Whitish	Tough & shrunken	Excellent	**9**
	SDS+T	Attractive turmeric odor dominant	Deep yellow	Tough & shrunken	Excellent	**9**
30	DS	Attractive salty odor dominant	Whitish	Tough & shrunken	Excellent	**8.9**
	PC	Attractive salty odor dominant	Whitish	Tough & shrunken	Excellent	**8.7**
	BS	Flavorful salty & slightly fishy odor	Whitish	Elastic	Excellent	**8.5**
	SDS	Attractive salty odor dominant	Whitish	Comparatively tougher & shrunken	Excellent	**8.9**
	SDS+T	Attractive turmeric odor dominant	Deep yellow	Comparatively tougher & shrunken	Excellent	**9**
45	DS	Attractive salty odor dominant	Whitish	Comparatively tougher & shrunken	Excellent	**8.8**
	PC	Flavorful salty & slightly fishy odor	Whitish	Semi elastic	Excellent	**8.6**
	BS	Characteristic salty & fishy odor	Whitish	Semi elastic	Moderately good	**8**
	SDS	Attractive salty odor dominant	Whitish	Firm & fragile	Excellent	**8.9**
	SDS+T	Attractive turmeric odor dominant	Deep yellow	Firm & fragile	Excellent	**9**

60	DS	Stimulating salty & slightly fishy odor	Whitish	Tough & firm	Excellent	**8.7**
	PC	Flavorful salty & fishy odor dominant	Whitish	Nearly firm	Excellent	**8.5**
	BS	Stimulating fishy & slightly salty odor	Whitish	Slightly elastic	Slightly good	**7.5**
	SDS	Attractive salty & slightly fishy odor	Whitish	Firm & fragile	Excellent	**8.8**
	SDS+T	Attractive turmeric odor dominant	Deep yellow	Firm & fragile	Excellent	**8.9**
75	DS	Stimulating fishy odor	Whitish	Tough	Excellent	**8.6**
	PC	Stimulating fishy & salty odor	Whitish	Nearly firm	Moderately good	**8**
	BS	Very low stimulating fishy odor	Whitish	Slightly soft	Moderately accepted	**7.0**
	SDS	Attractive salty & fishy odor dominant	Whitish	Firm & fragile	Excellent	**8.8**
	SDS+T	Attractive salty and turmeric odor	Deep yellow	Firm & fragile	Excellent	**8.9**
90	DS	Flavorful fishy odor	Fade	Nearly-firm	Excellent	**8.5**
	PC	Stimulating fishy odor dominant	Fade	Firm	Slightly good	**7.5**
	BS	Salty odor absent	Fade whitish	Slightly soft	Slightly accepted	**6.5**
	SDS	Stimulating salty &fishy odor	Whitish	Firm & fragile	Excellent	**8.7**
	SDS+T	Attractive salty and turmeric odor	Deep yellow	Firm & fragile	Excellent	**8.9**
105	DS	Distinctive fishy odor	Whitish	Firm	Moderately good	**8**
	PC	Characteristics fishy & slightly salty odor	Whitish	Firm	Moderately accepted	**7.0**
	BS	Slightly faded fishy odor	Fade	Comparatively soft	Just accepted	**6**
	SDS	Stimulating salty &fishy odor	Whitish	Firm & fragile	Excellent	**8.6**
	SDS+T	Attractive salty and turmeric odor	Deep yellow	Firm & fragile	Excellent	**8.8**
120	DS	Salty &characteristics fishy odor	Fade	Firm	Slightly good	**7.5**
	PC	Distinctive fishy & slightly salty odor	Whitish	Slightly firm.	Slightly accepted	**6.5**
	BS	Faded odor	Fade	Comparatively soft	Neither like or dislike	**5**
	SDS	Flavorful fishy odor	Whitish	Firm & fragile	Excellent	**8.6**
	SDS+T	Attractive salty and turmeric odor	Deep yellow	Firm & fragile	Excellent	**8.8**

135	DS	Slightly salty & characteristics fishy odor	Fade	Slightly soft	Moderately accepted	**7.0**
	PC	Faded salty odor	Fade	Comparatively soft.	Just accepted	**6**
	BS	Slightly Unpleasant odor	Fade	Soft & little slimy	Rejected	**4.5**
	SDS	Flavorful fishy odor	Whitish	Tough & shrunken	Excellent	**8.5**
	SDS+T	Stimulating fishy with turmeric odor	Deep yellow	Tough & shrunken	Excellent	**8.7**
150	DS	Faded salty odor	Fade	Slightly soft	Slightly accepted	**6.7**
	PC	Salty odor absent	Fade	Comparatively soft.	Just accepted	**5.8**
	BS	*	*	*	*	*
	SDS	Flavorful fishy odor	Whitish	Tough & shrunken	Excellent	**8.4**
	SDS+T	Stimulating fishy with turmeric odor	Deep yellow	Tough & shrunken	Excellent	**8.7**
165	DS	Distinctive fishy odor	Fade	Slightly soft	Slightly accepted	**6.5**
	PC	Fishy odor dominant	Fade	Comparatively soft.	Not so good	**5.5**
	BS	*	*	*	*	*
	SDS	Characteristic fishy odor	Whitish	Tough & shrunken	Very much good	**8.2**
	SDS+T	Stimulating fishy with turmeric odor	Deep yellow	Tough & shrunken	Excellent	**8.6**
180	DS	Fishy odor dominant	Fade white	Slightly soft	Just accepted	**6**
	PC	Slightly faded fishy odor	Fade	Comparatively soft	Neither like or dislike	**5**
	BS	*	*	*	*	*
	SDS	Characteristic fishy odor	Whitish	Tough	Moderately good	**8**
	SDS+T	Flavorful fishy with turmeric odor	Yellowish	Tough & shrunken	Excellent	**8.5**
195	DS	Salty odor absent	Fade white	Comparatively soft	Not so good	**5.5**
	PC	Slightly rancid odor	Fade	Comparatively soft & little slimy.	Rejected	**4.5**
	BS	*	*	*	*	*
	SDS	Distinctive fishy odor	Whitish	Tough	Slightly good	**7.7**
	SDS+T	Flavorful fishy with turmeric odor	Yellowish	Tough & shrunken	Very much good	**8.2**
210	DS	Slightly faded fishy odor	Fade	Comparatively soft	Nither like or dislike	**5**
	PC	*	*	*	*	*
	BS	*	*	*	*	*
	SDS	Distinctive fishy & slightly salty odor	Whitish	Firm	Slightly good	**7.5**
	SDS+T	Characteristic turmeric with fishy odor	Yellowish	Tough & shrunken	Moderately good	**8**

240	DS	Slightly unpleasant odor	Fade	Comparatively soft	Rejected	**4.5**
	PC	*	*	*	*	*
	BS	*	*	*	*	*
	SDS	Distinctive fishy & slightly salty odor	Whitish	Firm	Moderately accepted	**7**
	SDS+T	Characteristic turmeric with fishy odor	Yellowish	Tough	Slightly good	**7.7**
270	DS	*	*	*	*	*
	PC	*	*	*	*	*
	BS	*	*	*	*	*
	SDS	Salty odor absent	Whitish	Firm	Slightly accepted	**6.5**
	SDS+T	Stimulating turmeric & distinctive fishy odor	Yellowish	Tough	Slightly good	**7.5**
300	DS	*	*	*	*	*
	PC	*	*	*	*	*
	BS	*	*	*	*	*
	SDS	Slightly faded fishy odor	Whitish	Firm	Just accepted	**6**
	SDS+T	Slightly turmeric and distinctive fishy odor	Yellowish	Firm	Moderately accepted	**7**
330	DS	*	*	*	*	*
	PC	*	*	*	*	*
	BS	*	*	*	*	*
	SDS	Faded fishy odor	Whitish	Firm	Not so good	**5.5**
	SDS+T	Slightly turmeric and dominant fishy odor	Yellowish	Firm	Slightly accepted	**6.5**
360	DS	*	*	*	*	*
	PC	*	*	*	*	*
	BS	*	*	*	*	*
	SDS	Fishy odor dominant	Whitish	Nearly firm	Neither like or dislike	**5**
	SDS+T	Fishy & faded turmeric odor	Yellowish	Firm	Just accepted	**6**
390	DS	*	*	*	*	*
	PC	*	*	*	*	*
	BS	*	*	*	*	*
	SDS	Slightly Unpleasant odor	Fade	Comparatively soft	Rejected	**4.5**
	SDS+T	Turmeric odor absent	Fade yellow	Slightly soft	Not so good	**5.5**
420	DS	*	*	*	*	*
	PC	*	*	*	*	*
	BS	*	*	*	*	*
	SDS	*	*	*	*	*
	SDS+T	Slightly fishy odor	Fade	Comparatively soft	Neither like nor dislike	**5**
450	DS	*	*	*	*	*
	PC	*	*	*	*	*
	BS	*	*	*	*	*
	SDS	*	*	*	*	*
	SDS+T	Slightly unpleasant odor	Fade	Comparatively soft	Rejected	**4.5**

***= Rejected as per sensory evaluation, acceptability score, < 4.5 is considered as rejected or spoiled.**

Table: 4.4.1(F). Changes in sensory characteristics and scores of different salted Tengra (*M. tengra*) during observation at refrigeration (4^0C) storage (Peryam and Pilgrim, 1957) [DS = Dry salted, PC = Pickle cured, BS = Brine salted, SDS = Sun-dried salted, SDS+T = Turmeric treated sun-dried salted]

Months of storage	Product	Flavor(Smell/ Odor)	Color	Texture	Remarks	Hedonic scale score (0-9)
0	DS	Attractive salty odor	Whitish	Slightly tough	Excellent	**9**
	PC	Attractive salty odor	Whitish	Elastic	Excellent	**9**
	BS	Attractive salty odor	Whitish	Elastic	Excellent	**9**
	SDS	Attractive salty odor	Whitish	Tough	Excellent	**9**
	SDS+T	Attractive turmeric odor	Deep yellow	Tough	Excellent	**9**
4	DS	Attractive salty odor dominant	Whitish	Tough & shrunken	Excellent	**8.7**
	PC	Attractive salty odor dominant	Whitish	Tough & shrunken	Excellent	**8.6**
	BS	Attractive salty odor dominant	Whitish	Elastic	Excellent	**8.5**
	SDS	Attractive salty odor dominant	Whitish	Tough	Excellent	**8.8**
	SDS+T	Attractive turmeric odor dominant	Deep yellow	Tough	Excellent	**9**
8	DS	Stimulating fishy & salty odor	Whitish	Tough & shrunken	Excellent	**8.5**
	PC	Attractive salty odor dominant	Whitish	Tough & shrunken	Very much good	**8.2**
	BS	Characteristic salty odor	Whitish	Elastic	Slightly good	**7.5**
	SDS	Stimulating salty &fishy odor	Whitish	Tough & shrunken	Excellent	**8.6**
	SDS+T	Attractive salty and turmeric odor	Deep yellow	Tough & shrunken	Excellent	**8.7**
12	DS	Characteristic fishy odor	Whitish	Slightly firm	Very much good	**8.2**
	PC	Stimulating fishy & salty odor	Whitish	Nearly firm	Moderately accepted	**7**
	BS	Stimulating fishy & salty odor	Whitish	Slightly elastic	Slightly accepted	**6.5**
	SDS	Stimulating fishy odor	Whitish	Firm & Fragile	Excellent	**8.5**
	SDS+T	Salty and turmeric odor dominant	Deep Yellow	Firm & Fragile	Excellent	**8.6**
16	DS	Stimulating fishy odor	Whitish	Slightly firm	Moderately accepted	**7.2**
	PC	Characteristics fishy & salty odor	Whitish	Slightly firm	Slightly accepted	**6.5**
	BS	Fishy odor dominant & slightly salty odor	Fade whitish	Slightly soft	Just accepted	**6**
	SDS	Characteristic fishy odor dominant	Whitish	Firm & Fragile	Slightly good	**7.5**
	SDS+T	Salty and turmeric odor dominant	Deep Yellow	Firm & Fragile	Very much good	**8.2**

20	DS	Slightly salty and fishy odor	Fade	Slightly soft	Slightly accepted	**6.5**
	PC	Fishy odor dominant & Slightly salty odor	Fade	Slightly soft	Just accepted	**6**
	BS	Salty odor absent	Fade	Comparatively soft & little slimy	Not so good	**5.5**
	SDS	Slightly fishy odor	Whitish	Firm	Slightly accepted	**6.6**
	SDS+T	Characteristic salty and turmeric odor	Deep yellow	Comparatively Tougher & shrunken	Moderately accepted	**7.2**
24	DS	Fishy odor dominant and slightly salty odor	Fade white	Slightly soft	Just accepted	**6.0**
	PC	Salty odor absent	Fade	Slightly soft	Not so good	**5.5**
	BS	Slightly faded fishy odor	Fade	Slightly soft	Neither like or dislike	**5**
	SDS	Fishy odor dominant	Whitish	Firm & Fragile	Just accepted	**6.0**
	SDS+T	Slightly turmeric & fishy odor present	Yellowish	Firm & Fragile	Slightly accepted	**6.5**
28	DS	Salty odor absent	Fade white	Comparatively soft	Not so good	**5.5**
	PC	Fishy odor dominant	Fade	Comparatively soft	Neither like or dislike	**5**
	BS	Slightly putrid odor	Fade	soft	Rejected	**4.5**
	SDS	Salty odor absent & slightly fishy odor	Whitish	Slightly firm	Not so good	**5.5**
	SDS+T	Slightly turmeric odor still present	Yellowish	Firm & Fragile	Just accepted	**6**
32	DS	Fishy odor dominant	Fade	Comparatively soft	Neither like or dislike	**5**
	PC	Slightly rancid odor	Fade	Comparatively soft & little slimy	Rejected	**4.5**
	BS	*	*	*	*	*
	SDS	Fishy odor dominant	Fade	Comparatively soft	Neither like or dislike	**5**
	SDS+T	Turmeric odor absent	Fade yellow	Slightly soft	Not so good	**5.5**
36	DS	Slightly rancid odor	Fade	Comparatively soft	Rejected	**4.5**
	PC	*	*	*	*	*
	BS	*	*	*	*	*
	SDS	Unpleasant fishy odor	Fade	Comparatively soft	Rejected	**4.5**
	SDS+T	Slightly fishy odor	Fade	Slightly soft	Neither like or dislike	**5**
40	DS	*	*	*	*	*
	PC	*	*	*	*	*
	BS	*	*	*	*	*
	SDS	*	*	*	*	*
	SDS+T	Slightly off odor	Fade	Comparatively soft	Rejected	**4.5**

***= Rejected as per sensory evaluation, acceptability score, < 4.5 is considered as rejected or spoiled.**

For simplicity of evaluation, hedonic scale score was compared and changes in sensory scores revealing their characteristic changes in Dry salted (DS), Pickle cured (PC), Brine salted (BS), Sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish-products during different duration of storage both at (A) room temperature and (B) refrigeration temperature which is given in Figure 4.4.1.

From Figure 4.4.1 it is clear that all salted samples gradually decreased their quality and inversely related with storage period.

It could be also noticed from the figure that salted samples at zero day has received highest scores, followed by 15 days at room temperature and 2 month at refrigeration temperature in case of DS, PC, BS, SDS and SDS+T shol and taki fish-products whereas 30 days at room temperature and 4 month at refrigeration temperature in case of DS, PC, BS, SDS and SDS+T tengra fish-products. Samples, has received lowest scores of **4.5** when the product was rejected. This wide range indicated the diversity in the final quality and can be largely attributed to the effect of various conditions upon the salting agents and activities. It is seen that the main factor affecting the quality is time of storage.

It is in agreement with the findings of Ahmed *et al.* (2010a) who observed that sensory score was highest in '0' day and gradually decreased in salted Kass (*Hydrocynus forskalii*) during 6 month storage at room temperature (37^0C). In another study Akter *et al.* (2013a) observed that, turmeric treted sun-dried *M. vittatus* had sensory score of 4.0 at the end of 9 month while stored at room temperature which is more or less similar with present findings of SDS+T taki and tengra fish products.

Among the three experimental fishes (shol, taki and tengra) in room temperature, highest shelf life was observed in sun dried salt +Turmeric treated (SDS+T) tengra fish products which was 420 days (14 months) and lowest was observed in brine salted (BS) taki fish products which was 75 days. Whereas, in refrigerated

temperature, highest shelf life was observed in sun dried salt +Turmeric treated (SDS+T) tengra fish products which was 36 months and lowest was observed in brine salted (BS) taki fish products which was 10 months.

In all cases, Brine salted (BS) product has short shelf-life than other 4 types of salted products both room and refrigeration storage, whereas turmeric treated sun-dried salted (SDS+T) has longer shelf life than other four types of salted fish products.

Trim and Curran (1983) stated that, brined and dry salted products had a shelf life of approximately 100 days. No insect infestation or broken pieces were found around the salted-dried shol, taki and tengra fishes. It is also clear that the salted and salted-dried fish products stored at 4^0C temperature have been found to have longer shelf life than the salted and salted-dried fish products stored at room temperature. In room and refrigerator temperature, the quality of all 5 types of salt treated tengra fish products was found better than other fish products.

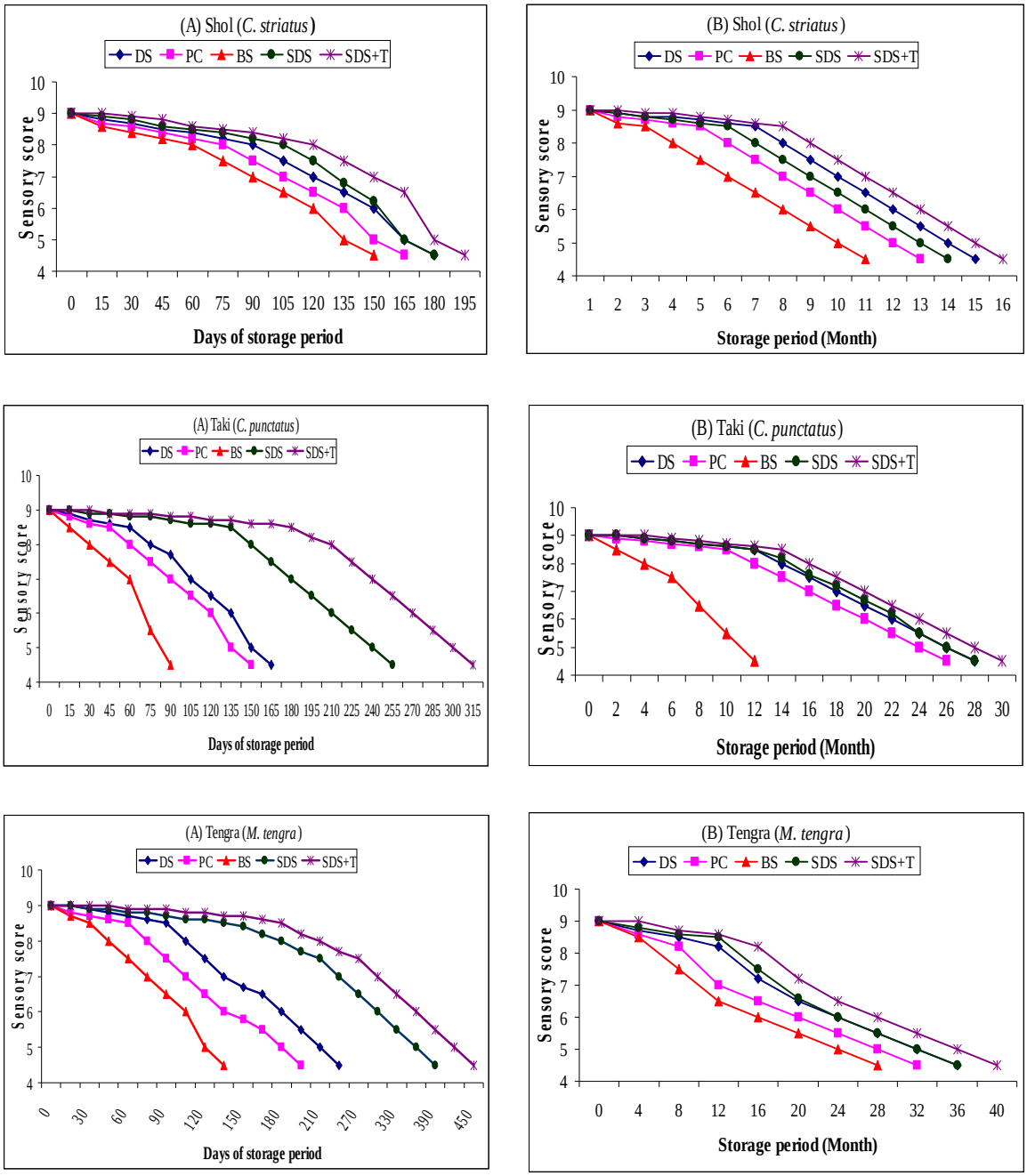


Figure. 4.4.1. Changes in sensory scores of five types of salted shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish during different duration of storage both at (A) room temperature and (B) refrigeration temperature (DS = Dry salted, PC = Pickle cured, BS= Brine salted, SDS=Sun-dried salted and SDS+T= turmeric treated sun-dried salted) [Peryam and Pilgrim, 1957]

4.4.2. Proximate composition analysis

4.4.2.1. Changes in proximate composition of 5 types of salted shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fishes in different storage condition

Analysis of four basic constituents in the edible portion of a fish namely moisture (water), protein, fat (lipid) and ash are referred to as proximate analysis (Love, 1988).

The summery of the changes of proximate composition of Dry salted (DS), Pickle cured (PC), Brine salted (BS), Sun-dried salted (SDS) and Turmeric treated sun-dried salted (SDS+T) shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fishes in different storage condition is shown in Table 4.4.2.1 (A) - (F).

Table: 4.4.2.1(A). Changes in proximate composition of Dry salted (DS), Pickle cured (PC), Brine salted (BS), Sun-dried salted (SDS) and Turmeric treated sun-dried salted (SDS+T) shol (*C. striatus*) during different duration of storage at room temperature (26-32^{0}C)

a) Dry salted (DS) Shol:

Days of storage period	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0*	48.84	28.21	3.99	18.98
15	49.64	27.94	3.89	18.82
30	50.30	27.66	3.84	18.69
45	50.44	27.57	3.78	18.49
60	50.71	27.38	3.71	18.41
75	51.17	27.11	3.66	18.29
90	51.40	27.0	3.58	18.11
105	52.09	26.75	3.52	17.97
120	52.15	26.60	3.48	17.88
135	53.11	26.06	3.41	17.76
150	53.45	25.89	3.35	17.61
165	53.69	25.52	3.30	17.58

b) Pickle cured (PC) Shol:

Days of storage period	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0*	50.29	27.64	3.79	18.69
15	51.08	27.13	3.73	18.62
30	51.65	26.79	3.68	18.51
45	52.68	25.97	3.63	18.42
60	52.95	25.72	3.57	18.33
75	53.02	25.65	3.52	18.21
90	53.06	25.57	3.48	18.17
105	53.80	25.33	3.41	18.00
120	54.00	25.28	3.36	17.84
135	54.60	24.99	3.20	17.59
150	54.94	24.82	3.11	17.45

c) Brine salted (BS) Shol:

Days of storage period	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0*	60.17	20.72	3.20	16.06
15	60.72	20.65	3.17	15.94
30	61.44	20.07	3.11	15.83
45	61.78	19.80	3.08	15.72
60	62.46	19.51	3.01	15.60
75	62.98	19.22	2.98	15.54
90	63.32	18.80	2.93	15.45
105	63.91	18.25	2.86	15.33
120	64.85	17.53	2.80	15.28
135	65.97	17.06	2.72	15.14

d) Sun-dried salted (SDS) Shol:

Days of storage period	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0*	29.77	41.48	5.10	22.80
15	29.98	41.39	5.02	22.74
30	30.24	41.16	4.93	22.68
45	30.83	40.77	4.89	22.61
60	31.15	40.61	4.74	22.53
75	31.61	40.35	4.62	22.47
90	32.02	39.97	4.59	22.44
105	32.58	39.65	4.44	22.39
120	33.37	39.06	4.36	22.22
135	33.82	38.91	4.21	22.09
150	34.61	38.75	4.08	21.71
165	35.26	38.62	3.81	21.44

e) Turmeric treated sun-dried salted (SDS+T) Shol:

Days of storage period	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0*	30.92	41.0	4.79	22.41
15	31.26	40.82	4.67	22.35
30	31.55	40.68	4.52	22.28
45	31.98	40.49	4.45	22.11
60	32.53	40.24	4.27	22.02
75	32.95	39.97	4.16	21.94
90	33.66	39.74	4.03	21.86
105	33.94	39.61	3.97	21.75
120	34.20	39.50	3.81	21.69
135	34.87	39.20	3.74	21.43
150	35.52	38.85	3.46	21.37
165	36.12	38.36	3.33	21.25
180	36.98	38.01	3.26	21.07

*= Immediate after ripening period

Table: 4.4.2.1(B). Changes in proximate composition of Dry salted (DS), Pickle cured (PC), Brine salted (BS), Sun-dried salted (SDS) and Turmeric treated sun-dried salted (SDS+T) shol (*C. striatus*) during different duration in refrigeration storage (4^0C)

a) Dry salted (DS) Shol:

Storage period (Month)	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0*	48.84	28.21	3.99	18.98
2	48.93	28.19	3.96	18.93
4	49.08	28.12	3.93	18.89
6	49.22	28.06	3.88	18.84
8	49.41	27.97	3.83	18.81
10	49.58	27.91	3.79	18.80
12	49.72	27.84	3.74	18.73
14	49.89	27.79	3.71	18.69
16	50.15	27.71	3.69	18.53
18	50.33	27.66	3.62	18.46
20	50.67	27.59	3.58	18.37
22	50.88	27.42	3.54	18.29
24	51.02	27.38	3.50	18.11
26	51.24	27.25	3.46	17.94

b) Pickle cured (PC) Shol:

Storage period (Month)	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0*	50.29	27.64	3.79	18.69
2	50.43	27.61	3.72	18.61
4	50.67	27.55	3.68	18.57
6	50.79	27.41	3.61	18.52
8	50.92	27.34	3.55	18.48
10	51.09	27.21	3.47	18.39
12	51.22	27.09	3.41	18.27
14	51.59	26.91	3.39	18.19
16	51.93	26.78	3.34	18.05
18	52.17	26.69	3.31	17.92
20	52.42	26.45	3.27	17.87
22	52.85	26.34	3.22	17.76

c) Brine salted (BS) Shol:

Storage period (Month)	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0*	60.17	20.72	3.20	16.06
2	60.43	20.51	3.17	16.02
4	60.87	20.36	3.15	15.96
6	61.19	20.02	3.12	15.90
8	61.57	19.85	3.09	15.84
10	61.94	19.69	3.05	15.77
12	62.13	19.38	3.01	15.71
14	62.76	19.13	2.98	15.64
16	63.19	18.77	2.94	15.56
18	63.48	18.50	2.89	15.49

d) Sun-dried salted (SDS) Shol:

Storage period (Month)	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0*	29.77	41.48	5.10	22.80
2	29.93	41.35	5.01	22.75
4	30.36	41.21	4.93	22.71
6	30.65	41.04	4.89	22.61
8	30.97	40.89	4.82	22.52
10	31.24	40.64	4.78	22.39
12	31.85	40.48	4.67	22.25
14	32.19	40.27	4.56	22.18
16	32.53	40.11	4.49	22.06
18	32.91	39.97	4.35	21.97
20	33.44	39.73	4.23	21.85
22	33.79	39.52	4.19	21.72
24	34.09	39.30	4.01	21.66

e) Turmeric treated sun-dried salted (SDS+T) Shol:

Storage period (Month)	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0*	30.92	41.0	4.79	22.41
2	31.11	40.94	4.71	22.38
4	31.36	40.88	4.66	22.33
6	31.67	40.79	4.58	22.29
8	31.99	40.71	4.52	22.24
10	32.24	40.59	4.45	22.20
12	32.48	40.35	4.39	22.15
14	32.71	40.13	4.24	22.08
16	33.02	39.87	4.19	21.98
18	33.58	39.69	4.13	21.91
20	33.76	39.52	4.01	21.82
22	34.24	39.34	3.94	21.78
24	34.81	39.13	3.82	21.66
26	35.07	39.04	3.75	21.57
28	35.48	38.81	3.69	21.41

*= Immediate after ripening period

Table: 4.4.2.1(C). Changes in proximate composition of Dry salted (DS), Pickle cured (PC), Brine salted (BS), Sun-dried salted (SDS) and Turmeric treated sun-dried salted (SDS+T) taki (*C. punctatus*) during different duration of storage at room temperature (26-32^{0}C)

a) Dry salted (DS) Taki:

Days of storage period	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0*	46.21	23.58	3.93	26.37
15	46.87	23.42	3.90	26.18
30	47.22	23.28	3.84	26.07
45	47.83	23.02	3.81	25.93
60	48.25	22.83	3.77	25.72
75	48.60	22.66	3.72	25.50
90	49.08	22.35	3.69	25.42
105	49.39	22.13	3.67	25.23
120	49.71	21.93	3.62	25.00
135	50.45	21.77	3.56	24.75
150	50.92	21.53	3.49	24.55

b) Pickle cured (PC) Taki:

Days of storage period	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0*	52.71	21.39	3.40	22.96
15	52.95	21.30	3.35	22.85
30	53.13	21.26	3.31	22.78
45	53.62	21.11	3.28	22.46
60	53.95	20.96	3.22	22.25
75	54.39	20.71	3.19	22.14
90	54.96	20.50	3.13	21.93
105	55.47	20.32	3.07	21.59
120	55.84	20.12	2.99	21.37
135	56.55	20.06	2.93	21.02

c) Brine salted (BS) Taki:

Days of storage period	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0*	62.28	18.02	2.76	17.24
15	62.71	17.81	2.71	17.02
30	63.46	17.58	2.67	16.74
45	63.94	17.34	2.62	16.48
60	64.53	17.13	2.54	16.14
75	65.08	16.92	2.48	15.99

d) Sun-dried salted (SDS) Taki:

Days of storage period	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0*	9.77	47.69	7.47	35.16
15	10.13	47.56	7.31	35.11
30	10.46	47.41	7.19	35.06
45	10.78	47.27	7.02	34.95
60	11.11	47.02	6.95	34.89
75	11.59	46.78	6.89	34.80
90	11.84	46.66	6.74	34.72
105	12.18	46.52	6.67	34.64
120	12.55	46.43	6.53	34.55
135	12.89	46.31	6.41	34.50
150	13.03	46.27	6.38	34.41
165	13.44	46.12	6.29	34.38
180	13.77	46.04	6.15	34.30
195	14.19	45.81	6.06	34.23
210	14.51	45.70	5.94	34.17
225	14.82	45.64	5.88	34.11
240	14.97	45.68	5.81	34.05

e) Turmeric treated sun-dried salted (SDS+T) Taki:

Days of storage period	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0*	12.72	46.06	7.09	34.29
15	12.98	45.97	7.02	34.21
30	13.11	45.92	6.88	34.15
45	13.37	45.86	6.51	34.08
60	13.59	45.79	6.39	34.02
75	13.78	45.73	6.22	33.97
90	13.93	45.67	6.16	33.82
105	14.21	45.59	6.09	33.79
120	14.46	45.43	6.01	33.70
135	14.82	45.32	5.92	33.63
150	15.13	45.26	5.89	33.58
165	15.44	45.11	5.81	33.53
180	15.81	44.99	5.77	33.47
195	16.25	44.78	5.68	33.41
210	16.77	44.64	5.59	33.33
225	17.04	44.57	5.52	33.28
240	17.41	44.42	5.47	33.15
255	17.76	44.36	5.38	33.07
270	18.12	44.25	5.24	33.00
285	18.59	44.11	5.13	32.96
300	18.88	43.92	5.01	32.90

*= Immediate after ripening period

Table: 4.4.2.1(D). Changes in proximate composition of Dry salted (DS), Pickle cured (PC), Brine salted (BS), Sun-dried salted (SDS) and Turmeric treated sun-dried salted (SDS+T) taki (*C. punctatus*) during different duration in refrigeration storage (4^0C)

a) Dry salted (DS) Taki:

Storage period (Month)	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0*	46.21	23.58	3.93	26.37
2	46.38	23.54	3.91	26.31
4	46.57	23.45	3.88	26.25
6	46.76	23.40	3.85	26.13
8	46.94	23.33	3.80	26.06
10	47.27	23.26	3.76	25.98
12	47.44	23.11	3.74	25.83
14	47.67	22.95	3.69	25.77
16	47.98	22.86	3.65	25.58
18	48.26	22.72	3.61	25.49
20	48.61	22.53	3.57	25.37
22	48.99	22.34	3.52	25.25
24	49.38	22.17	3.48	25.18
26	49.89	21.94	3.44	25.09

b) Pickle cured (PC) Taki:

Storage period (Month)	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0*	52.71	21.39	3.40	22.96
2	52.85	21.34	3.37	22.91
4	53.04	21.30	3.33	22.87
6	53.22	21.26	3.30	22.80
8	53.49	21.19	3.26	22.71
10	53.71	21.12	3.23	22.59
12	54.02	21.01	3.20	22.47
14	54.33	20.91	3.17	22.33
16	54.50	20.72	3.14	22.17
18	54.88	20.66	3.09	21.92
20	55.05	20.59	3.03	21.86
22	55.40	20.48	2.98	21.69
24	55.71	20.36	2.92	21.58

c) Brine salted (BS) Taki:

Storage period (Month)	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0*	62.28	18.02	2.76	17.24
2	62.44	17.92	2.73	17.13
4	62.82	17.80	2.70	17.01
6	63.09	17.71	2.67	16.89
8	63.63	17.63	2.64	16.67
10	64.02	17.45	2.59	16.50

d) Sun-dried salted (SDS) Taki:

Storage period (Month)	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0*	9.77	47.69	7.47	35.16
2	9.91	47.61	7.33	35.10
4	10.09	47.53	7.21	35.03
6	10.34	47.44	7.15	34.95
8	10.79	47.32	7.08	34.89
10	11.12	47.20	6.99	34.83
12	11.53	47.09	6.86	34.76
14	11.91	46.91	6.79	34.88
16	12.11	46.73	6.71	34.81
18	12.42	46.52	6.67	34.73
20	12.82	46.37	6.62	34.67
22	13.04	46.21	6.59	34.61
24	13.22	46.06	6.53	34.58
26	13.40	45.91	6.48	34.41

e) Turmeric treated sun-dried salted (SDS+T) Taki:

Storage period (Month)	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0*	12.72	46.06	7.09	34.29
2	12.81	45.92	7.00	34.24
4	12.99	45.83	6.92	34.19
6	13.22	45.79	6.86	34.11
8	13.58	45.68	6.80	34.06
10	13.84	45.57	6.71	33.93
12	14.13	45.45	6.67	33.88
14	14.52	45.33	6.61	33.79
16	14.81	45.21	6.55	33.70
18	15.12	45.15	6.41	33.61
20	15.41	45.06	6.35	33.57
22	15.74	44.94	6.27	33.42
24	16.01	44.81	6.14	33.38
26	16.34	44.75	6.09	33.29
28	17.02	44.53	6.00	33.13

*= Immediate after ripening period

Table: 4.4.2.1(E). Changes in proximate composition of Dry salted (DS), Pickle cured (PC), Brine salted (BS), Sun-dried salted (SDS) and Turmeric treated sun-dried salted (SDS+T) tengra (*M. tengra*) during different duration of storage at room temperature (26-32^{0}C)

a) Dry salted (DS) Tengra:

Days of storage period	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0*	41.41	22.05	10.65	26.15
15	41.68	21.96	10.60	26.08
30	41.91	21.91	10.53	25.92
45	42.19	21.88	10.48	25.89
60	42.40	21.80	10.41	25.81
75	42.62	21.74	10.33	25.73
90	42.80	21.69	10.26	25.69
105	43.11	21.64	10.20	25.50
120	43.29	21.51	10.13	25.44
135	43.56	21.41	10.02	25.36
150	43.90	21.32	9.93	25.28
165	44.22	21.23	9.81	25.20
180	44.43	21.17	9.70	25.14
195	44.61	21.05	9.64	25.09
210	44.82	20.99	9.59	25.00

b) Pickle cured (PC) Tengra:

Days of storage period	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0*	45.88	20.43	9.40	24.62
15	46.02	20.38	9.37	24.57
30	46.37	20.24	9.32	24.40
45	46.58	20.16	9.28	24.35
60	46.82	20.08	9.24	24.26
75	47.11	19.94	9.20	24.14
90	47.41	19.80	9.15	24.03
105	47.60	19.73	9.11	23.95
120	47.91	19.61	9.05	23.81
135	48.18	19.50	8.97	23.73
150	48.39	19.46	8.88	23.67
165	48.76	19.32	8.74	23.59
180	49.09	19.28	8.68	23.41

c) Brine salted (BS) Tengra:

Days of storage period	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0*	57.35	15.30	6.84	20.80
15	57.72	15.21	6.71	20.69
30	58.11	15.13	6.66	20.51
45	58.41	15.03	6.53	20.42
60	58.70	14.92	6.45	20.29
75	59.14	14.80	6.32	20.06
90	59.66	14.69	6.20	19.83
105	60.03	14.41	6.17	19.66
120	60.70	14.28	6.01	19.40

d) Sun-dried salted (SDS) Tengra:

Days of storage period	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0*	4.9	43.00	15.99	36.20
30	5.27	42.96	15.86	36.11
60	5.43	42.91	15.79	36.03
90	5.74	42.85	15.71	35.93
120	5.97	42.79	15.66	35.87
150	6.15	42.72	15.52	35.80
180	6.36	42.65	15.47	35.71
210	6.84	42.50	15.31	35.62
240	7.48	42.44	15.20	35.57
270	7.91	42.35	15.06	35.43
300	8.30	42.28	14.88	35.39
330	8.74	42.12	14.64	35.24
360	9.05	42.00	14.41	35.18

d) Turmeric treated sun-dried salted (SDS+T) Tengra:

Days of storage period	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0*	6.08	42.60	15.58	35.60
30	6.35	42.49	15.50	35.51
60	6.60	42.41	15.42	35.46
90	6.89	42.29	15.28	35.35
120	7.15	42.15	15.18	35.27
150	7.54	42.07	15.04	35.20
180	7.96	41.99	14.90	35.11
210	8.41	41.91	14.74	35.02
240	8.83	41.78	14.53	34.96
270	9.38	41.63	14.37	34.89
300	9.74	41.57	14.21	34.71
330	10.19	41.42	14.13	34.56
360	10.53	41.31	13.90	34.40
390	11.25	41.18	13.78	34.31
420	11.62	41.03	13.40	34.22

*= Immediate after ripening period

Table: 4.4.2.1(F). Changes in proximate composition of Dry salted (DS), Pickle cured (PC), Brine salted (BS), Sun-dried salted (SDS) and Turmeric treated sun-dried salted (SDS+T) tengra (*M. tengra*) during different duration in refrigeration (4^0C) storage

a) Dry salted (DS) Tengra:

Storage period (Month)	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0*	41.41	22.05	10.65	26.15
4	41.71	21.92	10.54	26.04
8	42.01	21.82	10.42	25.92
12	42.37	21.71	10.28	25.81
16	42.74	21.62	10.13	25.66
20	43.21	21.52	9.99	25.47
24	43.59	21.40	9.84	25.31
28	43.89	21.24	9.69	25.20
32	44.29	21.10	9.52	25.10

b) Pickle cured (PC) Tengra:

Storage period (Month)	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0*	45.88	20.43	9.40	24.62
4	46.13	20.33	9.31	24.54
8	46.34	20.17	9.22	24.43
12	46.71	20.05	9.10	24.35
16	47.31	19.89	8.94	24.24
20	47.70	19.74	8.78	24.02
24	48.17	19.60	8.56	23.89
28	48.79	19.41	8.41	23.59

c) Brine salted (BS) Tengra:

Storage period (Month)	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0*	57.35	15.30	6.84	20.80
4	57.72	15.18	6.76	20.69
8	58.21	14.93	6.67	20.55
12	58.79	14.70	6.58	20.29
16	59.36	14.51	6.43	20.09
20	59.92	14.30	6.27	19.78
24	60.57	14.10	6.09	19.48

d) Sun-dried salted (SDS) Tengra:

Storage period (Month)	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0*	4.9	43.00	15.99	36.20
4	5.14	42.93	15.90	36.13
8	5.39	42.86	15.82	36.01
12	5.55	42.79	15.74	35.95
16	5.73	42.70	15.67	35.90
20	5.95	42.62	15.54	35.83
24	6.19	42.53	15.39	35.71
28	6.40	42.47	15.20	35.67
32	6.74	42.40	15.09	35.50

e) Turmeric treated sun-dried salted (SDS+T) Tengra:

Storage period (Month)	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
0*	6.08	42.60	15.58	35.60
4	6.23	42.55	15.41	35.51
8	6.57	42.49	15.30	35.47
12	6.95	42.41	15.17	35.34
16	7.32	42.32	15.09	35.28
20	7.66	42.26	14.91	35.20
24	8.01	42.19	14.79	35.13
28	8.41	42.11	14.52	35.06
32	8.70	41.99	14.43	34.98
36	9.03	41.86	14.38	34.82

*= Immediate after ripening period

On the whole, processing of Shol, Taki and Tengra fish by salting and sun-dried salting had a significant effect on the proximate composition of these fishes. Hence it was observed, by comparison of shelf life values (storage both room and refrigeration) with the pre storage '0-Day' (fresh process) values that there was an increase in the moisture content of these fish-products whereas, there was a reduction in protein, fat and ash contents of these salted fish-products [Table 4.4.2.1 (A)- (F)]. This might be due to longer storage period. Ash decreased comparatively at lower rate than the protein and fat. These findings are similar with the findings of Siddique and Aktar (2011).

Among room temperature stored fish-products, lowest shelf-life period of about 135 days was observed in shol fish treated with brine salt (BS) whereas, turmeric treated sun-dried salted (SDS+T) shol showed a maximum period of 180 days. Similar results of lowest shelf-life were observed in taki (75 days) and tengra (120 days) treated with BS. Highest shelf-life of 300 and 420 days was recorded in taki and tengra treated with SDS+T [Table 4.4.2.1 (A), (C) & (E)].

On the other hand, among refrigeration stored fish-products, lowest shelf-life of about 18 months, 10 months and 24 months were observed in brine salted (BS) shol, taki and tengra fish-products whereas, turmeric treated sun-dried salted (SDS+T) shol, taki and tengra fish-products showed a maximum shelf-life of 28 months, 28 months and 36 months respectively [Table 4.4.2.1 (B), (D) & (F)].

A number of studies have been conducted on freshly stored salted and salted-dried fishes but these studies did not focus on the proximate compositions of longer time stored fish-products.

Monsur (2007) stated that, during 6 months storage at room temperature, moisture content increases in a range of 37.30 to 43.5%, 37.71 to 43.2% and 54.30 to 60.0% in case of dry salted, pickle cured and brine salted punti fish and 36.85 to 43.6%, 45.16 to 50.0% and 53.22 to 59.5% in case of dry salted, pickle cured and brine salted

chapila fish, whereas percent of protein decreases in a range of 27.14 to 21.8%, 25.32 to 21.0% and 21.40 to 15.8% in case of dry salted, pickle cured and brine salted punti fish and 25.66 to 19.5%, 22.17 to 17.8% and 23.28 to 16.9% in case of dry salted, pickle cured and brine salted chapila fish which is similar with present findings.

According to Kumar *et al.* (2013), moisture content increased from 7.14 to 15.07% and protein and fat content decreased from 51.32 to 46.33% and 9.48 to 5.78% respectively in case of 6 month stored 30% salted sun-dried *L. gonius* fish at room temperature whish is also similar with present findings.

The changing pattern of proximate composition of Dry salted (DS), Pickle cured (PC), Brine salted (BS), Sun-dried salted (SDS) and Turmeric treated sun-dried salted (SDS+T) shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fishes in different storage condition and different duration is shown in Figure 4.4.2.1 (A) - (F).

Changes in proximate composition in 5 different types of salted-products of all three fishes (shol, taki and tengra) stored both at room and refrigeration temperature indicated that, protein and fat content of sun-dried-salted products were much higher than other salted-products [Figure 4.4.2.1 (A) - (F)].

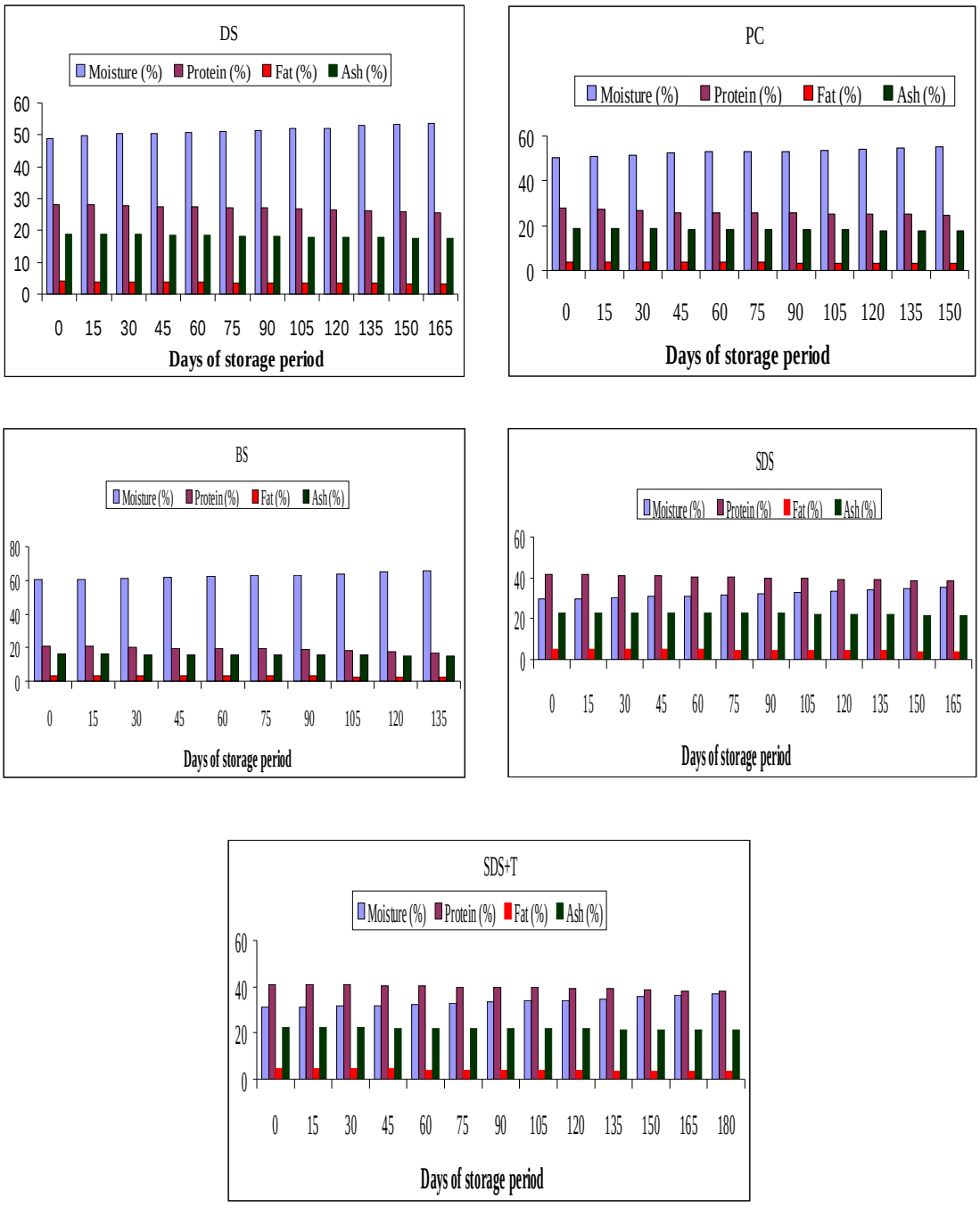


Figure. 4.4.2.1(A). The changing pattern in proximate composition of salted shol (*C. striatus*) during different days of observation, storage at room temperature (26-32^{0}C) in different treatments (DS= Dry salted, PC= Pickle cured, BS= Brine salted, SDS= Sun-dried salted, SDS+T= Turmeric treated sun-dried salted)

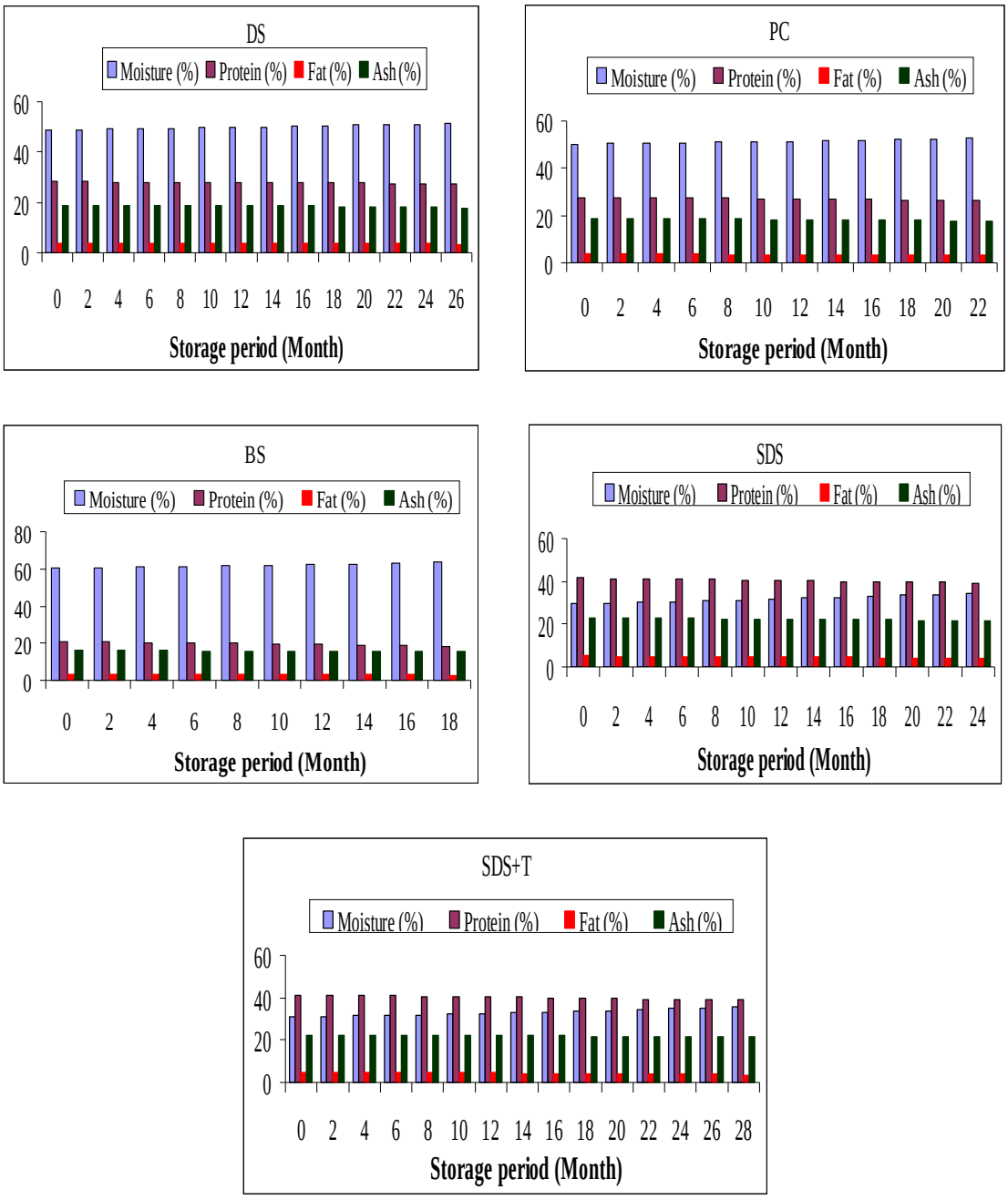


Figure. 4.4.2.1(B). The changing pattern in proximate composition of salted shol (*C. striatus*) during different observation, of storage at refrigeration temperature (4^0C) in different treatments (DS= Dry salted, PC= Pickle cured, BS= Brine salted, SDS= Sun-dried salted, SDS+T= Turmeric treated sun-dried salted)

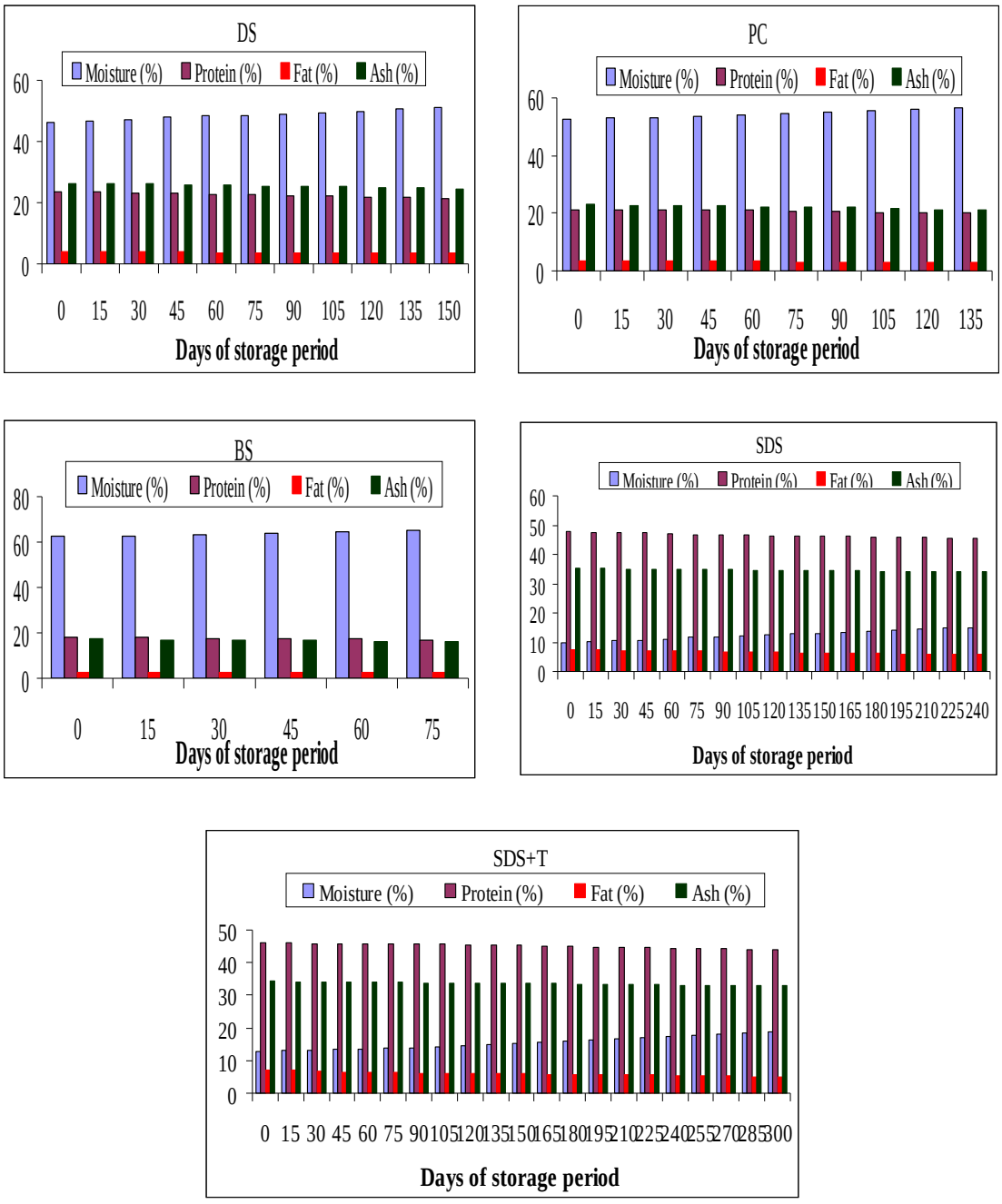


Figure. 4.4.2.1(C). The changing pattern in proximate composition of salted taki (*C. punctatus*) during different days of observation, storage at room temperature (26-32^{0}C) in different treatments (DS= Dry salted, PC= Pickle cured, BS= Brine salted, SDS= Sun-dried salted, SDS+T= Turmeric treated sun-dried salted)

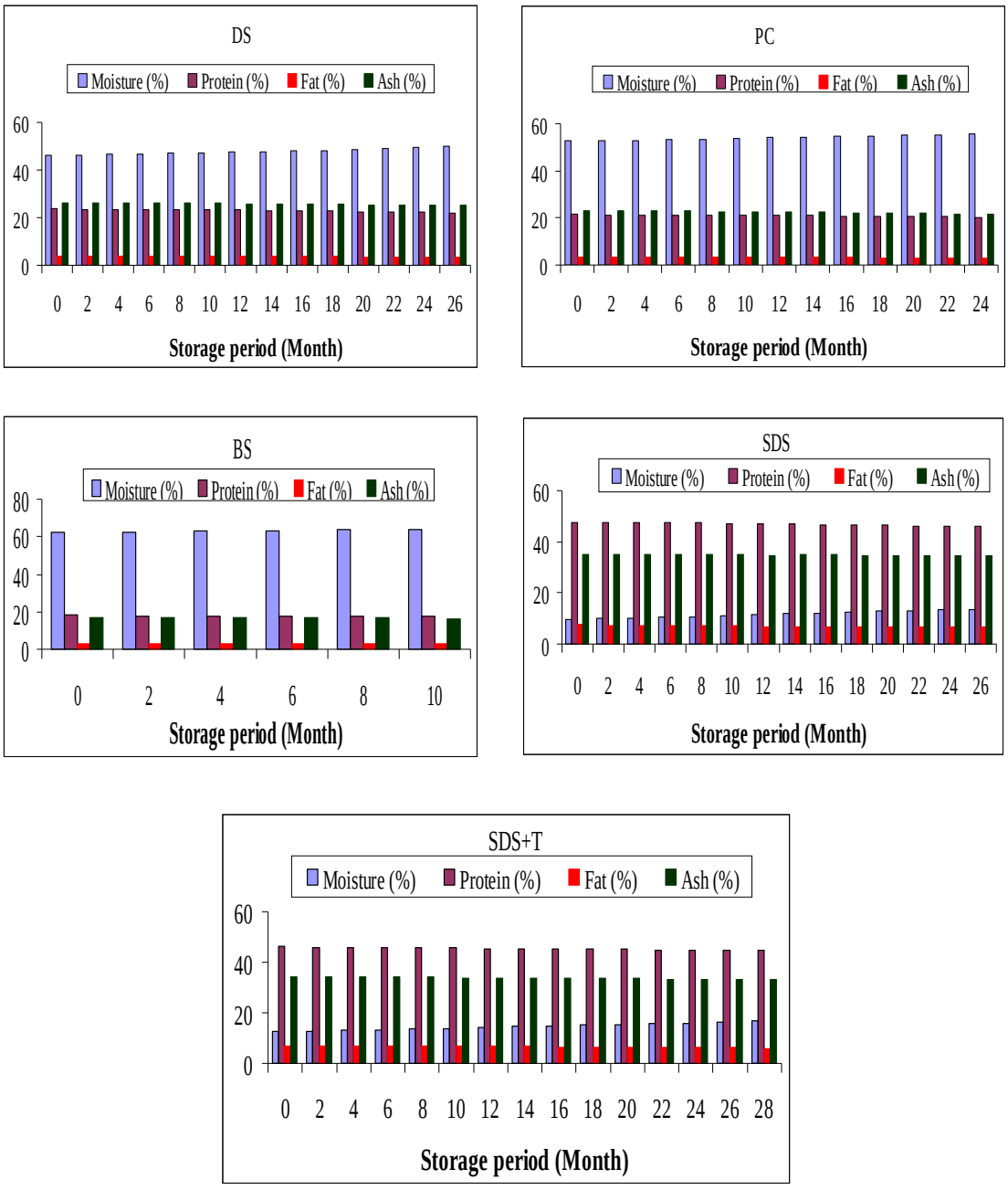


Figure. 4.4.2.1(D). The changing pattern in proximate composition of salted taki (*C. punctatus*) during different observation, storage at refrigeration temperature (4^0C) in different treatments (DS= Dry salted, PC= Pickle cured, BS= Brine salted, SDS= Sun-dried salted, SDS+T= Turmeric treated sun-dried salted)

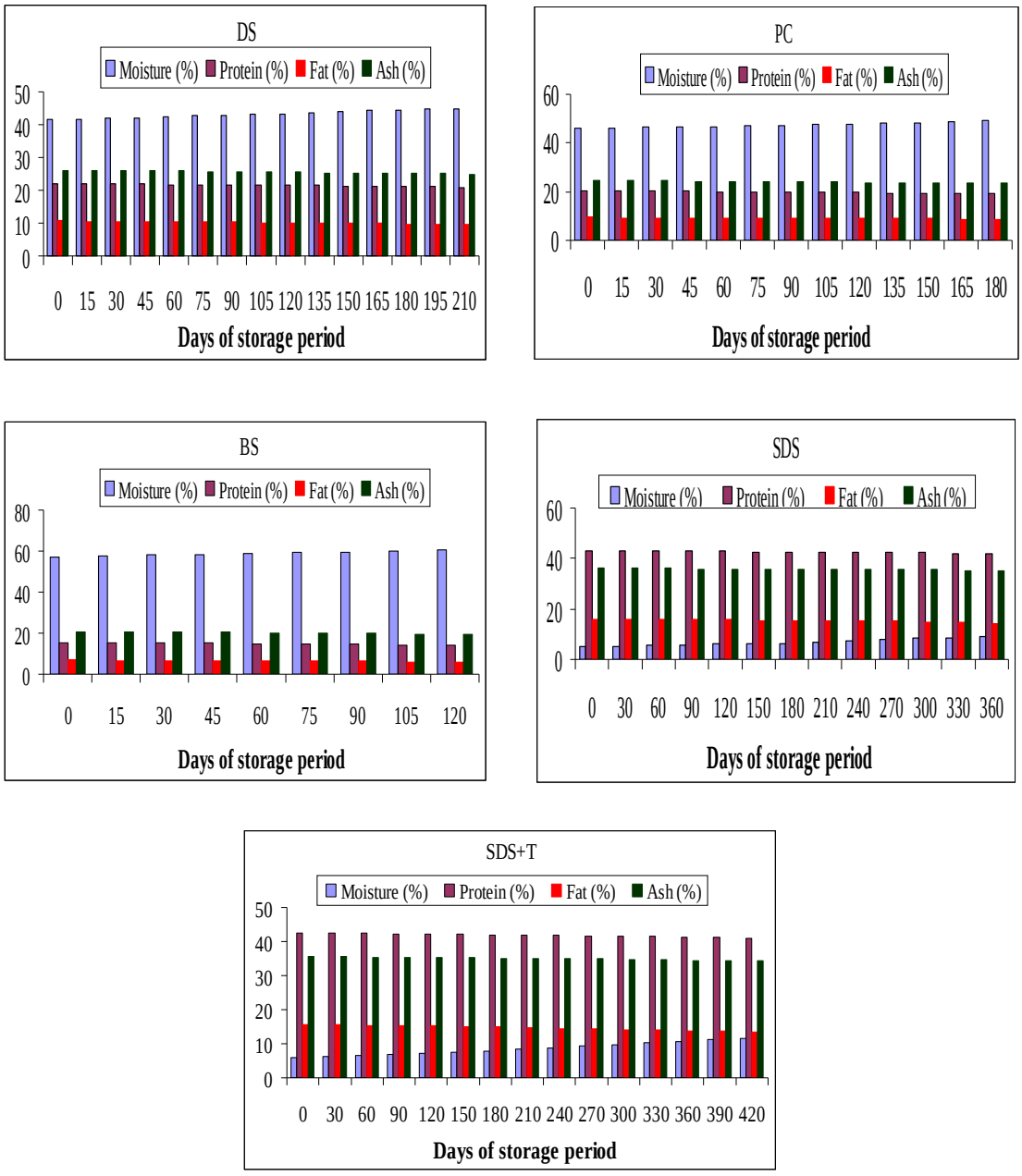


Figure. 4.4.2.1(E). The changing pattern in proximate composition of salted tengra (*M. tengra*) during different days of observation, storage at room temperature (26-32^{0}C) in different treatments (DS= Dry salted, PC= Pickle cured, BS= Brine salted, SDS= Sun-dried salted, SDS+T= Turmeric treated sun-dried salted)

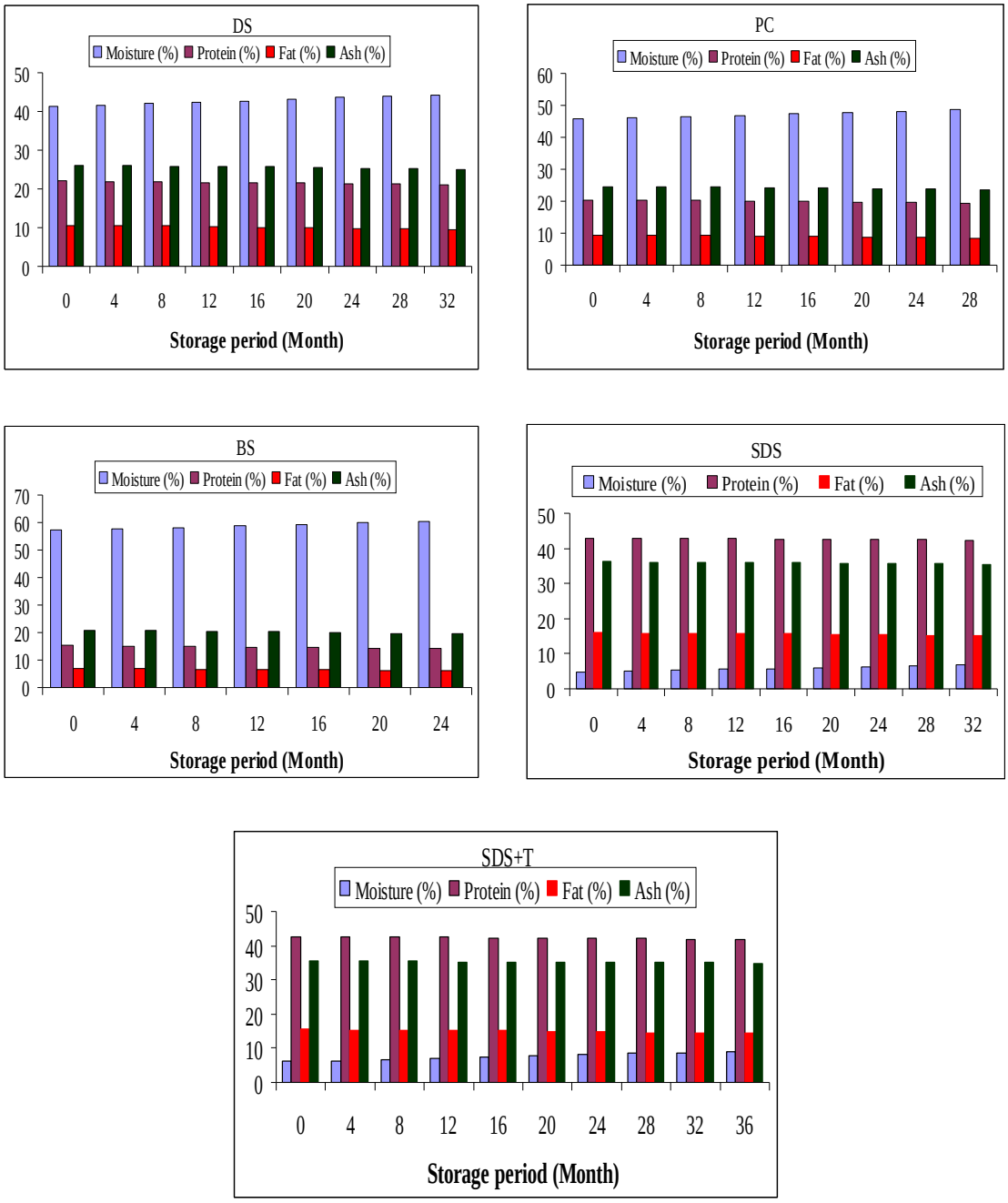


Figure. 4.4.2.1(F). The changing pattern in proximate composition of salted tengra (*M. tengra*) during different observation, storage at refrigeration temperature (4^0C) in different treatments (DS= Dry salted, PC= Pickle cured, BS= Brine salted, SDS= Sun-dried salted, SDS+T= Turmeric treated sun-dried salted)

4.4.2.2. Changes in moisture content

In case of shol fish, during storage at room temperature moisture content was varied in the range of 48.84% to 53.69% (165 days), 50.29% to 54.94% (150 days), 60.17% to 65.97% (135 days), 29.77% to 35.26% (165 days) and 30.92% to 36.98% (180 days) in dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) fish-products [Table-4.4.2.1(A)].

On the other hand, refrigeration stored dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) shol fish had moisture content varying in the range of 48.84% to 51.24% (26 months), 50.29% to 52.85% (22 months), 60.17% to 63.48% (18 months), 29.77% to 34.09% (24 months), and 30.92% to 35.48% (28 months) respectively [Table 4.4.2.1(B)].

In case of taki fish, during storage at room temperature moisture content was varied in the range of 46.21% to 50.92% (150 days), 52.71% to 56.55% (135 days), 62.28% to 65.08% (75 days), 9.77% to 14.97% (240 days) and 12.72% to 18.88% (300 days) in dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) fish-products [Table 4.4.2.1(C)].

In case of taki fish, during storage at refrigerated temperature moisture content was varied in the range of 46.21% to 49.89% (26 months), 52.71% to 55.71% (24 months), 62.28% to 64.02% (10 months), 9.77% to 13.40% (26 months) and 12.72% to 17.02% (28 months) in dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) fish-products [Table 4.4.2.1(D)].

It was observed from Table 4.4.2.1(E) that, dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) tengra fish-products had moisture content varying in the range of 41.41% to 44.82% (210 days), 45.88% to 49.09% (180 days), 57.35% to 60.70% (120 days), 4.90% to

9.05% (360 days) and 6.08% to 11.62% (420 days) during storage at room temperature.

It was also shown from Table 4.4.2.1(F) that, in case of tengra fish, during storage at refrigerated temperature moisture content was varied in the range of 41.41% to 44.29% (32 months), 45.88% to 48.79% (28 months), 57.35% to 60.57% (24 months), 4.90% to 6.74% (32 months), and 6.08% to 9.03% (36 months), in case of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) fish-products.

The moisture content is an exact indicator of the susceptibility of a product to undergo microbial spoilage (Troller and Christian, 1975). It has a potential effect on the chemical reaction rate and microbial growth rate of the food product (Labuze, 1970). Moisture also affects the stability and shelf life of the food product. Khan *et al.* (1997) reported that moisture varies because of species variation, size, processing method, temperature, season etc.

According to Poulter *et al*. (1982), a good quality salted and salted-dried fish with moisture content of below 20% could have a predicted mould-free shelf life between 100 and 450 days. This is in agreement with present research where moisture content bellow 20% has been found in sun-dried salted and turmeric treated sun-dried salted taki and tengra fish-products and their shelf life varied 240-420 days.

The moisture content also increased as storage progressed in refrigeration temperature, which agreed with earlier work of Indira *et al.* (2010) whose finding showed an increase in moisture content from an initial value of 79.35 to 82.60% for 3 weeks of storage at refrigeration temperature of C. *capio*.

The comparison of changes in moisture content of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) shol, taki and tengra fish samples stored both room and refrigeration temperature is presented in Figure 4.4.2.2.

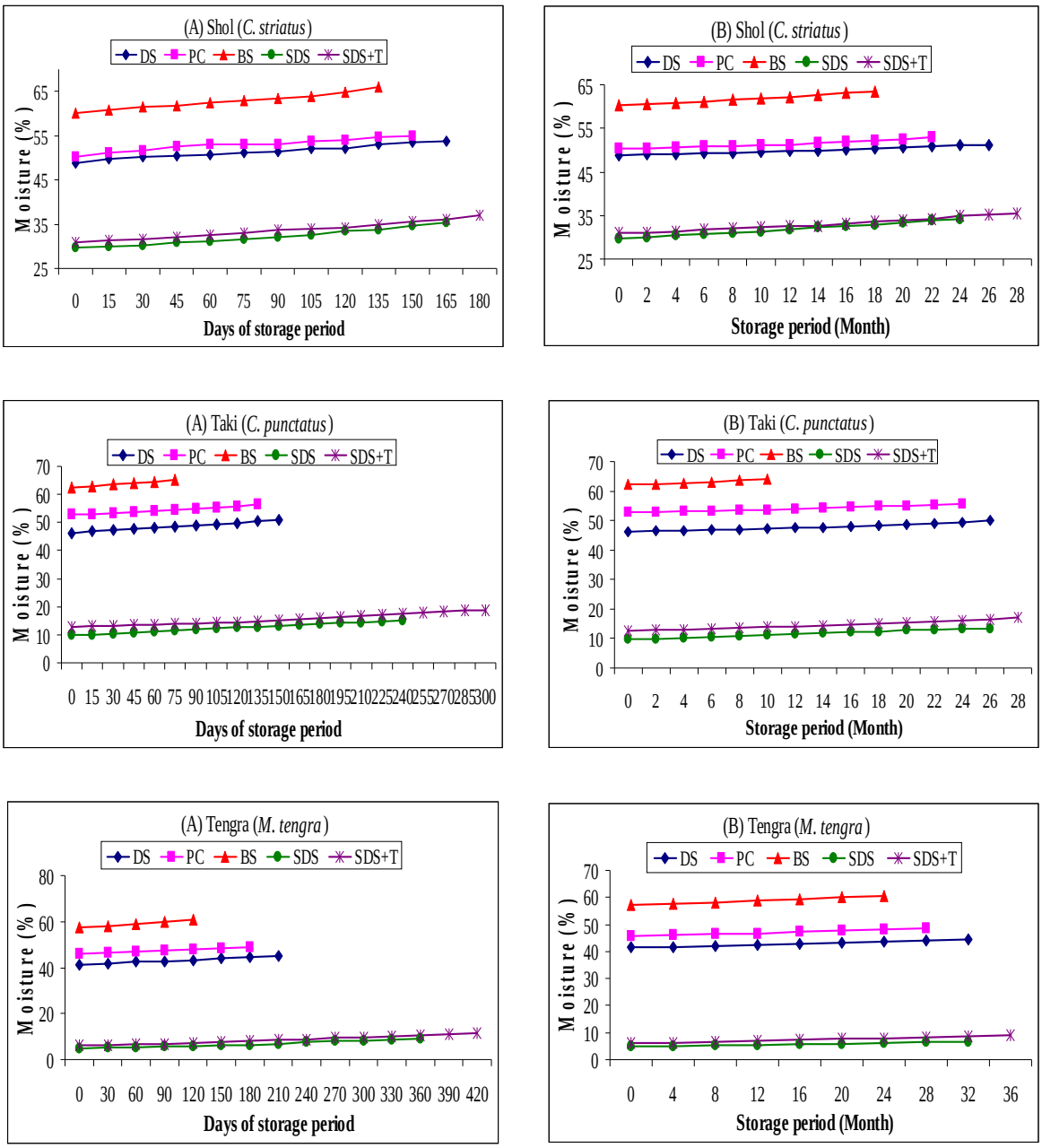


Figure. 4.4.2.2. Effects of different types of salting process on moisture content of DS, PC, BS, SDS, SDS+T treated shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish during different duration of storage both at (A) room temperature and (B) refrigeration temperature (DS= Dry salted, PC= Pickle cured, BS= Brine salted, SDS=Sun-dried salted and SDS+T= turmeric treated sun-dried salted)

From the figure it is shown that, the moisture content of brine salted (BS) shol, taki and tengra was highest than other samples whereas, sun-dried salted (SDS) shol, taki and tengra was lowest moisture content. Generally, the relative lower percentage moisture of the sun-dried salted (SDS) products could be as a result of the combined action effects of the salt and heat of sun, which enhanced greater dehydration.

During the storage period, moisture content increased both room and refrigerated temperature, but the increment in room temperature was more prominent than the product stored in refrigerated temperature. The phenomenon of increasing moisture content in room temperature is due to absorption of moisture from surrounding atmosphere. Increase in moisture content could be attributed to the difference in the moisture of the processed fish relative to the surroundings (Daramola *et al.,* 2007).

The polythene package used for the salted fish had low moisture vapor permeability and resistant to absorption of water which appeared to prevent migration of moisture from the product to the inside surface of the package as reported by Slavin (1963).

However, rapid increases in moisture content were found in 5 types of salted shol fish during storage at room temperature. This may be due to the fact that there was continuous raining during that period and the relative humidity was considerably high (about 90-100%, as recorded from weather forecast). The hygroscopic properties of salt may be influenced by the high relative humidity to show increased moisture content.

Ndakatu *et al.* (2011) stated that, the increase in the moisture content of all salted and salted-dried products could be as a result of moisture reabsorption during entire storage period. Similarly, Siddique and Akter (2011) observed percentage of moisture content ranged 13.81-20.50%, 22.22-34.99% and 20.76-32.65% in dried marine fishes (*Lepturacanthus savala, Harpodon nehereus* and *Johnius dussumieri*) stored at room temperature for 2 year which is similar with present investigation. Bhuiyan (1992) observed 6.9% - 14.2% moisture in dried marine fishes which is similar with the findings of SDS and SDS+T taki and tengra fish products.

4.4.2.3. Changes in protein content

In present study, the changes of protein content of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) **shol** fish products during storage at room temperature were 28.21 to 25.52%, 27.64 to 24.82%, 20.72 to 17.06%, 41.48 to 38.62% and 41.0 to 38.01% whereas at refrigeration storage, protein content of these fish-products varied from 28.21 to 27.25%, 27.64 to 26.34%, 20.72 to 18.50%, 41.48 to 39.30% and 41.0 to 38.81% respectively [Table 4.4.2.1(A) & Table 4.4.2.1(B)].

It is also shown in Table 4.4.2.1(C) & Table 4.4.2.1(D) that, dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) **taki** fish products had percentage of protein ranged from 23.58 to 21.53%, 21.39 to 20.06%, 18.02 to 16.92%, 47.69 to 45.68% and 46.06 to 43.92% during storage at room temperature and 23.58 to 21.94%, 21.39 to 20.36%, 18.02 to 17.45%, 47.69 to 45.91% and 46.06 to 44.53% respectively during storage at refrigeration temperature.

In case of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) **tengra** fish products, percentage of protein ranged from 22.05 to 20.99%, 20.43 to 19.28%, 15.30 to 14.28%, 43.00 to 42.00% and 42.60 to 41.03% during storage at room temperature and 22.05 to 21.10%, 20.43 to 19.41%, 15.30 to 14.10%, 43.00 to 42.40% and 42.60 to 41.86% respectively during storage at refrigeration temperature. [Table 4.4.2.1(E) & Table 4.4.2.1(F)]

The comparison of changes in protein content of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) shol, taki and tengra fish samples stored both room and refrigeration temperature is presented in Figure 4.4.2.3.

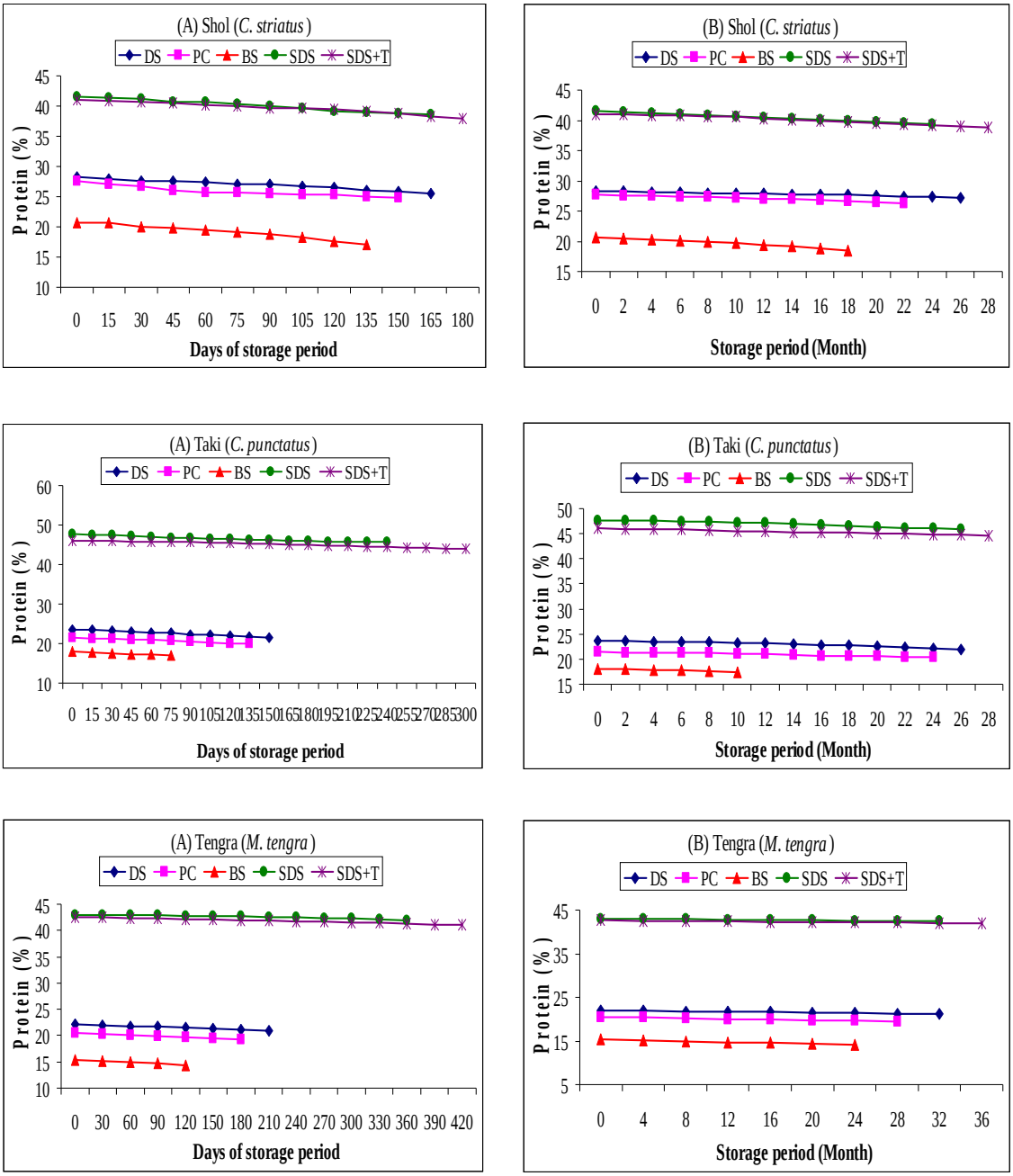


Figure. 4.4.2.3. Effects of different types of salting process on protein content of DS, PC, BS, SDS, SDS+T treated shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish during different duration of storage both at (A) room temperature and (B) refrigeration temperature (DS= Dry salted, PC= Pickle cured, BS= Brine salted, SDS=Sun-dried salted and SDS+T= turmeric treated sun-dried salted)

These above results are in accordance with the findings of Ghezala (1994); Abolagba and Osifo (2004); Fapohunda & Ogunkoya (2006) and Abolagba and Melle (2008) who reported that protein decompose with passing time.

From these result it is evident that the protein content of all salted fish-products has decreased during storage period. Loss of protein during storage period is extremely variable and it might be due to loss of low molecular weight nitrogenous substances along with the fluids diffused from fish body surface to outside salt particle.

It is also shown from this figure that, brine salted (BS) shol, taki and tengra fish-products has lower percentage of protein in comparison of other salted (DS, PC, SDS, SDS+T) fish-products, as a result of protein being dissolved in the brine and other nitrogenous substance including free amino acids (Clucas and Ward, 1996). On the other hand, highest percentage of protein was found in sun- dry salted (SDS) shol, taki and tengra fish-products.

According to Faturoti (1985) and Eyo (2001), reduction in the percentage crude protein of fish-species during the period of storage could be due to gradual degradation of the initial crude protein to more volatile products such as Total Volatile Bases (TVB), Hydrogen sulphide and Ammonia. Changes observed in protein content during storage may also have been due to leaching out of some extractable soluble protein fraction (Holma and Maalekuu, 2013).

Hassan *et al.* (2013) observed that, in storage condition the protein content decreased significantly with the time due to water soluble protein diffused out to the surrounding for exosmosis. However, Siddique and Aktar (2011) observed that protein level of three marine dried fishes (*Harpodon nehereus, Johnius dussumieri* and *Lepturacanthus savala*) were varied from 58.33% - 51.98%, 64.39% - 56.46% and 71.90% - 67.22% respectively during 2 years storage period which is similar with the present study.

In another study, Aktar (2009) observed that, percentage of protein decreased from 58.51% to 57.73% and 59.51% to 58.28% in case of 8 months refrigeration-stored sun-dried salted and turmeric treated sun-dried salted *M. vittatus* fish-product.

Eltom (1989) stated that, generally the quantity of protein loss depends on the exact nature and duration of the salting process and the conditions of fish when salted.

Abo-Taleb (1993) concluded that the decrease in protein content during storage might be due to protein hydrolysis by enzymes, which enhanced the loss of water soluble nitrogen with the separated drip. Protein decreased with storage of cured meat was attributed to some changes during storage that caused by 'maillard reaction and changes in pH (Lawrie, 1998).

According to Hamm (1994), salt causes the proteins in fish muscle to swell and salt lead the protein become denatured if increases in the muscle.

Decrease in protein during also at refrigeration storage is in agreement with the findings of Bhat *et al.* (2010) and Arekemase *et al.* (2012).

The above results indicate that sun-dried salted fish-products are best for consumption, from nutritional point of view.

4.4.2.4. Relationship between moisture & protein

From present study it is clear that, with the increase of storage period, there was an increase in moisture content and decrease in protein content in all salted fish samples. So, there exists an inverse relationship between the moisture content and protein content in the salted fish samples which was similar with the findings of Arekemase *et al*. (2012).

Begum *et al.* (2011) opeined that, the moisture content of the fish product gradually increased as the protein content of the products decreased.

According to Akter (2009), moisture content of salted-dried and salt and turmeric treated sun-dried tengra (*M. vitatus*) and kachki (*C. soborna*) gradually increased whereas protein content gradually decreased through entire period of storage both room and refrigerator temperature which is in harmony with present findings.

The relationship between moisture & protein contents of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) shol, taki and tengra fish samples stored both room and refrigeration temperature is presented in Figure 4.4.2.4(A)-(F).

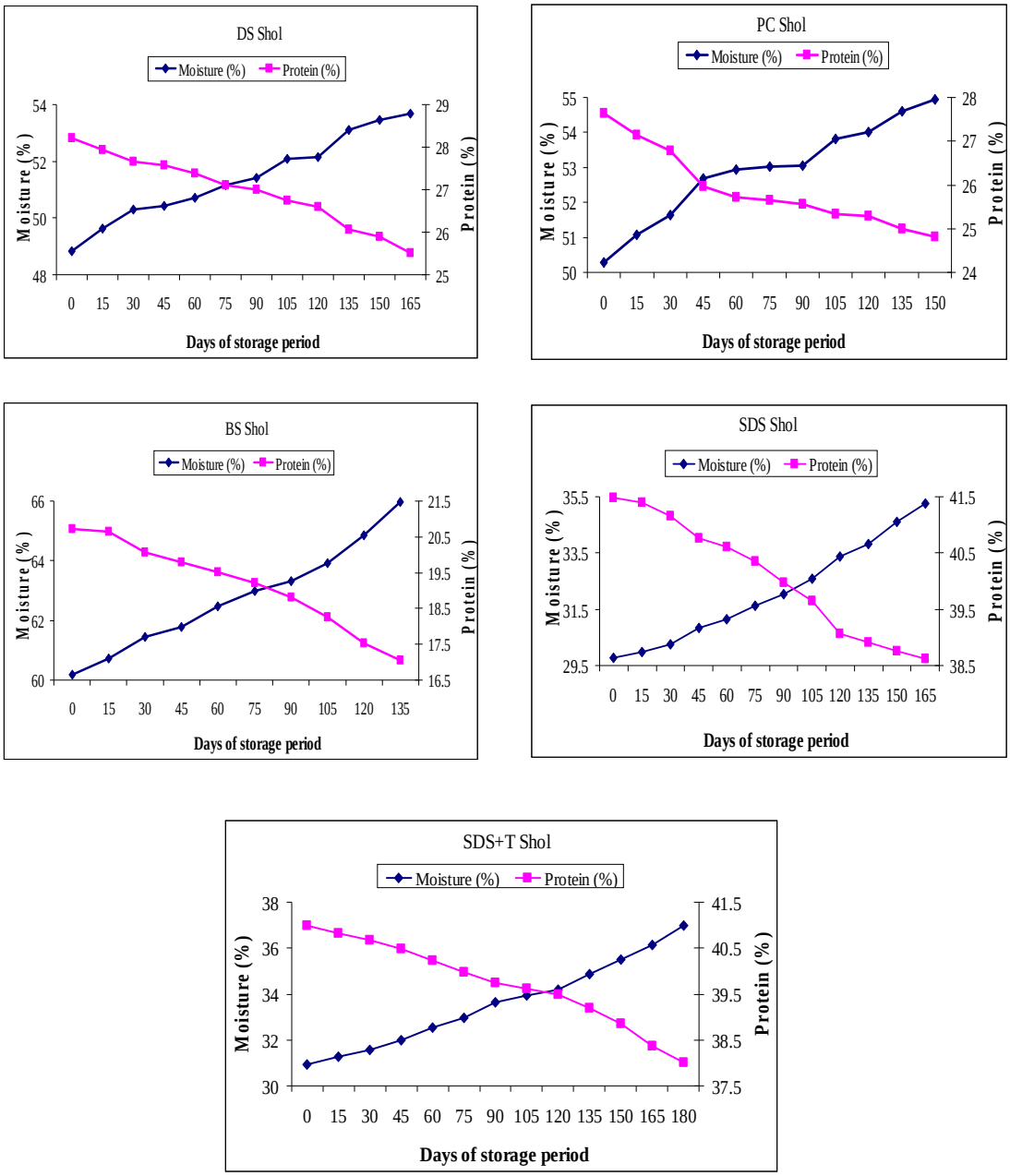


Figure. 4.4.2.4(A). Changing relationship between moisture & protein contents in different treatments (DS, PC, BS, SDS, SDS+T) of shol (*C. striatus*) fish during different duration of storage at room temperature. [DS= Dry salted, PC= Pickle cured, BS= Brine salted, SDS=Sun-dried salted and SDS+T= turmeric treated sun-dried salted]

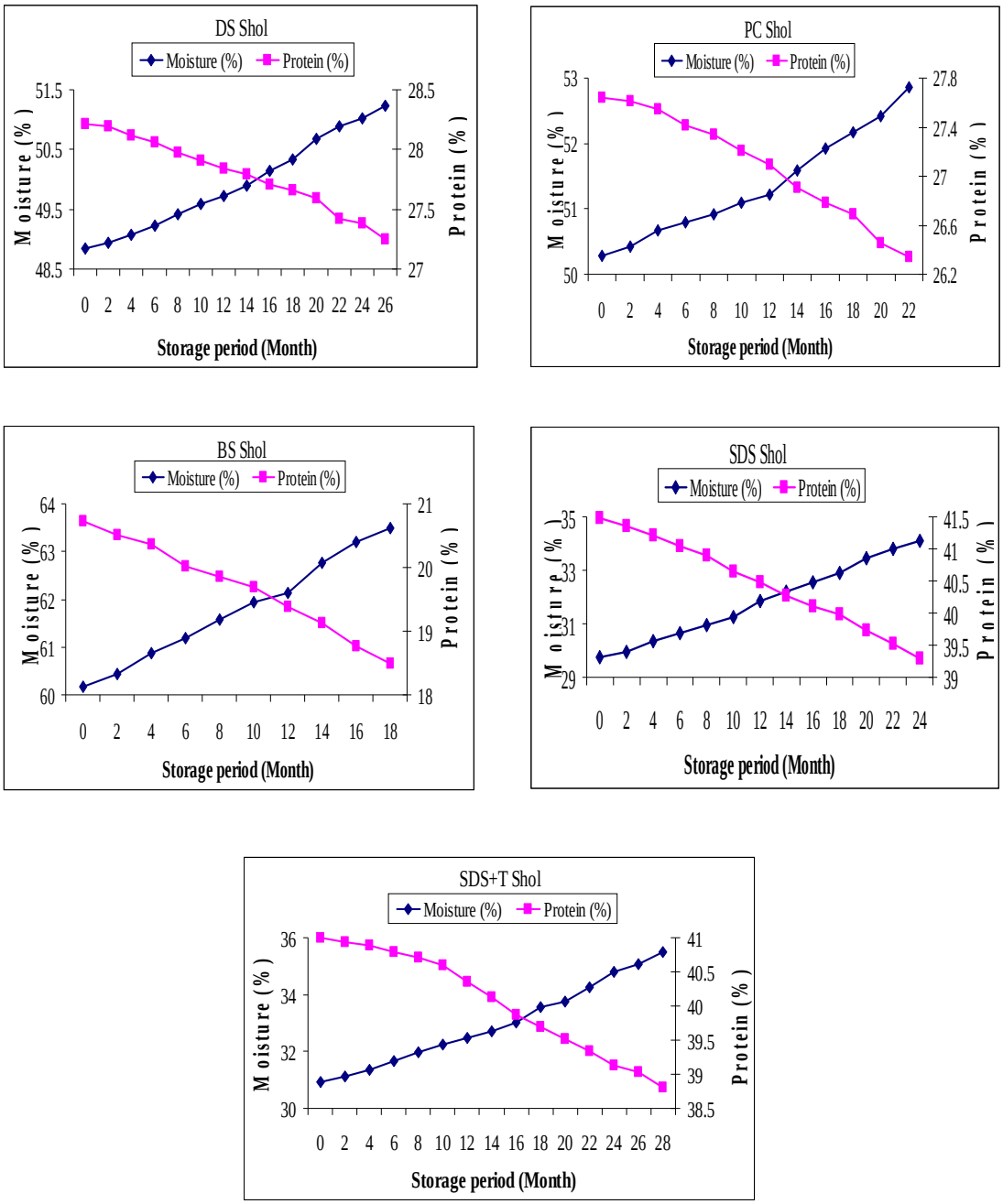


Figure. 4.4.2.4(B). Changing relationship between moisture & protein contents in different treatments (DS, PC, BS, SDS, SDS+T) of shol (*C. striatus*) fish during different duration of storage at refrigeration temperature [DS = Dry salted, PC = Pickle cured, BS= Brine salted, SDS=Sun-dried salted and SDS+T= turmeric treated sun-dried salted]

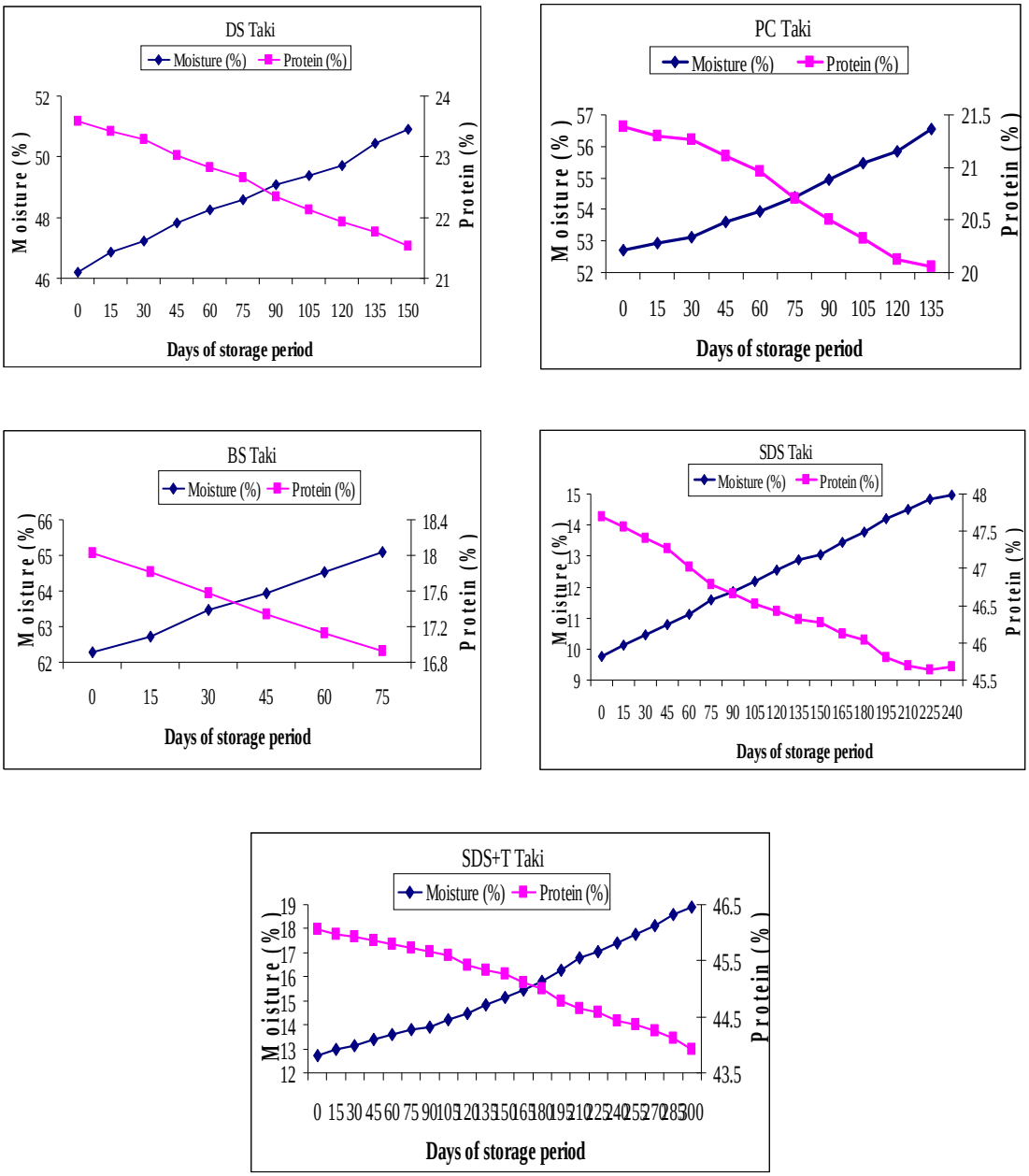


Figure. 4.4.2.4(C). Changing relationship between moisture & protein contents in different treatments (DS, PC, BS, SDS, SDS+T) of taki (*C. punctatus*) fish during different duration of storage at room temperature [DS = Dry salted, PC = Pickle cured, BS= Brine salted, SDS=Sun-dried salted and SDS+T= turmeric treated sun-dried salted]

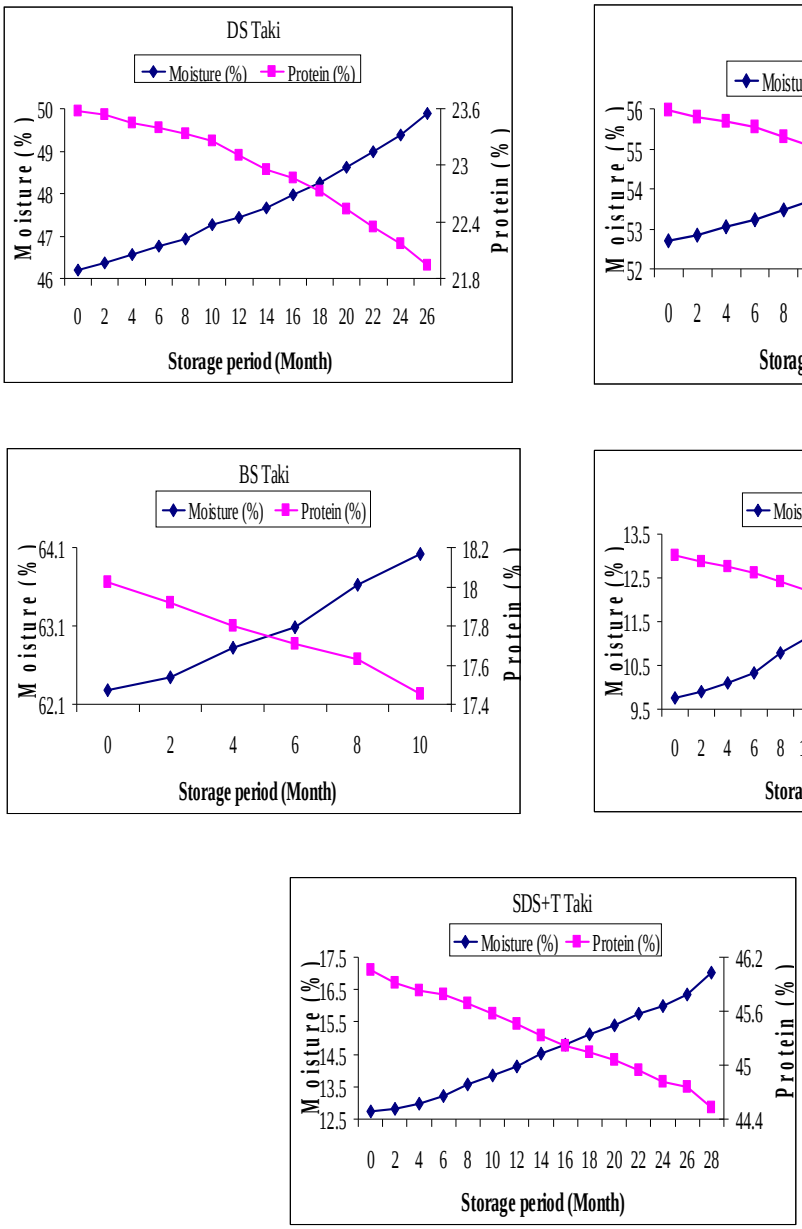


Figure. 4.4.2.4(D). Changing relationship between moisture & protein contents in different treatments (DS, PC, BS, SDS, SDS+T) of taki (*C. punctatus*) fish during different duration of storage at refrigeration temperature [DS = Dry salted, PC = Pickle cured, BS= Brine salted, SDS=Sun-dried salted and SDS+T= turmeric treated sun-dried salted]

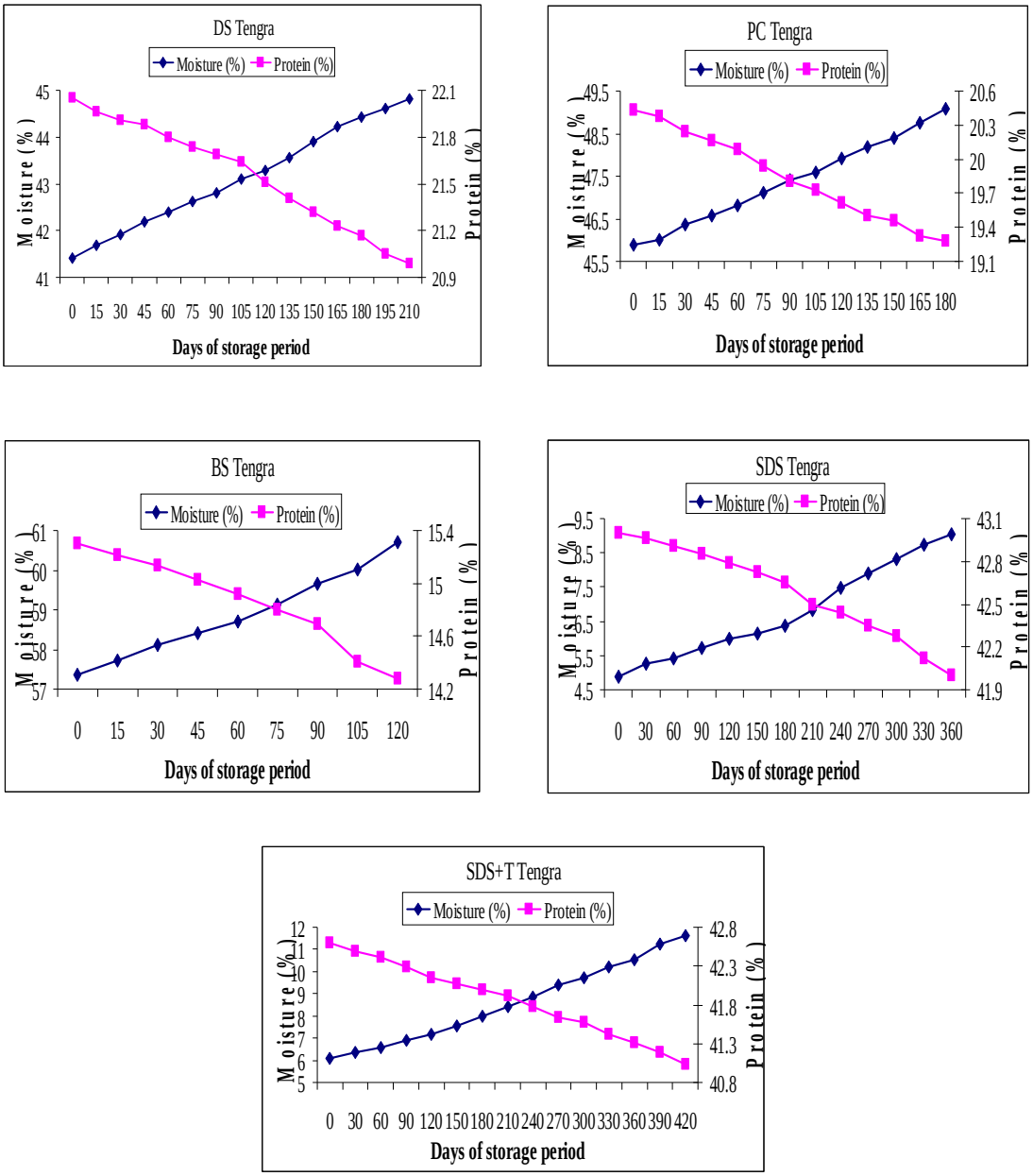
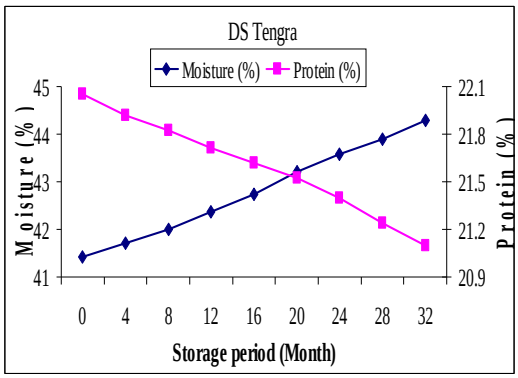
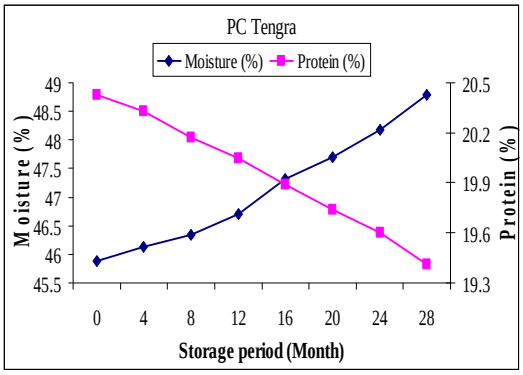
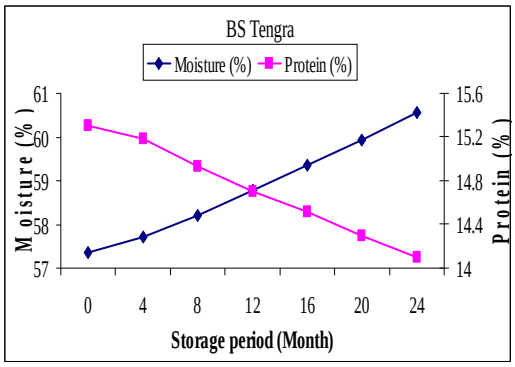
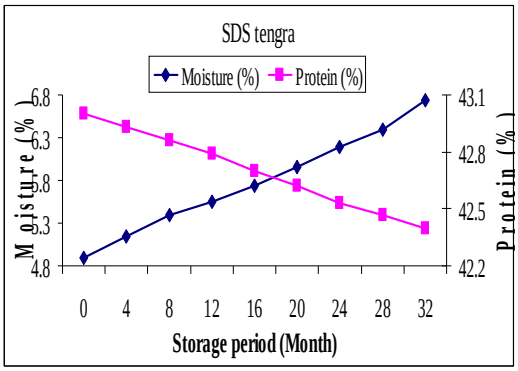


Figure. 4.4.2.4(E). Changing relationship between moisture & protein contents in different treatments (DS, PC, BS, SDS, SDS+T) of tengra (*M. tengra*) fish during different duration of storage at room temperature [DS = Dry salted, PC = Pickle cured, BS= Brine salted, SDS=Sun-dried salted and SDS+T= turmeric treated sun-dried salted]









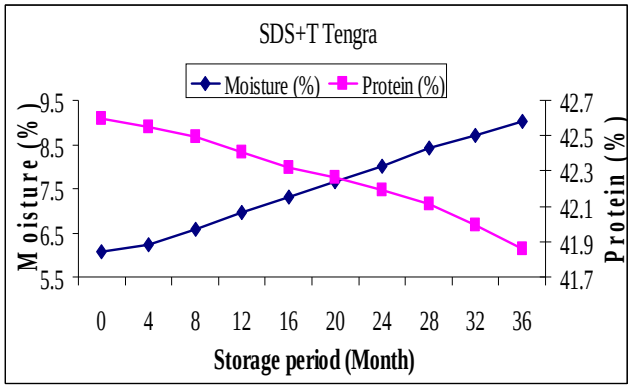


Figure. 4.4.2.4(F). Changing relationship between moisture & protein contents in different treatments (DS, PC, BS, SDS, SDS+T) of tengra (*M. tengra*) fish during different duration of storage at refrigeration temperature [DS = Dry salted, PC = Pickle cured, BS= Brine salted, SDS=Sun-dried salted and SDS+T= turmeric treated sun-dried salted]

4.4.2.5. Changes in fat content

In present study, the range of fat content of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) **shol** fish products during storage at room temperature were 3.99 to 3.30%, 3.79 to 3.11%, 3.20 to 2.72%, 5.10 to 3.81% and 4.79 to 3.26% whereas at refrigeration storage, fat content of these fish-products varied from 3.99 to 3.46%, 3.79 to 3.22%, 3.20 to 2.89%, 5.10 to 4.01% and 4.79 to 3.69% respectively [Table 4.4.2.1(A) & Table 4.4.2.1(B)].

It is also shown in Table 4.4.2.1(C) & Table 4.4.2.1(D) that, dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) **taki** fish products had percentage of fat content ranged from 3.93 to 3.49%, 3.40 to 2.93%, 2.76 to 2.48%, 7.47 to 5.81% and 7.09 to 5.01% during storage at room temperature and 3.93 to 3.44%, 3.40 to 2.92%, 2.76 to 2.59%, 7.47 to 6.48% and 7.09 to 6.00% respectively during storage at refrigeration temperature.

In case of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) **tengra** fish products, percentage of fat content ranged from 10.65 to 9.59%, 9.40 to 8.68%, 6.84 to 6.01%, 15.99 to 14.41% and 15.58 to 13.40% during storage at room temperature and 10.65 to 9.52%, 9.40 to 8.41%, 6.84 to 6.09%, 15.99 to 15.09% and 15.58 to 14.38% respectively during storage at refrigeration temperature [Table 4.4.2.1(E) & Table 4.4.2.1(F)].

The comparison of changes in fat content of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) shol, taki and tengra fish samples stored both room and refrigeration temperature is represented in Figure 4.4.2.5.

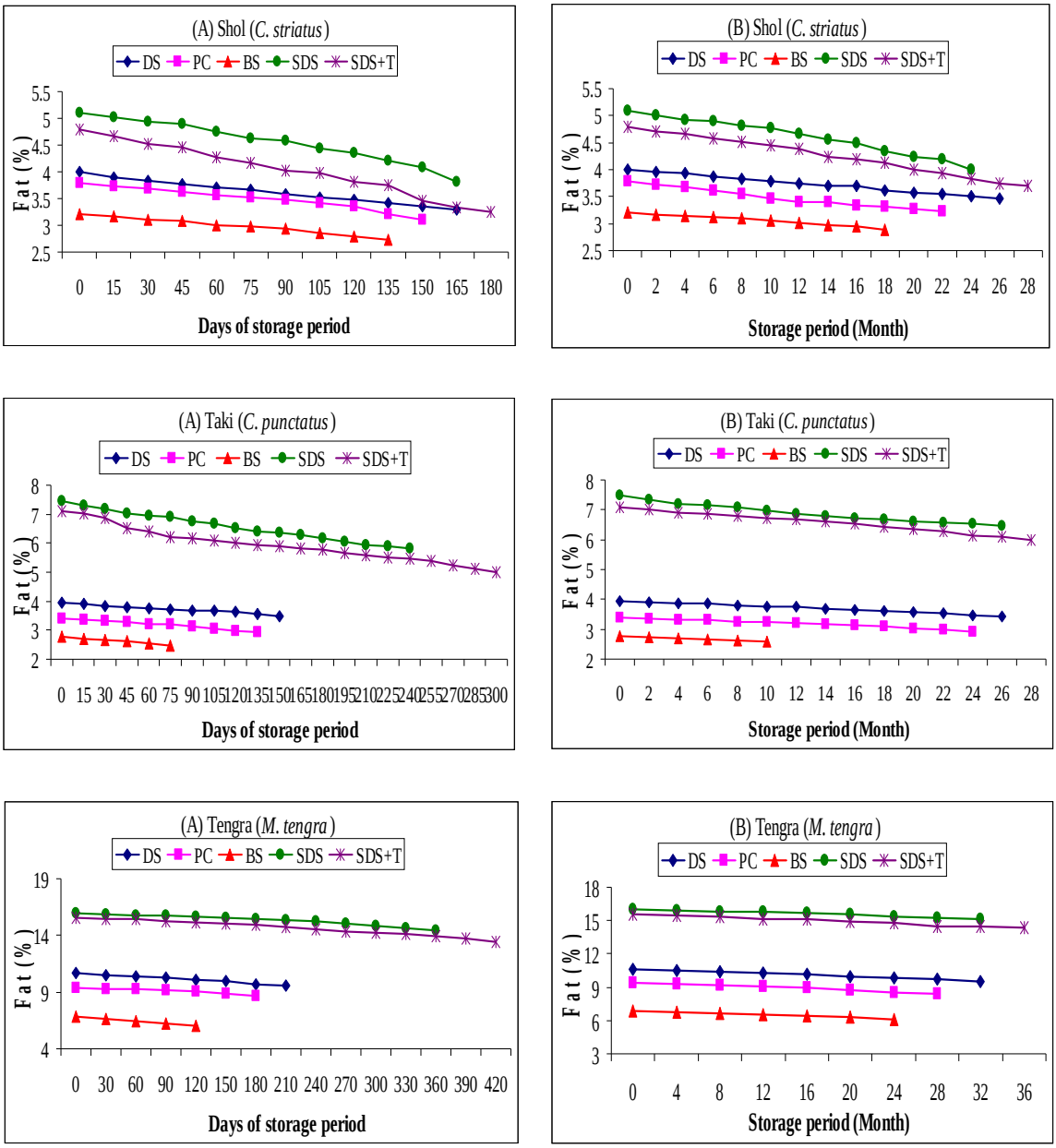


Figure. 4.4.2.5. Effects of different types of salting process on fat content of DS, PC, BS, SDS, SDS+T treated shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish during different duration of storage both at (A) room temperature and (B) refrigeration temperature (DS= Dry salted, PC= Pickle cured, BS= Brine salted, SDS=Sun-dried salted and SDS+T= turmeric treated sun-dried salted)

This figure indicates that, the fat content of 5 types of salted shol, taki and tengra fish-products decreases with the increase of storage period and the final fat content was comparatively highest in sun dried salted (SDS) shol, taki and tengra fish products both room and refrigeration temperature. Lowest amount of lipid content found in the brine salted (BS) fish-products which might be due to oxidation of fat associated with diffusion from inside the fish body along with fluids to the outer surface. The total lipid content in the figures showed a progressive decrease in the entire salted and sun-dried salted sample during storage period and it was probably due to oxidative deterioration, there-by affecting lipid extraction which is in agreement with Gandotra *et al.* (2012). Indira *et al.* (2010) observed, decrease in fat content of *C. caprio* from 3.13% to 2.32% during 21 days of refrigeration storage.

According to Obemeata *et al.* (2011), the high degree of unsaturation in the form of multiple double bonds in fatty acids renders fish highly susceptible to oxidative rancidity. Devadasan and Nair (1977) also observed lipid breakdown in oil sardine stored at refrigeration temperature. Arannilewa *et al.* (2005), Saliu (2008) and Egbal *et al*. (2010b) indicated that reduction in fat is associated with the oxidation of fat.

Decrease in the level of fat contents of small and large salted Bouri fish muscle (*Mugil cephalus*) was reported by El-Sebahy and Metwali (1988). Likewise, Rahman (1996) reported that lipid content of dry salted hilsa was 26.93% on '0' day and decreased in 15.67% on 42^{nd} day of storage respectively. Similarly, Sayed (1997) reported that during 18 days of observation of hilsa salting, lipid content decreased from 24.87% to 15.46% and 16.15% to 12.49% respectively.

Siddique and Aktar (2011) observed that the mean percentages of lipid of three types of marine dry fishes (*Harpodon nehereus*, *Johnius dussumieri* and *Lepturacanthus savala*) samples were decreased from 7.78% - 5.86%, 5.54% - 4.87% and 7.79% - 6.66% respectively during changes of 2 years of storage period.

The above findings are in close quarters with the present investigation.

Lovern (1950) reported that the variation in lipid content was influenced by the variation of species, diet, temperature, salinity, selective mobilization and distribution.

According to Horner (1992), reduction in lipid content could be attributed to oxidation of poly-unsaturated fatty acids (PUFA) contained in the fish tissue to products such as peroxides, aldehydes, ketones and the free fatty acids. However, the rate of fat deterioration was very gradual. Fish oil has been found to be more liable to spoilage than other oils due to their greater number of unsaturated fatty acids. The greater the degree of unsaturation, the greater would be the tendency for fat oxidation (rancidity) (Eyo, 1993).

Lipid deterioration limits shelf life of fish (Aubourg and Medina, 1999). Lipid oxidation causes loss of flavor and nutrition and creates toughening and texture problems (Aubourg and Medina, 1999). The result is unpleasant odor and flavor called rancidity. The oxidation of lipids plays a very important role in the spoilage of both lean and fatty fishes.

It is a generally agreed phenomenon that fat content decreases in fish product during the increase of storage time and lose quality. According to Pegg (1999), the oxidative stability of an oil or fat depends on the degree and nature of the unsaturation of its triglycerides, its antioxidants content, the presence of prooxidants and storage conditions such as temperature.

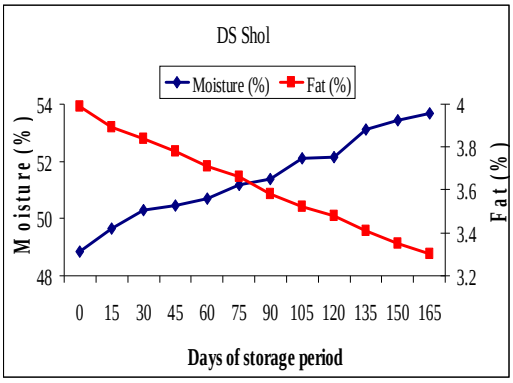
4.4.2.6. Relationship between moisture & fat

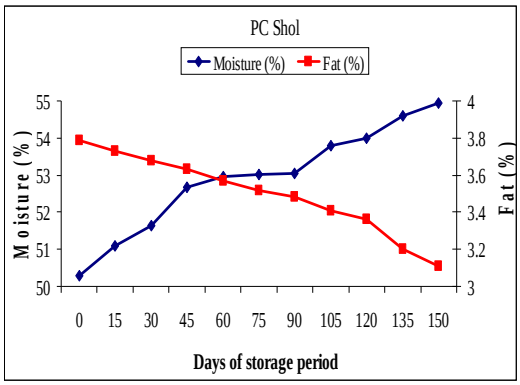
The percentage of oil (fat) and water (moisture) varied inversely with one another and this is more or less in agreement with the general rule established (Stansby, 1962).

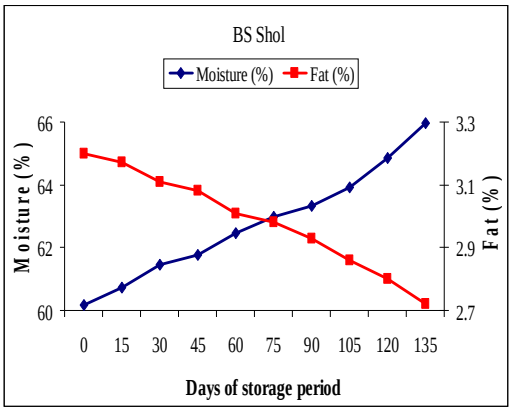
Nagakura and Sachithananthan (1971) reported that the crude fat showed an inverse relationship with moisture content during salting.

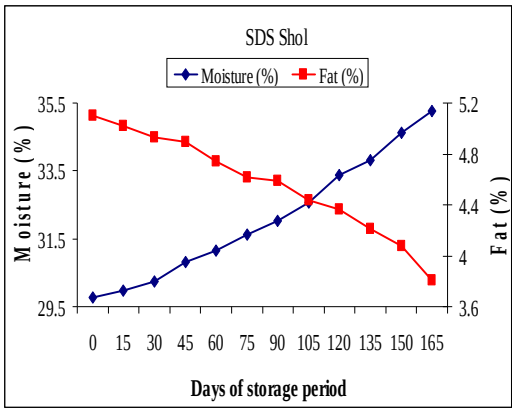
Present results showed that the moisture and lipid contents in all five types of salted shol, taki and tengra fish-products are inversely related, as recorded for other fish species (Chowdhury, 1981; FAO, 1999).

The inverse relationship between moisture & fat contents of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) shol, taki and tengra fish samples stored both room and refrigeration temperature is presented in Figure 4.4.2.6 (A)-(F).









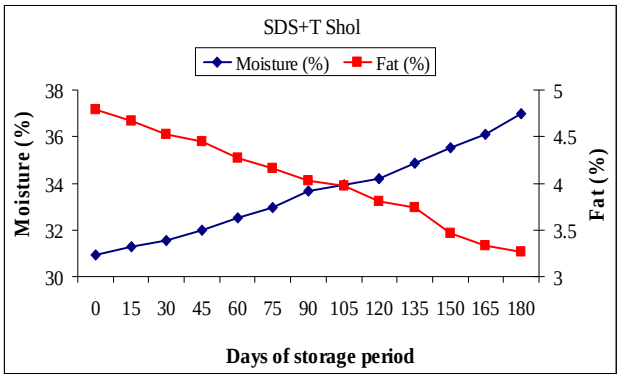


Figure. 4.4.2.6(A). Changing relationship between moisture & fat contents in different treatments (DS, PC, BS, SDS, SDS+T) of shol (*C. striatus*) fish during different duration of storage at room temperature [DS = Dry salted, PC = Pickle cured, BS= Brine salted, SDS=Sun-dried salted and SDS+T= turmeric treated sun-dried salted]

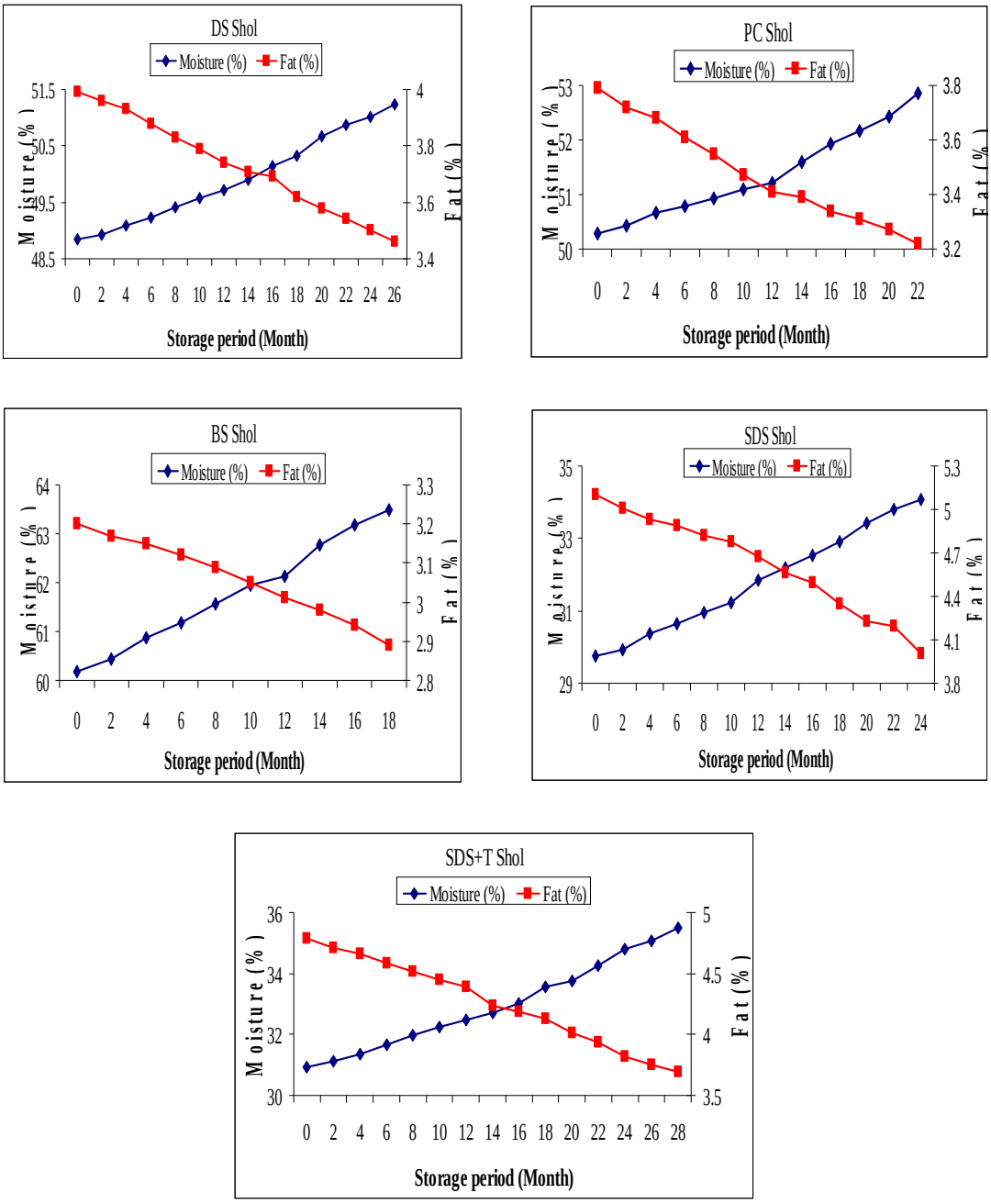


Figure. 4.4.2.6(B). Changing relationship between moisture & fat contents in different treatments (DS, PC, BS, SDS, SDS+T) of shol (*C. striatus*) fish during different duration of storage at refrigeration temperature [DS = Dry salted, PC = Pickle cured, BS= Brine salted, SDS=Sun-dried salted and SDS+T= turmeric treated sun-dried salted]

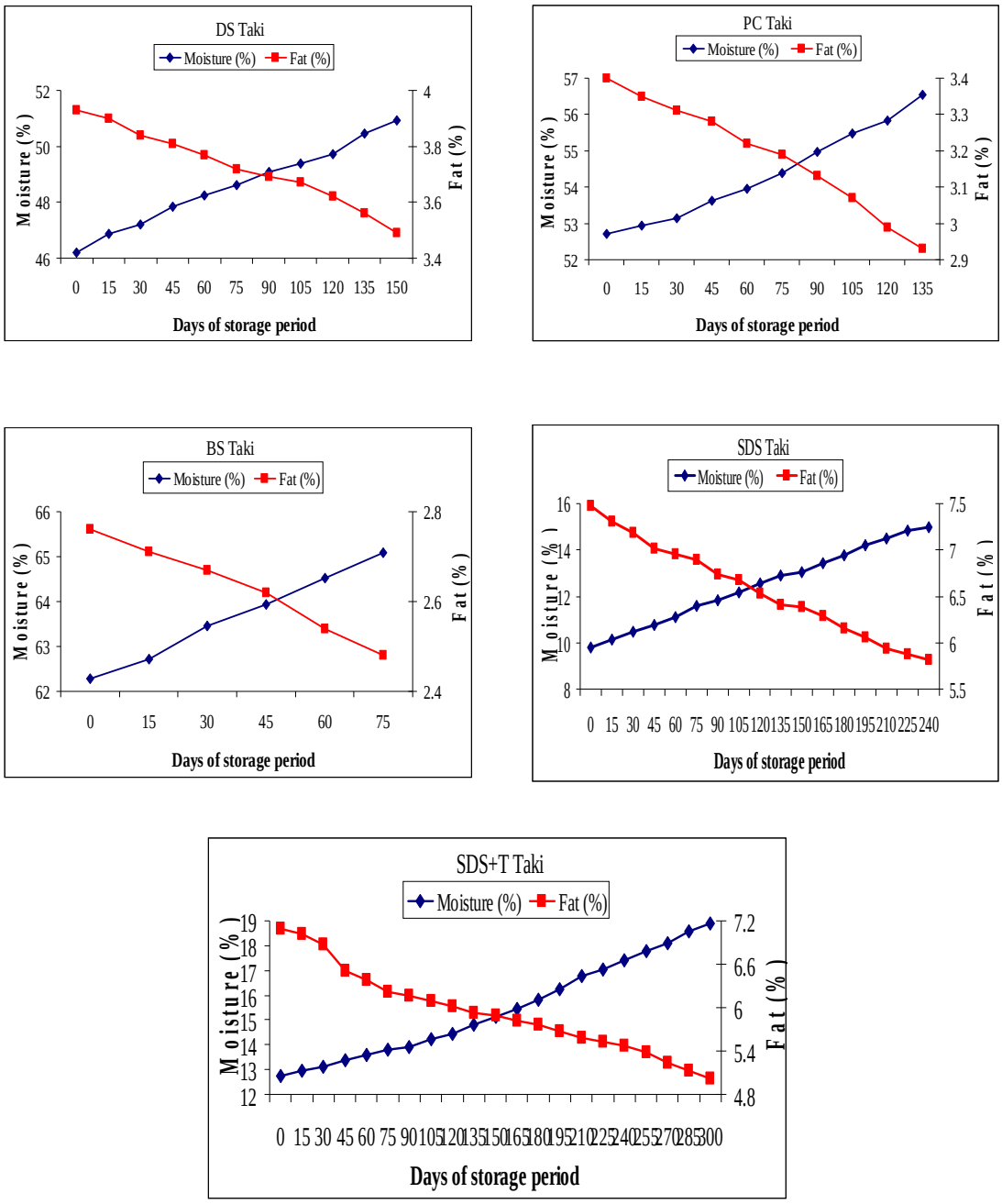


Figure. 4.4.2.6(C). Changing relationship between moisture & fat contents in different treatments (DS, PC, BS, SDS, SDS+T) of taki (*C. punctatus*) fish during different duration of storage at room temperature [DS = Dry salted, PC = Pickle cured, BS= Brine salted, SDS=Sun-dried salted and SDS+T= turmeric treated sun-dried salted]

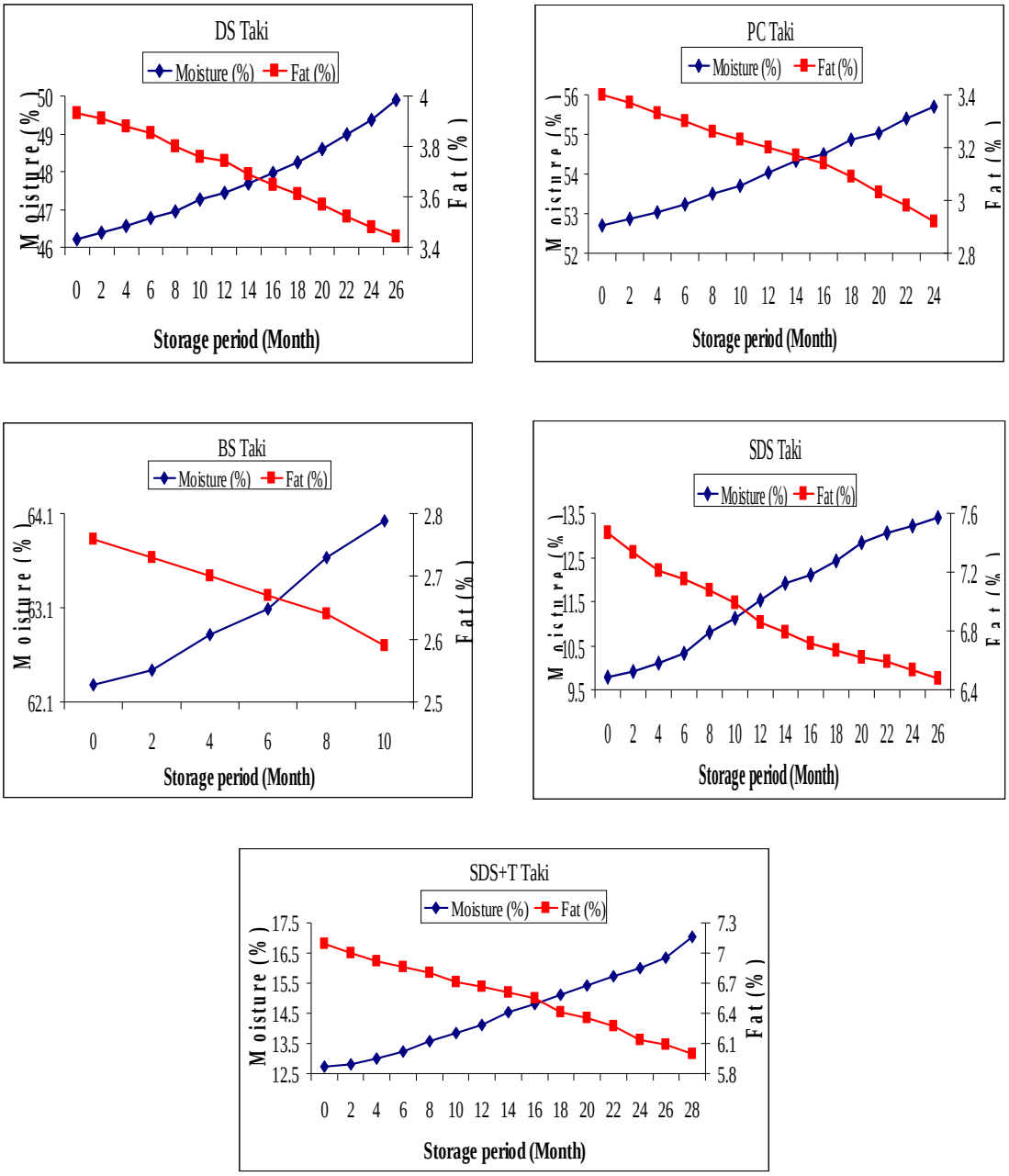


Figure. 4.4.2.6(D). Changing relationship between moisture & fat contents in different treatments (DS, PC, BS, SDS, SDS+T) of taki (*C. punctatus*) fish during different duration of storage at refrigeration temperature [DS = Dry salted, PC = Pickle cured, BS= Brine salted, SDS=Sun-dried salted and SDS+T= turmeric treated sun-dried salted]

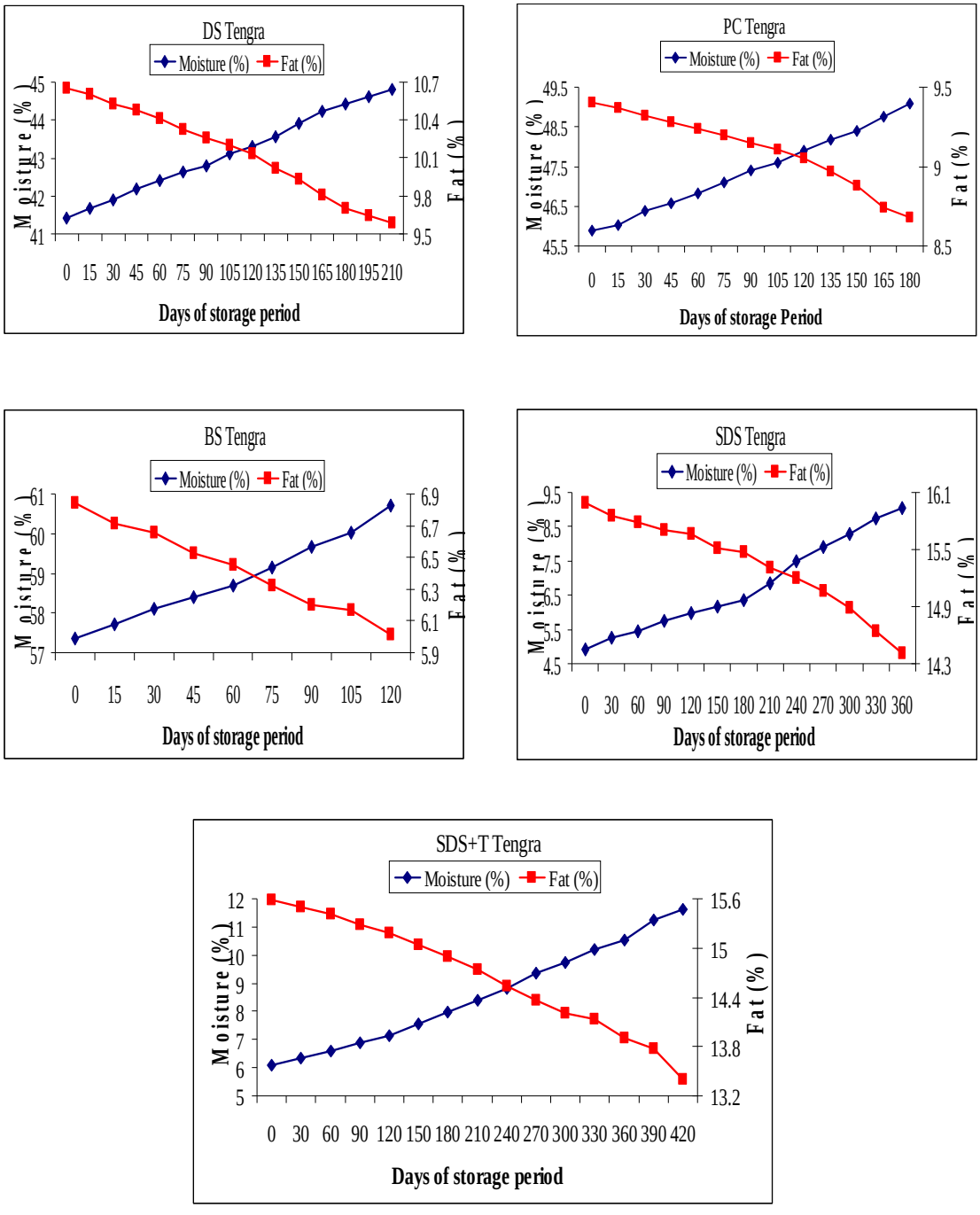


Figure. 4.4.2.6(E). Changing relationship between moisture & fat contents in different treatments (DS, PC, BS, SDS, SDS+T) of tengra (*M. tengra*) fish during different duration of storage at room temperature [DS = Dry salted, PC = Pickle cured, BS= Brine salted, SDS=Sun-dried salted and SDS+T= turmeric treated sun-dried salted]

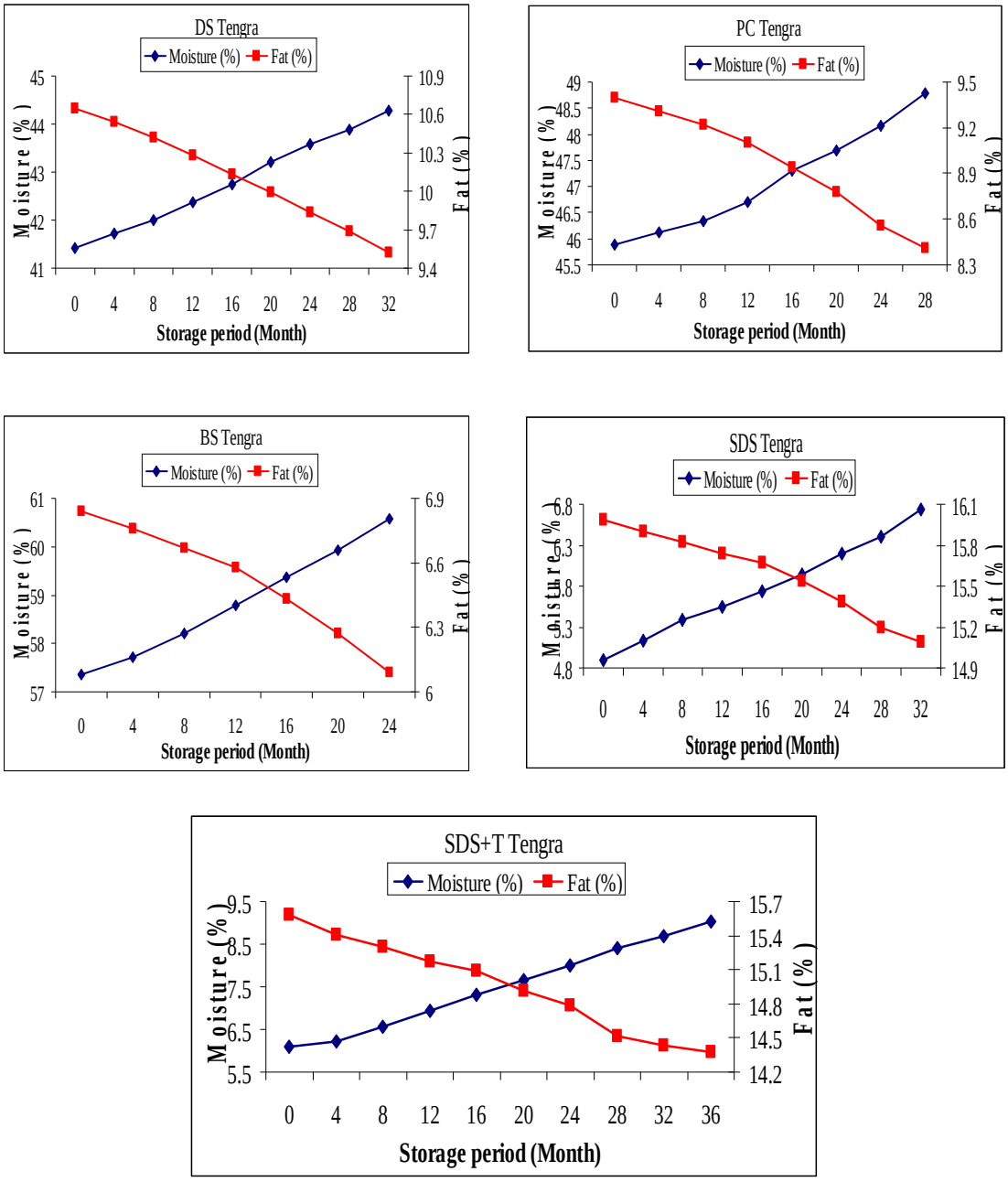


Figure. 4.4.2.6(F). Changing relationship between moisture & fat contents in different treatments (DS, PC, BS, SDS, SDS+T) of tengra (*M. tengra*) fish during different duration of storage at refrigeration temperature [DS = Dry salted, PC = Pickle cured, BS= Brine salted, SDS=Sun-dried salted and SDS+T= turmeric treated sun-dried salted]

4.4.2.7. Changes in ash content

In Shol fish, during storage at room temperature ash content varied in the range of 18.98% to 17.58%, 18.69% to 17.45%, 16.06% to 15.14%, 22.80% to 21.44% and 22.41% to 21.07% in case of dry salting (DS), pickle curing (PC), brine salting (BS), sun-dried salting (SDS) and turmeric treated sun-dried salting (SDS+T) [Table 4.4.2.1(A)]. On the other hand, during storage at refrigerated temperature ash content varied in the range of 18.98% to 17.94%, 18.69% to 17.76%, 16.06% to 15.49%, 22.80% to 21.66% and 22.41% to 21.41% in case of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) shol fish-products [Table 4.4.2.1(B)].

It is also observed from Table 4.4.2.1(C) that, during storage at room temperature ash content varied in the range of 26.37% to 24.55%, 22.96% to 21.02%, 17.24% to 15.99%, 35.16% to 34.05% and 34.29% to 32.90% in case of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) taki fish-products. Whereas, in refrigerated temperature taki fish showed a variable ash content in the range of 26.37% to 25.09%, 22.96% to 21.58%, 17.24% to 16.50%, 35.16% to 34.41% and 34.29% to 33.13% in case of dry salting (DS), pickle curing (PC), brine salting (BS), sun-dried salting (SDS) and turmeric treated sun-dried salting (SDS+T) [Table 4.4.2.1(D)].

Likewise, Dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) tengra fish-products had ash content varying in the range of 26.15% to 25.00%, 24.62% to 23.41%, 20.80% to 19.40%, 36.20% to 35.18% and 35.60% to 34.22% during storage at room temperature whereas 26.15% to 25.10%, 24.62% to 23.59%, 20.80% to 19.48%, 36.20% to 35.50% and 35.60% to 34.82% during storage at refrigerated temperature [Table 4.4.2.1(E) & Table 4.4.2.1(F)]. The comparison of changes in ash content of five types of salted shol, taki and tengra fish samples stored both room and refrigeration temperature is presented in Figure 4.4.2.7.

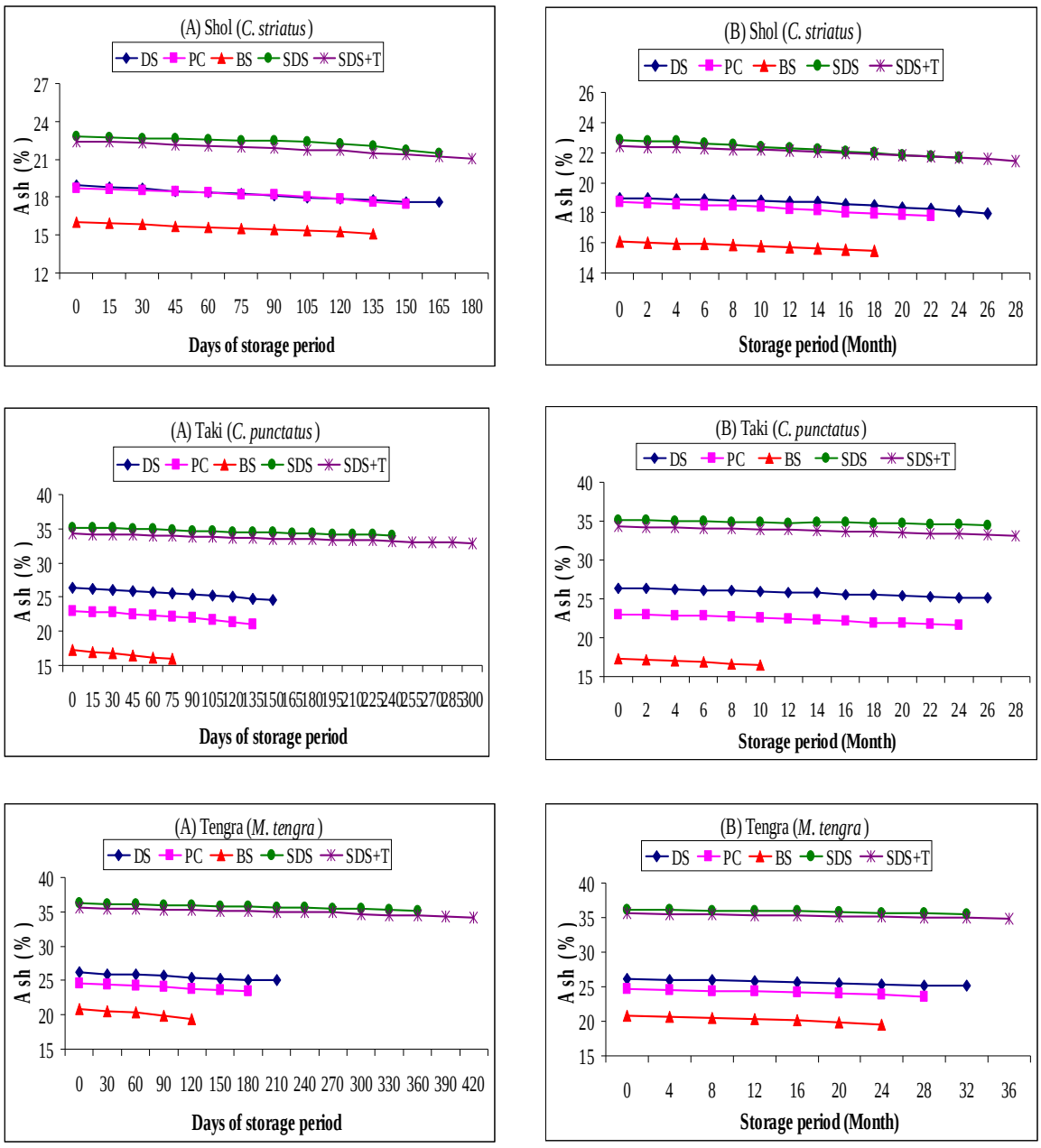


Figure. 4.4.2.7. Effects of different types of salting process on ash content of DS, PC, BS, SDS, SDS+T treated shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish during different duration of storage both at (A) room temperature and (B) refrigeration temperature (DS = Dry salted, PC = Pickle cured, BS= Brine salted, SDS=Sun-dried salted and SDS+T= turmeric treated sun-dried salted)

Figure 4.4.2.7 shows that the ash content in sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) shol, taki and tengra was slightly higher than other 3 types of fish products both room and refrigeration storage. This was because of the penetration of salt into the fish tissue and diffusion of moisture from the tissue was comparatively highest in sun-dried salted fish products.

There were significant differences in ash contents among DS, PC, BS, SDS, and SDS+T shol taki and tengra fish-products during different days of observation both room and refrigeration storage. On the other hand, ash content in the brine salted (BS) product was found significantly lower than that in the other 4 because there was no salt crystal stacked on the muscle surface.

From three fish species, comparatively higher ash content was found in five types (DS, PC, BS, SDS, and SDS+T) of tengra fish-products which is in accordance with Daramola *et al.* (2007), who stated that smaller sized fish species has higher ash content due to the higher bone of flesh ratio.

Siddique and Aktar (2011) observed that, ash level in marine dried *Harpodon nehereus, Johnius dussumieri* and *Lepturacanthus savala* fish samples were decreased from 7.56% to 4.76%, 6.37% to 4.89% and 4.86% to 4.64% respectively during 2 year of storage period at room temperature. Similarly, Hassan *et al.* (2013) stated that, the ash content changes with the time of storage due to absorbance of moisture and loss of protein.

These above findings are in close quarters with the present investigation.

4.4.3. Chemical composition analysis for determining quality changes

The keeping quality and wholesomeness of salted fish products are influenced by chemical composition of fish. On the other hand, composition of fish varies considerably with season, sexual development, feeding composition and movement of it.

Chemical methods are reliable measures of freshness or state of deterioration of products, because the concentrations of chemicals are dependent on storage time and temperature. Chemical composition may vary widely, not only for fish of the same species, but also within an individual fish, according to age, sex, and environmental conditions (FAO, 2005). Abu Gideiri *et al*., (2004) found a significant change in some chemical constituents of salted fish (*Oreocheomis niloticus*).

Knowledge on chemical composition of fish will help the processors to define the optimum processing and storage conditions for premium quality products.

In present research work, the following four chemical parameters have been assessed to find out the shelf life of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) shol, taki and tengra fish-products.

1. Changes in salt content
2. Changes in TVB-N (Total volatile base nitrogen)
3. Changes in FFA (Free fatty acid) value
4. Changes in pH value

The detail findings of these chemical parameters are given below under respective headings.

4.4.3.1. Changes in salt content

Salt is added more or less in all food items. Therefore the determination of salt content is very important. Salt is the main concentration of body fluid and regulates the electrolyte movements through cellular movement during living state. The salt content was shown to vary in different treatments depending on salting procedure.

After salting (end of ripening period) salt content of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) **shol** fish products were 16.00%, 16.08%, 15.20%, 14.90% and 14.30% respectively. End of storage at room temperature these value decreased in 14.36%, 14.60%, 13.73%, 12.15% and 12.02% whereas end of refrigeration storage these value decreased 15.20%, 15.26%, 14.36%, 12.74% and 12.28% respectively.

In case of **taki** fish, the salt content decreased from 16.06 to 15.05%, 16.22 to 15.34%, 15.5 to 14.42%, 14.72 to 12.89% and 13.77 to 12.20 % in dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) fish-products stored at room temperature whereas end of refrigeration storage salt content decreased considerably at 15.18%, 15.29%, 14.96%, 13.09% and 12.82% in dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) taki fish-products.

On the other hand, **tengra** fish had salt content ranged from 16.80 to 15.41%, 17.10 to 15.90%, 16.50 to 15.02%, 15.20 to 13.56% and 14.80 to 13.0 % in dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) fish-products stored at room temperature. However, end of refrigeration storage salt content changed considerably at 15.63%, 15.98%, 15.27%, 13.81% and 13.21**%** in dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) tengra fish-products.

The changes in salt (%) content of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) shol, taki and tengra fish-products during storage both at ambient and refrigeration temperature is presented in Table 4.4.3.1.

Table: 4.4.3.1. Changes in salt (%) contents of salted shol, taki and tengra stored both at room and refrigeration temperature in different treatments (DS = Dry salted, PC = Pickle cured, BS= Brine salted, SDS=Sun-dried salted and SDS+T= Turmeric treated sun-dried salted)

Fish	Treatment	Salt (%) content in fresh process product	Salt (%) content in end product (room temperature)	Salt (%) content in end product (refrigeration temperature)
Shol (*C. striatus*)	**DS**	16.00	14.36	15.20
	PC	16.08	14.60	15.26
	BS	15.20	13.73	14.36
	SDS	14.90	12.15	12.74
	SDS+T	14.30	12.02	12.28
Taki (*C. punctatus*)	**DS**	16.06	15.05	15.18
	PC	16.22	15.34	15.29
	BS	15.50	14.42	14.96
	SDS	14.72	12.89	13.09
	SDS+T	13.77	12.20	12.82
Tengra (*M. tengra*)	**DS**	16.80	15.41	15.63
	PC	17.10	15.90	15.98
	BS	16.50	15.02	15.27
	SDS	15.20	13.56	13.81
	SDS+T	14.80	13.00	13.21

As far as salt content is concerned, there was a decreasing trend in 5 types of salted shol, taki and tengra fish-products during the storage period both room and refrigeration temperature. The decrease in salt content can be attributed to uptake of moisture due to hydrostatic nature of salt during the storage period which is in accordance with the findings of Dewi *et al.* (2011).

Both room and refrigeration storage, the highest salt content was shown in pickle cured (PC) and lowest was found in turmeric treated sun-dried salted (SDS+T) shol, taki and tengra fish-products.

Salt content of dry salted Mackerel and Pink perch stored at ambient temperature were reported as 17.5% and > 21.0%, respectively (Srikar *et al.*, 1993). According to Kumar *et al.* (2013), salt content decreased from 17.50 to 12.74% in case of 6 month stored 30% salted sun-dried *L. gonius* fish at room temperature.

Dutta *et al.* (1992) have reported a decrease of salt content in salted-sun dried freshwater fish during eight months of storage. Sharma *et al.* (2013) also found a decreasing trend of salt with a value of 10.25% to 6.60% in 6 months stored sun-dried salted *G. chapra* fish-products. In another study, Al-Reza *et al.* (2015) have found a decrease of salt content in salted-sundried Chela fish with a value of 17.54% to 15.67% after 60 days of storage.

The above findings of decreasing trend of salt content with increase in storage time are similar with present findings.

4.4.3.2. Changes in Total Volatile Base Nitrogen (TVB-N)

Effects of different types of salting process on TVB-N (mg/100g) of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish during different duration of storage both at room and refrigeration temperature is presented in Table 4.4.3.2(A)-(F).

In present study, the range of TVB-N value of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) **shol** fish products during storage at room temperature were 5.25 to 31.33, 5.28 to 34.99, 3.64 to 34.52, 4.89 to 30.86 and 5.17 to 31.44 mg/100g of fish whereas at refrigeration storage, this value varied from 5.25 to 19.06, 5.28 to 19.74, 3.64 to 21.14, 4.89 to 19.15 and 5.17 to 20.02 mg/100g of fish [Table 4.4.3.2(A) & Table 4.4.3.2(B)].

TVB-N values in dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) **taki** fish products ranged from 4.18 to 30.76, 3.79 to 30.01, 5.70 to 30.96, 2.27 to 31.02 and 3.58 to 31.25 mg/100g of fish during storage at room temperature and 4.18 to 20.05, 3.79 to 20.58, 5.70 to 24.48, 2.27 to 20.27 and 3.58 to 20.31 mg/100g of fish respectively during storage at refrigeration temperature [Table 4.4.3.2(C) & Table 4.4.3.2(D)].

However, the TVB-N values of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) **tengra** fish products ranged from 3.90 to 30.22, 4.92 to 31.04, 2.59 to 30.45, 1.90 to 30.15 and 2.52 to 30.24 mg/100g of fish during storage at room temperature and 3.90 to 20.23, 4.92 to 20.59, 2.59 to 20.98, 1.90 to 20.75 and 2.52 to 21.07 mg/100g of fish respectively during storage at refrigeration temperature [Table 4.4.3.2(E) & Table 4.4.3.2(F)].

Table: 4.4.3.2(A). Changes in TVB-N (mg/100g) value of 5 types of salted shol during different duration of storage period at room temperature (DS = Dry salted, PC = Pickle cured, BS = Brine salted, SDS = Sun-dried salted, SDS+T = Turmeric treated sun-dried salted)

Days of storage period	DS	PC	BS	SDS	SDS+T
0	5.25	5.28	3.64	4.89	5.17
15	6.64	7.27	5.27	6.59	7.07
30	7.07	8.81	9.18	7.71	7.13
45	8.36	10.13	12.75	9.08	7.79
60	10.79	14.98	14.04	9.53	8.29
75	13.23	18.15	16.27	10.02	9.68
90	14.71	21.55	20.84	10.14	9.93
105	17.29	26.48	27.63	16.43	12.21
120	21.48	30.47	30.33	19.37	15.12
135	25.28	32.92	34.52	23.49	20.05
150	28.62	34.99	-	27.35	24.23
165	31.33	-	-	30.86	29.57
180	-	-	-	-	31.44

Table: 4.4.3.2(B). Changes in TVB-N (mg/100g) value of 5 types of salted shol during different duration of storage period at refrigeration temperature (4^0C) (DS = Dry salted, PC = Pickle cured, BS = Brine salted, SDS = Sun-dried salted, SDS+T = Turmeric treated sun-dried salted)

Storage period (Month)	DS	PC	BS	SDS	SDS+T
0	5.25	5.28	3.64	4.89	5.17
2	5.68	5.85	5.39	5.24	5.66
4	6.12	6.53	7.99	5.38	5.98
6	6.85	7.34	9.17	5.77	6.34
8	7.15	9.22	10.87	6.12	6.87
10	7.46	10.57	11.30	6.98	7.01
12	9.27	11.82	17.41	8.36	7.33
14	10.26	12.76	18.71	9.12	8.96
16	11.48	15.56	20.31	13.11	9.24
18	12.25	17.44	21.14	14.33	10.24
20	16.28	18.68	-	17.14	12.62
22	17.45	19.74	-	18.54	14.98
24	18.51	-	-	19.15	16.38
26	19.06	-	-	-	18.75
28	-	-	-	-	20.02

Table: 4.4.3.2(C). Changes in TVB-N (mg/100g) value of 5 types of salted taki during different duration of storage period at room temperature (DS = Dry salted, PC = Pickle cured, BS = Brine salted, SDS = Sun-dried salted, SDS+T = Turmeric treated sun-dried salted)

Days of storage period	DS	PC	BS	SDS	SDS+T
0	4.18	3.79	5.70	2.27	3.58
15	5.84	5.61	8.99	4.56	5.05
30	6.05	6.04	11.33	5.93	6.63
45	9.99	11.99	14.70	7.44	8.07
60	11.86	17.19	20.78	10.37	11.02
75	16.05	19.05	30.96	11.89	13.33
90	22.17	23.14	-	12.78	14.99
105	25.32	26.26	-	14.04	16.16
120	27.22	29.44	-	15.51	18.03
135	29.84	30.01	-	18.85	19.92
150	30.76	-	-	22.63	21.73
165	-	-	-	25.71	22.49
180	-	-	-	28.42	23.86
195	-	-	-	29.50	23.97
210	-	-	-	30.23	25.62
225	-	-	-	30.74	26.11
240	-	-	-	31.02	27.26
255	-	-	-	-	29.87
270	-	-	-	-	30.54
285	-	-	-	-	30.99
300	-	-	-	-	31.25

Table: 4.4.3.2(D). Changes in TVB-N (mg/100g) value of 5 types of salted taki during different duration of storage period at refrigeration temperature (4^0C) (DS = Dry salted, PC = Pickle cured, BS = Brine salted, SDS = Sun-dried salted, SDS+T = Turmeric treated sun-dried salted)

Storage period (Month)	DS	PC	BS	SDS	SDS+T
0	4.18	3.79	5.70	2.27	3.58
2	5.45	5.89	7.92	3.18	4.12
4	6.01	7.64	9.11	4.21	5.38
6	8.36	8.27	14.54	5.67	7.57
8	9.13	9.54	22.79	7.03	9.16
10	10.01	11.79	24.48	8.98	10.69
12	11.59	12.58	-	10.17	12.52
14	12.39	13.32	-	13.33	13.87
16	13.74	14.88	-	15.74	15.22
18	14.28	16.25	-	16.62	16.98
20	15.33	17.93	-	17.19	17.56
22	17.17	19.69	-	18.85	18.04
24	18.78	20.58	-	19.96	18.79
26	20.05	-	-	20.27	19.33
28	-	-	-	-	20.31

Table: 4.4.3.2(E). Changes in TVB-N (mg/100g) value of 5 types of salted tengra during different duration of storage period at room temperature (DS = Dry salted, PC = Pickle cured, BS = Brine salted, SDS = Sun-dried salted, SDS+T = Turmeric treated sun-dried salted)

Days of storage period	DS	PC	BS	SDS	SDS+T
0	3.90	4.92	2.59	1.90	2.52
30	5.23	5.73	6.61	4.24	4.86
60	8.29	8.42	12.56	6.21	7.13
90	12.08	15.93	20.38	8.42	9.35
120	16.38	24.17	30.45	11.12	12.73
150	22.07	28.08	-	14.91	16.54
180	27.74	31.04	-	17.82	18.48
210	30.22	-	-	19.25	20.81
240	-	-	-	21.89	22.69
270	-	-	-	23.18	24.41
300	-	-	-	25.92	25.15
330	-	-	-	28.46	26.62
360	-	-	-	30.15	27.41
390	-	-	-	-	29.53
420	-	-	-	-	30.24

Table: 4.4.3.2(F). Changes in TVB-N (mg/100g) value of 5 types of salted tengra during different duration of storage period at refrigeration temperature (4^0C) (DS = Dry salted, PC = Pickle cured, BS = Brine salted, SDS = Sun-dried salted, SDS+T = Turmeric treated sun-dried salted)

Storage period (Month)	DS	PC	BS	SDS	SDS+T
0	3.90	4.92	2.59	1.90	2.52
4	4.95	6.33	6.27	5.02	4.18
8	7.57	8.94	8.25	7.63	7.29
12	10.13	11.71	11.42	9.26	9.07
16	12.21	13.98	14.53	11.74	10.94
20	15.31	16.02	17.86	14.01	13.33
24	16.76	18.65	20.98	15.99	14.79
28	19.87	20.59	-	18.04	16.36
32	20.23	-	-	20.75	18.22
36	-	-	-	-	21.07

TVB-N is a group of biogenic amines formed in non-fermented food products during storage (Horsfall *et al.,* 2006). TVB-N produced by decomposition of proteins into simpler substances (ammonia, trimethylamine, creatine, purine bases and free amino acids) (Viji *et al.,* 2014). This TVB-N measurement can be used as a parameter for the determination of microbiological and enzymatic spoilage of fish product (Gram *et al.,* 2002). Total volatile base nitrogen (TVB-N) is important compound provide a measure of the progress of spoilage that is independent of sensory assessment.

According to Connell (1990) the limiting level for rejection of TVB-N is 30-40 mg/100g for storage at ambient temperature and 20 mg/100g for storage at refrigeration temperature while Kirk and Sawyer (1991) suggested a value of 30-40 mg/100g as the upper limit. On the other hand, according to Silva *et al.* (1998), TVB-N level below 20 to 36 mg/100g indicates that the fish is fresh. The European commission regulations (EC) set the range of TVB-N value for the acceptable fish/fishery products from 2-35 mg/100g of fish (Orban *et al.,* 2011). According to Ghaly *et al.* (2010), 35 mg/100 g of TVB-N value has been suggested as border line.

In present investigation, TVB-N values of the salted products either stored at refrigeration temperature or at room temperature showed linearly increasing pattern throughout storage period but neither of the value exceeded the recommended value set by different researchers for various fish and fish products as acceptable condition.

The TVB-N (mg/100g) increased in a characteristics pattern to a certain level of storage period of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) shol, taki and tengra fish-products which is mounted in Figure 4.4.3.2.

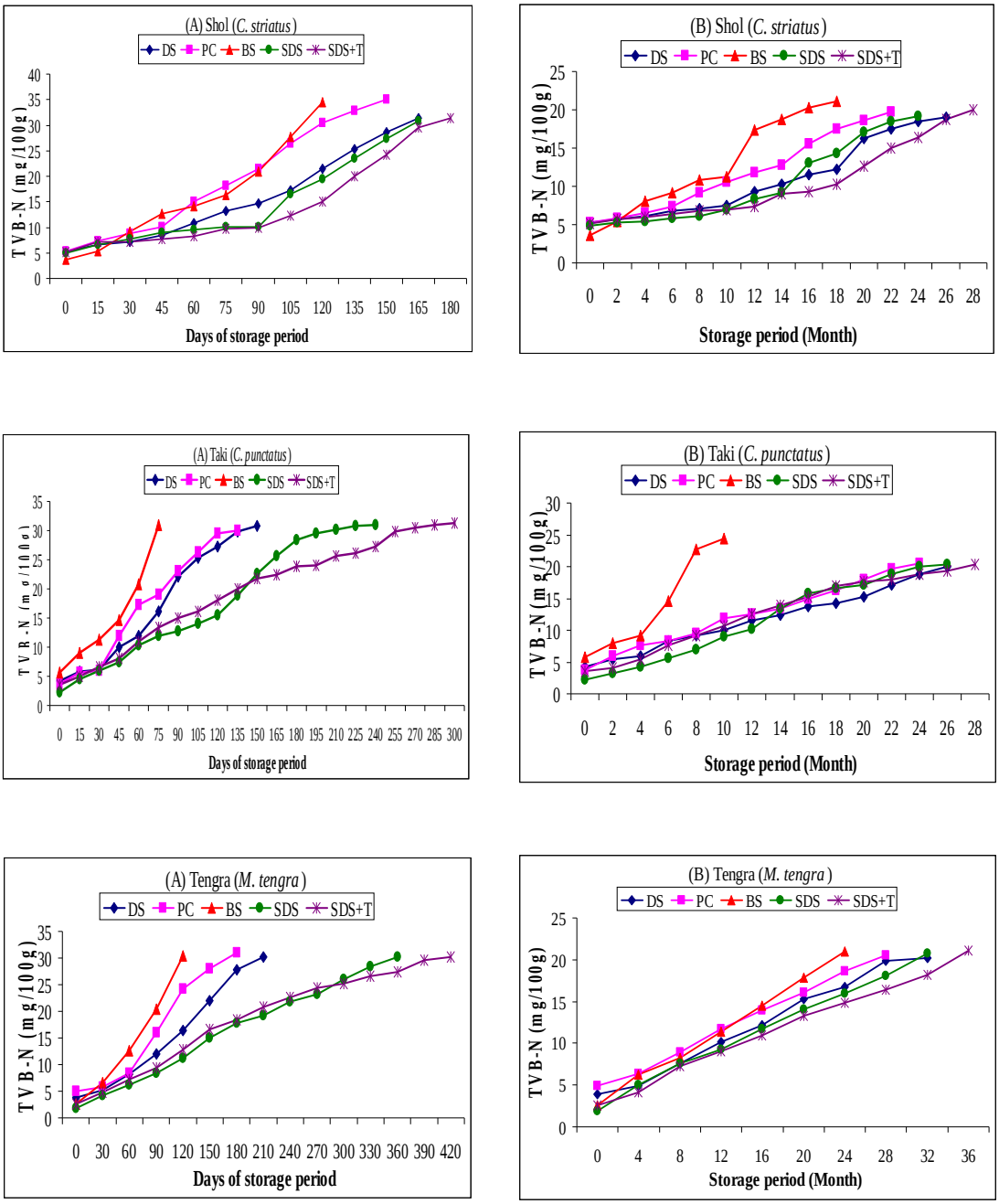


Figure. 4.4.3.2. Effects of different types of salting process on TVB-N (mg/100g) value of DS, PC, BS, SDS, SDS+T treated shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish during different duration of storage both at (A) room temperature and (B) refrigeration temperature (DS = Dry salted, PC = Pickle cured, BS= Brine salted, SDS=Sun-dried salted and SDS+T= Turmeric treated sun-dried salted)

As expected, a significant increase in TVB-N values was observed in all salt-cured fish samples. A statistically significant but moderate increase was observed in TVB-N values of the refrigeration storage salted fish-samples, while a sharp increase was observed in the same values of the fish-samples during storage at room temperature which is in agreement with the findings of Estrada *et al.* (1985). In early storage, spoilage rate become slower than later storage time, it would appear from the figure 4.4.3.2. This is also supported by Reed *et al*. (1929) and Muslemuddin (1970). Brine salted (BS) fish-products had the most rapid increase in TVB-N value compared to other salted samples.

At the end of shelf life it was found that, in case of **shol** fish, the highest TVB-N (mg/100g) value of end products was in PC shol and lowest value was in SDS shol in room temperature, whereas in refrigeration temperature the highest TVB-N (mg/100g) value was in BS shol and lowest value was in DS shol fish products.

In case of **taki** fish, it has been found that the highest TVB-N (mg/100g) value was in SDS+T taki and lowest value was in PC taki in room temperature, whereas in refrigeration temperature the highest TVB-N (mg/100g) content was in BS taki and lowest value was in DS taki fish products at the end of shelf life.

The highest TVB-N (mg/100g) content in **tengra** fish, has been found in PC tengra and lowest value was in SDS tengra in room temperature, whereas in refrigeration temperature the highest TVB-N (mg/100g) content was in SDS+T tengra and lowest value was in DS tengra fish products at the end of storage period.

The TVB-N value that helps for the determination of level of fish spoilage has an inverse relationship with the sensory score of salted and sun dried-salted fish-products. It is evident from present findings that the total sensory score come down while the TVB-N value increased, which have been noted by Shewan (1938, 1942); Herzberg *et al.* (1976); Ahmed *et al.* (1981b) and Rubbi *et al.* (1987). The combined total amount of ammonia (NH_3), dimethylamine (DMA) and trimethylamine (TMA) in fish is called the total volatile base nitrogen (TVB-N) content of the fish and is

commonly used as an index for the determination of freshness of fish (Huss, 1988; Wu and Bechtel, 2008). According to Truelstrup *et al.* (1995), the increase in TVB-N was caused by a combination of microbiological and autolytic deamination of amino acids and the complete microbial reduction of TMAO to TMA. According to Connell (1995), increase in TVB-N value is related to bacterial spoilage. In this aspect, Chaijan *et al.* (2006) opined that TVB-N is mainly contributed by ammonia in the muscle produced by de-amination of muscle proteins. Similarly, Wallace (2000) has pointed out that TVB-N is better index of spoilage. Therefore in the present study, increase in TVB-N values throughout the storage period may be due to microbial activity, absorption of moisture and relative decrease in salt content.

Aksu *et al.* (1997) reported that, TVB-N value of 8.3 mg/100g in anchovy marinated using acetic acid of 2% increased to 15.1 mg/100g at the storage of 150 days. On the other hand, Gökoğlu, *et al.* (1998) found that TVB-N value in fresh sardine increased from 13.2 mg/100g to 64.8 mg/100g during refrigeration storage. In another report, the TVB-N value in anchovy marinated with 4% acetic acid increased from 9.8 mg/100 g to 14 mg/100g during the storage of 8 months in 4°C (Dokuzlu, 2000). A much higher TVB-N level (60.5 mg/100 g) was reported by day 42 in vacuum-packaged, salted sea bream stored under refrigeration (4ºC) temperature (Chouliara *et al.*, 2004). Likewise, a comparable pattern of the increase in TVB-N has been reported in brined anchovies (Karaçam *et al.*, 2002) and brined chub mackerel (Goulas and Kontominas 2005) during refrigeration storage. According to Begum *et al.* (2012b), the range of TVB-N value of 6 months room temperature stored sun-dried salted punti fish was 18.02-42.09 mg/100g respectively which is much higher in present findings.

From the above observation, it was concluded that TVB-N value also increased in refrigeration storage which is in accordance with present research. While the initial TVB-N values in our samples were more or less similar to the findings of other researchers, the increase in TVB-N values during the storage was lower than others. The probable reason for these differences may be due to differences in fish species and different salting process.

4.4.3.3. Changes in Free Fatty Acid (FFA) value

Effects of different types of salting process on FFA (%) value of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish during different duration of storage both at room and refrigeration temperature is presented in Table 4.4.3.3(A)-(F).

In case of Shol fish, during storage at room temperature, FFA (%) value was varied in the range of 1.5% to 13.5% (165 days), 1.3% to 13% (150 days), 1.8% to 14% (135 days), 2.3% to 10.5% (165 days) and 2.4% to 11.1% (180 days) in case of dry salting (DS), pickle curing (PC), brine salting (BS), sun-dried salting (SDS) and turmeric treated sun-dried salting (SDS+T). On the other hand, at the end of refrigeration storage, these value was 10.1% (26 months) in dry salted (DS) shol, 11.9% (22 months) in pickle cured (PC), shol, 12.2% (18 months) in brine salted (BS) shol, 11.0% (24 months) in sun-dried salted (SDS) shol and 11.4% (28 months) in case of turmeric treated sun-dried salted shol (SDS+T) fish-products [Table 4.4.3.3(A) & Table 4.4.3.3(B)].

During storage of taki fish at room temperature, FFA (%) value varied in the range of 1.9% to 11.5% (150 days), 2.4% to 12.8% (135 days), 2.6% to 13.3% (75 days), 1.8% to 12.2% (240 days) and 2.3% to 13.1% (300 days) in case of dry salting (DS), pickle curing (PC), brine salting (BS), sun-dried salting (SDS) and turmeric treated sun-dried salting (SDS+T) whereas at the end of refrigeration storage, these value was 12.5% (26 months) in dry salted (DS) taki, 12.7% (24 months) in pickle cured (PC) taki, 12.8% (10 months) in brine salted (BS) taki, 12.1% (26 months) in sun-dried salted (SDS) taki and 12.3% (28 months) in case of turmeric treated sun-dried salted taki (SDS+T) fish-products [Table 4.4.3.3(C) & Table 4.4.3.3(D)].

It is also shown in Table 4.4.3.3(E) that, during storage at room temperature FFA (%) value was varying in the range of 2.8% to 12.1% (210 days), 3.2% to 12.7% (180

days), 1% to 13% (120 days), 1.8% to 13.1% (360 days) and 1.6% to 13.2% (420 days) in case of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) tengra fish-products. On the other hand, at the end of refrigeration storage, these value was 11.0% (32 months) in dry salted (DS) tengra, 11.4% (28 months) in pickle cured (PC) tengra, 11.6% (24 months) in brine salted (BS) tengra, 11.1% (32 months) in sun-dried salted (SDS) tengra and 11.2% (36 months) in case of turmeric treated sun-dried salted (SDS+T) tengra fish-products [& 4.4.3.3(F)].

A rise in free fatty acid value in fish during storage, from 1.1% to 2.5% in 24 hours of storage, 7.8 or 8.1% after 120 hours of storage kept at room temperature was reported by Lassen *et al*. (1951). On the other hand, Ranke *et al.* (1959) reported that, after 6 months and 24 months of storage (at-13^{0}C to -25^{0}C) the appearance of considerable contents of FFA with certain bad odors and off flavors presumably derived from the degradation of fats, greatly reduced the consumer appeal of trout. According to Srikar *et al.* (1993), FFA contents increased significantly throughout the period of storage of Mackerel and Pink perch at ambient temperature and reached 21.1% respectively after 35 days of storage. Rahman (1996) reported that, the FFA value of raw hilsa fish was 1.16% and after 8 weeks of observation, the FFA value of dry salted hilsa products storage at room temperature (26-30^{0}C) reached to 11.7%. Similarly, Chakraborty *et al.* (1997) found that, free fatty acid (FFA) value ranged from 2.9% of total lipid on the start day to 7.58, 7.15 and 8.56% on the 60 days of observation in dry salting, wet salting and sun-dry salted fishes respectively. Likewise, Mansur *et al*. (1998) reported that, the initial value of raw pre-spawning and post-spawning hilsa was 1.15% and 1.21% respectively which after dry salting increased to 7.86% and 8.35% respectively on 18th days of storage at 28^{0}C to 32^{0}C. A higher FFA (%) of 31.84% has been reported in salted Anchovy after 9 week of storage (Hernandaz- Herrero *et al.*, 1999). Sharma *et al.* (2013) also found an increasing trend of FFA with a value of 5.24% to 28.34% in 6 months stored sun-dried salted *G. chapra* fish-products.

The present study denoted that the contents of free fatty acid values are similar with the above mentioned study.

Table: 4.4.3.3(A). Changes in FFA% (Free Fatty Acid) value of 5 types of salted shol during different duration of storage period at room temperature (DS = Dry salted, PC = Pickle cured, BS = Brine salted, SDS = Sun-dried salted, SDS+T = Turmeric treated sun-dried salted)

Days of storage period	DS	PC	BS	SDS	SDS+T
0	1.5	1.3	1.8	2.3	2.4
15	2.2	2.4	2.9	2.8	2.9
30	2.7	2.8	3.5	3.1	3.2
45	3.5	4.4	6.7	4.1	3.8
60	5.5	6.1	9.2	4.8	4.5
75	6.8	7.4	10.5	6.1	5.7
90	7.3	8.5	11.2	7.2	6.6
105	9.1	9.2	12.7	8.2	7.8
120	9.8	10.5	13.3	8.9	8.5
135	11.4	12.2	14	9.3	9.1
150	12	13	-	9.9	9.4
165	13.5	-	-	10.5	10.2
180	-	-	-	-	11.1

Table: 4.4.3.3(B). Changes in FFA% (Free Fatty Acid) value of 5 types of salted shol during different duration of storage period at refrigeration temperature (4^0C) (DS = Dry salted, PC = Pickle cured, BS = Brine salted, SDS = Sun-dried salted, SDS+T = Turmeric treated sun-dried salted)

Storage period (Month)	DS	PC	BS	SDS	SDS+T
0	1.5	1.3	1.8	2.3	2.4
2	1.7	1.9	2.2	2.4	2.5
4	2.0	2.1	2.6	2.5	2.6
6	2.6	3.4	4.1	3.2	3.9
8	3.1	4.6	5.9	4.5	4.2
10	3.9	5.8	7.5	5.1	5.5
12	4.4	6.9	8.1	6.7	6.4
14	5.2	7.5	9.7	7.9	7.3
16	6.9	8.1	10.9	8.2	8.1
18	7.6	9.7	12.2	8.9	8.7
20	8.1	10.8	-	9.1	9.2
22	8.7	11.9	-	10.3	9.9
24	9.3	-	-	11.0	10.7
26	10.1	-	-	-	11.1
28	-	-	-	-	11.4

Table: 4.4.3.3(C). Changes in FFA% (Free Fatty Acid) value of 5 types of salted taki during different duration of storage period at room temperature (DS = Dry salted, PC = Pickle cured, BS = Brine salted, SDS = Sun-dried salted, SDS+T = Turmeric treated sun-dried salted)

Days of storage period	DS	PC	BS	SDS	SDS+T
0	1.9	2.4	2.6	1.8	2.3
15	2.5	3.1	5.7	2.2	2.9
30	3.1	3.7	7.3	3.1	3.1
45	3.8	4.4	9.9	3.7	3.8
60	4.6	5.2	12	4.9	4.6
75	5.8	6.9	13.3	5.5	5.2
90	6.2	7.5	-	6.8	5.9
105	7.4	9.5	-	7.2	6.7
120	8.1	11.9	-	8.5	7.3
135	9.8	12.8	-	9.1	7.9
150	11.5	-	-	9.5	8.6
165	-	-	-	10.0	9.0
180	-	-	-	10.4	9.8
195	-	-	-	10.8	10.2
210	-	-	-	11.3	10.5
225	-	-	-	11.9	10.9
240	-	-	-	12.2	11.4
255	-	-	-	-	12.0
270	-	-	-	-	12.6
285	-	-	-	-	12.9
300	-	-	-	-	13.1

Table: 4.4.3.3(D). Changes in FFA% (Free Fatty Acid) value of 5 types of salted taki during different duration of storage period at refrigeration temperature (4^0C) (DS = Dry salted, PC = Pickle cured, BS = Brine salted, SDS = Sun-dried salted, SDS+T = Turmeric treated sun-dried salted)

Storage period (Month)	DS	PC	BS	SDS	SDS+T
0	1.9	2.4	2.6	1.8	2.3
2	2.2	3.1	5.4	2.1	2.9
4	2.7	3.9	7.3	2.8	3.3
6	3.6	4.4	9.9	3.9	4.2
8	4.5	5.2	11.1	4.2	4.6
10	5.4	6.7	12.8	4.7	5.1
12	6.1	7.3	-	5.4	5.9
14	7.3	8.5	-	6.7	6.6
16	8.7	9.6	-	7.3	7.2
18	9.8	10.1	-	8.2	8.4
20	10.3	11.3	-	9.5	9.8
22	11.7	11.9	-	10.4	10.5
24	12.1	12.7	-	11.3	11.2
26	12.5	-	-	12.1	11.9
28	-	-	-	-	12.3

Table: 4.4.3.3(E). Changes in FFA% (Free Fatty Acid) value of 5 types of salted tengra during different duration of storage at room temperature (DS = Dry salted, PC = Pickle cured, BS = Brine salted, SDS = Sun-dried salted, SDS+T = Turmeric treated sun-dried salted)

Days of storage period	DS	PC	BS	SDS	SDS+T
0	2.8	3.2	1	1.8	1.6
30	3.8	4.7	5.1	2.4	2.1
60	5.3	6.8	8.7	3.9	3.8
90	6.9	8.5	11	5.1	4.9
120	7.8	10.1	13	6.7	6.1
150	9.2	11.9	-	7.9	7.7
180	11.7	12.7	-	8.6	8.2
210	12.1	-	-	9.3	8.9
240	-	-	-	10.4	9.6
270	-	-	-	11.1	10.2
300	-	-	-	11.9	10.9
330	-	-	-	12.2	11.3
360	-	-	-	13.1	12.1
390	-	-	-	-	12.6
420	-	-	-	-	13.2

Table: 4.4.3.3(F). Changes in FFA% (Free Fatty Acid) value of 5 types of salted tengra during different duration of storage at refrigeration temperature (4^0C) (DS = Dry salted, PC = Pickle cured, BS = Brine salted, SDS = Sun-dried salted, SDS+T = Turmeric treated sun-dried salted)

Storage period (Month)	DS	PC	BS	SDS	SDS+T
0	2.8	3.2	1	1.8	1.6
4	3.5	4.1	2.9	2.8	2.1
8	4.8	5.3	3.8	3.7	3.3
12	5.7	6.7	5.3	5.1	4.2
16	7.1	7.2	7.5	6.6	5.4
20	8.2	8.9	9.1	7.9	6.7
24	9.5	10.5	11.6	9.1	8.3
28	10.3	11.4	-	10.2	9.9
32	11.0	-	-	11.1	10.1
36	-	-	-	-	11.2

Free fatty acid (FFA), a tertiary product of rancidity, increased during storage. The FFA is a measure of hydrolytic rancidity-the extent of lipid hydrolysis by lipase action. The FFA value which indicates the rancidity of fat, increased gradually with the passing of storage period. FFA value is a measure of the extent of oxidative deterioration in oily fish, but it can fall further at latter stages of fish spoilage (FAO/SIFAR, 2001).

A high level of FFA is characteristic of product that have undergone both microbial and biochemical spoilage (Horner, 1997; Tungkawachara *et al.,* 2003). This may indicate that greater proportions of unsaturated fatty acids were liberated and were subjected to oxidative splitting at the double bonds. The resulting substances, mostly ketones and aldehydes, appear to be largely responsible for flavor, odor and taste of the salted fish product (El-Sebahy and Metwali, 1988).

An assessment of changes in FFA could provide an objective method for measuring the maturation of salted fish (Roldan *et al.,* 1985). Bernardez *et al.* (2005) stated that double bonds of unsaturated fatty acids are highly susceptible to oxidation and this leads to the production of carbonyls and other secondary oxidation products which impart the characteristic rancid off flavor to the product. The accumulation of free fatty acids (FFA) in fish oils in undesirable amount due to secondary reaction catalyzed, such as increased susceptibility to oxidation and consequent development of off flavors (Nair and Suseela, 2000).

Fat oxidation is a self catalyzing reaction, which is affected by the age of the raw material as well as oxidation of fats during processing and storage (AOAC, 1993). Salt does not inhibit lipases responsible for liberation of free fatty acids (Roldan *et al.*, 1985; Perez- Villarreal and Pozo, 1992).

The FFA value increased in a characteristic pattern to a certain level of storage period both room and refrigeration temperature of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) shol, taki and tengra fish-products which is mounted in Figure 4.4.3.3.

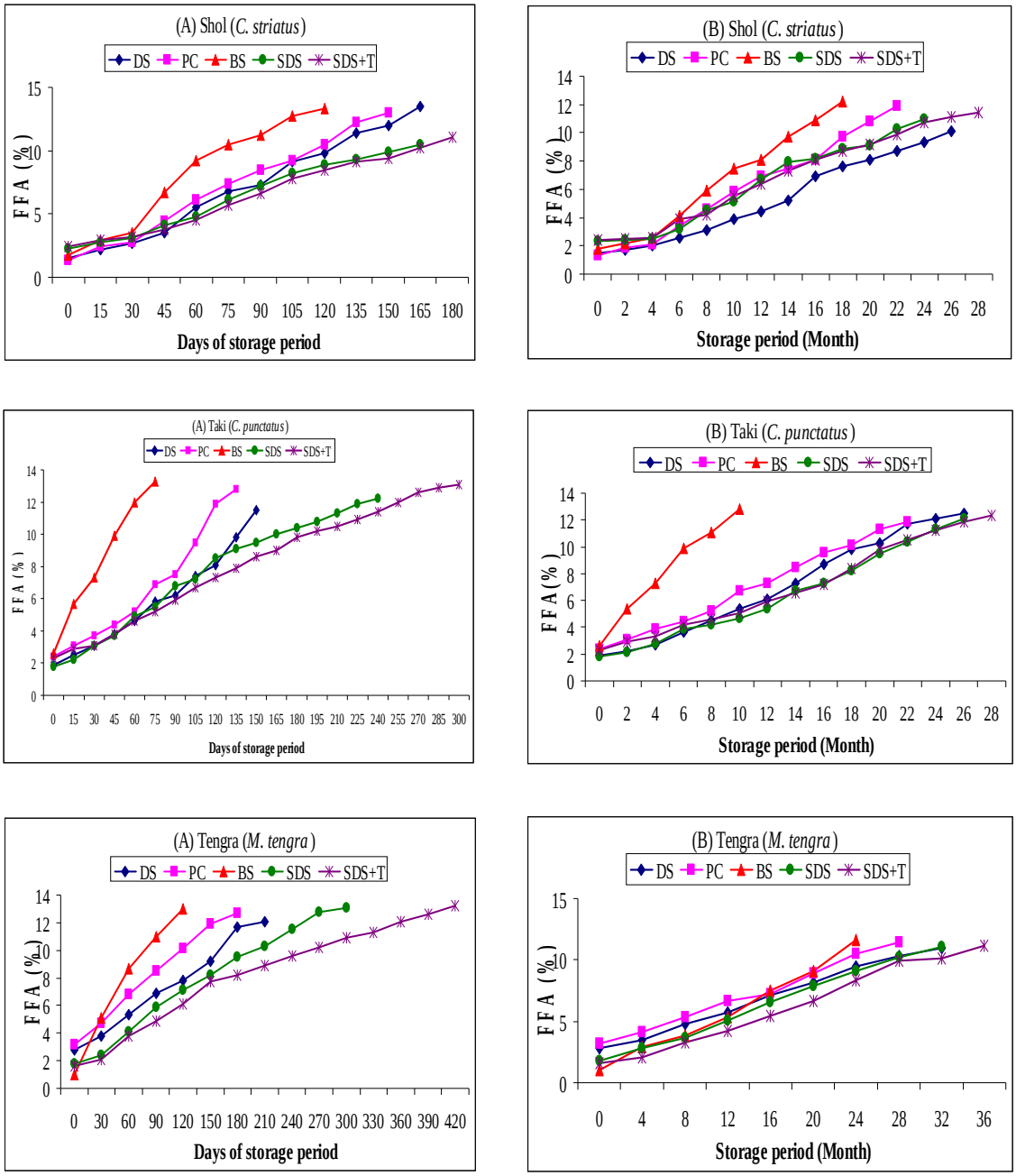


Figure. 4.4.3.3. Effects of different types of salting process on FFA (%) value of DS, PC, BS, SDS, SDS+T treated shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish during different duration of storage both at (A) room temperature and (B) refrigeration temperature (DS = Dry salted, PC = Pickle cured, BS= Brine salted, SDS=Sun-dried salted and SDS+T= Turmeric treated sun-dried salted)

During the storage period, FFA value increased both at room and refrigerated temperature, but the increment in room temperature was more prominent than the product stored at refrigeration temperature.

From the Figure 4.4.3.3, it is showed that, at the end of room and refrigeration storage, FFA (%) value was prominently highest in BS taki and shol. In room temperature at the end of shelf life the lowest FFA (%) value was in SDS shol and DS taki whereas at the end of refrigeration storage this value was lowest in DS shol and SDS taki fish products respectively.

However in tengra fish, at the end of shelf life in room temperature the highest FFA (%) value was reported in SDS+T tengra and lowest value was in DS tengra whereas at the end of refrigeration storage, the highest FFA (%) value was in BS tengra and lowest value was in DS tengra fish products.

However, the brine salted (BS) shol taki and tengra fish-products stored whether at room or refrigeration temperature showed a comparatively higher FFA (%) value with a short shelf life.

4.4.3.4. Changes in pH value

Effects of different types of salting process on pH value of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish during different duration of storage both at room and refrigeration temperature is presented in Table 4.4.3.4(A)-(F).

In case of shol fish, during storage at room temperature pH value was varied in the range of 6.3 to 8.1 (165 days), 6.4 to 8.3 (150 days), 6.5 to 8.3 (135 days), 6.2 to 8.3 (165 days) and 6.3 to 8.2 (180 days) respectively in case of dry salting (DS), pickle curing (PC), brine salting (BS), sun-dried salting (SDS) and turmeric treated sun-dried salting (SDS+T) [Table 4.4.3.4(A)].

In case of shol fish, during storage at refrigeration temperature pH value was varied in the range of 6.3 to 7.1 (26 months), 6.4 to 7.2 (22 months), 6.5 to 7.2 (18 months), 6.2 to 7.1 (24 months) and 6.3 to 7.2 (28 months) respectively in case of dry salting (DS), pickle curing (PC), brine salting (BS), sun-dried salting (SDS) and turmeric treated sun-dried salting (SDS+T) [Table 4.4.3.4(B)].

During storage of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) taki fish at room temperature, the pH values varied in the range of 6.5 to 7.9 (150 days), 6.6 to 8.3 (135 days), 6.8 to 8.3 (75 days), 5.9 to 7.9 (240 days) and 5.9 to 8.0 (300 days) respectively whereas at refrigeration storage, this value was varied in the range of 6.5 to 7.9 (26 months), 6.6 to 7.8 (24 months), 6.8 to 7.8 (10 months), 5.9 to 7.6 (26 months) and 5.9 to 7.8 (28 months) respectively [Table 4.4.3.4(C) & 4.4.3.4(D)].

It is also shown in Table 4.4.3.4(E) that, during storage at room temperature dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) tengra fish had pH value varying in the range of 6.0

to 7.9 (210 days) , 6.0 to 8.0 (180 days), 5.9 to 7.9 (120 days), 6.3 to 7.1 (360 days) and 6.4 to 7.2 (420 days) respectively.

However, when Tengra was stored at refrigerated temperature, pH value varying in the range of 6.0 to 6.9 (32 months), 6.0 to 6.9 (28 months), 5.9 to 6.9 (24 months), 6.3 to 7.0 (32 months) and 6.4 to 7.1 (36 months) respectively in case of dry salting (DS), pickle curing (PC), brine salting (BS), sun-dried salting (SDS) and turmeric treated sun-dried salting (SDS+T) [Table 4.4.3.4(F)].

pH value of fresh shol, taki and tengra fish was 6.9, 7.0 and 7.0 in present study (Table 4.1.2), but when salt was added with the fish, pH value decreased (Table 4.3.1) and after that among shelf life period pH value increased in the time interval. These variations in pH values may be attributed to the lipid and protein decomposition and the ratio between both those formed products which affect on the final pH value (Awad, 1999).

According to Huss (1988), the fish products are acceptable up to a pH of 6.8 but are considered to be spoiled above a pH of 7.0. While the initial pH values in present investigation were similar to findings of other researchers, the increase in pH values during the storage of room temperature was higher than the findings of Huss, 1988; Shenderyuk and Bykowski, 1989; Eyo, 1993 and Erkan *et al.*, 2011. The probable reason of these differences is differences in fish species and different methods of salting.

Shenderyuk and Bykowski (1989) opained that, in the post-mortem period, decomposition of nitrogenous compounds leads to an increase in pH in the fish flesh. The increase in pH indicates the loss of quality.

Eyo (1993) stated that pH is an indicator of the extent of microbial spoilage in fish and some proteolytic microbes produce acid after decomposition of carbohydrate, thereby increasing the acid level of the medium. The pH value is a reliable indicator of the degree of freshness or spoilage.

Table: 4.4.3.4(A). Changes in pH value of 5 types of salted shol during different duration of storage period at room temperature (DS = Dry salted, PC = Pickle cured, BS = Brine salted, SDS = Sun-dried salted, SDS+T = Turmeric treated sun-dried salted)

Days of storage period	DS	PC	BS	SDS	SDS+T
0	6.3	6.4	6.5	6.2	6.3
15	6.4	6.5	6.7	6.4	6.4
30	6.5	6.6	6.9	6.5	6.5
45	6.7	6.8	7.2	6.7	6.7
60	6.9	6.9	7.5	6.9	6.9
75	7.1	7.1	7.7	7.2	7.1
90	7.3	7.3	7.9	7.5	7.3
105	7.5	7.5	8.1	7.7	7.4
120	7.7	7.7	8.2	7.8	7.6
135	7.9	8.0	8.3	8.0	7.8
150	8.0	8.3	-	8.1	7.9
165	8.1	-	-	8.3	8.0
180	-	-	-	-	8.2

Table: 4.4.3.4(B). Changes in pH value of 5 types of salted shol during different duration of storage period at refrigeration temperature (4^0C) (DS = Dry salted, PC = Pickle cured, BS = Brine salted, SDS = Sun-dried salted, SDS+T = Turmeric treated sun-dried salted)

Storage period (Month)	DS	PC	BS	SDS	SDS+T
0	6.3	6.4	6.5	6.2	6.3
2	6.3	6.4	6.5	6.2	6.3
4	6.4	6.5	6.6	6.3	6.3
6	6.4	6.5	6.6	6.3	6.4
8	6.5	6.6	6.7	6.4	6.4
10	6.5	6.7	6.8	6.4	6.4
12	6.6	6.8	6.9	6.5	6.5
14	6.7	6.9	7.0	6.6	6.5
16	6.7	6.9	7.1	6.7	6.6
18	6.8	7.0	7.2	6.8	6.7
20	6.9	7.1	-	6.9	6.8
22	7.0	7.2	-	7.0	6.9
24	7.0	-	-	7.1	7.0
26	7.1	-	-	-	7.1
28	-	-	-	-	7.2

Table: 4.4.3.4(C). Changes in pH value of 5 types of salted taki during different duration of storage period at room temperature (DS = Dry salted, PC = Pickle cured, BS = Brine salted, SDS = Sun-dried salted, SDS+T = Turmeric treated sun-dried salted)

Days of storage period	DS	PC	BS	SDS	SDS+T
0	6.5	6.6	6.8	5.9	5.9
15	6.6	6.9	7.0	6.0	6.0
30	6.6	7.1	7.3	6.1	6.1
45	6.7	7.3	7.7	6.2	6.2
60	6.8	7.5	8.0	6.4	6.3
75	7.0	7.8	8.3	6.7	6.4
90	7.2	7.9	-	6.9	6.7
105	7.3	8.1	-	7.0	6.9
120	7.5	8.2	-	7.3	7.0
135	7.7	8.3	-	7.4	7.2
150	7.9	-	-	7.5	7.3
165	-	-	-	7.6	7.4
180	-	-	-	7.7	7.5
195	-	-	-	7.7	7.6
210	-	-	-	7.8	7.7
225	-	-	-	7.8	7.7
240	-	-	-	7.9	7.8
255	-	-	-	-	7.8
270	-	-	-	-	7.9
285	-	-	-	-	7.9
300	-	-	-	-	8.0

Table: 4.4.3.4(D). Changes in pH value of 5 types of salted taki during different duration of storage period at refrigeration temperature (4^0C) (DS = Dry salted, PC = Pickle cured, BS = Brine salted, SDS = Sun-dried salted, SDS+T = Turmeric treated sun-dried salted)

Storage period (Month)	DS	PC	BS	SDS	SDS+T
0	6.5	6.6	6.8	5.9	5.9
2	6.5	6.6	6.9	6.0	6.1
4	6.6	6.7	7.0	6.0	6.2
6	6.6	6.8	7.3	6.1	6.4
8	6.7	7.0	7.6	6.3	6.5
10	6.8	7.1	7.8	6.4	6.6
12	7.0	7.2	-	6.6	6.8
14	7.1	7.3	-	6.8	6.9
16	7.3	7.4	-	6.9	7.0
18	7.4	7.5	-	7.1	7.3
20	7.5	7.6	-	7.3	7.3
22	7.6	7.7	-	7.3	7.4
24	7.8	7.8	-	7.5	7.5
26	7.9	-	-	7.6	7.7
28	-	-	-	-	7.8

Table: 4.4.3.4(E). Changes in pH value of 5 types of salted tengra during different duration of storage period at room temperature (DS = Dry salted, PC = Pickle cured, BS = Brine salted, SDS = Sun-dried salted, SDS+T = Turmeric treated sun-dried salted)

Days of storage period	DS	PC	BS	SDS	SDS+T
0	6.0	6.0	5.9	6.3	6.4
30	6.2	6.4	6.2	6.3	6.4
60	6.6	6.8	7.1	6.3	6.4
90	6.8	7.0	7.5	6.4	6.5
120	7.2	7.5	7.9	6.4	6.5
150	7.5	7.8	-	6.5	6.6
180	7.7	8.0	-	6.5	6.6
210	7.9	-	-	6.6	6.7
240	-	-	-	6.7	6.7
270	-	-	-	6.8	6.8
300	-	-	-	6.9	6.9
330	-	-	-	7.0	7.0
360	-	-	-	7.1	7.0
390	-	-	-	-	7.1
420	-	-	-	-	7.2

Table: 4.4.3.4(F). Changes in pH value of 5 types of salted tengra during different duration of storage period at refrigeration temperature (4^0C) (DS = Dry salted, PC = Pickle cured, BS = Brine salted, SDS = Sun-dried salted, SDS+T = Turmeric treated sun-dried salted)

Storage period (Month)	DS	PC	BS	SDS	SDS+T
0	6.0	6.0	5.9	6.3	6.4
4	6.1	6.1	6.2	6.3	6.4
8	6.1	6.2	6.3	6.4	6.5
12	6.2	6.4	6.4	6.5	6.5
16	6.3	6.5	6.5	6.6	6.6
20	6.4	6.6	6.7	6.7	6.7
24	6.6	6.8	6.9	6.8	6.8
28	6.7	6.9	-	6.9	6.9
32	6.9	-	-	7.0	7.0
36	-	-	-	-	7.1

Gümüş *et al.* (2008) investigated, changes in pH value of 20% brine salted red mullet stored in 4^0C (refrigeration temperature) and found that pH of raw fish was 6.67 but after salting this value slightly decreased to 6.51 whereas at the end of 11 days of storage, this value slightly increased in a range of 6.51 to 6.59 which indicate that pH value of salted fish also increased during refrigeration storage.

The results of pH value of this study also showed similarity with findings of Eltom (1989); Gökoğlu *et al.* (1994); Al-Reza *et al.* (2015).

The findings of increased pH value is parallel to those of Reilly *et al.* (1985), who made comment that the increased pH value can be attributed to the higher levels of volatile nitrogen compounds produced by bacteria and tissue enzymes in fish.

Oehlenschlaeger (1991) investigated on the pH in freshly homogenized sea fish and observed increase of the pH with increasing length of storage time.

The pH is an important factor that affects microbial growth and spoilage of foods (Jay, 1998). Benjakul *et al.* (2002) reported that the decomposition of nitrogenous compounds caused an increase in pH in fish flesh.

According to Nurullah *et al.* (2002), a good relationship between changes in pH and organoleptic qualities of fish was observed, where the quality greatly decreased along with the increase of pH. This finding is similar with present study.

According to Salaudeen *et al.* (2010), the increase in pH was postulated to be due to an increase in volatile bases.

The pH value increased in a characteristic pattern to a certain level of storage period of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) shol, taki and tengra fish-products which is mounted in Figure 4.4.3.4.

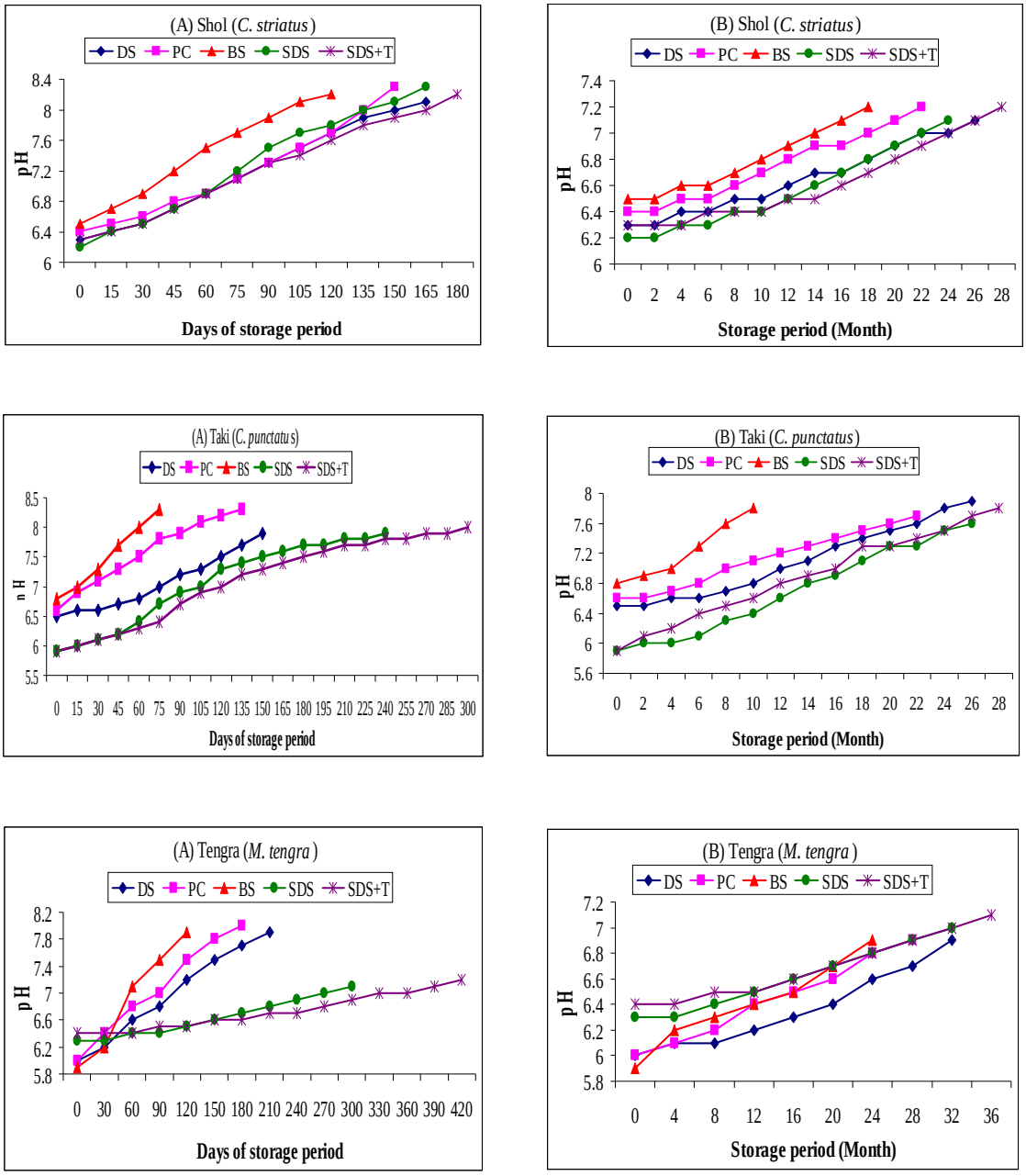


Figure. 4.4.3.4. Effects of different types of salting process on pH value of DS, PC, BS, SDS, SDS+T treated shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish during different duration of storage both at (A) room temperature and (B) refrigeration temperature (DS= Dry salted, PC= Pickle cured, BS= Brine salted, SDS=Sun-dried salted and SDS+T= Turmeric treated sun-dried salted)

The entire figure shows that, pH values of the fish-products increased both at room and refrigeration temperature, but the increment of values in room temperature were more prominent than the product stored in refrigeration temperature.

It is also shown from the Figure that, in case of Shol fish, pH value was prominently highest in BS shol and lowest in SDS+T shol during whole period of shelf life in room and refrigeration storage. Similarly, in case of Taki fish, the highest pH value was in BS taki both room and refrigeration storage and lowest was in SDS+T taki in room temperature, whereas in refrigeration temperature the lowest value was in SDS taki fish products. Likewise, tengra had the highest pH value in BS and lowest was in SDS+T tengra in room temperature, whereas in refrigeration temperature the highest pH value was in SDS+T tengra and lowest value was in DS tengra fish products.

Comparatively higher pH value was observed in five types of salted and sun-dried salted shol and taki fish stored at room temperature is similar with the findings of Azam *et al.* (2003a) who observed that, pH value of stored sun-dried Parshe and Bombay duck was 8.03 and 8.13 respectively. Increase of pH value in stored salted and sun-dried salted shol, taki and tengra fish-samples agrees with the findings of Yatsunami and Takenaka (1996) and Erkan and Ozden (2008) that an increase in pH value during storage period is due to the increase in volatile bases from decomposition of nitrogenous compounds by endogenous or microbial enzymes.

4.4.4. Bacteriological study

The bacteriological examination of fish products was to evaluate the possible presence of bacteria in terms of quantity of public health significance and to give an impression of the hygienic quality including temperature abuse and hygiene during handling and processing (WHO, 1999).

In the trial experiment, there was found no significant variation in the short interval. So, to find out the possible differences, it was scheduled to take data of standard plate count (SPC) and Halophilic bacteria count (HBC) at every 1 month interval at room temperature storage shol, taki and tengra fish-products. On the other hand in case of refrigeration storage fish-products, these data were taken every 2 month interval in shol and taki fish and every 4 month interval in tengra fish.

4.4.4.1. Estimation of standard plate count (SPC)

The standard plate count (SPC) of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) shol, taki and tengra fish-products during different duration of storage period both at room and refrigeration temperature was studied and the results are presented in Table 4.4.4.1(A) - (F).

In present study, the range of SPC of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) **shol** fish products during storage at room temperature were 2.7×10^3 to 1.7×10^6, 3.6×10^3 to 2.0×10^6, 2.1×10^4 to 3.7×10^6, 2.4×10^3 to 1.3×10^6 and 2.1×10^3 to 1.6×10^6 cfu/g of fish

whereas at refrigeration storage, this value varied from 2.7×10^3 to 1.9×10^6, 3.6×10^3 to 2.2×10^6, 2.1×10^4 to 4.3×10^6, 2.4×10^3 to 1.6×10^6 and 2.1×10^3 to 2.2×10^6 cfu/g of fish.

In case of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) **taki** fish products, SPC ranged from 4.0×10^3 to 2.9×10^6, 3.0×10^4 to 2.5×10^6, 4.3×10^4 to 3.9×10^6, 3.9×10^3 to 2.1×10^6 and 2.8×10^3 to 1.7×10^6 cfu/g of fish during storage at room temperature and 4.0×10^3 to 3.3×10^6, 3.0×10^4 to 4.2×10^6, 4.3×10^4 to 5.1×10^6, 3.9×10^3 to 3.0×10^6 and 2.8×10^3 to 2.5×10^6 cfu/g of fish respectively during storage at refrigeration temperature.

On the other hand, in dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) **tengra** fish products this value ranged from 3.8×10^3 to 1.8×10^6, 1.2×10^4 to 2.0×10^6, 1.5×10^4 to 4.0×10^6, 2.8×10^3 to 1.3×10^6 and 2.5×10^3 to 1.4×10^6 cfu/g of fish during storage at room temperature and 3.8×10^3 to 2.1×10^6, 1.2×10^4 to 3.3×10^6, 1.5×10^4 to 3.0×10^6, 2.8×10^3 to 1.8×10^6 and 2.5×10^3 to 1.7×10^6 cfu/g of fish respectively during storage at refrigeration temperature.

According to Hobbs (1962), when total viable bacterial counts reaches to 10^6 cfu/g or more in processed food and food products, it is considered that these food items are spoiled. In another report, when aerobic plate counts reach 10^6 cfu/g, the food product was assumed to be at or near spoilage (Pascual and Calderón 2000; Sivertsvik, *et al.*, 2002; Arashisar *et al*., 2004; Ozogul *et al*., 2004).

In Bangladesh, DOF and BSTI recommended the SPC of processed fish to be not more than 10^6 cfu/g. But according to Ojagh *et al.* (2010) if any sample contains more than 10^8 cfu/g bacterial counts then these microbes can cause spoilage of that product. In this study, the total viable count of salted samples was varied during storage time but evaluation of these salted products were within the limits of 10^7cfu/g specified for quality grading of fish by the International Commission of Microbiological Standards for Foods (ICMSF, 1996).

Table: 4.4.4.1(A). Changes in standard plate count (SPC) (cfu/g) of 5 types of salted shol (*C. striatus*) during different duration of storage at room temperature (DS= Dry salted, PC= Pickle cured, BS= Brine salted, SDS= Sun-dried salted and SDS+T= Turmeric treated sun-dried salted)

Storage period (Month)	DS	PC	BS	SDS	SDS+T
0	2.7×10^3	3.6×10^3	2.1×10^4	2.4×10^3	2.1×10^3
1	1.1×10^4	0.9×10^4	8.2×10^4	0.7×10^4	5.1×10^3
2	2.1×10^4	3.3×10^4	3.4×10^5	2.6×10^4	2.4×10^4
3	7.2×10^4	6.7×10^4	9.0×10^5	6.6×10^4	7.0×10^4
4	3.5×10^5	5.5×10^5	3.7×10^6	3.1×10^5	3.5×10^5
5	1.7×10^6	2.0×10^6	-	1.3×10^6	8.3×10^5
6	-	-	-	-	1.6×10^6

Table: 4.4.4.1(B). Changes in standard plate count (SPC) (cfu/g) of 5 types of salted shol (*C. striatus*) during different duration of storage at refrigeration temperature (4^0C) (DS= Dry salted, PC= Pickle cured, BS= Brine salted, SDS= Sun-dried salted and SDS+T= Turmeric treated sun-dried salted)

Storage period (Month)	DS	PC	BS	SDS	SDS+T
0	2.7×10^3	3.6×10^3	2.1×10^4	2.4×10^3	2.1×10^3
2	3.3×10^3	4.4×10^3	2.9×10^4	3.1×10^3	2.8×10^3
4	4.2×10^3	5.2×10^3	4.1×10^4	4.5×10^3	3.9×10^3
6	5.7×10^3	7.6×10^3	4.6×10^4	6.6×10^3	5.1×10^3
8	8.2×10^3	1.0×10^4	5.8×10^4	8.8×10^3	8.3×10^3
10	1.4×10^4	1.5×10^4	7.2×10^4	1.1×10^4	1.3×10^4
12	2.2×10^4	2.4×10^4	9.5×10^4	1.7×10^4	2.1×10^4
14	3.7×10^4	4.3×10^4	3.5×10^5	2.7×10^4	3.5×10^4
16	4.4×10^4	6.6×10^4	6.3×10^5	4.0×10^4	4.7×10^4
18	6.2×10^4	2.3×10^5	4.3×10^6	7.7×10^4	5.2×10^4
20	1.9×10^5	8.0×10^5	-	2.1×10^5	6.8×10^4
22	3.5×10^5	2.2×10^6	-	8.3×10^5	1.5×10^5
24	7.2×10^5	-	-	1.6×10^6	4.1×10^5
26	1.9×10^6	-	-	-	9.0×10^5
28	-	-	-	-	2.2×10^6

Table: 4.4.4.1(C). Changes in standard plate count (SPC) (cfu/g) of 5 types of salted taki (*C. punctatus*) during different duration of storage at room temperature (DS= Dry salted, PC= Pickle cured, BS= Brine salted, SDS= Sun-dried salted and SDS+T= Turmeric treated sun-dried salted)

Storage period (Month)	DS	PC	BS	SDS	SDS+T
0	4.0×10^{3}	3.0×10^{4}	4.3×10^{4}	3.9×10^{3}	2.8×10^{3}
1	2.5×10^{4}	7.5×10^{4}	5.5×10^{5}	6.6×10^{3}	3.4×10^{3}
2	6.8×10^{4}	2.1×10^{5}	3.9×10^{6}	1.1×10^{4}	4.0×10^{3}
3	3.3×10^{5}	8.3×10^{5}	-	2.2×10^{4}	6.2×10^{3}
4	7.3×10^{5}	2.5×10^{6}	-	5.6×10^{4}	0.9×10^{4}
5	2.9×10^{6}	-	-	8.3×10^{4}	1.6×10^{4}
6	-	-	-	2.4×10^{5}	3.5×10^{4}
7	-	-	-	8.0×10^{5}	7.3×10^{4}
8	-	-	-	2.1×10^{6}	1.9×10^{5}
9	-	-	-	-	6.8×10^{5}
10	-			-	1.7×10^{6}

Table: 4.4.4.1(D). Changes in standard plate count (SPC) (cfu/g) of 5 types of salted taki (*C. punctatus*) during different duration of storage at refrigeration temperature (4^{0}C) (DS= Dry salted, PC= Pickle cured, BS= Brine salted, SDS= Sun-dried salted and SDS+T= Turmeric treated sun-dried salted)

Storage period (Month)	DS	PC	BS	SDS	SDS+T
0	4.0×10^{3}	3.0×10^{4}	4.3×10^{4}	3.9×10^{3}	2.8×10^{3}
2	4.9×10^{3}	3.6×10^{4}	7.2×10^{4}	4.5×10^{3}	4.2×10^{3}
4	5.6×10^{3}	4.9×10^{4}	1.6×10^{5}	6.1×10^{3}	5.0×10^{3}
6	7.0×10^{3}	6.0×10^{4}	4.0×10^{5}	7.3×10^{3}	6.5×10^{3}
8	9.1×10^{3}	7.5×10^{4}	8.5×10^{5}	1.3×10^{4}	8.6×10^{3}
10	1.0×10^{4}	8.8×10^{4}	5.1×10^{6}	2.0×10^{4}	1.2×10^{4}
12	1.3×10^{4}	1.1×10^{5}	-	2.8×10^{4}	1.6×10^{4}
14	2.3×10^{4}	1.9×10^{5}	-	3.7×10^{4}	2.0×10^{4}
16	3.0×10^{4}	2.2×10^{5}	-	4.6×10^{4}	2.7×10^{4}
18	4.5×10^{4}	4.2×10^{5}	-	7.3×10^{4}	3.3×10^{4}
20	1.8×10^{5}	5.5×10^{5}	-	1.6×10^{5}	5.1×10^{4}
22	3.4×10^{5}	7.9×10^{5}	-	3.3×10^{5}	9.0×10^{4}
24	8.1×10^{5}	4.2×10^{6}	-	6.1×10^{5}	2.3×10^{5}
26	3.3×10^{6}	-	-	3.0×10^{6}	7.2×10^{5}
28	-	-	-	-	2.5×10^{6}

Table: 4.4.4.1(E). Changes in standard plate count (SPC) (cfu/g) of 5 types of salted tengra (*M. tengra*) during different duration of storage at room temperature (DS= Dry salted, PC= Pickle cured, BS= Brine salted, SDS= Sun-dried salted and SDS+T= Turmeric treated sun-dried salted)

Storage period (Month)	DS	PC	BS	SDS	SDS+T
0	3.8×10^3	1.2×10^4	1.5×10^4	2.8×10^3	2.5×10^3
1	5.5×10^3	2.1×10^4	2.5×10^5	3.3×10^3	4.1×10^3
2	9.1×10^3	3.3×10^4	5.5×10^5	6.2×10^3	6.5×10^3
3	2.3×10^4	6.2×10^4	7.2×10^5	7.7×10^3	7.7×10^3
4	6.5×10^4	1.8×10^5	4.0×10^6	1.1×10^4	9.4×10^3
5	2.9×10^5	7.3×10^5	-	2.0×10^4	1.3×10^4
6	7.2×10^5	2.0×10^6	-	4.4×10^4	2.5×10^4
7	1.8×10^6	-	-	8.5×10^4	4.2×10^4
8	-	-	-	1.8×10^5	5.9×10^4
9	-	-	-	3.0×10^5	8.2×10^4
10	-	-	-	4.3×10^5	1.5×10^5
11	-	-	-	7.0×10^5	3.2×10^5
12	-	-	-	1.3×10^6	4.6×10^5
13	-	-	-	-	7.8×10^5
14	-	-	-	-	1.4×10^6

Table: 4.4.4.1(F). Changes in standard plate count (SPC) (cfu/g) of 5 types of salted tengra (*M. tengra*) during different duration of storage at refrigeration temperature (4^0C) (DS= Dry salted, PC= Pickle cured, BS= Brine salted, SDS= Sun-dried salted and SDS+T= Turmeric treated sun-dried salted)

Storage period (Month)	DS	PC	BS	SDS	SDS+T
0	3.8×10^3	1.2×10^4	1.5×10^4	2.8×10^3	2.5×10^3
4	6.2×10^3	2.6×10^4	4.1×10^4	4.1×10^3	4.8×10^3
8	8.2×10^3	5.1×10^4	7.2×10^4	1.8×10^4	6.6×10^3
12	1.3×10^4	8.7×10^4	2.4×10^5	3.1×10^4	1.4×10^4
16	2.6×10^4	1.6×10^5	3.8×10^5	5.1×10^4	2.0×10^4
20	4.6×10^4	6.2×10^5	7.5×10^5	2.3×10^5	4.0×10^4
24	2.5×10^5	9.0×10^5	3.0×10^6	4.2×10^5	5.8×10^4
28	7.7×10^5	3.3×10^6	-	8.0×10^5	2.6×10^5
32	2.1×10^6	-	-	1.8×10^6	5.5×10^5
36	-	-	-	-	1.7×10^6

The present study showed that, in case of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) shol fish products, stored at room temperature, the highest SPC was found 3.7×10^6 cfu/g in brine salted (BS) products in 4 month of storage period which was in acceptable limit of microbiological standard and the lowest SPC was found 1.3×10^6 in 5 months stored sun-dried salted (SDS) products, whereas in refrigeration storage, the highest and lowest SPC was also found in brine and sun dried salted shol but at the end of 18 and 24 months of storage period respectively [Table 4.4.4.1(A) & Table 4.4.4.1(B)].

It was also shown in Table 4.4.4.1(C) that, among 5 types of salted taki fish products, both room and refrigeration storage, the highest SPC was found in brine salted (BS) products at the end of 2 months (3.9×10^6 cfu/g) and 10 months (5.1×10^6 cfu/g) whereas the lowest SPC was found sun dried salted (SDS) products at the end of shelf life of 10 months (1.7×10^6 cfu/g) and 28 months (2.5×10^6 cfu/g) respectively [Table4.4.4.1(D)] .

On the other hand, in case of 5 types of salted tengra fish products, the highest SPC was found in brine salted (BS) products in 4 months (4.0×10^6 cfu/g) of storage and the lowest SPC was found in sun dried salted (SDS) products in 12 months (1.3×10^6 cfu/g) of storage at room temperature whereas during storage at refrigeration temperature the highest and lowest SPC was found in pickle cured (PC) and turmeric treated sun-dried salted (SDS+T) products at the end of 28 months and 36 months of shelf life respectively [Table 4.4.4.1(E) & Table 4.4.4.1(F)].

Effects of different types of salting process on standard plate count (cfu/g) of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish-products during different duration of storage both at room and refrigeration temperature is presented in Figure 4.4.4.1.

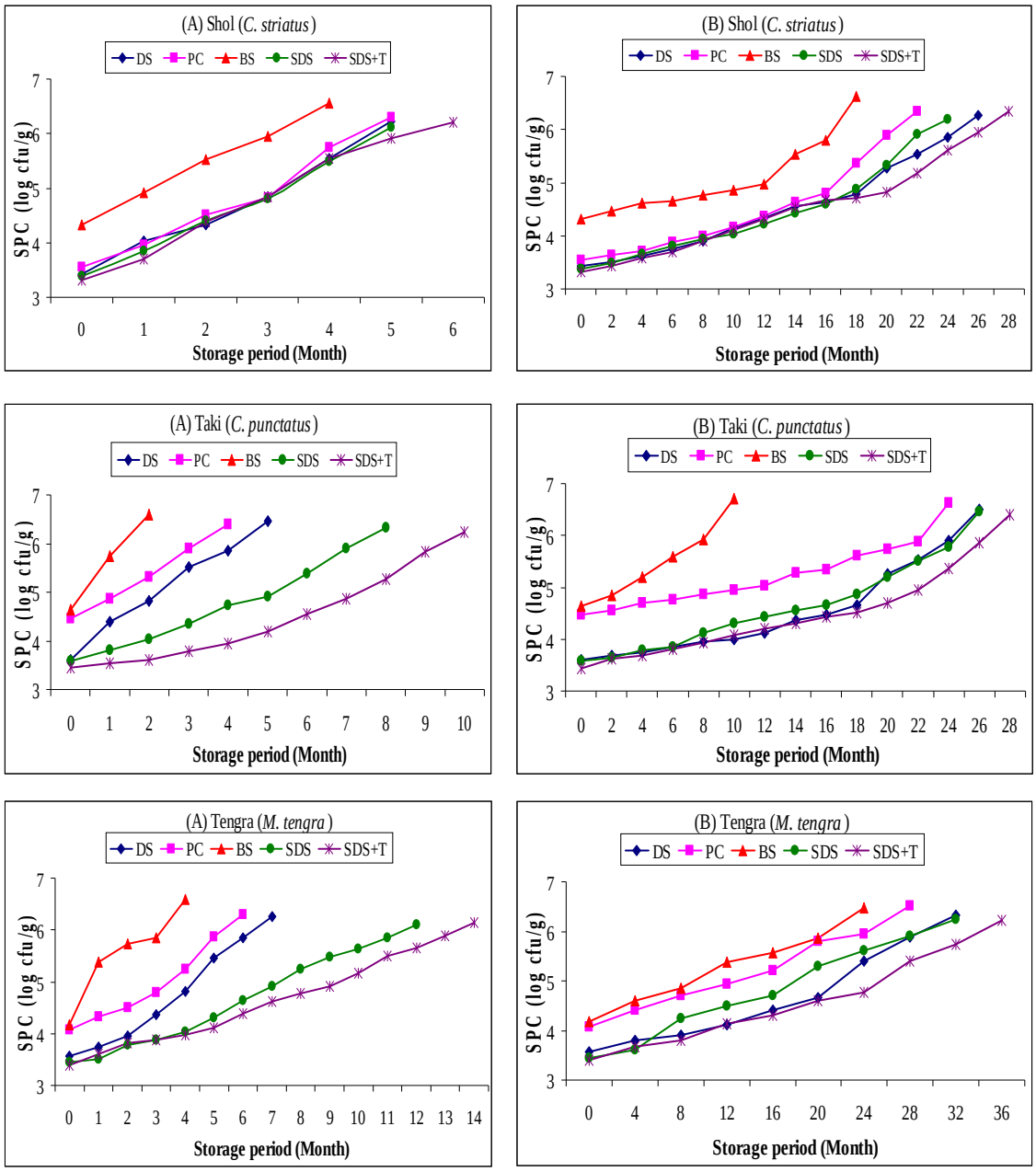


Figure. 4.4.4.1. Changes in standard plate count (cfu/g) of shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish of five different treatments (DS- dry salting, PC- pickle curing, BS- brine salting, SDS- sun dried salting, SDS+T- turmeric treated sun-dried salting) during different duration of storage both at (A) room and (B) refrigeration temperature

From all these figures, it was shown that total number of bacteria varied from one sampling to another. Gradual increase of SPC count of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) shol, taki and tengra fish during different duration of storage both room and refrigeration temperature was observed due to increase in moisture content of the product.

It was also observed from Figure 4.4.4.1 that, the sun dried salted (SDS+T) and turmeric treated sun dried salted (SDS+T) shol, taki and tengra fish products had relatively low bacterial count and higher shelf life. This result reveals that use of salt and turmeric together yielded best outcome compared to the salt alone. Salt removed water from fish body and thus helped through salting, whereas the salt and turmeric not only removed water from fish but also added some nutrients that prevented the growth of moulds and bacteria due to the creation of an unfavorable expansion medium retaining the good taste. The antimicrobial property of turmeric may have contributed to the acceptable microbial quality of the turmeric treated sun-dried salted samples at the end of storage period.

According to Akande *et al*, (1991) the presence of spices which are bactericidal in nature may have accounted for the low counts obtained. The introduction of heat during Sun-dried salting (SDS) and turmeric treated sun-dried salting (SDS+T) methods employed would not only kill microorganisms but will also reduce the moisture content of the fish muscles making the environment less favorable for microbial growth. However, heat does not destroy all organisms as thermophiles may survive in dried fish after heating. The inclusion of salt and turmeric along with heating of sun (in case of SDS+T) usually provides a more efficient method of salting.

Jones (1962) stated that fish enzyme activities occur at higher temperature in tropical fish. So, it is evident that enzymatic activities for fish spoilage were higher in case of fish kept at room temperature. It is in harmony with present study. Lower temperatures (refrigeration temperature) gave lower count regardless of relative humidity of drying. According to Lopez, *et al*. (1998); Juneja and Marmer (1998), the

microbial deactivation kinetics depends on several factors: variety, water content (i.e., water activity), temperature, composition of the medium (acidity, types of solids, pH, etc.), as well as salting and drying method.

According to Gümüş *et al.* (2008) the total mesophilic aerobic count of 20% brine salted red mullet was 2.84 log cfu/g at the beginning of the storage at 4^0C and increased in 5.70 log cfu/g in 11^{th} day. From this research, it was showed that SPC increased during refrigeration storage which is similar with my present study. The absence of pathogenic microbes in salted fishes is reported by Kakatkar *et al.* (2010). Lilabati *et al.* (1999) and Prakash *et al.* (2011) reported the direct relationship between the microbial count and moisture content of salted-dried fish sample.

Temperature changes have greater impact on microbiological growth than on enzymatic activity and many bacteria are unable to grow at temperatures below 10°C (Huss, 1995). Magnusson *et al.* (2006) opeined that at 5^0 C temperature, 20% salted cod was a stable product microbiologically till 11 months of refrigeration storage.

SPC of fish muscles increased significantly with increase in the duration of storage. As the duration of storage increase, processed fish samples may absorb small amounts of moisture from surrounding atmosphere providing enabling environment for microbial growth (Eyo, 2006). According to Kumar *et al.* (2013), SPC increased from 2.94×10^2 to 2.10×10^3 cfu/g respectively in case of 6 month stored 30% salted sun-dried *L. gonius* fish at room temperature and showed a positive correlation of total plate count (TPC) with TVB-N but was significantly negatively correlated with overall acceptability which is similar with present findings. The increase of SPC in fish flesh during storage has been demonstrated by Bahmani *et al.* (2011).

According to the findings of Nurullah *et al.* (2007), the initial APC of 5 sun-dried SIS species was in the range of 5.3×10^4 -7.3×10^4 cfu/g. The bacterial load increased slowly and at the end of the 60 days storage reached to the range 6.6×10^6-8.6×10^7 cfu/g. This finding was similar with present findings. Hood *et al.* (1983) reported that microbial load increases with duration of storage and temperature.

No yeast or mould was detected in our salted and salted-dried samples till the end of storage. Similar result was revealed by Bahri *et al*. (2006) from salted Grey Mullet.

Thus, the relative increase in microbial load of 5 types of salted shol taki and tengra fish products stored both at room (26-32^{0}C) and refrigeration (4°C) temperature is an important indicator of quality and influences the time for which salted fish can be kept.

The results of the study on bacterial load in 3 types of fish products showed that total bacterial load were comparatively low. The ranges were within the acceptable limit and there is a positive relationship between moisture content and bacterial growth in fish. However, the number of total bacteria will be related to the remaining shelf life which is in agreement with Huss (1995) and Gram *et al*. (1987, 1990).

According to Ólafsdóttir *et al.* (1997) an estimation of the total viable counts (TVC) or total aerobic plate count is usually used as an acceptability index in standards, guidelines and specification. According to Frazier and Westhoff (1998) micro-organisms proliferate in the presence of water which is in agreement with present study. Various reports in the literature (Simidu, 1961) described that a fair percent of the total nitrogen is being utilized for bacterial metabolism.

According to Mansur *et al.* (2014), a correlation was found between bacterial count and total volatile base nitrogen. The sample with high total volatile base nitrogen showed maximum bacterial count. Comparatively samples with low total volatile base nitrogen had minimum bacterial count. On the other hand, high moisture content promoted the growth of microorganisms and accelerated the formation of total volatile base nitrogen irrespective of the samples analyzed. This finding is in accordance with present study.

4.4.4.2. Estimation of halophilic bacterial count (HBC)

In case of Shol fish, during storage at room temperature HBC (halophilic bacterial count) was varying in the range of 3.0×10^2 to 2.1×10^5 (5 months), 4.1×10^2 to 2.4×10^5 (5 months), 4.0×10^3 to 3.4×10^5 (4 months), 2.5×10^2 to 1.5×10^5 (5 months) and 2.0×10^2 to 1.8×10^5 cfu/g (6 months) respectively in case of dry salting (DS), pickle curing (PC), brine salting (BS), sun-dried salting (SDS) and sun-dried salting + turmeric (SDS+T) [Table 4.4.4.2(A)].

In case of Shol fish, during storage at refrigerated temperature HBC (halophilic bacterial count) was varying in the range of 3.0×10^2 to 2.6×10^5 (26 months), 4.1×10^2 to 3.1×10^5 (22 months), 4.0×10^3 to 3.8×10^5 (18 months), 2.5×10^2 to 2.0×10^5 (24 months) and 2.0×10^2 to 2.3×10^5 cfu/g (28 months) respectively in case of dry salting (DS), pickle curing (PC), brine salting (BS), sun-dried salting (SDS) and sun-dried salting + turmeric (SDS+T) [Table 4.4.4.2(B)].

It is also shown in Table 4.4.4.2(C) that, during storage at room temperature Taki fish had HBC (halophilic bacterial count), varying in the range of 3.0×10^2 to 1.9×10^5 (5 months), 3.3×10^3 to 2.9×10^5 (4 months), 4.1×10^3 to 3.2×10^5 (2 months), 2.4×10^2 to 2.5×10^5 (8 months) and 2.1×10^2 to 3.0×10^5 cfu/g (10 months) respectively in case of dry salting (DS), pickle curing (PC), brine salting (BS), sun-dried salting (SDS) and sun-dried salting + turmeric (SDS+T) whereas at refrigeration storage, this value was varying in the range of 3.0×10^2 to 4.1×10^5 (26 months), 3.3×10^3 to 3.5×10^5 (24 months), 4.1×10^3 to 3.9×10^5 (10 months), 2.4×10^2 to 3.2×10^5 (26 months) and 2.1×10^2 to 4.5×10^5 cfu/g (28 months) respectively [Table 4.4.4.2(D)].

In case of Tengra fish, during storage at room temperature HBC (halophilic bacterial count) was varying in the range of 3.2×10^2 to 2.6×10^5 (7 months), 1.4×10^3 to 6.0×10^5 (6 months), 1.8×10^3 to 7.8×10^5 (4 months), 1.5×10^2 to 5.0×10^5 (12 months) and 1.2×10^2 to 5.5×10^5 cfu/g (14 months) respectively in case of dry salting (DS), pickle

curing (PC), brine salting (BS), sun-dried salting (SDS) and sun-dried salting + turmeric (SDS+T) [Table 4.4.4.2(E)]. On the other hand, at refrigeration storage, HBC (halophilic bacterial count) was varying in the range of 3.2×10^2 to 3.2×10^5 (32 months), 1.4×10^3 to 3.1×10^5 (28 months), 1.8×10^3 to 5.0×10^5 (24 months), 1.5×10^2 to 2.8×10^5 (32 months) and 1.2×10^2 to 5.5×10^5 cfu/g (36 months) respectively in case of dry salting (DS), pickle curing (PC), brine salting (BS), sun-dried salting (SDS) and sun-dried salting + turmeric (SDS+T) [Table 4.4.4.2(F)].

Effect of different types of salting process on Halophilic bacterial count (HBC) of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish during different duration of storage both at room and refrigeration temperature is presented in Table 4.4.4.2(A)-(F).

Table: 4.4.4.2(A). Changes in halophilic bacterial count (HBC) (cfu/g) of 5 types of salted shol (*C. striatus*) during different duration of storage at room temperature (DS= Dry salted, PC= Pickle cured, BS= Brine salted, SDS= Sun-dried salted and SDS+T= Turmeric treated sun-dried salted)

Storage period (Month)	DS	PC	BS	SDS	SDS+T
0	3.0×10^{2}	4.1×10^{2}	4.0×10^{3}	2.5×10^{2}	2.0×10^{2}
1	1.1×10^{3}	8.1×10^{2}	2.5×10^{4}	5.5×10^{2}	5.1×10^{2}
2	4.5×10^{3}	2.0×10^{3}	7.8×10^{4}	3.0×10^{3}	2.2×10^{3}
3	2.2×10^{4}	2.6×10^{4}	2.6×10^{5}	7.8×10^{3}	6.5×10^{3}
4	7.1×10^{4}	8.8×10^{4}	3.4×10^{5}	4.5×10^{4}	3.1×10^{4}
5	2.1×10^{5}	2.4×10^{5}	-	1.5×10^{5}	7.5×10^{4}
6	-	-	-	-	1.8×10^{5}

Table: 4.4.4.2(B). Changes in halophilic bacterial count (HBC) (cfu/g) of 5 types of salted shol (*C. striatus*) during different duration of storage at refrigeration temperature (4^{0}C) (DS= Dry salted, PC= Pickle cured, BS= Brine salted, SDS= Sun-dried salted and SDS+T= Turmeric treated sun-dried salted)

Storage period (Month)	DS	PC	BS	SDS	SDS+T
0	3.0×10^{2}	4.1×10^{2}	4.0×10^{3}	2.5×10^{2}	2.0×10^{2}
2	4.8×10^{2}	7.2×10^{2}	4.8×10^{3}	4.4×10^{2}	3.1×10^{2}
4	8.0×10^{2}	1.1×10^{3}	5.3×10^{3}	1.3×10^{3}	6.1×10^{2}
6	1.2×10^{3}	1.9×10^{3}	7.3×10^{3}	3.1×10^{3}	1.1×10^{3}
8	2.3×10^{3}	4.4×10^{3}	1.7×10^{4}	6.3×10^{3}	1.8×10^{3}
10	2.8×10^{3}	6.5×10^{3}	3.3×10^{4}	7.6×10^{3}	2.2×10^{3}
12	3.9×10^{3}	1.7×10^{4}	8.8×10^{4}	0.9×10^{4}	3.4×10^{3}
14	6.6×10^{3}	2.4×10^{4}	2.1×10^{5}	1.6×10^{4}	4.8×10^{3}
16	1.6×10^{4}	4.3×10^{4}	3.1×10^{5}	3.1×10^{4}	8.0×10^{3}
18	2.9×10^{4}	8.0×10^{4}	3.8×10^{5}	4.4×10^{4}	1.3×10^{4}
20	5.5×10^{4}	1.6×10^{5}	-	7.3×10^{4}	2.3×10^{4}
22	9.0×10^{4}	3.1×10^{5}	-	1.1×10^{5}	3.9×10^{4}
24	1.6×10^{5}	-	-	2.0×10^{5}	5.5×10^{4}
26	2.6×10^{5}	-	-	-	9.2×10^{4}
28	-	-	-	-	2.3×10^{5}

Table: 4.4.4.2(C). Changes in halophilic bacterial count (HBC) (cfu/g) of 5 types of salted taki (*C. punctatus*) during different duration of storage at room temperature (DS= Dry salted, PC= Pickle cured, BS= Brine salted, SDS= Sun-dried salted and SDS+T= Turmeric treated sun-dried salted)

Storage period (Month)	DS	PC	BS	SDS	SDS+T
0	3.0×10^{2}	3.3×10^{3}	4.1×10^{3}	2.4×10^{2}	2.1×10^{2}
1	5.5×10^{3}	2.0×10^{4}	5.5×10^{4}	4.0×10^{2}	3.0×10^{2}
2	7.3×10^{3}	6.7×10^{4}	3.2×10^{5}	1.1×10^{3}	4.4×10^{2}
3	2.0×10^{4}	2.6×10^{5}	-	1.9×10^{3}	5.9×10^{2}
4	6.5×10^{4}	2.9×10^{5}	-	6.6×10^{3}	1.2×10^{3}
5	1.9×10^{5}	-	-	1.5×10^{4}	2.6×10^{3}
6	-	-	-	3.3×10^{4}	7.2×10^{3}
7	-	-	-	5.6×10^{4}	2.1×10^{4}
8	-	-	-	2.5×10^{5}	4.6×10^{4}
9	-	-	-	-	8.1×10^{4}
10	-			-	3.0×10^{5}

Table: 4.4.4.2(D). Changes in halophilic bacterial count (HBC) (cfu/g) of 5 types of salted taki (*C. punctatus*) during different duration of storage at refrigeration temperature (4^{0}C) (DS= Dry salted, PC= Pickle cured, BS= Brine salted, SDS= Sun-dried salted and SDS+T= Turmeric treated sun-dried salted)

Storage period (Month)	DS	PC	BS	SDS	SDS+T
0	3.0×10^{2}	3.3×10^{3}	4.1×10^{3}	2.4×10^{2}	2.1×10^{2}
2	3.9×10^{2}	3.9×10^{3}	2.5×10^{4}	2.9×10^{2}	2.8×10^{2}
4	4.4×10^{2}	8.2×10^{3}	6.2×10^{4}	3.5×10^{2}	3.4×10^{2}
6	6.4×10^{2}	0.9×10^{4}	1.7×10^{5}	1.0×10^{3}	3.9×10^{2}
8	0.81×10^{3}	1.3×10^{4}	2.2×10^{5}	1.6×10^{3}	4.6×10^{2}
10	1.3×10^{3}	1.7×10^{4}	3.9×10^{5}	2.0×10^{3}	0.91×10^{3}
12	2.1×10^{3}	2.4×10^{4}	-	3.9×10^{3}	1.5×10^{3}
14	5.0×10^{3}	4.4×10^{4}	-	5.3×10^{3}	2.0×10^{3}
16	1.5×10^{4}	5.1×10^{4}	-	6.1×10^{3}	3.3×10^{3}
18	6.9×10^{4}	7.3×10^{4}	-	8.1×10^{3}	5.3×10^{3}
20	9.3×10^{4}	2.0×10^{5}	-	1.8×10^{4}	1.7×10^{4}
22	2.2×10^{5}	2.9×10^{5}	-	2.6×10^{4}	2.5×10^{4}
24	3.8×10^{5}	3.5×10^{5}	-	7.1×10^{4}	4.8×10^{4}
26	4.1×10^{5}	-	-	3.2×10^{5}	6.8×10^{4}
28	-	-	-	-	4.5×10^{5}

Table: 4.4.4.2(E). Changes in halophilic bacterial count (HBC) (cfu/g) of 5 types of salted tengra (*M. tengra*) fish during different duration of storage at room temperature (DS= Dry salted, PC= Pickle cured, BS= Brine salted, SDS= Sun-dried salted and SDS+T= Turmeric treated sun-dried salted)

Storage period (Month)	DS	PC	BS	SDS	SDS+T
0	3.2×10^2	1.4×10^3	1.8×10^3	1.5×10^2	1.2×10^2
1	6.8×10^2	5.3×10^3	3.1×10^4	3.1×10^2	2.1×10^2
2	2.1×10^3	1.1×10^4	6.1×10^4	5.4×10^2	2.9×10^2
3	5.4×10^3	2.3×10^4	2.5×10^5	0.8×10^3	5.2×10^2
4	1.6×10^4	4.8×10^4	7.8×10^5	2.2×10^3	1.1×10^3
5	3.6×10^4	9.3×10^4	-	3.6×10^3	1.8×10^3
6	7.6×10^4	6.0×10^5	-	1.3×10^4	3.6×10^3
7	2.6×10^5	-	-	2.4×10^4	5.5×10^3
8	-	-	-	4.8×10^4	8.1×10^3
9	-	-	-	7.3×10^4	3.1×10^4
10	-	-	-	1.9×10^5	6.1×10^4
11	-	-	-	3.8×10^5	8.8×10^4
12	-	-	-	5.0×10^5	1.5×10^5
13	-	-	-	-	3.3×10^5
14	-	-	-	-	5.5×10^5

Table: 4.4.4.2(F). Changes in halophilic bacterial count (HBC) (cfu/g) of 5 types of salted tengra (*M. tengra*) during different duration of storage at refrigeration temperature (4^0C) (DS= Dry salted, PC= Pickle cured, BS= Brine salted, SDS= Sun-dried salted and SDS+T= Turmeric treated sun-dried salted)

Storage period (Month)	DS	PC	BS	SDS	SDS+T
0	3.2×10^2	1.4×10^3	1.8×10^3	1.5×10^2	1.2×10^2
4	4.2×10^2	3.6×10^3	4.1×10^3	3.5×10^2	2.5×10^2
8	7.0×10^2	7.1×10^3	6.6×10^3	2.5×10^3	1.8×10^3
12	2.5×10^3	1.6×10^4	2.5×10^4	4.1×10^3	3.6×10^3
16	4.1×10^3	3.4×10^4	5.1×10^4	6.5×10^3	7.8×10^3
20	8.8×10^3	5.1×10^4	3.2×10^5	3.3×10^4	2.2×10^4
24	1.9×10^4	1.8×10^5	5.0×10^5	5.5×10^4	4.8×10^4
28	3.9×10^4	3.1×10^5	-	8.1×10^4	7.6×10^4
32	3.2×10^5	-	-	2.8×10^5	3.5×10^5
36	-	-	-	-	5.5×10^5

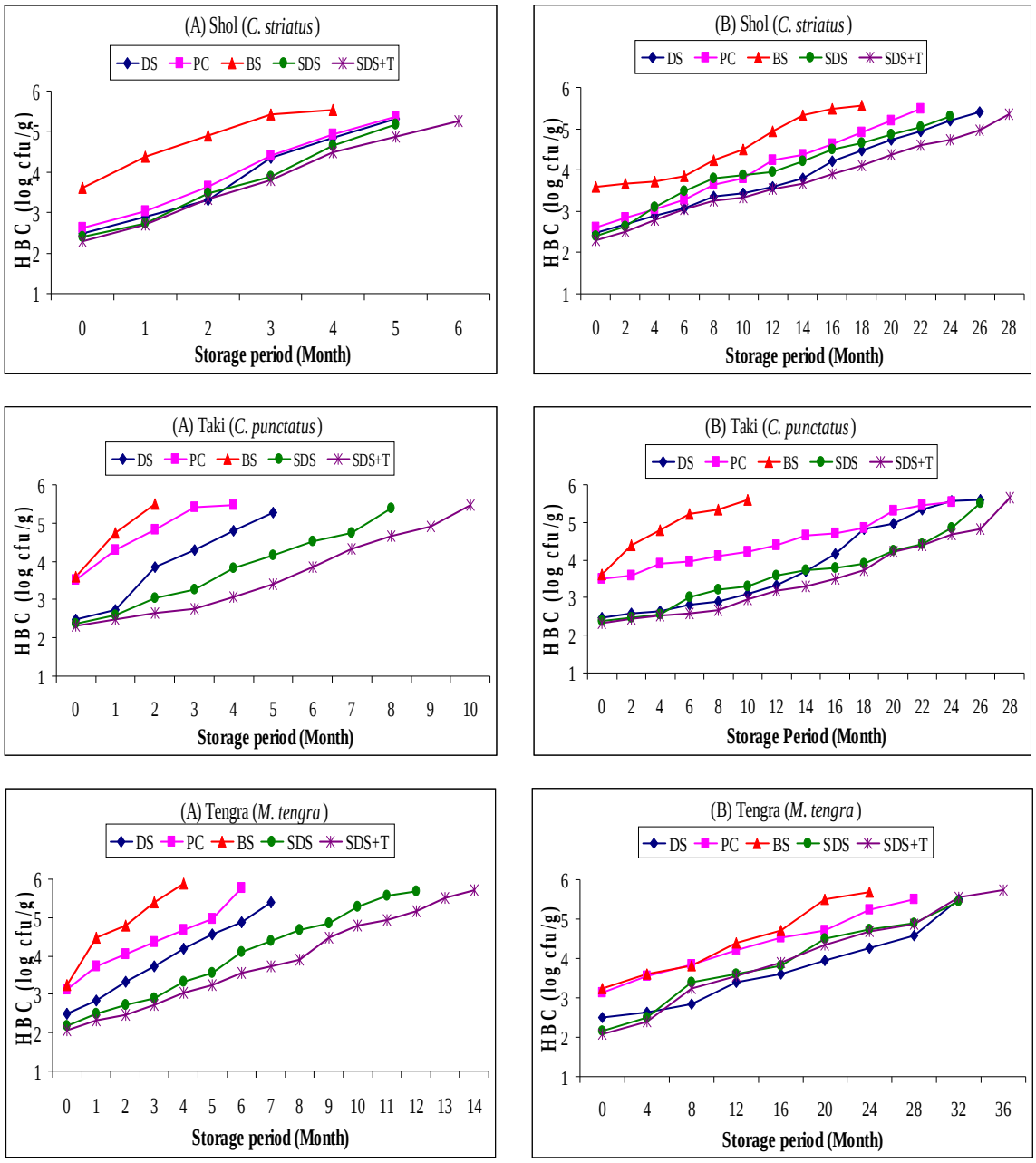


Figure. 4.4.4.2. Changes in halophilic bacterial count (cfu/g) of shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish of five different treatments (DS- dry salting, PC- pickle curing, BS- brine salting, SDS- sun dried salting, SDS+T- turmeric treated sun-dried salting) during different duration of storage both at (A) room and (B) refrigeration temperature

Changes in halophilic bacterial count (cfu/g) of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) shol (*C. striatus*), taki (*C. punctatus*) and tengra (*M. tengra*) fish during different duration of storage both at room and refrigeration temperature is presented in Figure 4.4.4.2.

Gradual increases of HBC was observed during storage both room and refrigeration temperature may be due to increase in moisture content of the product and this value was prominently highest in BS shol and taki and lowest in SDS+T shol and taki fish. In case of Tengra fish, the highest HBC was in BS tengra and lowest was in SDS+T tengra in room temperature, whereas in refrigeration temperature the highest HBC was in BS tengra but lowest value was in DS tengra fish products (Figure 4.4.4.2).

According to Lupin *et al.* (1981), the water activity was more suitable for growth of the most halophilic bacteria. Although salt prevents the growth of spoilage bacteria, but other microorganisms such as high salt tolerant and halophiles are not affected by the presence of salt. A number of authors reported that solar salt contains a large number of halophiles and counts up to 10^5cells/g (Brain *et al*. 1958; Clucas, 1984; Frazier, 1958).

Halophelic or halotolerant bacteria are those that can tolerate a broad range of NaCl concentration (Onishi and Kamekura, 1972; Novitsky and Kushner, 1975; Kushner and Kamekura, 1988; Larsen, 1986). Halophiles actually require salt for growth and will not grow unless salt is present and grow mostly in salted dried fish products (Khan *et al.,* 2005; Kumar *et al.,* 2013). Tolerance of salt concentration generally indicates the ability of the organisms to survive under adverse environmental condition and resistance against chlorine and other antibacterial agents. The aroma in salted and fermented fish product has been claimed to be derived from the activity of various types of halophilic bacteria (Van Veen, 1953). During ripening of salted Anchovy, the microflora was found to be dominated by halophilic and halotolerant bacteria (Perez-Villarreal and Pozo, 1992). According to Gram and Huss (1996) the most common type of spoilage is characterized by the presence of putrid off-odors

and off-flavors, and is caused by growth of a Gram-negative, halophilic and obligate anaerobic rod (up to 10^6-10^7 cfu/g). The Gram-positive were largely halophilic types and come from the salt used to treat the fish (Sefa-Dedeh and Youngs, 1976).

According to Kumar *et al.* (2013), halophilic bacterial count (HBC) increased from 1.0×10^1 to 1.52×10^2 cfu/g respectively in case of 6 month stored 30% salted sun-dried *L. gonius* fish at room temperature which is similar with present findings.

AOAC (2005) observed that aw for a saturated salt solution is 0.75 and if the equilibrium relative humidity is greater than 75% of the salted products it will take up moisture from the atmosphere increasing the O_2 and consequently introducing the possibility of storage by group of microbes.

Can (2011) observed that, halophilic bacterial populations increased in 3.59 to 7.12 log cfu/g in dry salted whole sardine and 3.59 to 6.11 log cfu/g in dry salted fillet sardine when 5 months stored in 4^0C (refrigeration temperature). This finding is similar with present results.

4.4.5. Statistical analysis

Statistical analysis among different biochemical compositions of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) Shol, taki and tengra fish during different duration of storage both at room temperature (26-32^{0}C) and refrigeration temperature (4^{0}C)

To see the degree and nature of relationship between moisture & protein, moisture & fat, protein & fat and moisture & TVB-N of the salted shol, taki and tengra fish after salting by 5 different treatment (DS, PC, BS, SDS, SDS+T) and stored both room and refrigeration temperature to the end day till spoilage, paired 't' test was done with the help of **SPSS 20** data analysis program.

The calculated coefficient of correlation 'r' and paired 't' test values and their significance level were represented in the Table 4.4.5(A)-(C) [Appendix- E, F, G].

From the table it was found that, the value of "moisture & protein" and "moisture & fat" are negatively correlated and "protein & fat" and "moisture & TVB-N" are positively correlated.

The (-) 'r' value from the table indicates negative correlation between moisture & protein" and "moisture & fat".

Table: 4.4.5(A). Statistical analysis among different biochemical compositions of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) shol fish during different duration of storage both at room temperature (26-32^{0}C) and refrigeration temperature (4^{0}C) [Appendix- E]

Relationship between	Treatment	Temp.	'r' value	p-value	't' value	p-value
1) Moisture & protein	DS	Room temp.	-0.993	0.000**	35.777	0.000**
		Refrigeration temp.	-0.995	0.000**	74.550	0.000**
	PC	Room temp.	-0.987	0.000**	38.213	0.000**
		Refrigeration temp.	-0.994	0.000**	66.112	0.000**
	BS	Room temp.	-0.993	0.000**	44.902	0.000**
		Refrigeration temp.	-0.996	0.000**	70.824	0.000**
	SDS	Room temp.	-0.987	0.000**	-9.560	0.000**
		Refrigeration temp.	-0.998	0.000**	-14.287	0.000**
	SDS+T	Room temp.	-0.997	0.000**	-7.746	0.000**
		Refrigeration temp.	-0.992	0.000**	-12.304	0.000**
2) Moisture & fat	DS	Room temp.	-0.992	0.000**	94.608	0.000**
		Refrigeration temp.	-0.994	0.000**	176.719	0.000**
	PC	Room temp.	-0.956	0.000**	99.301	0.000**
		Refrigeration temp.	-0.958	0.000**	164.903	0.000**
	BS	Room temp.	-0.997	0.000**	95.210	0.000**
		Refrigeration temp.	-0.994	0.000**	149.809	0.000**
	SDS	Room temp.	-0.994	0.000**	42.641	0.000**
		Refrigeration temp.	-0.994	0.000**	54.212	0.000**
	SDS+T	Room temp.	-0.994	0.000**	43.879	0.000**
		Refrigeration temp.	-0.995	0.000**	60.578	0.000**
3) Protein & fat	DS	Room temp.	0.987	0.000**	129.832	0.000**
		Refrigeration temp.	0.993	0.000**	662.797	0.000**
	PC	Room temp.	0.911	0.000**	103.521	0.000**
		Refrigeration temp.	0.970	0.000**	301.614	0.000**
	BS	Room temp.	0.996	0.000**	46.912	0.000**
		Refrigeration temp.	0.997	0.000**	82.024	0.000**
	SDS	Room temp.	0.972	0.000**	183.449	0.000**
		Refrigeration temp.	0.993	0.000**	345.673	0.000**
	SDS+T	Room temp.	0.991	0.000**	288.787	0.000**
		Refrigeration temp.	0.994	0.000**	343.740	0.000**
4) Moisture & TVB-N	DS	Room temp.	0.974	0.000**	16.407	0.000**
		Refrigeration temp.	0.987	0.000**	34.618	0.000**
	PC	Room temp.	0.945	0.000**	11.832	0.000**
		Refrigeration temp.	0.991	0.000**	31.725	0.000**
	BS	Room temp.	0.987	0.000**	16.241	0.000**
		Refrigeration temp.	0.979	0.000**	29.672	0.000**
	SDS	Room temp.	0.978	0.000**	8.746	0.000**
		Refrigeration temp.	0.966	0.000**	19.366	0.000**
	SDS+T	Room temp.	0.954	0.000**	9.622	0.000**
		Refrigeration temp.	0.972	0.000**	24.584	0.000**

**p-value <0.01

Table: 4.4.5(B). Statistical analysis among different biochemical compositions of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) taki fish during different duration of storage both at room temperature (26-32^{0}C) and refrigeration temperature (4^{0}C) [Appendix- F]

Relationship between	Treatment	Temperature	'r' value	p value	't' value	p value
1) Moisture & protein	DS	Room temp.	-0.993	0.000**	39.437	0.000**
		Refrigeration temp.	-0.997	0.000**	54.883	0.000**
	PC	Room temp.	-0.993	0.000**	58.614	0.000**
		Refrigeration temp.	-0.994	0.000**	88.619	0.000**
	BS	Room temp.	-0.999	0.000**	76.305	0.000**
		Refrigeration temp.	-0.986	0.000**	125.984	0.000**
	SDS	Room temp.	-0.993	0.000**	-59.866	0.000**
		Refrigeration temp.	-0.988	0.000**	-70.158	0.000**
	SDS+T	Room temp.	-0.999	0.000**	-51.297	0.000**
		Refrigeration temp.	-0.997	0.000**	-65.026	0.000**
2) Moisture & fat	DS	Room temp.	-0.995	0.000**	91.362	0.000**
		Refrigeration temp.	-0.994	0.000**	124.856	0.000**
	PC	Room temp.	-0.995	0.000**	110.203	0.000**
		Refrigeration temp.	-0.995	0.000**	160.103	0.000**
	BS	Room temp.	-0.990	0.000**	127.558	0.000**
		Refrigeration temp.	-0.991	0.000**	200.073	0.000**
	SDS	Room temp.	-0.998	0.000**	11.122	0.000**
		Refrigeration temp.	-0.986	0.000**	11.049	0.000**
	SDS+T	Room temp.	-0.953	0.000**	17.102	0.000**
		Refrigeration temp.	-0.994	0.000**	18.095	0.000**
3) Protein & fat	DS	Room temp.	0.990	0.000**	111.256	0.000**
		Refrigeration temp.	0.988	0.000**	192.557	0.000**
	PC	Room temp.	0.988	0.000**	159.709	0.000**
		Refrigeration temp.	0.987	0.000**	311.194	0.000**
	BS	Room temp.	0.992	0.000**	116.600	0.000**
		Refrigeration temp.	0.999	0.000**	257.581	0.000**
	SDS	Room temp.	0.992	0.000**	932.244	0.000**
		Refrigeration temp.	0.963	0.000**	481.401	0.000**
	SDS+T	Room temp.	0.951	0.000**	826.011	0.000**
		Refrigeration temp.	0.995	0.000**	1155.260	0.000**
4) Moisture & TVB-N	DS	Room temp.	0.976	0.000**	11.902	0.000**
		Refrigeration temp.	0.989	0.000**	34.952	0.000**
	PC	Room temp.	0.980	0.000**	13.457	0.000**
		Refrigeration temp.	0.992	0.000**	34.765	0.000**
	BS	Room temp.	0.954	0.003**	14.436	0.000**
		Refrigeration temp.	0.982	0.000**	16.429	0.000**
	SDS	Room temp.	0.988	0.000**	-2.578	0.020*
		Refrigeration temp.	0.995	0.000**	-.051	0.960
	SDS+T	Room temp.	0.962	0.000**	-2.670	0.015*
		Refrigeration temp.	0.973	0.000**	1.451	0.169

**p-value <0.01

* p-value <0.05

Table: 4.4.5(C). Statistical analysis among different biochemical compositions of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) tengra fish during different duration of storage both at room temperature (26-32^{0}C) and refrigeration temperature (4^{0}C) [Appendix-G]

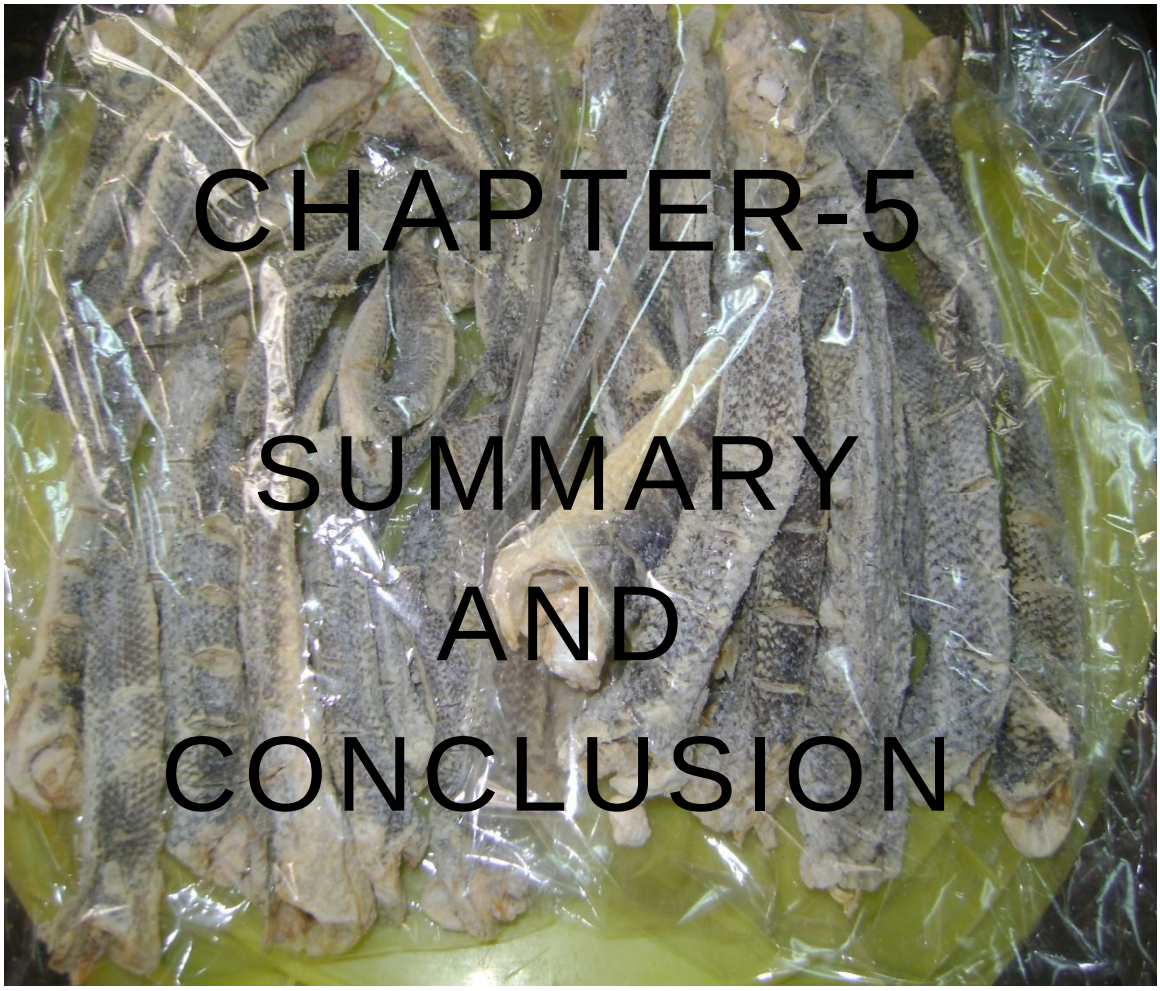
Relationship between	Treatment	Temperature	'r' value	p value	't' value	p value
1) Moisture & protein	DS	Room temp.	-0.997	0.000**	38.545	0.000**
		Refrigeration temp.	-0.995	0.000**	48.131	0.000**
	PC	Room temp.	-0.996	0.000**	46.663	0.000**
		Refrigeration temp.	-0.995	0.000**	55.094	0.000**
	BS	Room temp.	-0.995	0.000**	57.627	0.000**
		Refrigeration temp.	-0.998	0.000**	71.964	0.000**
	SDS	Room temp.	-0.994	0.000**	-75.435	0.000**
		Refrigeration temp.	-0.995	0.000**	-136.535	0.000**
	SDS+T	Room temp.	-0.996	0.000**	-55.839	0.000**
		Refrigeration temp.	-0.993	0.000**	-85.798	0.000**
2) Moisture & fat	DS	Room temp.	-0.997	0.000**	58.481	0.000**
		Refrigeration temp.	-0.999	0.000**	70.167	0.000**
	PC	Room temp.	-0.987	0.000**	72.815	0.000**
		Refrigeration temp.	-0.998	0.000**	77.287	0.000**
	BS	Room temp.	-0.995	0.000**	71.277	0.000**
		Refrigeration temp.	-0.994	0.000**	95.714	0.000**
	SDS	Room temp.	-0.991	0.000**	-16.524	0.000**
		Refrigeration temp.	-0.991	0.000**	-32.281	0.000**
	SDS+T	Room temp.	-0.997	0.000**	-9.443	0.000**
		Refrigeration temp.	-0.993	0.000**	-16.164	0.000**
3) Protein & fat	DS	Room temp.	0.997	0.000**	1050.551	0.000**
		Refrigeration temp.	0.998	0.000**	439.163	0.000**
	PC	Room temp.	0.970	0.000**	151.097	0.000**
		Refrigeration temp.	0.994	0.000**	793.028	0.000**
	BS	Room temp.	0.984	0.002**	206.706	0.000**
		Refrigeration temp.	0.986	0.000**	117.232	0.000**
	SDS	Room temp.	0.995	0.000**	596.983	0.000**
		Refrigeration temp.	0.986	0.000**	732.494	0.000**
	SDS+T	Room temp.	0.995	0.000**	526.947	0.000**
		Refrigeration temp.	0.983	0.000**	460.188	0.000**
4) Moisture & TVB-N	DS	Room temp.	0.992	0.000**	8.704	0.000**
		Refrigeration temp.	0.994	0.000**	17.821	0.000**
	PC	Room temp.	0.975	0.000**	8.425	0.000**
		Refrigeration temp.	0.988	0.000**	20.912	0.000**
	BS	Room temp.	0.996	0.000**	10.088	0.001**
		Refrigeration temp.	0.997	0.000**	23.244	0.000**
	SDS	Room temp.	0.978	0.000**	-4.302	0.001**
		Refrigeration temp.	0.997	0.000**	-3.108	0.014*
	SDS+T	Room temp.	0.965	0.000**	-5.154	0.000**

**p-value <0.01
* p-value <0.05

From Table 4.4.5(A) it is shown that during storage at room temperature ‘r’ value between moisture and protein content of DS shol fish was -0.993 and its ‘p’ value was 0.000 which means moisture and protein content of DS shol was highly significantly correlated ($p<0.01$)** but their relation is negative (inverse). On the other hand, at room temperature, ‘r’ value between protein and fat content of PC shol fish was 0.911 and its ‘p’ value was 0.000 which means protein and fat content of PC shol was highly significantly correlated ($p<0.01$)** but their relation is positive. Other results from this Table can be interpreted in the same way.

From Table 4.4.5(B) it is shown that during storage at room temperature moisture and TVB-N content of SDS taki fish have ‘r’ value 0.988 and its ‘p’ value was 0.000 which means moisture and TVB-N content of SDS Taki was highly significantly correlated ($p<0.01$)** but its ‘t’ value of -2.578 and its ‘p’ value was 0.020 which means that these two parameters were significantly correlated ($p<0.05$)* according to its ‘t’ value. It was also shown from this table that moisture and TVB-N content of refrigeration stored SDS taki fish have ‘r’ value 0.995 and its ‘p’ value was 0.000** which means moisture and TVB-N content of SDS Taki was highly significantly correlated ($p<0.01$)** but its ‘t’ value of -0.051 and its ‘p’ value was 0.960 which means that there was insignificant difference ($p>0.05$) present between these two parameters according to its ‘t’ value.

Results of Table 4.4.5(C) can be interpreted in the same way.



CHAPTER-5

SUMMARY AND CONCLUSION

CHAPTER-5

SUMMARY AND CONCLUSION

In Bangladesh, different processing methods like salting, drying, fermentation, icing, freezing etc, have been practicing from a long time to preserve the fish from spoilage or to increase the shelf life. Among them, salt curing of fish is oldest and widely used method of fish preservation. The main features of salting are the removal of sufficient water from the fish tissue and its partial replacement by salt. As a result, a condition is arrived when spoilage activity are slowed down.

Present research on "Performance and quality assessment of salt curing of Taki (*Channa punctatus*), Shol (*Channa striatus*) and Tengra (*Mystus tengra*)" have designed and planned on technological aspect and performance of five different types of salt-curing method viz., Dry salting (DS), Pickle curing (PC), Brine salting (BS), Sun-dried salting (SDS) and Turmeric treated sun-dried salting (SDS+T), with view to give updated idea about salt curing of fish in Bangladesh. Biochemical composition and Bacteriological study of fishes in fresh condition, in freshly processed salted condition and in different storage condition was measured analytically and was recorded for making comparative study.

Biochemical composition and standard plate count (SPC) of fresh shol, taki and tengra have been studied. The value of moisture, protein, fat, ash, TVB-N (Total Volatile base-Nitrogen), FFA (Free Fatty Acid), pH and SPC of fresh shol fish was 77.03±0.12%, 19.52±0.07%, 1.93±0.07%, 1.44±0.11%, 4.41±0.01 mg/100g, 0.6±0.06%, 6.9±0.06 and 2×10^5 cfu/g; fresh taki fish was 78.65±0.07%, 16.89±0.10%, 2.50±0.06%, 1.36±0.11%, 3.43±0.02 mg/100g, 0.5±0.10%, 7.0±0.06 and 1.1×10^5 cfu/g and fresh tengra fish was 74.27±0.07%, 16.96±0.07%, 6.04±0.06%, 2.67±0.08%, 4.27±0.02 mg/100g, 0.9±0.21%, 7.0±0.10 and 5×10^5 cfu/g respectively.

Fresh shol, taki and tengra fish had Ca 11.2, 16.35 and 22.025; Mg 10.125, 9.425 and 11.4; Fe 1.475, 1.275 and 2.25; Cu 0.7, 0.65 and 0.55; Zn 0.25, 0.425 and 1.275; Mn 0.1, 0.05 and 0.125 mg/100g of fish respectively.

During salting process, some changes in chemical and physico-chemical characteristics took place and in certain stage, the original characteristics of the raw fish were found virtually absent. This stage was considered as 'salt ripening' of fish. Salt ripening of five different types of salted shol, taki and tengra was done by assessing changes in salt penetration rate and its effect on moisture content, fluid loss pattern in fish muscles and percent weight loss of fishes during ripening period.

In Bangladesh, shol, taki and tengra is mainly consumed as fresh and there was found very limited valid reports on using these fishes for producing salted products. For this reason, consumer preferability study was done after preparing five different types of salted (dry salted, pickle cured, brine salted, sun-dried salted and turmeric treated sun-dried salted) shol, taki and tengra fish curry with traditional 'South Asian recipe' and these fish-curries was subjected to panel tests with 5 panel members for considering its taste.

After salt-ripening, Dry salted (DS), Pickle cured (PC), Brine salted (BS), Sun-dried salted (SDS) and Turmeric treated sun-dried salted (SDS+T) shol fish-products had Ca content of 438.75, 201.5, 196.25, 468.75 and 219.5; Mg content of 83.69, 32.19, 14.75, 85.69 and 43.687; Fe content of 2.65, 1.82, 1.5, 3.32 and 2.8; Cu content of 0.275, 0.225, 0.15, 0.375 and 0.30; Zn content of 2.125, 1.725, 1.45, 2.675 and 2.275; Mn content of 0.45, 0.31, 0.225, 0.775 and 0.525 mg/100g of fish; taki fish-products had Ca content of 600, 497.5, 123, 700.75 and 578.25; Mg content of 147.75, 65.5, 52.5, 157.5 and 126.5; Fe content of 3.75, 2.95, 2.07, 5.35 and 4.99; Cu content of 0.425, 0.25, 0.2, 0.84 and 0.62; Zn content of 1.525, 1.43, 1.175, 3.50 and 3.0; Mn content of 0.625, 0.60, 0.55, 0.925 and 0.775 mg/100g of fish whereas tengra fish products had Ca content of 1250, 910, 310.75, 1405 and 976.25; Mg content of 520.5, 77.375, 52.5, 795.5 and 331; Fe content of 4.3, 3.75, 2.425, 6.95 and 5.025; Cu content of 0.435, 0.40, 0.3, 0.625 and 0.475; Zn content of 2.175, 1.625, 1.275, 3.6

and 3.575**;** Mn content of 0.65, 0.45, 0.375, 1.02 and 0.722 mg/100g of fish respectively.

To extend the shelf life, all five types of (DS, PC, BS, SDS and SDS+T) salted shol, taki and tengra fish-products were stored in two different temperature system viz. (i) room temperature (26-32^{0}C) and (ii) refrigeration temperature (4^{0}C). For determining the performance of shelf life quality of these salted fish-products, some parameters like Sensory evaluation (sensory characteristics and sensory scores); Chemical composition (Salt, TVB-N, FFA, pH value) analysis and bacteriological study (SPC and HBC) were conducted. Besides these parameters, proximate composition (moisture, protein, fat, ash) of all salted fish products through entire storage period were also studied periodically.

Performance of quality of Dry salted (DS), Pickle cured (PC), Brine salted (BS), Sun-dried salted (SDS) and Turmeric treated sun-dried salted (SDS+T) shol, taki and tengra fish-products while stored in sealed polythene bag at room and refrigeration temperature were found to have variation in their shelf life and in accordance with storage condition and method of salting of fish.

Sensory evaluation of five different salted shol, taki and tengra fish-products were analyzed on 9 point hedonic scale (according to Peryam and Pilgrim, 1957) for their acceptability through their entire shelf life period. At the beginning of storage, the sensory quality of DS, PC, BS, SDS and SDS+T shol, taki and tengra fish products were considered excellent which turned to slightly acceptable to the consumer at the end of shelf life and the acceptability score <4.5 was considered as rejected or spoiled for all salted fish samples both at room and refrigeration storage.

Dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun dried salted (SDS+T) shol fish products turned to slightly acceptable to the panel members with sensory score of 5 (lower limit for acceptable score) in the end of 165 days, 150 days, 135 days, 165 days and 180 days respectively in room temperature stored products whereas 26 months, 22 months, 18 months, 24

months and 28 months in case of refrigeration storage products. Similarly, room temperature stored dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun dried salted (SDS+T) taki fish products turned into sensory score of 5 at the end of 150 days, 135 days, 75 days, 240 days and 300 days and at the end of 26 months, 24 months, 10 months, 26 months and 28 months in case of refrigeration storage products. On the other hand, in room temperature, the highest shelf life of dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun dried salted (SDS+T) tengra fish products was 210 days, 180 days, 120 days, 360 days and 420 days respectively whereas 32 months, 28 months, 24 months, 32 months and 36 months in case of refrigeration storage products with sensory score of 9 to 5 in entire storage period.

Dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) Shol fish-products (from zero day to end period) had moisture content, in the range of 48.84% to 53.69%, 50.29% to 54.94%, 60.17% to 65.97%, 29.77% to 35.26% and 30.92% to 36.98%; protein content, in the range of 28.21% to 25.52%, 27.64% to 24.82%, 20.72% to 17.06%, 41.48% to 38.62% and 41.0% to 38.01%; fat content in the range of 3.99% to 3.30%, 3.79% to 3.11%, 3.20% to 2.72%, 5.10% to 3.81% and 4.79% to 3.26% and ash content in the range of 18.98% to 17.58%, 18.69% to 17.45%, 16.06% to 15.14%, 22.80% to 21.44% and 22.41% to 21.07% during storage at room temperature. Whereas, in refrigeration storage these values varied (from zero day to end period) in a range of 48.84% to 51.24%, 50.29% to 52.85%, 60.17% to 63.48%, 29.77% to 34.09% and 30.92% to 35.48% (moisture); 28.21% to 27.25%, 27.64% to 26.34%, 20.72% to 18.50%, 41.48% to 39.30% and 41.0% to 38.81% (protein); 3.99% to 3.46%, 3.79% to 3.22%, 3.20% to 2.89%, 5.10% to 4.01% and 4.79% to 3.69% (fat); 18.98% to 17.94%, 18.69% to 17.76%, 16.06% to 15.49%, 22.80% to 21.66% and 22.41% to 21.41% (ash) respectively.

On the other hand, dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) Taki fish-products had moisture content varied in the range of 46.21% to 50.92%, 52.71% to 56.55%,

62.28% to 65.08%, 9.77% to 14.97% and 12.72% to 18.88%**;** protein content, in the range of 23.58% to 21.53%, 21.39% to 20.06%, 18.02% to 16.92%, 47.69% to 45.68% and 46.06% to 43.92%**;** fat content in the range of 3.93% to 3.49%, 3.40% to 2.93%, 2.76% to 2.48%, 7.47% to 5.81% and 7.09% to 5.01% and ash content in the range of 26.37% to 24.55%, 22.96% to 21.02%, 17.24% to 15.99%, 35.16% to 34.05% and 34.29% to 32.90% during storage at room temperature. Whereas, in refrigeration storage these values varied in a range of 46.21% to 49.89%, 52.71% to 55.71%, 62.28% to 64.02%, 9.77% to 13.40% and 12.72% to 17.02% for moisture; 23.58% to 21.94%, 21.39% to 20.36%, 18.02% to 17.45%, 47.69% to 45.91% and 46.06% to 44.53% for protein; 3.93% to 3.44%, 3.40% to 2.92%, 2.76% to 2.59%, 7.47% to 6.48% and 7.09% to 6.00% for fat and 26.37% to 25.09%, 22.96% to 21.58%, 17.24% to 16.50%, 35.16% to 34.41% and 34.29% to 33.13% for ash respectively.

Similarly, dry salted (DS), pickle cured (PC), brine salted (BS), sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) Tengra fish-products had moisture content in the range of 41.41% to 44.82%, 45.88% to 49.09%, 57.35% to 60.70%, 4.90% to 9.05% and 6.08% to 11.92%; protein content, in the range of 22.05% to 20.99%, 20.43% to 19.28%, 15.30% to 14.28%, 43.00% to 42.00% and 42.60% to 41.03%; fat content in the range of 10.65% to 9.59%, 9.40% to 8.68%, 6.84% to 6.01%, 15.99% to 14.41% and 15.58% to 13.40% and ash content in the range of 26.15% to 25.00%, 24.62% to 23.41%, 20.80% to 19.40%, 36.20% to 35.18% and 35.60% to 34.22% during storage at room temperature. Whereas, in refrigeration storage these values varied in a range of 41.41% to 44.29%, 45.88% to 48.79%, 57.35% to 60.57%, 4.90% to 6.74% and 6.08% to 9.03% for moisture; 22.05% to 21.10%, 20.43% to 19.41%, 15.30% to 14.10%, 43.00% to 42.40% and 42.60% to 41.86% for protein; 10.65% to 9.52%, 9.40% to 8.41%, 6.84% to 6.09%, 15.99% to 15.09% and 15.58% to 14.38% for fat and 26.15% to 25.10%, 24.62% to 23.59%, 20.80% to 19.48%, 36.20% to 35.50% and 35.60% to 34.82% for ash respectively.

Food quality of the salted shol, taki and tengra fish-products also conducted through determining their TVB-N, pH and FFA value during entire storage period both at room and refrigeration temperature. While the TVB-N and FFA values of room temperature stored salted-samples were found similar to findings of other researchers, the increase in pH values during the storage of room temperature was higher than others. The probable reason of these differences might be due to the differences in fish species and different methods of salting.

During storage period, moisture, TVB-N, FFA, pH, SPC, HBC (Halophilic Bacterial Count) value increased and protein, fat, ash, salt and sensory score value decreased both at room and refrigeration temperature, but the increment in room temperature was more prominent than the products stored in refrigerated temperature. There was a direct relationship between moisture and TVB-N value but inverse relationship was found between sensory score and TVB-N value. The overall quality of all salted fish-products were excellent in fresh process condition and five types of salted products stored at refrigeration temperature were found in better condition as was determined by organoleptic, biochemical and bacteriological analysis.

The initial value of chemical composition (salt, TVB-N, FFA and pH) were 16.00%, 5.25 mg/100g, 1.5% and 6.3 in DS shol**;** 16.08%, 5.28 mg/100g, 1.3% and 6.4 in PC shol**;** 15.20%, 3.64 mg/100g, 1.8% and 6.5 in BS shol**;** 14.90%, 4.89 mg/100g, 2.3% and 6.2 in SDS shol**;** 14.30%, 5.17 mg/100g, 2.4% and 6.3 in SDS+T shol respectively whereas, in case of taki fish-products these values were 16.06%, 4.18 mg/100g, 1.9% and 6.5 in DS; 16.22%, 3.79 mg/100g, 2.4% and 6.6 in PC; 15.50%, 5.70 mg/100g, 2.6% and 6.8 in BS; 14.72%, 2.27 mg/100g, 1.8% and 5.9 in SDS; 13.77%, 3.58 mg/100g, 2.3% and 5.9 in SDS+T respectively. On the other hand, tengra fish-products had these values of 16.80%, 3.90 mg/100g, 2.8% and 6.0 in DS; 17.10%, 4.92 mg/100g, 3.2% and 6.0 in PC; 16.50%, 2.59 mg/100g, 1.0% and 5.9 in BS; 15.20%, 1.9 mg/100g, 1.8% and 6.3 in SDS and 14.80%, 2.52 mg/100g, 1.6% and 6.4 in SDS+T respectively.

Salt (%), TVB-N (mg/100g), FFA (%), and pH value were found 14.36%, 31.33 mg/100g, 13.5% and 8.1 in DS; 14.60%, 34.99 mg/100g, 13% and 8.3 in PC; 13.73%, 34.52 mg/100g, 14% and 8.3 in BS; 12.15%, 30.86 mg/100g, 10.5% and 8.3 in SDS; 12.02%, 31.44 mg/100g, 11.1% and 8.2 in SDS+T treated room temperature stored shol fish-products during the shelf life period whereas, these values were 15.20%, 19.06 mg/100g, 10.1% and 7.1 in DS; 15.26%, 19.74 mg/100g, 11.9% and 7.2 in PC; 14.36%, 21.14 mg/100g, 12.2% and 7.2 in BS; 12.74%, 19.15 mg/100g, 11.0% and 7.1 in SDS; 12.28%, 20.02 mg/100g, 11.4% and 7.2 in SDS+T treated refrigeration stored shol fish-products before just sensory rejection.

Similarly, taki fish-products kept at room temperature showed these values as 15.05%, 30.76 mg/100g, 11.5% and 7.9 in DS; 15.34%, 30.01 mg/100g, 12.8% and 8.3 in PC; 14.42%, 30.96 mg/100g, 13.3% and 8.3 in BS; 12.89%, 31.02 mg/100g, 12.2% and 7.9 in SDS; 12.20%, 31.25 mg/100g, 13.1% and 8.0 in SDS+T during their shelf life period. However, these values in taki fish-products when kept at refrigeration temperature showed the values as 15.18%, 20.05 mg/100g, 12.5% and 7.9 in DS; 15.29%, 20.58 mg/100g, 12.7% and 7.8 in PC; 14.96%, 24.48 mg/100g, 12.8% and 7.8 in BS; 13.09%, 20.27 mg/100g, 12.1% and 7.6 in SDS; 12.82%, 20.31 mg/100g, 12.3% and 7.8 in SDS+T at the end of shelf life.

Similarly, in case of tengra fish-products, these values were found 15.41%, 30.22 mg/100g, 12.1% and 7.9 in DS; 15.90%, 31.04 mg/100g, 12.7% and 8.0 in PC; 15.02%, 30.45 mg/100g, 13% and 7.9 in BS; 13.56% 30.15 mg/100g, 13.1% and 7.1 in SDS; 13.0%, 30.24 mg/100g, 13.2% and 7.2 in SDS+T at the end of room temperature storage and 15.63%, 20.23 mg/100g, 11.0% and 6.9 in DS; 15.98%, 20.59 mg/100g, 11.4% and 6.9 in PC; 15.27%, 20.98 mg/100g, 11.6% and 6.9 in BS; 13.81%, 20.75 mg/100g, 11.1% and 7.0 in SDS; 13.21%, 21.07 mg/100g, 11.2% and 7.1 respectively in SDS+T treated refrigeration stored tengra fish-products before just sensory rejection.

At room temperature storage condition, the range of SPC (Standard Plate Count) of DS, PC, BS, SDS and SDS+T shol fish products were 2.7×10^3 to 1.7×10^6, 3.6×10^3 to

2.0×10^6, 2.1×10^4 to 3.7×10^6, 2.4×10^3 to 1.3×10^6 and 2.1×10^3 to 1.6×10^6 cfu/g; taki fish products were 4.0×10^3 to 2.9×10^6, 3.0×10^4 to 2.5×10^6, 4.3×10^4 to 3.9×10^6, 3.9×10^3 to 2.1×10^6 and 2.8×10^3 to 1.7×10^6 cfu/g ; tengra fish products were 3.8×10^3 to 1.8×10^6, 1.2×10^4 to 2.0×10^6, 1.5×10^4 to 4.0×10^6, 2.8×10^3 to 1.3×10^6 and 2.5×10^3 to 1.4×10^6 cfu/g of fish whereas at refrigeration storage, this value varied from 2.7×10^3 to 1.9×10^6, 3.6×10^3 to 2.2×10^6, 2.1×10^4 to 4.3×10^6, 2.4×10^3 to 1.6×10^6 and 2.1×10^3 to 2.2×10^6 cfu/g in case of shol fish products; 4.0×10^3 to 3.3×10^6, 3.0×10^4 to 4.2×10^6, 4.3×10^4 to 5.1×10^6, 3.9×10^3 to 3.0×10^6 and 2.8×10^3 to 2.5×10^6 cfu/g in case of taki fish products; 3.8×10^3 to 2.1×10^6, 1.2×10^4 to 3.3×10^6, 1.5×10^4 to 3.0×10^6, 2.8×10^3 to 1.8×10^6 and 2.5×10^3 to 1.7×10^6 cfu/g of fish in case of tengra fish products respectively.

Halophilic Bacterial count (HBC) of DS, PC, BS, SDS and SDS+T in shol fish products stored at room temperature were 3.0×10^2 to 2.1×10^5, 4.1×10^2 to 2.4×10^5, 4.0×10^3 to 3.4×10^5, 2.5×10^2 to 1.5×10^5 and 2.0×10^2 to 1.8×10^5 cfu/g; taki fish products were 3.0×10^2 to 1.9×10^5, 3.3×10^3 to 2.9×10^5, 4.1×10^3 to 3.2×10^5, 2.4×10^2 to 2.5×10^5 and 2.1×10^2 to 3.0×10^5 cfu/g ; tengra fish products were 3.2×10^2 to 2.6×10^5, 1.4×10^3 to 6.0×10^5, 1.8×10^3 to 7.8×10^5, 1.5×10^2 to 5.0×10^5 and 1.2×10^2 to 5.5×10^5 cfu/g of fish whereas at refrigeration storage, this values varied from 3.0×10^2 to 2.6×10^5, 4.1×10^2 to 3.1×10^5, 4.0×10^3 to 3.8×10^5, 2.5×10^2 to 2.0×10^5 and 2.0×10^2 to 2.3×10^5 cfu/g in case of shol fish products; 3.0×10^2 to 4.1×10^5, 3.3×10^3 to 3.5×10^5, 4.1×10^3 to 3.9×10^5, 2.4×10^2 to 3.2×10^5 and 2.1×10^2 to 4.5×10^5 cfu/g in case of taki fish products; of 3.2×10^2 to 3.2×10^5, 1.4×10^3 to 3.1×10^5, 1.8×10^3 to 5.0×10^5, 1.5×10^2 to 2.8×10^5 and 1.2×10^2 to 5.5×10^5 cfu/g of fish in case of tengra fish products respectively.

Visually there was found no fungal attack in all of 5 types of salted shol, taki and tengra fish products during the storage period. This is most probably because, the moisture content and the higher salt content in the products prevented the occurrence of fungal growth which kept these salted fish-products for a long storage period.

The results of the whole study indicated that despite some changes in the biochemical properties during prolonged refrigeration storage at 4^0C, the treated fishes appeared to be suitable for refrigeration storage.

It was observed that processing and storage significantly affected the proximate and chemical composition of different salted Shol, Taki and Tengra fish-products. The most important is their general reduction effect on the moisture content which is an index of spoilage. But however during storage with the exception of sun-dried salted (SDS) Shol, Taki and Tengra fish-products, moisture content of other fish-products prominently increased after storage.

The proximate composition of different salted shol, taki and tengra fishes revealed that processed fish have very good nutritional value. These processed samples were very rich in protein and lipid. However, the nutritional value of these salted fishes is gradually decreased due to the longer storage. Therefore, the fish processing industries should be kept more precautionary steps during storage of processed fish samples in the warehouse and in the sales centre.

The present research also revealed that the application of salt and turmeric gave the best performance in comparison to other treatments as it retained the more beneficial nutrient property; lower moisture, TVB-N, FFA, pH value and bacterial load and prolong shelf life. The salt used in five different treatments to remove water from the fish body and thus to help the fish through salting whereas the turmeric treated sun-dried salting (SDS+T) are used not only to remove water from the fish body but also added some antibiotic factors that prevented the growth of moulds and bacteria due to the formation of an unfavorable growth medium but with a good taste.

In conclusion, fish samples (shol, taki and tengra) processed by five types of salting underwent some loss in quality with time of storage. However, turmeric treated sun-dried samples were most organoleptically and chemically accepted with minimum deterioration. The maximum shelf life was extended by salt and turmeric treated sun dried samples in all three fishes. So, from the present study it can be concluded that

salt in combination with turmeric with sun-drying stored at 4^0C temperature is the best method for long time preservation of freshwater fishes-taki, shol and tengra.

There happens lot of lacking during processing of traditional salted fresh water fishery products. Good manufacturing practice is a prerequisite for high quality product, but our traditional salted fishery products are not up to the mark of standard of high quality and shows lower shelf life period and consequently we are deprived of earning large amount of business money. So, commercial traders those who produce market salted fish in our country may be asked to follow the suggestions made over here on the basis of the findings of the present study.

Conclusion

On the basis of the results, following conclusions can be drawn –

1) Quality assessment of salted and sun-dried salted fish-products while stored in sealed polythene bag at ambient/room temperature (26-32^{0}C) and refrigerated temperature (4^{0}C) have been found to have variation in their shelf life in accordance with storage condition and method of salting of fish.

2) Different salting methods have significant influences on the proximate, chemical and mineral composition of shol, taki and tengra fishes and making these fishes nutritionally suitable for consumption.

3) Stored product-quality in refrigeration temperature was found to be better than those stored at ambient temperature.

4) Turmeric treated sun-dried salted shol, taki and tengra fish-samples were most organoleptically accepted, highest shelf-life as well as deteriorated at a very slow rate.

5) Brine salted sample deteriorated faster in odor and flavor than other salted samples.

6) On the basis of quality-standard and performances of the five salting methods, Turmeric treated sun dried salting (SDS+T) was found best followed by Sun-dried salting (SDS), Dry salting (DS), Pickle curing (PC) and Brine salting (BS) respectively.

7) Among the three species having five categories each, the salted tengra fish in all categories showed better performance in respects of shelf life period.

8) Salted fish is a rich source of minerals. Thus salted fish can be considered an important supplement of the majority of minerals in the diet.

9) Due to the use of salt, the growth of pathogenic micro organisms was controlled.

Thus the present study provides information about the suitability of different salting methods of three commercially important fresh water fishes to produce a very stable and safe product with long shelf life. By assessing the nutritional quality as well as the feasibility of the method, it can be recommended to explore the turmeric treated sun-dried salting (SDS+T) method in commercial scale which further will contribute in national economy of Bangladesh.

Recommendation

- The present experiment has been devised for the first time of its kind. In the present experiment, studies have been made in order to find out the shelf lives of the fishes under consideration. As different fish species have different shelf lives in different treatments, the end products have not been manually tasted or consumed elaborately during the whole study period. Hopefully future researchers may concentrate on this aspect and will make extensive studies.

- It is traditionally believed that only fatty fishes, like, hilsa is suitable for salting. However, this study reveals that, many other important freshwater fish eg. taki, shol, tengra may successfully be used for salting with longer shelf life.

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APPENDICES

Appendix-A

Table: Ingredients of Super Salt (Iodized, vacuum evaporated, free flowing salt)

Ingredients	Amount
NaCl	99.50
Iodine content	20-50 ppm
Water	0.03%
pH	7.00

Appendix-B

Table: 1. Fluid loss pattern (Reduction of fluid) in shol fish muscles during different days of ripening period

Days of ripening period	DS	SDS	SDS+T
1	500	580	520
2	410	330	210
3	130	110	30
4	75	50	20
5	50	20	10
6	25	-	5
7	10	-	-

Table: 2. Fluid loss pattern (Reduction of fluid) in Taki fish muscles during different days of ripening period

Days of ripening period	DS	SDS	SDS+T
1	400	400	300
2	300	250	250
3	170	80	60
4	50	20	20
5	30	5	10
6	20	-	5
7	5	-	-

Table: 3. Fluid loss pattern (Reduction of fluid) in Tengra fish muscles during different days of ripening period

Days of ripening period	DS	SDS	SDS+T
1	750	740	760
2	100	100	180
3	80	20	30
4	25	5	20
5	10	-	5
6	5	-	-
7	2	-	-

Appendix-C

Table. Micro/trace element concentrations in standard guidelines

Micro / Trace elements	Recommended levels (ppm)
Cu	30 (FAO/WHO, 1983)
Fe	100 (FAO/WHO, 1989)
Mn	1.0 (FAO/WHO, 1989)
Zn	100 (FAO/WHO, 1989)

Appendix-D

Score Sheet of Taste of 'Fish-curry' prepared from 5 different types of salted Shol, Taki and Tengra Fish-products

Name: Prof. Dr. Gulshan Ara Latifa, Department of Zoology, Dhaka University (Supervisor)

Score	Description	DS Shol	PC Shol	BS Shol	SDS Shol	SDS+T Shol	DS Taki	PC Taki	BS Taki	SDS Taki	SDS+T Taki	DS Tengra	PC Tengra	BS Tengra	SDS Tengra	SDS+T Tengra
9	Like extremely					✓					✓					✓
8	Like very much	✓	✓	✓	✓		✓	✓		✓		✓	✓	✓	✓	
7	Like moderately								✓							
6	Like slightly															
5	Neither like nor dislike															
4	Dislike slightly															
3	Dislike moderately															
2	Dislike very much															
1	Dislike extremely															

[DS=Dry-salted, PC=Pickle-cured, BS=Brine-salted, SDS=Sun-dried salted, SDS+T=Turmeric treated sun-dried salted]

GA Latifa

Signature

Score Sheet of Taste of 'Fish-curry' prepared from 5 different types of salted Shol, Taki and Tengra Fish-products

Name: Prof. Dr. Subhash Chandra Chakraborty, Dean, Faculty of Fisheries, Bangladesh Agricultural University (Co-Supervisor)

Score	Description	DS Shol	PC Shol	BS Shol	SDS Shol	SDS+T Shol	DS Taki	PC Taki	BS Taki	SDS Taki	SDS+T Taki	DS Tengra	PC Tengra	BS Tengra	SDS Tengra	SDS+T Tengra
9	Like extremely					✓					✓					✓
8	Like very much	✓	✓	✓	✓		✓	✓		✓		✓	✓	✓	✓	
7	Like moderately								✓							
6	Like slightly															
5	Neither like nor dislike															
4	Dislike slightly															
3	Dislike moderately															
2	Dislike very much															
1	Dislike extremely															

[DS=Dry-salted, PC=Pickle-cured, BS=Brine-salted, SDS=Sun-dried salted, SDS+T=Turmeric treated sun-dried salted]

Signature

Score Sheet of Taste of 'Fish-curry' prepared from 5 different types of salted Shol, Taki and Tengra Fish-products

Name: Mohajira Begum, SSO, IFST, BCSIR

Score	Description	DS Shol	PC Shol	BS Shol	SDS Shol	SDS+T Shol	DS Taki	PC Taki	BS Taki	SDS Taki	SDS+T Taki	DS Tengra	PC Tengra	BS Tengra	SDS Tengra	SDS+T Tengra
9	Like extremely					✓					✓					✓
8	Like very much	✓	✓	✓	✓		✓	✓		✓		✓	✓	✓	✓	
7	Like moderately								✓							
6	Like slightly															
5	Neither like nor dislike															
4	Dislike slightly															
3	Dislike moderately															
2	Dislike very much															
1	Dislike extremely															

[DS=Dry-salted, PC=Pickle-cured, BS=Brine-salted, SDS=Sun-dried salted, SDS+T=Turmeric treated sun-dried salted]

Signature

Score Sheet of Taste of 'Fish-curry' prepared from 5 different types of salted Shol, Taki and Tengra Fish-products

Name: Md. Abdul Khaleque, Lab-Technician, IFST, BCSIR

Score	Description	DS Shol	PC Shol	BS Shol	SDS Shol	SDS+T Shol	DS Taki	PC Taki	BS Taki	SDS Taki	SDS+T Taki	DS Tengra	PC Tengra	BS Tengra	SDS Tengra	SDS+T Tengra
9	Like extremely					✓					✓					✓
8	Like very much	✓	✓	✓	✓		✓	✓	·	✓		✓	✓	✓	✓	
7	Like moderately								✓							
6	Like slightly															
5	Neither like nor dislike															
4	Dislike slightly															
3	Dislike moderately															
2	Dislike very much															
1	Dislike extremely															

[DS=Dry-salted, PC=Pickle-cured, BS=Brine-salted, SDS=Sun-dried salted, SDS+T=Turmeric treated sun-dried salted]

M.A. Khaleque
Signature

Score Sheet of Taste of 'Fish-curry' prepared from 5 different types of salted Shol, Taki and Tengra Fish-products

Name: Mosarrat Nabila Nahid, Ph.D Researcher, Department of Zoology, Dhaka University.

Score	Description	DS Shol	PC Shol	BS Shol	SDS Shol	SDS+T Shol	DS Taki	PC Taki	BS Taki	SDS Taki	SDS+T Taki	DS Tengra	PC Tengra	BS Tengra	SDS Tengra	SDS+T Tengra
9	Like extremely					✓					✓					✓
8	Like very much	✓	✓	✓	✓		✓	✓		✓		✓	✓	✓	✓	
7	Like moderately								✓							
6	Like slightly															
5	Neither like nor dislike															
4	Dislike slightly															
3	Dislike moderately															
2	Dislike very much															
1	Dislike extremely															

[DS=Dry-salted, PC=Pickle-cured, BS=Brine-salted, SDS=Sun-dried salted, SDS+T=Turmeric treated sun-dried salted]

Signature

Appendix-E

Shol (*Channa striatus*) - Room Temperature

1. Dry-salted (DS) Shol

T-Test-Moisture &Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	51.4158	12	1.52905	.44140
	Protein	26.9742	12	.84128	.24286

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & Protein	12	-.993	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture - Protein	24.44167	2.36659	.68318	22.93801	25.94533	35.777	11	.000

T-Test-Moisture & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	51.4158	12	1.52905	.44140
	Fat	3.6258	12	.22232	.06418

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & Fat	12	-.992	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture - Fat	47.79000	1.74984	.50514	46.67820	48.90180	94.608	11	.000

T-Test-Protein & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Protein	26.9742	12	.84128	.24286
	Fat	3.6258	12	.22232	.06418

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Protein & Fat	12	.987	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Protein - Fat	23.34833	.62297	.17984	22.95252	23.74415	129.832	11	.000

T-Test-Moisture &TVB-N

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	51.4158	12	1.52905	.44140
	TVB-N	15.8375	12	8.99261	2.59594

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & TVB-N	12	.974	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture – TVB-N	35.57833	7.51203	2.16854	30.80542	40.35125	16.407	11	.000

2. Pickle-cured (PC) Shol

T-Test- Moisture & Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	52.9155	11	1.44384	.43534
	Protein	25.8991	11	.90802	.27378

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & Protein	11	-.987	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture - Protein	27.01636	2.34481	.70699	25.44110	28.59163	38.213	10	.000

T-Test Moisture & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	52.9155	11	1.44384	.43534
	Fat	3.4982	11	.21489	.06479

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & Fat	11	-.956	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture - Fat	49.41727	1.65052	.49765	48.30844	50.52610	99.301	10	.000

T-Test Protein & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Protein	25.8991	11	.90802	.27378
	Fat	3.4982	11	.21489	.06479

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Protein & Fat	11	.911	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Protein - Fat	22.40091	.71768	.21639	21.91876	22.88306	103.521	10	.000

T-Test- Moisture & TVB-N

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	52.9155	11	1.44384	.43534
	TVB-N	19.1845	11	10.80885	3.25899

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & TVB-N	11	.945	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture – TVB-N	33.73091	9.45544	2.85092	27.37866	40.08316	11.832	10	.000

3. Brine-salted (BS) Shol

T-Test -Moisture & Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	62.7600	10	1.82633	.57753
	Protein	19.1610	10	1.24909	.39500

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & Protein	10	-.993	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture - Protein	43.59900	3.07053	.97099	41.40248	45.79552	44.902	9	.000

T-Test - Moisture & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	62.7600	10	1.82633	.57753
	Fat	2.9860	10	.15946	.05042

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & Fat	10	-.997	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture - Fat	59.77400	1.98533	.62781	58.35378	61.19422	95.210	9	.000

T-Test-Protein & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Protein	19.1610	10	1.24909	.39500
	Fat	2.9860	10	.15946	.05042

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Protein & Fat	10	.996	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Protein - Fat	16.17500	1.09034	.34480	15.39501	16.95499	46.912	9	.000

T-Test-Moisture & TVB-N

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	62.7600	10	1.82633	.57753
	TVB-N	17.4470	10	10.61956	3.35820

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & TVB-N	10	.987	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture – TVB-N	45.31300	8.82266	2.78997	39.00165	51.62435	16.241	9	.000

4. Sun-dried salted (SDS) Shol

T-Test- Moisture & Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	32.1033	12	1.83976	.53109
	Protein	40.0600	12	1.05203	.30369

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & Protein	12	-.987	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture - Protein	-7.95667	2.88317	.83230	-9.78855	-6.12478	-9.560	11	.000

T-Test- Moisture & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	32.1033	12	1.83976	.53109
	Fat	4.5658	12	.39946	.11532

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & Fat	12	-.994	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture - Fat	27.53750	2.23712	.64580	26.11610	28.95890	42.641	11	.000

T-Test- Protein & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Protein	40.0600	12	1.05203	.30369
	Fat	4.5658	12	.39946	.11532

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Protein & Fat	12	.972	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Protein - Fat	35.49417	.67024	.19348	35.06831	35.92002	183.449	11	.000

T-Test-Moisture & TVB-N

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	32.1033	12	1.83976	.53109
	TVB-N	14.6217	12	8.71278	2.51516

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & TVB-N	12	.978	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture – TVB-N	17.48167	6.92395	1.99877	13.08240	21.88093	8.746	11	.000

5. Turmeric treated sun-dried salted (SDS+T) Shol

T-Test- Moisture & Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	33.5754	13	1.92782	.53468
	Protein	39.7285	13	.93794	.26014

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & Protein	13	-.997	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture - Protein	-6.15308	2.86412	.79437	-7.88385	-4.42230	-7.746	12	.000

T-Test- Moisture & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	33.5754	13	1.92782	.53468
	Fat	4.0354	13	.50181	.13918

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & Fat	13	-.994	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture - Fat	29.54000	2.42729	.67321	28.07321	31.00679	43.879	12	.000

T-Test-Protein & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Protein	39.7285	13	.93794	.26014
	Fat	4.0354	13	.50181	.13918

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Protein & Fat	13	.991	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Protein - Fat	35.69308	.44563	.12360	35.42378	35.96237	288.787	12	.000

T-Test-Moisture & TVB-N

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	33.5754	13	1.92782	.53468
	TVB-N	14.4369	13	8.98794	2.49281

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & TVB-N	13	.954	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture – TVB-N	19.13846	7.17156	1.98903	14.80473	23.47219	9.622	12	.000

Shol (*Channa striatus*)-Refrigeration Temperature

1. Dry-salted (DS) Shol

T-Test-Moisture & Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	49.9257	14	.80459	.21504
	Protein	27.7929	14	.30726	.08212

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & Protein	14	-.995	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture - Protein	22.13286	1.11084	.29689	21.49147	22.77424	74.550	13	.000

T-Test- Moisture & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	49.9257	14	.80459	.21504
	Fat	3.7300	14	.17436	.04660

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & Fat	14	-.994	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture - Fat	46.19571	.97810	.26141	45.63098	46.76045	176.719	13	.000

T-Test- Protein & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Protein	27.7929	14	.30726	.08212
	Fat	3.7300	14	.17436	.04660

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Protein & Fat	14	.993	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Protein - Fat	24.06286	.13584	.03631	23.98442	24.14129	662.797	13	.000

T-Test- Moisture & TVB-N

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	49.9257	14	.80459	.21504
	TVB-N	10.9336	14	5.00670	1.33810

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & TVB-N	14	.987	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture – TVB-N	38.99214	4.21439	1.12634	36.55882	41.42546	34.618	13	.000

2. Pickle-cured (PC) Shol

T-Test- Moisture & Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	51.3642	12	.82431	.23796
	Protein	27.0850	12	.44966	.12980

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & Protein	12	-.994	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture - Protein	24.27917	1.27217	.36724	23.47087	25.08746	66.112	11	.000

T-Test- Moisture & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	51.3642	12	.82431	.23796
	Fat	3.4800	12	.18800	.05427

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & Fat	12	-.958	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture - Fat	47.88417	1.00590	.29038	47.24505	48.52328	164.903	11	.000

T-Test-Protein & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Protein	27.0850	12	.44966	.12980
	Fat	3.4800	12	.18800	.05427

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Protein & Fat	12	.970	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Protein - Fat	23.60500	.27111	.07826	23.43275	23.77725	301.614	11	.000

T-Test-Moisture & TVB-N

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	51.3642	12	.82431	.23796
	TVB-N	11.7325	12	5.14286	1.48462

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & TVB-N	12	.991	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture – TVB-N	39.63167	4.32739	1.24921	36.88217	42.38116	31.725	11	.000

3. Brine-salted (BS) Shol

T-Test- Moisture & Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	61.7730	10	1.13632	.35934
	Protein	19.6930	10	.74452	.23544

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & Protein	10	-.996	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture - Protein	42.08000	1.87886	.59415	40.73595	43.42405	70.824	9	.000

T-Test- Moisture & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	61.7730	10	1.13632	.35934
	Fat	3.0600	10	.10360	.03276

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & Fat	10	-.994	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture - Fat	58.71300	1.23936	.39192	57.82642	59.59958	149.809	9	.000

T-Test- Protein & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Protein	19.6930	10	.74452	.23544
	Fat	3.0600	10	.10360	.03276

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Protein & Fat	10	.997	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Protein - Fat	16.63300	.64125	.20278	16.17428	17.09172	82.024	9	.000

T-Test-Moisture & TVB-N

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	61.7730	10	1.13632	.35934
	TVB-N	12.5930	10	6.34918	2.00779

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & TVB-N	10	.979	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture – TVB-N	49.18000	5.24133	1.65745	45.43058	52.92942	29.672	9	.000

4. Sun-dried salted (SDS) Shol

T-Test-Moisture & Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	31.8246	13	1.46691	.40685
	Protein	40.4608	13	.71355	.19790

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & Protein	13	-.998	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture - Protein	-8.63615	2.17954	.60450	-9.95324	-7.31907	-14.287	12	.000

T-Test- Moisture & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	31.8246	13	1.46691	.40685
	Fat	4.6177	13	.34424	.09548

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & Fat	13	-.994	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture - Fat	27.20692	1.80947	.50186	26.11347	28.30038	54.212	12	.000

T-Test- Protein & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Protein	40.4608	13	.71355	.19790
	Fat	4.6177	13	.34424	.09548

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Protein & Fat	13	.993	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Protein - Fat	35.84308	.37386	.10369	35.61715	36.06900	345.673	12	.000

T-Test- Moisture & TVB-N

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	31.8246	13	1.46691	.40685
	TVB-N	10.3177	13	5.40361	1.49869

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & TVB-N	13	.966	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture – TVB-N	21.50692	4.00405	1.11052	19.08730	23.92654	19.366	12	.000

5. Turmeric treated sun-dried salted (SDS+T) Shol

T-Test- Moisture & Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	32.9627	15	1.47483	.38080
	Protein	40.0527	15	.76091	.19647

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & Protein	15	-.992	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture - Protein	-7.09000	2.23171	.57622	-8.32588	-5.85412	-12.304	14	.000

T-Test-Moisture & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	32.9627	15	1.47483	.38080
	Fat	4.2580	15	.36175	.09340

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & Fat	15	-.995	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture - Fat	28.70467	1.83520	.47385	27.68837	29.72096	60.578	14	.000

T-Test- Protein & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Protein	40.0527	15	.76091	.19647
	Fat	4.2580	15	.36175	.09340

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Protein & Fat	15	.994	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Protein - Fat	35.79467	.40331	.10413	35.57132	36.01801	343.740	14	.000

T-Test-Moisture & TVB-N

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	32.9627	15	1.47483	.38080
	TVB-N	10.3700	15	4.97552	1.28467

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & TVB-N	15	.972	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture – TVB-N	22.59267	3.55921	.91898	20.62164	24.56369	24.584	14	.000

Appendix-F

Taki (*Channa punctatus*)-Room Temperature

1. Dry-salted (DS) Taki

T-Test-Moisture & Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	48.5936	11	1.49050	.44940
	Protein	22.5909	11	.69952	.21091

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & Protein	11	-.993	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture - Protein	26.00273	2.18683	.65936	24.53359	27.47186	39.437	10	.000

T-Test- Moisture & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	48.5936	11	1.49050	.44940
	Fat	3.7273	11	.13886	.04187

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & Fat	11	-.995	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture - Fat	44.86636	1.62874	.49108	43.77216	45.96057	91.362	10	.000

T-Test- Protein & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Protein	22.5909	11	.69952	.21091
	Fat	3.7273	11	.13886	.04187

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Protein & Fat	11	.990	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Protein - Fat	18.86364	.56234	.16955	18.48585	19.24142	111.256	10	.000

T-Test-Moisture & TVB-N

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	48.5936	11	1.49050	.44940
	TVB-N	17.2073	11	10.19506	3.07393

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & TVB-N	11	.976	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture – TVB-N	31.38636	8.74597	2.63701	25.51074	37.26199	11.902	10	.000

2. Pickle-cured (PC) Taki

T-Test- Moisture & Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	54.3570	10	1.31312	.41525
	Protein	20.7730	10	.50140	.15856

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & Protein	10	-.993	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture - Protein	33.58400	1.81189	.57297	32.28785	34.88015	58.614	9	.000

T-Test- Moisture & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	54.3570	10	1.31312	.41525
	Fat	3.1870	10	.15585	.04928

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & Fat	10	-.995	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture - Fat	51.17000	1.46832	.46433	50.11962	52.22038	110.203	9	.000

T-Test- Protein & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Protein	20.7730	10	.50140	.15856
	Fat	3.1870	10	.15585	.04928

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Protein & Fat	10	.988	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Protein - Fat	17.58600	.34821	.11011	17.33691	17.83509	159.709	9	.000

T-Test-Moisture & TVB-N

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	54.3570	10	1.31312	.41525
	TVB-N	17.2520	10	10.00290	3.16320

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & TVB-N	10	.980	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture – TVB-N	37.10500	8.71931	2.75729	30.86758	43.34242	13.457	9	.000

3. Brine-salted (BS) Taki

T-Test- Moisture & Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	63.6667	6	1.06755	.43583
	Protein	17.4667	6	.41596	.16982

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & Protein	6	-.999	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture - Protein	46.20000	1.48308	.60546	44.64361	47.75639	76.305	5	.000

T-Test-Moisture & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	63.6667	6	1.06755	.43583
	fat	2.6300	6	.10545	.04305

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & fat	6	-.990	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - fat	61.03667	1.17209	.47850	59.80664	62.26670	127.558	5	.000

T-Test-Protein & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	protein	17.4667	6	.41596	.16982
	fat	2.6300	6	.10545	.04305

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	protein & fat	6	.992	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	protein - fat	14.83667	.31168	.12724	14.50957	15.16376	116.600	5	.000

T-Test-Moisture & TVB-N

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Moisture	63.6667	6	1.06755	.43583
	TVB-N	15.4100	6	9.20098	3.75629

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Moisture & TVB-N	6	.954	.003

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Moisture – TVB-N	48.25667	8.18842	3.34291	39.66345	56.84988	14.436	5	.000

4. Sun-dried Salted (SDS) Taki

T-Test-Moisture & Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	12.4724	17	1.67307	.40578
	protein	46.5241	17	.67563	.16386

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & protein	17	-.993	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - protein	-34.05176	2.34522	.56880	-35.25757	-32.84596	-59.866	16	.000

T-Test-Moisture & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	12.4724	17	1.67307	.40578
	fat	6.5700	17	.51593	.12513

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & fat	17	-.998	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - fat	5.90235	2.18815	.53070	4.77731	7.02739	11.122	16	.000

T-Test-Protein & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	protein	46.5241	17	.67563	.16386
	fat	6.5700	17	.51593	.12513

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	protein & fat	17	.992	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	protein - fat	39.95412	.17671	.04286	39.86326	40.04497	932.244	16	.000

T-Test-Moisture & TVB-N

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	12.4724	17	1.67307	.40578
	TVB-N	17.7582	17	10.10192	2.45008

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & TVB-N	17	.988	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture – TVB-N	-5.28588	8.45253	2.05004	-9.63177	-.93999	-2.578	16	.020

5. Turmeric treated sun-dried salted (SDS+T) Taki

T-Test- Moisture& Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	15.4367	21	1.97357	.43067
	protein	45.1310	21	.67976	.14834

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & protein	21	-.999	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - protein	-29.69429	2.65274	.57888	-30.90180	-28.48677	-51.297	20	.000

T-Test- Moisture & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	15.4367	21	1.97357	.43067
	fat	5.9419	21	.59223	.12924

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & fat	21	-.953	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - fat	9.49476	2.54419	.55519	8.33666	10.65287	17.102	20	.000

T-Test- Protein & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	protein	45.1310	21	.67976	.14834
	fat	5.9419	21	.59223	.12924

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	protein & fat	21	.951	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	protein - fat	39.18905	.21741	.04744	39.09008	39.28801	826.011	20	.000

T-Test-Moisture & TVB-N

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	15.4367	21	1.97357	.43067
	TVB-N	19.5462	21	8.93327	1.94940

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & TVB-N	21	.962	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture – TVB-N	-4.10952	7.05443	1.53940	-7.32066	-.89839	-2.670	20	.015

Taki (*Channa punctatus*)-Refrigeration Temperature

1. Dry-salted (DS) Taki

T-Test- Moisture & Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	47.7393	14	1.15809	.30951
	protein	22.9414	14	.53353	.14259

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & protein	14	-.997	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - protein	24.79786	1.69059	.45183	23.82174	25.77398	54.883	13	.000

T-Test- Moisture & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	47.7393	14	1.15809	.30951
	fat	3.7021	14	.16244	.04341

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & fat	14	-.994	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - fat	44.03714	1.31969	.35270	43.27517	44.79911	124.856	13	.000

T-Test- Protein & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	protein	22.9414	14	.53353	.14259
	fat	3.7021	14	.16244	.04341

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	protein & fat	14	.988	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	protein - fat	19.23929	.37385	.09991	19.02343	19.45514	192.557	13	.000

T-Test- Moisture & TVB-N

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	47.7393	14	1.15809	.30951
	TVB-N	11.8907	14	4.97957	1.33085

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & TVB-N	14	.989	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture – TVB-N	35.84857	3.83764	1.02565	33.63278	38.06436	34.952	13	.000

2. Pickle-cured (PC) Taki

T-Test-Moisture & Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	54.0700	13	.99655	.27639
	protein	20.9485	13	.35256	.09778

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & protein	13	-.994	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - protein	33.12154	1.34758	.37375	32.30721	33.93587	88.619	12	.000

T-Test-Moisture & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	54.0700	13	.99655	.27639
	fat	3.1862	13	.15003	.04161

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & fat	13	-.995	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - fat	50.88385	1.14592	.31782	50.19138	51.57632	160.103	12	.000

T-Test- Protein & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	protein	20.9485	13	.35256	.09778
	fat	3.1862	13	.15003	.04161

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	protein & fat	13	.987	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	protein - fat	17.76231	.20580	.05708	17.63795	17.88667	311.194	12	.000

T-Test- Moisture & TVB-N

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	54.0700	13	.99655	.27639
	TVB-N	12.4731	13	5.30093	1.47021

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & TVB-N	13	.992	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture – TVB-N	41.59692	4.31410	1.19652	38.98994	44.20391	34.765	12	.000

3. Brine-salted (BS) Taki

T-Test- Moisture & Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	63.0467	6	.67775	.27669
	protein	17.7550	6	.20501	.08370

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & protein	6	-.986	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - protein	45.29167	.88060	.35950	44.36753	46.21580	125.984	5	.000

T-Test- Moisture & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	63.0467	6	.67775	.27669
	fat	2.6817	6	.06178	.02522

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & fat	6	-.991	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - fat	60.36500	.73905	.30171	59.58942	61.14058	200.073	5	.000

T-Test- Protein & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	protein	17.7550	6	.20501	.08370
	fat	2.6817	6	.06178	.02522

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	protein & fat	6	.999	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	protein - fat	15.07333	.14334	.05852	14.92291	15.22376	257.581	5	.000

T-Test- Moisture & TVB-N

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	63.0467	6	.67775	.27669
	TVB-N	14.0900	6	7.96374	3.25118

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & TVB-N	6	.982	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture – TVB-N	48.95667	7.29903	2.97982	41.29680	56.61653	16.429	5	.000

4. Sun-dried salted (SDS) Taki

T-Test- Moisture & Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	11.6050	14	1.28359	.34305
	protein	46.8993	14	.60353	.16130

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & protein	14	-.988	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - protein	-35.29429	1.88231	.50307	-36.38110	-34.20748	-70.158	13	.000

T-Test- Moisture & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	11.6050	14	1.28359	.34305
	fat	6.8914	14	.31603	.08446

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & fat	14	-.986	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - fat	4.71357	1.59615	.42659	3.79198	5.63516	11.049	13	.000

T-Test- Protein & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	protein	46.8993	14	.60353	.16130
	fat	6.8914	14	.31603	.08446

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	protein & fat	14	.963	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	protein - fat	40.00786	.31096	.08311	39.82832	40.18740	481.401	13	.000

T-Test- Moisture & TVB-N

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	11.6050	14	1.28359	.34305
	TVB-N	11.6764	14	6.51411	1.74097

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & TVB-N	14	.995	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture – TVB-N	-.07143	5.23841	1.40002	-3.09600	2.95314	-.051	13	.960

5. Turmeric treated sun-dried salted (SDS+T) Taki

T-Test- Moisture & Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	14.5507	15	1.36764	.35312
	protein	45.3387	15	.46710	.12061

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & protein	15	-.997	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - protein	-30.78800	1.83375	.47347	-31.80350	-29.77250	-65.026	14	.000

T-Test-Moisture & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	14.5507	15	1.36764	.35312
	fat	6.5647	15	.34332	.08865

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & fat	15	-.994	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - fat	7.98600	1.70933	.44135	7.03940	8.93260	18.095	14	.000

T-Test- Protein & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	protein	45.3387	15	.46710	.12061
	fat	6.5647	15	.34332	.08865

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	protein & fat	15	.995	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	protein - fat	38.77400	.12999	.03356	38.70201	38.84599	1155.260	14	.000

T-Test- Moisture & TVB-N

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	14.5507	15	1.36764	.35312
	TVB-N	12.8747	15	5.79388	1.49597

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & TVB-N	15	.973	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture – TVB-N	1.67600	4.47423	1.15524	-.80174	4.15374	1.451	14	.169

Appendix-G

Tengra (*Mystus tengra*) - Room Temperature

1. Dry-salted (DS) Tengra

T-Test--Moisture & Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	43.1200	8	1.21022	.42788
	protein	21.5550	8	.37302	.13188

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & protein	8	-.997	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - protein	21.56500	1.58246	.55948	20.24203	22.88797	38.545	7	.000

T-Test-DS-Moisture & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	43.1200	8	1.21022	.42788
	fat	10.1500	8	.38526	.13621

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & fat	8	-.997	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - fat	32.97000	1.59458	.56377	31.63690	34.30310	58.481	7	.000

T-Test- Protein & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	protein	21.5550	8	.37302	.13188
	fat	10.1500	8	.38526	.13621

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	protein & fat	8	.997	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	protein - fat	11.40500	.03071	.01086	11.37933	11.43067	1050.551	7	.000

T-Test-Moisture & TVB-N

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	43.1200	8	1.21022	.42788
	TVB-N	15.7387	8	10.09733	3.56995

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & TVB-N	8	.992	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture – TVB-N	27.38125	8.89770	3.14581	19.94259	34.81991	8.704	7	.000

2. Pickle-cured (PC) Tengra

T-Test-- Moisture & Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	47.4100	7	1.14042	.43104
	protein	19.8429	7	.42390	.16022

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & protein	7	-.996	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - protein	27.56714	1.56304	.59077	26.12157	29.01271	46.663	6	.000

T-Test-Moisture & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	47.4100	7	1.14042	.43104
	fat	9.1029	7	.25421	.09608

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & fat	7	-.987	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - fat	38.30714	1.39190	.52609	37.01985	39.59444	72.815	6	.000

T-Test-Protein & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	protein	19.8429	7	.42390	.16022
	fat	9.1029	7	.25421	.09608

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	protein & fat	7	.970	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	protein - fat	10.74000	.18806	.07108	10.56607	10.91393	151.097	6	.000

T-Test- Moisture & TVB-N

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	47.4100	7	1.14042	.43104
	TVB-N	16.7557	7	10.73545	4.05762

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & TVB-N	7	.975	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture – TVB-N	30.65429	9.62697	3.63865	21.75082	39.55775	8.425	6	.000

3. Brine-salted (BS) Tengra

T-Test--Moisture & Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	58.9040	5	1.31188	.58669
	protein	14.8640	5	.39853	.17823

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & protein	5	-.995	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - protein	44.04000	1.70884	.76422	41.91819	46.16181	57.627	4	.000

T-Test-Moisture & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	58.9040	5	1.31188	.58669
	fat	6.4320	5	.33566	.15011

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & fat	5	-.995	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - fat	52.47200	1.64612	.73617	50.42807	54.51593	71.277	4	.000

T-Test-Protein & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	protein	14.8640	5	.39853	.17823
	fat	6.4320	5	.33566	.15011

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	protein & fat	5	.984	.002

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	protein - fat	8.43200	.09121	.04079	8.31874	8.54526	206.706	4	.000

T-Test-Moisture & TVB-N

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	58.9040	5	1.31188	.58669
	TVB-N	14.5180	5	11.14494	4.98417

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & TVB-N	5	.996	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture – TVB-N	44.38600	9.83874	4.40002	32.16959	56.60241	10.088	4	.001

4. Sun-dried salted (SDS) Tengra

T-Test--Moisture & Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	6.7800	13	1.38514	.38417
	protein	42.5823	13	.32762	.09087

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & protein	13	-.994	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - protein	-35.80231	1.71124	.47461	-36.83640	-34.76822	-75.435	12	.000

T-Test-Moisture & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	6.7800	13	1.38514	.38417
	fat	15.3462	13	.48725	.13514

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & fat	13	-.991	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - fat	-8.56615	1.86911	.51840	-9.69564	-7.43666	-16.524	12	.000

T-Test-Protein & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	protein	42.5823	13	.32762	.09087
	fat	15.3462	13	.48725	.13514

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	protein & fat	13	.995	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	protein - fat	27.23615	.16450	.04562	27.13675	27.33556	596.983	12	.000

T-Test-Moisture & TVB-N

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	6.7800	13	1.38514	.38417
	TVB-N	16.4208	13	9.43004	2.61542

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & TVB-N	13	.978	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture – TVB-N	-9.64077	8.08057	2.24115	-14.52381	-4.75773	-4.302	12	.001

5. Turmeric treated sun-dried salted (SDS+T) Tengra

T-Test- -Moisture & Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	8.5680	15	1.81954	.46980
	protein	41.8553	15	.49062	.12668

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & protein	15	-.996	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - protein	-33.28733	2.30880	.59613	-34.56590	-32.00876	-55.839	14	.000

T-Test-Moisture & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	8.5680	15	1.81954	.46980
	fat	14.6640	15	.68225	.17616

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & fat	15	-.997	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - fat	-6.09600	2.50033	.64558	-7.48064	-4.71136	-9.443	14	.000

T-Test-Protein & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	protein	41.8553	15	.49062	.12668
	fat	14.6640	15	.68225	.17616

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	protein & fat	15	.995	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	protein - fat	27.19133	.19985	.05160	27.08066	27.30201	526.947	14	.000

T-Test- Moisture & TVB-N

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	8.5680	15	1.81954	.46980
	TVB-N	18.5647	15	9.25178	2.38880

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & TVB-N	15	.965	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture – TVB-N	-9.99667	7.51191	1.93957	-14.15662	-5.83671	-5.154	14	.000

Tengra (*Mystus tengra*) -Refrigeration Temperature

1. Dry-salted (DS) Tengra

T-Test--Moisture & Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	42.8022	9	1.00829	.33610
	protein	21.5978	9	.31459	.10486

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & protein	9	-.995	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - protein	21.20444	1.32167	.44056	20.18852	22.22037	48.131	8	.000

T-Test-Moisture & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	42.8022	9	1.00829	.33610
	fat	10.1178	9	.38942	.12981

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & fat	9	-.999	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - fat	32.68444	1.39743	.46581	31.61029	33.75860	70.167	8	.000

T-Test-Protein & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	protein	21.5978	9	.31459	.10486
	fat	10.1178	9	.38942	.12981

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	protein & fat	9	.998	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	protein - fat	11.48000	.07842	.02614	11.41972	11.54028	439.163	8	.000

T-Test-Moisture & TVB-N

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	42.8022	9	1.00829	.33610
	TVB-N	12.3256	9	6.13197	2.04399

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & TVB-N	9	.994	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture – TVB-N	30.47667	5.13040	1.71013	26.53309	34.42024	17.821	8	.000

2. Pickle-cured (PC) Tengra

T-Test--Moisture & Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	47.1288	8	1.03906	.36736
	protein	19.9525	8	.35756	.12642

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & protein	8	-.995	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - protein	27.17625	1.39518	.49327	26.00985	28.34265	55.094	7	.000

T-Test-Moisture & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	47.1288	8	1.03906	.36736
	fat	8.9650	8	.35825	.12666

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & fat	8	-.998	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - fat	38.16375	1.39666	.49379	36.99611	39.33139	77.287	7	.000

T-Test-Protein & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	protein	19.9525	8	.35756	.12642
	fat	8.9650	8	.35825	.12666

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	protein & fat	8	.994	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	protein - fat	10.98750	.03919	.01386	10.95474	11.02026	793.028	7	.000

T-Test-Moisture & TVB-N

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	47.1288	8	1.03906	.36736
	TVB-N	12.6425	8	5.68848	2.01118

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & TVB-N	8	.988	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture – TVB-N	34.48625	4.66433	1.64909	30.58677	38.38573	20.912	7	.000

3. Brine-salted (BS) Tengra

T-Test--Moisture & Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	58.8457	7	1.17639	.44463
	protein	14.7171	7	.44672	.16884

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & protein	7	-.998	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - protein	44.12857	1.62238	.61320	42.62812	45.62902	71.964	6	.000

T-Test-Moisture & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	58.8457	7	1.17639	.44463
	fat	6.5200	7	.27129	.10254

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & fat	7	-.994	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - fat	52.32571	1.44641	.54669	50.98801	53.66342	95.714	6	.000

T-Test-Protein & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	protein	14.7171	7	.44672	.16884
	fat	6.5200	7	.27129	.10254

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	protein & fat	7	.986	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	protein - fat	8.19714	.18500	.06992	8.02605	8.36824	117.232	6	.000

T-Test-Moisture & TVB-N

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	58.8457	7	1.17639	.44463
	TVB-N	11.7000	7	6.53874	2.47141

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & TVB-N	7	.997	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture – TVB-N	47.14571	5.36642	2.02831	42.18261	52.10882	23.244	6	.000

4. Sun-dried salted (SDS) Tengra

T-Test--Moisture & Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	5.7767	9	.60150	.20050
	protein	42.7000	9	.21059	.07020

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & protein	9	-.995	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - protein	-36.92333	.81130	.27043	-37.54695	-36.29972	-136.535	8	.000

T-Test-Moisture & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	5.7767	9	.60150	.20050
	fat	15.5933	9	.31273	.10424

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & fat	9	-.991	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - fat	-9.81667	.91230	.30410	-10.51793	-9.11541	-32.281	8	.000

T-Test-Protein & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	protein	42.7000	9	.21059	.07020
	fat	15.5933	9	.31273	.10424

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	protein & fat	9	.986	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	protein - fat	27.10667	.11102	.03701	27.02133	27.19200	732.494	8	.000

T-Test-Moisture & TVB-N

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	5.7767	9	.60150	.20050
	TVB-N	11.5933	9	6.21408	2.07136

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & TVB-N	9	.997	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture – TVB-N	-5.81667	5.61457	1.87152	-10.13241	-1.50093	-3.108	8	.014

5. Turmeric treated sun-dried salted (SDS+T) Tengra

T-Test-Moisture & Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	7.4960	10	1.03943	.32870
	protein	42.2780	10	.24399	.07715

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & protein	10	-.993	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - protein	-34.78200	1.28197	.40539	-35.69906	-33.86494	-85.798	9	.000

T-Test-Moisture & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	7.4960	10	1.03943	.32870
	fat	14.9580	10	.42240	.13357

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & fat	10	-.993	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture - fat	-7.46200	1.45985	.46165	-8.50632	-6.41768	-16.164	9	.000

T-Test-Protein & Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	protein	42.2780	10	.24399	.07715
	fat	14.9580	10	.42240	.13357

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	protein & fat	10	.983	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	protein - fat	27.32000	.18774	.05937	27.18570	27.45430	460.188	9	.000

T-Test- Moisture & TVB-N

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	moisture	7.4960	10	1.03943	.32870
	TVB-N	11.7770	10	6.07302	1.92046

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	moisture & TVB-N	10	.995	.000

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	moisture – TVB-N	-4.28100	5.04023	1.59386	-7.88657	-.67543	-2.686	9	.025

PUBLISHED PAPERS

List of Published Papers

1. **Farid, F.B.** Latifa, G.A. Nahid, M.N. and Begum, M. 2014. Comparative study of the sensory scores, quality and shelf life study of dry and pickle salted shoal (*C. striatus;* Bloch, 1801) at room temperature (27^0-31^0C). *Int. J. of Fish. and Aquat. Studies.* **2** (1): XX-XX.157-163.

2. **Farid, F.B.** Latifa, G.A. Nahid, M.N. and Begum, M. 2014. Effects of Salting on the shelf lives extension of Sun-dried Shoal (*Channa striatus* Bloch, 1801) and Taki (*C. punctatus;* Bloch, 1793) fish-products stored at Room Temperature (27^0-30^0C). *Int. J. of Multidis. Res. and Dev*. **1** (7): 45-50.

3. **Farid, F.B.** Latifa, G.A. Nahid, M.N. and Begum, M. 2014. Effect of Sun-drying on proximate composition and pH of Shoal fish (*C. striatus*; Bloch, 1801) treated with Salt and Salt-turmeric storage at Room Temperature (27^0 - 30^0C). *IOSR J. of Agric. and Vet. Sci. (IOSR-JAVS).* **7**(9), Ver. III: 01-08.

4. **Farid, F.B.** Latifa, G.A. Nahid, M.N. and Begum, M. 2014. Investigation on the shelf-life quality of dry-salted and sun-dried salted snake-head shoal fish (*Channa striatus* bloch, 1801) stored at refrigerator temperature (4^0c). *Asian Academic Res. J. of Multidis. (AARJMD).* **1** (26):158-171.

5. **Farid, F.B.** Latifa, G.A. Chakraborty, S.C. Nahid, M.N. and Begum, M. 2014. Comparative study on shelf life quality of Brine Salted Taki (*Channa punctatus* Bloch, 1793) and Shoal (*Channa striatus* Bloch, 1801) at Refrigerator Temperature (4^0C). *IOSR J. of Agric. and Vet. Sci. (IOSR-JAVS).* **7** (10): 63-69.

6. **Farid, F.B.** Latifa, G.A. Chakraborty, S.C. Nahid, M.N. and Begum, M. 2014. Nutritional quality analysis of Bangladeshi fish species, *M. tengra* (Hamilton-Buchanan, 1822) preserved with different salt curing methods in laboratory condition. *American J. of Food and Nut.* **2** (6): 100-107.

7. **Farid, F.B.** Latifa, G.A. Nahid, M.N. and Begum, M. 2014. Protective effect of Brine-salting on physicochemical attributes of Taki (*Channa punctatus*; Bloch, 1793) and Tengra (*Mystus tengra*; Hamilton-Buchanan, 1822) fish at room temperature (26^0-32^0C). *Bang. J. of Zool*. **42** (2): 271-276.

8. **Farid, F.B.** Latifa, G.A. Nahid, M.N. and Begum, M. 2014. Comparison of the changes in phisico-chemical characteristics of Dry Salted Snake-head Shoal (*Channa striatus* Bloch, 1801) and Taki (*Channa punctatus* Bloch, 1793) at Room temperature (27^0-31^0C). *Res. J. of Ani. Vet. and Fish. Sci.* **2** (9): 1-6.

9. **Farid, F.B.** Latifa, G.A. Chakraborty, S.C. Nahid, M.N. and Begum, M. 2015. Protective effect of salt (sodium chloride) and turmeric (*Curcuma longa*) on physicochemical attributes of sun-dried Tengra fish *(Mystus tengra*; Hamilton-Buchanan, 1822) at Laboratory condition. *Int. Res. J. of Biol. Sci.* **4** (2): 33-40.

10. **Farid, F.B.** Latifa, G.A. Chakraborty, S.C. Nahid, M.N. and Begum, M. 2015. Biochemical and mineral composition and bacteriological study in some selected fresh water fishes in Meghna river of Bangladesh. *Int. J. of Multidis. Res. and Dev*. **2** (11): 125-130.

11. **Farid, F.B.** Latifa, G.A. Chakraborty, S.C. Nahid, M.N. and Begum, M. 2016. Shelf-life study of Pickle-salted *C. striatus*, *C. punctatus* and *M. tengra* during refrigeration (4^0C) storage. *Int. J. of Sci. Res. and Manag.* **4** (1): 3852-3856.

12. **Farid, F.B.** Latifa, G.A. Chakraborty, S.C. Nahid, M.N. and Begum, M. 2016. Effects of Dry, Pickle and Brine Salting on biochemical and mineral composition and bacterial load of freshwater snakehead fish Taki (*Channa punctatus*). *Int. J. of Adv. Res.* **4** (1): 150-156.

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Farzana Binte Farid
Department of Zoology, University of Dhaka, Dhaka 1000, Bangladesh.

Gulshan Ara Latifa
Department of Zoology, University of Dhaka, Dhaka 1000, Bangladesh.

Mosarrat Nabila Nahid
Department of Zoology, University of Dhaka, Dhaka 1000, Bangladesh.

Mohajira Begum
Institute of Food Science and Technology, BCSIR, Dhaka 1205, Bangladesh

Correspondence:
Farzana Binte Farid
Department of Zoology, University of Dhaka, Dhaka 1000, Bangladesh.

Comparative study of the sensory scores, quality and shelf life study of dry and pickle salted shoal (*C. striatus;* Bloch, 1801) at room temperature (27-31 ^{0}C).

Farzana Binte Farid, Gulshan Ara Latifa, Mosarrat Nabila Nahid and Mohajira Begum

Abstract
A clear understanding on the difference between qualities of Dry-salted and Pickle-salted shoal fish-product has been assessed by analyzing physical changes by acceptability technique and changes in chemical index of Total Volatile base Nitrogen, pH and Free Fatty Acid value at Room temperature (27-31 ^{0}C). In fresh-process condition the values of Total Volatile base Nitrogen , pH and Free Fatty Acid were 5.25 mgN/100 g, 6.3, 1.5% in case of dry-salted and 5.28 mgN/100 g, 6.4 and 1.3% in case of Pickle-salted shoal respectively. This value increased significantly ($p<0.05$)with the time of storage and between this two salted-products these values rapidly increased in pickle-salted than dry-salted shoal fish and the end of 150 days, the pickle-salted shoal fish-product became spoiled whereas dry-salted shoal still in fresh condition. Experimentally it has been proved that the dry-salted shoal fish has longer shelf life (165 days) and has found better way for preservation in laboratory condition.

Keywords: Dry-salting, pickle-salting, Shoal, sensory-scores, quality, shelf-life

1. Introduction
Fish is one of the best sources of proteins, vitamins and minerals and are essential nutrients required for supplementing both infant and adult diets [1]. Fishes contain 72% water, 19% protein and 5% calcium [2]. In terms of weight of food consumed, fish ranks third after rice and vegetables [3, 4]. The inland fisheries resources of Bangladesh are among the richest in the world with only China and India producing more inland fish than Bangladesh. Situated in the delta of the enormous Brahmaputra, Ganges and Meghna river system, the country's water, climatic and soil conditions are highly favorable to inland fisheries and aquaculture. Fish, as soon as it is caught is susceptible to damage and the facilities for processing, storing and distributing the fish caught are inadequate or non-existent in most cases. Microbial action has been known to play a large part in the spoilage of fish [5]. Bacterial spoilage is characterized by softening of the muscle tissue and the production of slime and offensive odors [6]. In the tropics at ambient temperature, spoilage is rapid; fish will spoil within 12-20 h depending on species, method of capture [7]. There is therefore, need for preservation of fish which generally slows down spoilage. These methods are done so as to increase its shelf-life [8].
Salting is a popular procedure for preserving fish. Salting methods are simple and involve salt crystals or brine. There are three types of salting of fish: dry salting, wet salting and a combination of the two methods. Length of salting period as well as salt concentration depends on the expected final product [9]. Sodium chloride diffuses to the outside from muscles due to difference in osmotic pressure between the brine and fish muscle. This process did not continue indefinitely: Sodium and chlorine ions form a water binding complex with protein which itself exerts an osmotic pressure and eventually equilibrium is reached [10]. In salted fish, where the salt concentration reaches ≈20%, high ionic strength causes contraction of the myofibrils and dehydration of proteins. Also, pH of the medium and the type of salts used for salting can influence the degree of protein denaturation [11]. Salting fish is likely to remain in good demand by those who value tradition and taste but it has also gained acceptance in innovative products that provide convenience.
Among the freshwater fish species Shoal (*C. striatus*) is very important due to commercial purpose. This fish bear high market price and are delicious, nutritious and popular to the

consumers. The people of Bangladesh are habituated and preferred to take fish in fresh condition. But it is difficult to reach the fish in fresh condition to the consumers all over the season. More over the peak of snake-headed shoal fish catch in Bangladesh is seasonal. During catching season, the catch is much higher than the consumers need. So it is necessary to take some steps for their proper preservation and marketing and during this period maintain proper quality. The aim of the present study was to determine the quality of dry and pickle salted shoal fish (*C. striatus*) in laboratory condition.

2. Materials and methods

The fresh Shoal fish had been collected from the river Meghana in the early hours of the day and the fishes were brought to the Fish Technology Section, IFST, BCSIR, Dhaka where fishes were carefully washed with cooled tap water. Head, scales, fins, gills and viscera were removed and washed to remove blood, slime and unnecessary flesh. The experiment was conducted for a period of 6 months between January, 2014 and July, 2014.

A fresh flesh sample of shoal fish specimens was taken (6 to 7 slice) randomly which represented the parts from whole body of the fish. Then the slices were chopped with skin and bone and finally ground with an electric blender to make a homogenous sample before being sampled for analysis. Therefore, total cleaned fishes were grouped into 2 batches.

2.1. Method of salting

Sodium chloride (NaCl), also called common salt, and table salt, is generally recognized as a safe, antimicrobial and incidental food additive [12]. Salt has been used as a seasoning and flavor enhancer as well as a preservative or curing agent, had been purchased from the local market. Salt is a powerful depressor of water activity (a^w) of the food [13]. Moreover, it is known that chloride ions are toxic for some microorganisms [14].

2.1.1. Dry salting (DS)

The fresh fishes were enrolled by dry commercial salt (NaCl) of about 30% by weight of the dressed fish (fish weight: salt weight 3:1), stacked in containers and stored for a salting or curing period, at room temperature. In this method, the extracted water of the fish due to salt action had been removed from the container. Thus the fishes are always allowed to remain in dry condition for the production of dry salt cured fish.

2.1.2. Pickle salting: (PS)

The fresh fishes were enrolled by dry commercial salt (NaCl) of about 30% by weight of the dressed fish (fish weight: salt weight 3:1), stacked in containers and stored for a salting or curing period, at room temperature. The salt reacts with the fish and water is extracted out from the fish-body and a salt-solution is formed. Thus in this method, the fishes are always, allowed to remain in such solution for the production of pickle-cured fish.

During salting process, moisture content decreased and salt content increased considerably during the first 6 to 7 days which is called ripening period.

2.2. Storage of the product

At the end of the ripening period, dry and pickle salted product of Shoal fishes was packaging with plastic bag maintaining aseptic condition as far as possible and was stored at room temperature (27-31 ^{0}C). The preservation period of product is linked to the amount of salt added; therefore a straight proportion is present between the amount of salt used and the preservation period [15].

Evaluation of quality changes in dry salted and pickle salted Shoal fish was carried out 15 days interval for room temperature, until the fish become spoil or inedible condition. Two duplicate experiments were conducted at regular time intervals during storage period. Salt crystal was removed from the dry salted and pickle salted product by tissue paper before being sampled for analysis.

There are some parameters which determine the quality of salted fish during storage condition, such as- freshness test by sensory scores, TVB-N value, pH, FFA etc. Freshness test of the fishes indicate the quality test in term of odor, color and appearance in different species. TVB-N value, pH, FFA of fishes is important indicator to determine the quality of fish.

2.3. Estimation of Sensory score

Determination of the quality of Dry and Pickle salted shoal fish was made by trained panel of six judges in BCSIR, IFST Lab. following 9-point hedonic scales. Comparison was carried out in terms of sensory characteristics, such as color, flavor, end texture and general appearance. The panel was requested to rate each sensory feature of the salted product. The average score of 5 was considered to be the borderline of acceptability (9. Like extremely; 8. Like very much; 7. Like moderately; 6. Like slightly ;< 5. Bad.) [16].

Data were analyzed by using SPSS for windows-20 statistical programme. Significance was established at $p< 0.05$ [17].

2.4. Estimation of TVB-N (Total Volatile base Nitrogen)

TVB-N was determined by Conway modified micro-duffision technique [18]. 25 ml of 10% Trichloroacetic Acid (TCA) was added to 2 gm of ground fish sample and kept overnight and then filtrated with known volume. 2% boric acid, TCA, K_2CO_3 and the solutions made from the fish samples were taken into the Conway dishes. After the addition of Potassium carbonate (K_2CO_3), each dish was covered by a piece of glass that was stacked with glue (Paraffin soft white) initially. Then it was kept for 24 hours. The samples and Potassium carbonate (K_2CO_3) reacts to form NH_3 which was absorbed by the boric acid and then the solution of each Conway dish had been titrated by N/70 H_2SO_4 with the help of a micro-burette.

Table: Showing the amount of chemicals taken in the inner and outer chamber of the Conway dishes.

Conway dishes	Inner Chamber		Outer Chamber	
	Chemical	Amount	Chemical	Amount
Blank	2% Boric acid	1 ml	K_2CO_3	1 ml
With Sample	2% Boric acid	1 ml	Sample Solution	1 ml
			K_2CO_3	1 ml

Finally TVB-N was calculated.

Calculation

$$\text{TVB-N} = (\text{titration reading - blank reading}) \times \text{Strength of acid} \times 0.2 \times \frac{\text{Volume of extract}}{\text{Volume of extract taken}} \times \frac{100}{\text{Weight of sample taken}}$$

2.5. Estimation of pH

A 1 g sample of the fish flesh was homogenized in 10 ml of distilled water and the mixture was filtered. The pH of the filtrate was measured using a pH meter (Mettler Toledo 320-s, Shanghi, China) [19].

2.6. Estimation of Free fatty acid (FFA)

Oil sample used throughout the work was prepared by extracting the salted fish by folch reagent (chloroform and methanol in the ratio of 2:1 v/v).The salted fish was first cut into small pieces and then ground. The ground material was then mixed with folch reagent in a large wide mouthed stopper glass bottle for 24 hours at room temperature after 15 minutes stirring with glass rod. Extraction was facilitated by occasional shaking. The mixture was filtered through a Buchner funnel and the filtrate was evaporated in batches under heater or oven at 60 ^{0}C.

Seven gram of well-mixed oil was taken into 250 ml flask and 50 ml ethanol was added, previously neutralized by adding 2 ml phenolphthalein solution. Titration was done by 0.25 N sodium hydroxide with vigorous shaking until permanent final faint pink color appeared and persisted for at least one minute. The value was reported as percentage (%) of free fatty acid expressed as oleic acid. Milli litre of 0.25 N NaOH required for titration corresponds to the percentage of free fatty acid.

3. Results and Discussions

TVB-N, pH value, FFA (Quality parameters) of fresh Shoal fish was 4.41 mgN/100 g, 6.9 and 0.6% respectively (Figure 1).

3.1. Sensory evaluation (Score)

According to the panel's evaluation, the sensory properties of Dry salted (DS) and Pickle salted (PS) shoal fish products were in acceptable condition throughout storage period though, statistically there was significant difference ($p<0.05$) in the sensory evaluation during storage period based on the panel's score (Figure 2). The initial score of the sensory evaluation of DS and PS shoal was 9. But during storage period this score rapidly decreased and at the end of the storage period, the score was 5 in case of DS (165 days) and PS (150 days) Shoal**.** This hedonic rating-scale was applied to evaluate the acceptability of the sun dried fishes by their external morphological and quality changes [20]. This hedonic rating scale was also applied by using 9- points for the sensory evaluation of the dried and dehydrated fish [21].

3.2. Changes in TVB-N (Total Volatile Base Nitrogen) value

TVB-N has been used as an index for the determination of freshness of fish [22, 23]. Volatile nitrogenous bases increase in concentration during the spoilage of fish [24].

Total volatile base nitrogen (TVB-N) is important compound provide a measure of the progress of spoilage that is independent of sensory assessment. The level of TVB-N in fish & fish products are mostly used as spoilage indicator through bacterial activity [25]. The same result have been evident in the present study. TVB-N values were found to vary from 5.25 (o day) to 31.33 mgN/100 g (165 days) for DS and 5.28 (o day) to 34.99 mgN/100 g (165 days) for PS Shoal. Significant statistical differences were found between the initial product and end product ($P<0.05$) after storage period. TVB-N values of the products storage at room temperature showed linearly increasing pattern throughout storage period but neither of the value exceeded the recommended value set for fish regarded as acceptable condition. The limiting level for rejection of TVB-N is 30-40 mgN/100 g for storage at ambient temperature and 20 mgN/100 g for storage at refrigerator temperature [26]. Present findings are in close association with him. The rate of spoilage increases with the time and is closely interrelated (Fig. 3).

The same result had been evident from studys of many researchers [27, 28]. This stage perhaps due to autolysis in the tissue [29].

3.3. Changes in pH value

pH is an indicator of the Extent of microbial spoilage in fish and some proteolytic microbes produce acid after decomposition of carbohydrate, thereby increasing the acid level of the medium [30]. pH value is a reliable indicator of the degree of freshness or spoilage.

The pH in fresh condition fresh- water fish flesh is almost neutral [31]. In the post-mortem period, decomposition of nitrogenous compounds leads to an increase in pH in the fish flesh [32]. The increase in pH indicates the loss of quality. The pH value of Dry-salted (DS) and Pickle-salted (PS) shoal fish-product was increased significantly ($P<0.05$) with storage period. pH value of fresh Shoal fish was 6.9 in our study . But when salt is added with the fish, pH value decrease due to increase of acidic compound and after that among shelf life study pH value increases in the time interval due to increase of basic compounds. In the present study pH value were found to vary from 6.3 (0 day) to 8.1 (165 days) for DS and 6.4 (0 day) to 8.3 (150 days) for PS Shoal (Figure 4).

The fish products are acceptable up to a pH of 6.8 but are considered to be spoiled above a pH of 7.0. [23] The limit of acceptability is usually 6.8 to 7.0, [33] while the initial pH values in the samples were similar to findings of other researchers; the increase in pH values during the storage of room temperature (30-34 ^{0}C) was higher than others. The probable reason of these differences is differences in fish species and different methods of salting.

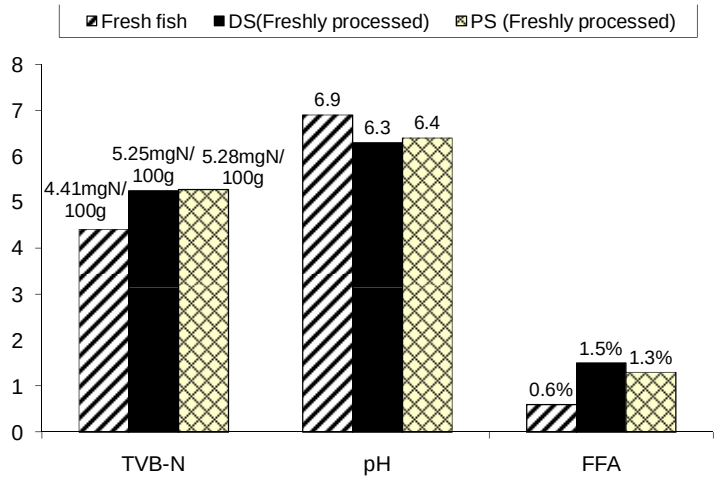

Fig 1: Chemical composition of fresh fish, freshly processed dry (DS) and pickle (PS) salted shoal fish.

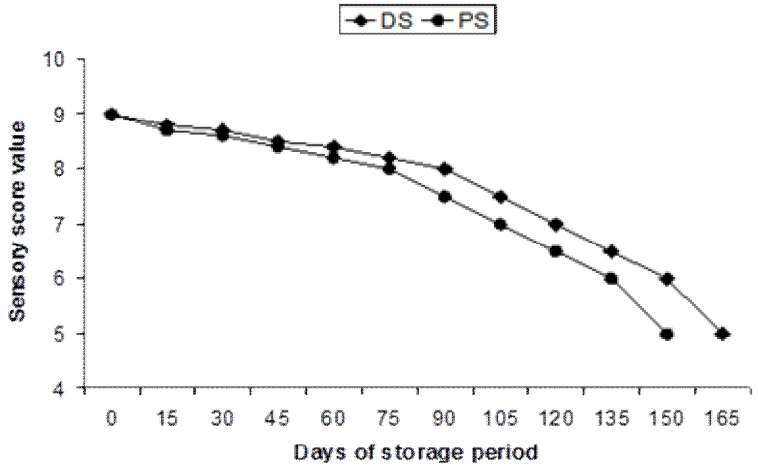

Fig 2: Changes in sensory score of DS and PS Shoal at Room temperature during different days of observation.

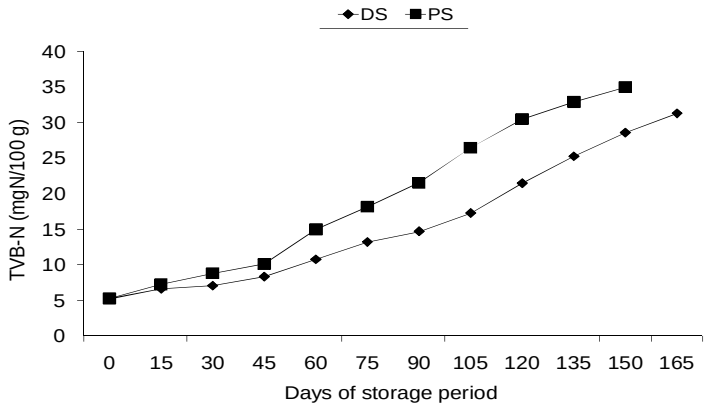

Fig 3: Changes in TVB-N (mgN/100 g) contents of 2 types of salt treated Shoal fish during different duration of storage at Room Temperature.

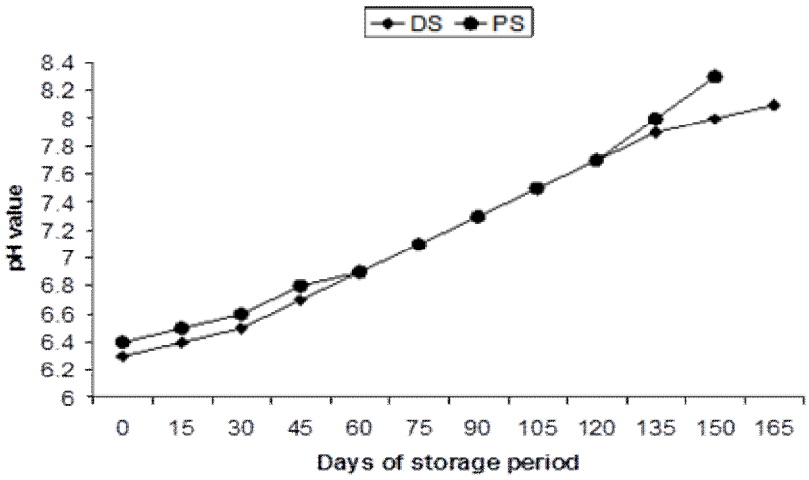

Fig 4: Changes in pH value of Dry and pickle salted Shoal fish during different duration of storage at Room Temperature.

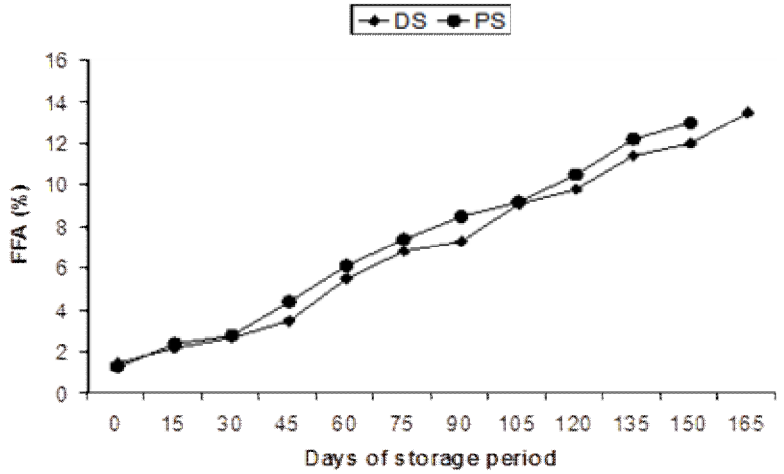

Fig 5: Changes in FFA (Free Fatty Acid) value of 2 types of salt treated Shoal fish during different duration of storage at Room Temperature.

3.4. Changes in FFA (Free Fatty Acid) value

Among the various parameters to assess the extent of deterioration in fish, determination of free fatty acid (FFA) content has been widely used.

Free fatty acid (FFA), a tertiary product of rancidity, increased during storage. The FFA is a measure of hydrolytic rancidity-the extent of lipid hydrolysis by lipase action. In most fish oils, rancidity is noticeable when the FFA (calculated as oleic acid) is in between 0.5%-1.5% [30]. It produced as a result of fat oxidation (rancidity). FFA value is a measure of the extent of oxidative deterioration in oily fish, but it can fall further at latter stages of fish spoilage [34]. A high level of FFA is characteristics of product that have undergone both microbial and biochemical spoilage [10, 35]. This may indicate that greater proportions of unsaturated fatty acids were liberated and were subjected to oxidative splitting at the double bonds. The resulting substances, mostly ketones and aldehydes, appear to be largely responsible for flavor, odor and taste of the salted fish product [36]. The same result was found in the present study.

The FFA value of dry and pickle salted shoal increased gradually with the passing of storage period (figure 5). Significant statistical differences were found between the initial product and end product ($P<0.05$) after storage period. It was vary from 1.5% (o day) to 13.5% (165 day) for DS and 1.3% (o day) to 13% (150 day) for PS Shoal respectively.

Free fatty acid (FFA) value ranged from 2.9% of total lipid on the start day to 7.58, 7.15 and 8.56% on the 60 days of observation in dry salting, wet salting and sundry salted fishes respectively [37]. while the initial value of raw pre-spawning and post-spawning hilsa was 1.15% and 1.21% respectively which after dry salting increased to 7.86% and 8.35% respectively on 18th days of storage at 28 ^{0}C to 32 ^{0}C [38].

The present study denoted that the contents of free fatty acid values are similar with the above mentioned studies. The FFA value increased in a characteristics pattern to a certain level of storage period.

High level of FFA is an indication of microbial spoilage activity [23]. Most fat acidity begins to be noticeable to the palate when the FFA value calculated as Oleic acid is about 0.5-1.5% [39].

4. Conclusion

The wide range of storage period indicates the diversity in the final quality and can be largely attributed to the effect of various conditions upon the salting agents and activities. It is seen that the main factor affecting the quality is time of storage and storage condition.

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Farzana Binte Farid
Department of Zoology, University of Dhaka, Dhaka-1000, Bangladesh

Gulshan Ara Latifa
Department of Zoology, University of Dhaka, Dhaka-1000, Bangladesh

Mosarrat Nabila Nahid
Department of Zoology, University of Dhaka, Dhaka-1000, Bangladesh

Mohajira Begum
Institute of Food Science and Technology, Bangladesh Council of Scientific and industrial Research (BCSIR), Dhaka-1205, Bangladesh

Correspondence:
Farzana Binte Farid
Department of Zoology, University of Dhaka, Dhaka-1000, Bangladesh.

Effects of Salting on the shelf lives extension of sun-dried Shoal (*Channa striatus* Bloch, 1801) and Taki (*C. punctatus*; Bloch, 1793) fish-products stored at room temperature (27^0C - 30^0C)

Farzana Binte Farid, Gulshan Ara Latifa, Mosarrat Nabila Nahid and Mohajira Begum

Abstract
Keeping the quality of fish and fish products at its best is the most important issue in any kind of fish processing. Among them, Drying of fish (dried fish-product called 'shutki' in Bangladesh) is widely used as a traditional method for preservation and considered as a concentrated source of nutrients to Bangladeshi people. So, an experiment was carried out to assess the sensory-evaluation and changes in chemical-compositions of sun-dried salted (SDS) Shoal and Taki fish products stored at room temperature. TVB-N, pH and FFA value increased significantly ($p<0.05$) with the time of storage period and between these two salted products, these parameters rapidly increased in SDS Shoal than SDS Taki fish and at the end of 5 months SDS Shoal fish product became spoiled whereas SDS Taki fish still in fresh condition. TVB-N value was found below the range suggested by various researchers for fish and fish- products.

Keywords: Sun-dried-Salting, Shoal, Taki, Quality-evaluation

1. Introduction
It has been estimated that about 80% of the animal protein in the diet of most people in Bangladesh comes from fish alone [1]. Fresh and dried fish are a popular food in the world, including Bangladesh [2].

The purpose of processing and preservation fish is to get fish to an ultimate consumer in good and acceptable condition. Inadequate preservation techniques would imply a substantial shortfall in fish availability thereby affecting the protein intake [3]. In order to reduce the wastage and spoilage of fish during periods of oversupply, and to enhance long storage, it is necessary to adopt appropriate as well as affordable processing and preservation techniques for fish. Traditional fish processing is an important livelihood activity for large number of people in Bangladesh. The common fish preservation methods are salting, smoking, icing, freezing, drying etc. Of these methods of preservations, drying is one of the most important methods throughout the world. It is a common practice in meat, fish and other animal protein based industry, because it preserved the quality for an extended time and offers several advantages such as insignificant alterations and minimum deterioration in the product. Drying is a traditional method of preserving fish being they have been used for centuries and dried salted products are popular in many areas, particularly in Africa, SE Asia and Latin America. In Bangladesh, About 20% of the artesian catch is sun dried and consumed in the domestic market [4]. Bangladesh earns a significant amount of foreign currency by exporting dried fish and fishery products which ranks third in the export item.

Now a day, the physical and organoleptic qualities of most of the sun-dried products available in the market are not satisfactory for human consumption. There are frequent complaints from the consumers about the quality of the products. To avoid this, in this research we use salt as a preservative before sun-drying. Sodium chloride has traditionally been used in curing and preservation of meat and fish due to its capacity to improve the water holding capacity of proteins. Brining reduces the microorganisms count on dry fish [5]. If the moisture content of fresh fish is reduced during drying to around 25%, bacteria cannot grow and autolytic activity will be greatly reduced, but to prevent mould growth, the moisture content must be reduced to 15% or lower. The presence of salt in the muscle tissue

In addition, it aids the removal of water by osmosis. When salt is added to fish before drying, final moisture content of 35% in the flesh, depending on the salt concentration may be sufficiently low to inhibit bacteria [7]. Sodium chloride diffuses to the outside from muscles due to difference in osmotic pressure between the salt and fish muscle. This process does not continue indefinitely: sodium and chlorine ions form a water binding complex with protein which itself exerts an osmotic pressure and eventually equilibrium is reached [8].

Channa striatus and *Channa punctatus* are popular air-breathing fresh water snake head fishes belongs to Channidae family and have long been regarded as valuable fish in Far East. In Bangladesh, *C. striatus* is known as "Shoal" and *C. punctatus* is known as "Taki". These fish species are available in the fresh water stream in South Asia. Fish lipids are the main sources of polyunsaturated fatty acids (PUFAs) [9]. These fishes' *C. striatus* and *C. punctatus* contain polyunsaturated fatty acids (PUFA), which play important roles in cardiovascular system to reduce the risk of heart attack. Moderate fatty fish like- Shoal and Taki are suitable for sun-drying and dried taki fish is very popular food item among the local people of Bangladesh.

In the traditional storage of dried fish in Bangladesh, no proper measures are normally taken to protect the fish against unfavorable environmental conditions. Therefore, alternative affordable, safe, hygienic and environmental friendly methods must be developed and adopted for fish drying.

Considerable information are available about the nutritional aspects of different types of sun-dried product available in the market but very little is known about their quality whether this products are able to satisfy the consumer or not. The objective of this study was to find a suitable process for the drying environment for preserving fish by adding salt as preservative and to verify the curing method, quality changes during shelf life study and consumer acceptance of salt treated dried Shoal and taki fish products.

2. Materials and Methods

2.1. Collection of the fishes and location of the experiment

Fresh experimental fishes, shoal (*Channa striatus*) and Taki (*Channa punctatus*) had been collected from the river Meghna in the early hours of the day and the fishes were brought to the Fish Technology Section, IFST, BCSIR, Dhaka, Bangladesh for conducting the research activities, starts in the month of October, 2013. The whole experimental period covered 9 months of duration started from October, 2013 to June, 2014.

2.2. Preparation of fish

Fishes were carefully washed with cooled tap water. Head, scales, fins, gills and viscera were removed and again washed with tap water to remove blood, slime and unnecessary flesh. Due to the presence of hard shield like bony elements, bones of head are discarded as the waste.

2.3. Fresh Sample

A fresh flesh sample of Shoal and Taki fish species was taken to the laboratory for quality analysis of fresh experimental fish. About 6 or 7 slices was taken randomly which represented the parts from whole body of the fish. Then the slices were chopped with skin and finally ground with an electric blender to make a homogenous sample before being sampled for analysis.

2.4. Method of Sun-dried salting

Being a safe, antimicrobial and incidental food additive, toxic for some microorganisms, depressor of water activity (a^w) of the food, sodium chloride has been used as a seasoning and flavor enhancer as well as a preservative or curing agent [10, 11, 12].

During this experiment the raw fishes were enrolled by dry commercial salt (NaCl) of about 30% by weight of the dressed fish (fish weight: salt weight 3:1). They were kept on a plastic bade basket in the sun. They were kept in sun regularly during day time (12 a.m. to 3 p.m.) for 7 days as sometimes the sky was cloudy and until the ripening period was over. At the same time, temperature and relative humidity were also recorded. During sun-drying, they were kept covered by dense meshed nylon or mosquito net to avoid outside contamination and prevent bird attack and fly infestation.

During sun-dried salting process, moisture content decreased and salt content increased considerably during the first 7 days which is called ripening period. The ripening of the product was determined by observing the changes in sensory characteristics such as color, texture, flavor etc. and changes in moisture content and salt penetration rate.

2.5. Storage of the product

At the end of drying period (ripening), sun-dried salted product of two fishes was packaging with plastic bag maintaining aseptic condition as far as possible and was stored at room temperature (27-30^{0}C). The preservation period of product is linked to the amount of salt added; therefore a straight proportion is present between the amount of salt used and the preservation period [13].

2.6. Sampling procedures

Evaluation of physical and chemical changes in sun-dried-salted Shoal and Taki fishes were carried out 1 month interval for the room temperature (26 ^{0}C-30 ^{0}C) until the fish become spoil or inedible condition. The experiment was done for second time at regular intervals during storage period. Salt crystal was removed from sun-dried salted fish-product using dry tissue paper before being sampled for analysis.

2.7. Quality analysis

Analytical methods were applied for the determination of chemical composition of the fresh fish and shelf-life quality of processed fish products on experimental basis. The analytical methods are given below**:**

- Sensory score evaluation has been done by using 9-point hedonic scales as described by Peryan and Pilgrim (9. Like extremely; 8. Like very much; 7. Like moderately; 6. Like slightly ;< 5. Bad) [14].
- TVB-N was determined by Conway modified micro-diffusion technique [15].
- pH was used to measure quality deterioration of Shoal and Taki fish using a pH meter (Mettler Toledo 320-s, Shanghi, China) [16].
- FFA of the fish was determined by AOAC method [17].

Data were analysed using SPSS for windows-20 statistical programme. Significance was established at $p< 0.05$.

3. Results & Discussion

TVB-N, pH value, FFA (Quality parameters) of fresh Shoal and Taki fishes were 4.41 mgN/100g, 6.9 and 0.6% and 3.43mgN/100g, 7.0 and 0.5% respectively **(Figure 1)**.

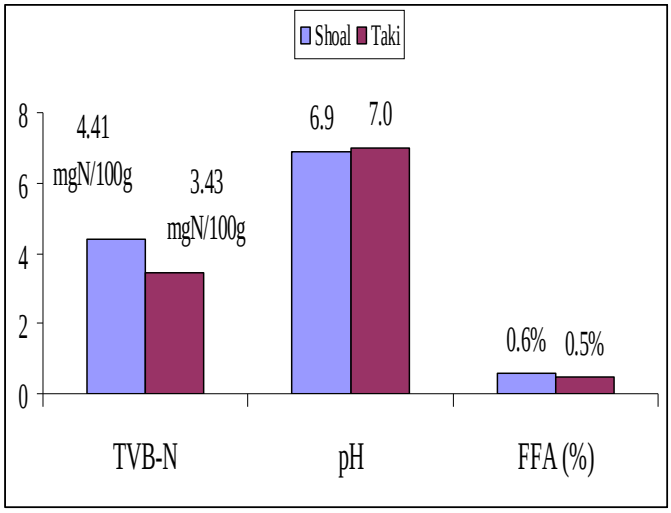

Fig 1: Initial quality of the fresh experimental fishes, Shoal (*Channa striatus*) and Taki (*Channa punctatus*).

3.1. Ripening period: Salting process starts when the surface of fish goes in contact with salt and is completed when all the fish reach the appropriate salinity, taste, consistency and odor. Many workers agree that maximum salt uptake takes place within 6-7 days of salting without further uptake during subsequent storage [18, 19, 20]. Similar results also obtained in the present study where the fish contained maximum salt content in 7 days of salting. Decrease in moisture content have found during salting, which is due to the fact that the osmotic migration of salt into and water out of the fish [8, 21]. This led to increase in salt content and consequently extend shelf life of the products [22, 23]. During salting process, the changes in physico-chemical characteristics takes place and in certain stage the original characteristics of the raw fishes is found virtually absent. This stage is regarded as salt ripening of fish**.** According to Voskresensky these changes are induced by enzyme which breakdown both proteins and fats [24]. Borgstrom reported that salt ripening is the autolytic phenomena caused by the enzyme of the muscle or gastrointestinal tract [25]. During ripening changes induced by enzyme led to breakdown of proteins in tissue structures of muscle and the body organs of the fish. As a result some of the nitrogenous substances chiefly of low molecular weight diffuse from the fish into the salt brine. In present study, a comparatively higher salt content was observed in SDS Taki than SDS Shoal in 7th day **(Table 1)**.

Table 1. Changes in salt penetration rate and its effect on moisture content in experimental fishes Shoal (*Channa striatus*) and Taki (*Channa punctatus*) during 7 days of ripening period

Days of ripening period	SDS Shoal		SDS Taki	
	Moisture (%)	Salt (%)	Moisture (%)	Salt (%)
0*	77.03	0	78.65	0
1	57.31	6.9	56.08	6.8
2	39.09	9.7	43.84	10.22
3	35.87	10.1	38.07	11.30
4	33.12	11.3	31.42	12.57
5	32.55	12.8	26.57	13.25
6	30.48	13.2	19.05	14.16
7	29.77	14.5	9.77	14.72

3.2. Sensory evaluation and score

During present study, high quality sun-dried salted fish products with excellent sensory and physical properties were obtained through this salting process. Now it is of interest to see how long these salted-dried products could be kept in acceptable condition during storage at room temperature. There were significant change in color, flavor, taste and texture in two fish species subjected to Sun-dried-salting methods. The table shows that with the lapse of storage time both products produce a salty taste with different degree of smell, color and texture. The changes of color from whitish to brownish may be due to lipid oxidation during storage period. This is quite clear from the present study that lipid oxidation presence of oxygen was more prominent than that of products stored in room temperature. In the present study there was no fungal attack in sun-dried salted Shoal and Taki fish products. According to the panel's evaluation, the sensory properties of sun-dried salted (SDS) Shoal and Taki fish-products were in acceptable condition throughout storage period though, statistically there was significant difference ($p<0.05$) in the sensory evaluation during storage period based on the panel's score. The initial score of the sensory evaluation of SDS Shoal and Taki was 9. But during storage period this score rapidly decreased and at the end of the storage period, the score was 5 in case of SDS Shoal (5 month) and SDS Taki (8 month). Yu applied this hedonic rating scale to evaluate the acceptability of sun dried fishes by their external morphological and quality changes [26]. Morshed applied the hedonic rating scale by using 9-points for the sensory evaluation of the dried and dehydrated fish [27].

The sensory analysis of salted- sun dried (SDS) Shoal and Taki fishes were done and reported that the quality of salted sun- dried Taki fish-product was much better **(Table 2)**.

Table 2: Sensory evaluation of Sun-dried salted Shoal and Taki fish-products during different days of observation at room temperature

Obser-vation period	Product	Flavor(Smell / Odor)	Color	Texture	Comment /Remarks	Hedonic scale score (0-9)
0 Day	Shoal	Attractive salty odor	Whitish	Tough	Excellent	9
	Taki	Attractive salty odor	Whitish	Comparatively Tougher & shrunken	Excellent	9
1 month	Shoal	Attractive salty and slightly fishy odor	Whitish	Comparatively Tougher& shrunken	Excellent	8.6
	Taki	Stimulating salty & fishy odor dominant	Whitish	Comparatively Tougher & shrunken	Excellent	8.7
2 month	Shoal	Attractive salty and fishy odor dominant	Whitish	Comparatively Tougher& shrunken	Excellent	8.5
	Taki	Flavorful salty & fishy odor	Whitish	Firm	Very good	8.6
3 month	Shoal	Slightly salty & characteristic fishy odor	Fade brown	Semi-Elastic	Very much good	8.0
	Taki	Characteristic fishy odor	Whitish	Firm	Good	8.5
4 month	Shoal	Slightly salty odor present	Yellowish brown	Elastic	Slightly Good	7.5
	Taki	Characteristic fishy odor dominant	Whitish	Firm	Moderately good	8.0
5 month	Shoal	Slightly fishy odor present	Yellowish brown	Slightly soft	Neither like or dislike	5
	Taki	Distinctive fishy & slightly salty odor	Whitish	Firm	Slightly good	7.5
6 month	Shoal	Faded odor	Faded radish	Comparatively soft.	Rejected	4.5
	Taki	Fishy odor dominant	Whitish	Nearly firm	Slightly accepted	6.5
7 month	Shoal	*	*	*	*	*
	Taki	Salty odor absent	Whitish	Nearly firm	Just accepted	6
8 month	Shoal	*	*	*	*	*
	Taki	Slightly faded fishy odor	Fade	Comparatively soft	Neither like or dislike	5
9 month	Shoal	*	*	*	*	*
	Taki	Slightly Rancid odor	Fade	Comparatively soft	Rejected	4.5

*= Rejected due to sensory evaluation

3.3. Changes in Total Volatile base Nitrogen (TVB-N) value: Total volatile base nitrogen (TVB-N) is important compound provide a measure of the progress of spoilage that is independent of sensory assessment. Wallace has pointed out that TVB-N is better index of spoilage [28]. The result obtained for TVB-N is presented in **figure-2**.

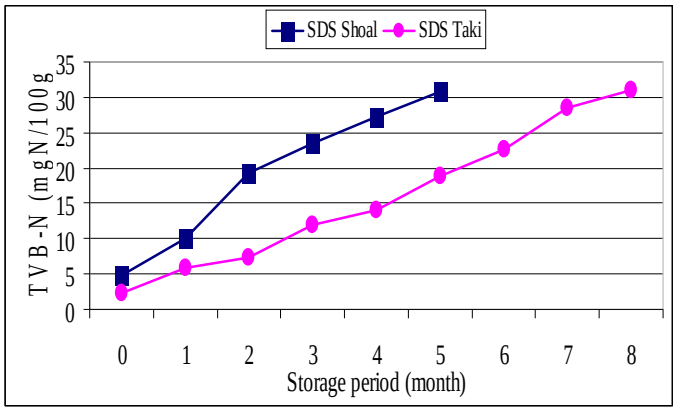

Fig 2: Changes in TVB-N (mgN/100g) contents of sun-dried salted (SDS) Shoal and Taki fish during different duration of storage at room temperature.

TVB-N values were found to vary from 4.89 (o day) to 30.86 mg N/100g (5 month) for SDS Shoal and 2.27 (o day) to 31.02 mg N/100g (8 month) for SDS Taki. TVB-N values of the products storage at room temperature showed linearly increasing pattern throughout storage period but neither of the value exceeded the recommended value set for fish regarded as acceptable condition. The limiting level for rejection of TVB-N is 30-40mgN/100g for storage at ambient temperature [29]. The present findings are in close association with him. The rate of spoilage increases with the time and is closely interrelated. The same result had been evident from other researchers [30, 31]. The stage perhaps due to autolysis in the tissue [32].

3.4. Changes in pH value: pH is an indicator of the extent of microbial spoilage in fish and some proteolytic microbes produce acid after decomposition of carbohydrate, thereby increasing the acid level of the medium [33]. The pH value is a reliable indicator of the degree of freshness or spoilage. The pH in fresh condition freshwater fish flesh is almost neutral [34]. In the post-mortem period, decomposition of nitrogenous compounds leads to an increase in pH in the fish flesh [35]. The increase in pH indicates the loss of quality. Although, it was

stated that a measurement of pH is unreliable for most species of fish because the end products of spoilage of both alkaline and acidic nature tend to neutralize each other [36]. Most microorganisms grow the best at pH values between 6.6 and 7.5, whereas only a few grow at a pH below 4.

pH value of fresh Shoal and Taki fish were found 6.9 and 7.0 respectively in the present study. But when salt was added with fish, pH value decreases rapidly 6.2 for SDS Shoal and 5.9 for SDS Taki. This result is higher than that reported by Riebroy, who found that the pH in Thai-fermented fish mince for fresh fish is 6.3 , while fermented is 4.6 [37]. This result is in the normal ranges reported by other researchers [38, 39, 40]. After that the pH value of SDS shoal and taki fish-product was increased significantly (P<0.05) with storage period at room temperature. In the present study pH value were found to vary from 6.2 (0 day) to 8.3 (5 month) for SDS Shoal and 5.9 (0 day) to 7.9 (8 month) for SDS Taki **(Figure 3)**.

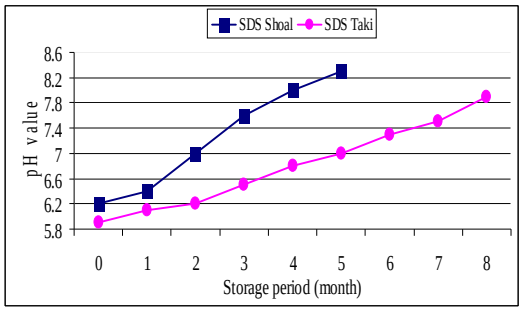

Fig 3: Changes in pH value of sun-dried salted (SDS) Shoal and Taki fish during different duration of storage at room temperature.

3.5. Changes in FFA (Free Fatty Acid) value: The FFA value which indicates the rancidity of fat of 2 types of sun-dried salted fishes during storage at room temperature was shown in **Figure 4**.

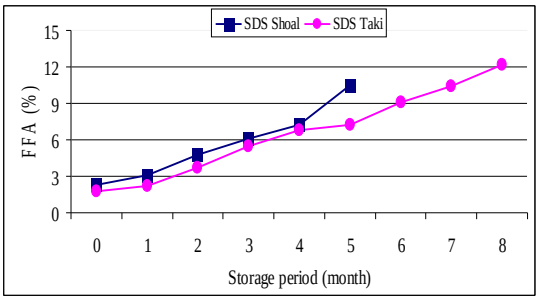

Fig 4: Changes in FFA (%) value of sun-dried salted (SDS) Shoal and Taki fish during different duration of storage at room temperature.

The FFA value of sun-dried salted shoal and taki fish increased gradually with the passing of storage period. FFA value is a measure of the extent of oxidative deterioration in oily fish, but it can fall further at latter stages of fish spoilage [41]. A high level of FFA is characteristics of product that have undergone both microbial and biochemical spoilage [8, 42]. This may indicate that greater proportions of unsaturated fatty acids were liberated and were subjected to oxidative splitting at the double bonds. The resulting substances, mostly ketones and aldehydes, appear to be largely responsible for flavor, odor and taste of the salted fish product [43]. The same result was found in the present study.

In the present study FFA values were found to vary from 2.3% (o day) to 10.5% (5 month) for SDS Shoal and 1.8 % (o day) to 12.2% (8 month) for SDS Taki respectively. After 6 and 24 months of storage (at-13^{0}C to -25^{0}C) the appearance of considerable contents of FFA with certain bad odors and off flavors presumably derived from the degradation of fats, greatly reduced the consumer appeal of trout [44]. The present finding is closely related with that of Rahman who reported the FFA value was 1.16% for raw hilsa fish and after 8 weeks of observation, the FFA value of dry salted hilsa products at room temperature (26 – 30 ^{0}C) reached to 11.7% [45]. Free fatty acid (FFA) value ranged from 2.9% of total lipid on the start day to 7.58, 7.15 and 8.56% on the 60 days of observation in dry salting, wet salting and sundry salted fishes respectively [46]. While **t**he initial value of raw pre-spawning and post-spawning hilsa was 1.15% and 1.21% respectively which after dry salting increased to 7.86% and 8.35% respectively on 18th days of storage at 28^{0}C to 32^{0}C [47]. Free fatty acid contents increased from 4.55-5.12% within 4 days of drying and then gradually increased up to 10 days of drying (6.86%) [48]. The present study denoted that the contents of free fatty acid values are similar with the above mentioned studies. The FFA value increased in a characteristics pattern to a certain level of storage period.

4. Conclusion

The present investigation revealed that, processing and storage significantly affected the quality of fish-products. The application of salt was much more effective in drying method and subsequent uses of salt are required to keep fish-products longer from deterioration. The results of chemical analysis and sensory evaluation carried out, proves that the overall shelf-life quality of sun-dried Taki fish product is best. Commercial traders those who produce market dry-fish in our country may be asked to follow the suggestions made over here on the basis of the findings of the present study.

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Effect of Sun-drying on proximate composition and pH of Shoal fish (C. striatus; Bloch, 1801) treated with Salt and Salt-turmeric storage at Room Temperature (27^0 - 30^0C)

Farzana Binte Farid*, Gulshan Ara Latifa, Mosarrat Nabila Nahid and Mohajira Begum[1]
Department of Zoology, University of Dhaka, Dhaka 1000, Bangladesh,
[1]Institute of Food Science and Technology, BCSIR, Dhaka 1205, Bangladesh.

Abstract: *Sun-drying is one of the most important low-cost methods of fish-preservation and the products provide nutrients to all categories of people through the world including Bangladesh. The study was conducted to obtain a better understanding difference between sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) Shoal fish-product in laboratory-condition by analyzing proximate-composition and pH using standard methods of analyses. In fresh-process condition, the values of moisture (%), protein (%), fat (%, ash (%)and pH value were 29.77%, 41.48%, 5.10%, 22.80% and 6.2 in case of SDS Shoal fish and 30.92%, 41.0%, 4.79%, 22.41% and 6.3 respectively in case of SDS+T Shoal fish product. During storage period, the values of moisture (%) and pH were significantly ($p<0.05$) increased 35.26% and 8.3 in SDS (165 days) and 36.98% and 8.2 in SDS+T (180 days) respectively. The values of protein (%), fat (%) and ash (%) content were significantly ($p<0.05$) decreased 38.62%, 3.81% and 21.44 % respectively in case of SDS Shoal (165 days) and 38.01%, 3.26% and 21.07% (180 days) respectively in case of SDS +T Shoal fish. Experimentally it has been proved that the fishes preserved in SDS +T has longer shelf life and has found better way for preservation.*

Key Words: *Proximate-composition, pH, Sun-dried-salting, Shoal, Turmeric-treated-sun-dried-salting.*

I. Introduction

Nutritional studies have proved that fish protein rank in the same class as chicken protein and are superior to beef protein, milk and egg albumin [1]. Furthermore, fish is rich in protein with amino acid composition which is essential for the maintenance of a healthy body.[2, 3, 4].

Among the good quality animal protein sources, fish is the most perishable. Fish is the most susceptible animal to autolysis, oxidation and hydrolysis of fats and microbial spoilage [5]. The deterioration is believed to cause mainly by the bacterial activity, which brings about very noticeable changes in the texture, flavor, odor and general appearance of the product. The microorganisms present on fish include those which are associated with the raw material and acquired during harvesting, handling and processing [6, 7]. Fishes are in constant interaction with microorganisms which cause spoilage and their metabolic activities result in the appearance of slime, gross discoloration and strong odors and which become offensive to customers [8, 9].

Fish processing has been practiced in Bangladesh for a long time, simplest method employed are - drying, salting and semi-fermentation. The process of fish drying involves the removal of moisture from fish flesh; this could be achieved through sun-drying, smoke-drying, application of pressure and use of absorbent pads. Sun drying is presumably the oldest method of fish preservation employing hot heat from sun and atmospheric air [10]

A bulk of the total artisanal marine sector is being utilized for production of traditional dried products. Drying, the oldest, easiest and excellent way of fish processing has been introduced in our country by the Arabian saints and businessmen who have been believed to be pioneer in the production and marketing of dried fish products throughout the world since the Egyptian civilization [11]. Presently this low-cost, simple and popular fish processing method is being practiced extensively in our country as well as throughout the world. Although dried fishes do not give similar flavor, taste or texture of fresh fish, it is liked and consumed by a large number of people of the world due to characteristic taste and flavor developed in the dried fishery products during drying process. Dried fish is an important source of animal protein is providing nutrition of the poor and economically disadvantaged people of Bangladesh. Today, dried fish has become accepted in most people's diets. It is no longer considered a poor man's dish and some dried fish are served as a snack in bars and hotels.

According to local estimate artisanal catch comprises 95% of the total marine catch in our country of which 20% is being sun-dried and consumed in the domestic market [12]. Bangladesh earns a significant amount of foreign currency by exporting dried fish and fishery products which ranks third in the export item.

* To whom communication should be made, e-mail: farzanafarid79@gmail.com

Bangladesh earned about taka 3.71 crore as foreign currency by exporting about 272 tons of dried fish and fishery products in 2004-2005[13].

In addition, the annual fish harvest fluctuates seasonally, with periods of high and low supply. During the periods of high supply, a lot of fish is spoiled and washed. It is estimated that about 30% (about 3,07,500 mt) of the freshly harvested fish are spoiled every year due to lack of poor processing and preservation at artisanal fishermen level, while acute shortage and increase costs of fish are experienced in periods of low harvest [14]**.**

In order to reduce the wastage and spoilage of fish during periods of oversupply, and to enhance long storage, it is necessary to adopt appropriate as well as affordable processing and preservation techniques for fish especially in the artisanal fisherman's environment.

In the traditional storage of dried fish in Bangladesh, no proper measures are normally taken to protect the fish against unfavorable environmental conditions. Dry fishes are normally kept in gunny bags or in bamboo baskets. In the humid months during the monsoon, the average relative humidity in Bangladesh is over 85%. Dried fish in this period absorbs moisture from air, resulting in a higher water activity which is favorable for microbial growth.

Therefore, alternative affordable, safe, hygienic and environmental friendly methods must be developed and adopted for fish drying. In this study salt and turmeric are also used with sun drying as a preservative. Sodium chloride (NaCl), also called common salt, and table salt, is generally recognized as a safe, antimicrobial and incidental food additive [15]. Salt has been used as a seasoning and flavor enhancer as well as a preservative or curing agent, had been purchased from the local market. Salt is a powerful depressor of water activity (a^w) of the food [16]**.** Moreover, it is known that chloride ions are toxic for some microorganisms [17]. Although, large amounts of salt give fish a very salty taste. At the same time many of the nutrients are lost if too much salt is used.

Turmeric is one of the oldest known anti-bacterial ingredients used by the ancient civilizations. In Bangladesh, turmeric is cheap, easily available and is considered as one of the important ingredient for cooking any kind of dish. Even in some parts of Bangladesh, rural people usually use turmeric for short time preservation of small sized fishes. But the scientific information's about the use of turmeric in fish preservation are totally absent.

Fresh, dried and powered form of turmeric is used as a newly introduced element in salt curing method with a hypothesis that it would work as protection against insect, pest, fungus and other pathogens and also a longer term preserver.

Biochemical quality assessments are necessary to ensure the food safety of any processed products. Only few studies on the biochemical and nutritional changes are available during dehydration [18]**.** But, no literature is found about sun-dried salted and turmeric treated sun-dried salted fresh water fish.

II. Materials And Methods

2.1. Collection of the fishes and location of the experiment:

The raw fish (Shoal) had been collected from the river Meghan in the early hours of the day and the fishes were brought to the Fish Technology Section, IFST, BCSIR, Dhaka for conducting the research activities, starts in the month of January, 2014.The whole experimental period covered 6 months of duration started from January, 2014 to July, 2014.

2.2. Preparation of fish:

The fishes were carefully washed with cooled tap water. Head, scales, fins, gills and viscera were removed and again washed with tap water to remove blood, slime and unnecessary flesh. Because of hard and large bones of the head, the bones and head of Shoal are included as the waste.

2.3. Fresh Sample:

A fresh flesh sample of shoal fish species was taken to the laboratory for quality analysis. 6 or 7 slice was taken randomly which represented the parts from whole body of the fish. Then the slices were chopped with skin and bone and finally ground with an electric blender to make a homogenous sample before being sampled for analysis.

2.4. Sun-dried-salt curing method:

During this experiment the raw fishes were enrolled by dry commercial salt (NaCl) of about 30% by weight of the dressed fish (fish weight: salt weight 3:1). They were kept on a plastic bade basket in the sun. They were kept in sun regularly during day time (12 a.m. to 3 p.m.) for 2-7 days as sometimes the sky was cloudy and until the ripening period was over. At the same time, temperature and relative humidity were also recorded. During sun-drying, they were kept covered by dense meshed nylon or mosquito net to avoid outside contamination and prevent bird attack and fly infestation.

2.5. Turmeric treated sun-dried-salt curing method:

During this experiment the raw fishes were enrolled by dry commercial salt (NaCl) of about 30% by weight of the dressed fish (fish weight: salt weight 3:1) and Turmeric powder of about 1% of dressed fish weight. They were kept on a plastic bade basket in the sun. They were kept in sun regularly during day time (12 a.m. to 3 p.m.) for 2-7 days until the ripening period was over. At the same time, temperature and relative humidity were also recorded. During sun-drying, they were kept covered by dense meshed nylon or mosquito net to avoid outside contamination and prevent bird attack and fly infestation.

2.6. Sampling procedures:

Evaluation of physical and biochemical changes in sun-dried- salted and turmeric treated Sun-Dried Salted shoal fishes were carried out 15 days interval for the room temperature (27^0C-30^0C) until the fish become spoil or inedible condition. Two duplicate experiments were conducted at regular time intervals during salting period.

6 or 7 slice was taken randomly which represented the parts from whole body of the fish. Salt crystal were removed from sun dried salted fishes and mixture of Salt and turmeric were removed from turmeric treated sun dried salted product by tissue paper. Then the slices were chopped with skin and bone and finally ground with an electric blender to make a homogenous sample before being sampled for analysis.

2.7. Proximate composition analysis:

Proximate compositions of fish were determined by conventional method of AOAC (Association of Official Analytical Chemicals) on weight basis [19].

2.7.1. Estimation of moisture: About 5 gram of previously prepared fairly minced samples were taken into each known weight basin and weighed in a digital balance (Toledo, Switzerland). The samples were allowed to dry into the oven (Memmet 854 Schwabach) at 105^0C for 24 hours in order to remove the moisture until constant weight. After that, the basins are taken out of the oven, cooled in a desiccators and were weighed in a digital balance.

Calculation

$$\% \text{ of Moisture} = \frac{\text{Weight Loss}}{\text{Original Weight of Sample Taken}} \times 100$$

2.7.2. Estimation of protein: The protein content of the fish was determined by micro-kjeldahl method [20]. It involves conversion of organic nitrogen to ammonium sulphate by digestion with concentrated sulphuric acid in a microkjeldahl flask. The digest was diluted, made alkaline with sodium hydroxide and distilled. The liberated ammonia was collected in a boric acid solution and was determined titrametrically.

Calculation

$$\frac{\% \text{ of } N_2(\text{titration reading - blank reading}) \times \text{strength of acid} \times 0.002 \times \frac{100}{5} \times 100}{\text{weight of sample taken}}$$

For most routine purposes the percent of protein in the sample is then calculated by multiplying the % of N_2 with an empirical factor of 6.25 for fish.

% of protein = % of total $N_2 \times 6.25$

2.7.3. Estimation of fat: About 5 g of the homogenous sample was taken into conical flasks and 10 ml of folch reagent (Chloroform: Methanol = 2:1) was added into the sample and homogenized properly and kept in air-tight condition for 24 hours. Fat contents of the fish muscle react with that solvent and remains in the solution. After 24 hours the solution of the flask was filtered in another weighed conical flask through a filter paper. Then these flasks were given in a hot water bath to dry up and removed the solvent. After that the flasks were kept into an oven for an hour to get the actual fat content. Then the flasks were weighed in an electronic balance to get the amount of fat content.

Calculation

$$\% \text{ of Fat} = \frac{\text{Weight of the residue}}{\text{Weight of sample taken}} \times 100$$

2.7.4. Estimation of ash: About 4-5 g fish sample was weighed into a pre-weighed crucible. The crucible with the contents was heated first over a long flame till all the material was completely churned. Then it was transferred in the Muffle Furnace held at dark red at a rate of 600^0C for 5 hours until the residue become white. The crucible were cooled in desiccators and weighed. Finally the % of ash content was calculated.

Calculation

$$\% \text{ of ash} = \frac{\text{Weight of fish}}{\text{Weight of sample taken}} \times 100$$

2.8. Estimation of pH:

A 1 g sample of the fish flesh was homogenized in 10 ml of distilled water and the mixture was filtered. The pH of the filtrate was measured using a pH meter (Mettler Toledo 320-s, Shanghi, China) [21].

2.9. Statistical analysis:

Data were analyzed by using SPSS for windows-20 statistical programme. Significance was established at $p< 0.05$ [22].

III. Results And Discussion

The bio-chemical composition (proximate composition and pH value) of fish is an important aspect in fish processing as influences both the keeping quality and the technological characteristics of the fish. The proximate composition (moisture, protein, fat and ash) and pH of Shoal fish in fresh condition as well as sun-dried-salted (SDS) and turmeric treated sun-dried-salted (SDS+T) condition were determined. The proximate composition of fish varied from species to species and even within the same species from one individual to another [23]. The body composition of fish seems to depend on sex, season and diet [24]. Starvation alters body constituents. Fat and protein are used as a source of energy, decreasing progressively during starvation while water content increases proportionally [25, 26].

Moisture, protein, fat, ash and pH value was 77.03%, 17.32%, 4.62%, 1.44% and 6.9 in case of fresh shoal fish respectively (Fig. 1). Fresh fish samples presented a high moisture and low protein content, similar to previously reported [27]. Fish species with low levels of fat are suitable to be processed [28].

Changes in proximate composition in sun-dried-salted and turmeric treated sun-dried-salted shoal fish products during different days of observation period through shelf life study were shown in Fig. 2 & 3.

3.1. Moisture (%): In case of SDS and SDS+T the decrease in moisture content become emphasize during the sun drying. This step which is normally combined with salting has duel effects such as the lowering of the water activity (aw) level and a specific inhibitory effect on the growth of some species of microorganisms through the Na+ ion. So the two steps (Salting and Drying) are interrelated to reduce the moisture sufficiently. The decrease in moisture is due to osmotic migration of salt into and water out of the fish [29, 30]. Decrease in moisture led to increase in salt content and consequently extend shelf life of the products [31, 32].

In present experiment, moisture content was found 29.77% and 30.92% in freshly processed SDS and SDS+T shoal and after completing the duration of storage period, it was found 35.26% (165 days) and 36.98% (180 days) respectively (fig. 2 & 3).

In case of sun- dried salting and sun-dried salt +turmeric fish, significant reduction of moisture content was assumed to be increased due to action of salting and sunlight, because, the salt uptake consequently moisture loss is temperature dependent [33]. Similar opinion about the influence of air temperature in drying was described by [34]. They opined that even a small increase of only a few degrees in the intensity of sun-light may appreciably improve the over-all efficiency of the operation. This is certainly a surprising distinctive feature in this study. Fish dried after salting with 15 to 30% salt or brining in 9.5 to 15% brine solution for 18 hours showed higher degree of retention of all the materials than plain dried fish [35].

3.2. Protein (%): The product with highest protein and fat content was referred to as a nutritious food. In general dried fish contains more nutrient than fresh fish [36]. Nutritionally dehydrated products are very good and neither the nutritive value nor the digestibility of the protein is adversely affected due to dehydration process [37, 38]. Protein is the major nutrient factor contributed by animals to human diet. Fish is a major supplier of this factor. Fish as a source of protein supplies protein of high class quality compared to protein of other animal sources. Fish is however a highly bio-perishable material postmortem. Different technologies, including the two applied in this research have been used to preserve both the intrinsic and extrinsic qualities of fish soon after harvest. In this work therefore, an effort to find out which of these preservation methods; Sun-dried salting and turmeric treated Sun-dried salting, ensures better proximate composition and shelflife storage at Room Temperature (27^0 - 30^0C).

In SDS and SDS+T shoal fish products, protein content was found 41.48%, and 41.0%, in fresh-process condition and after completing the duration of storage period it was decreased into 38.62% (165 days) and 38.01 (% (180 days) respectively (Fig 2 & 3). The decrease of protein level was found to be significantly, proportional ($P<0.05$). Protein decreased with storage of cured meat and this was attributed to some changes during storage that caused by 'maillard reaction and changes in pH [39]. Salt causes the proteins in fish muscle to swell and salt lead the protein become denatured if increases in the muscle [40].

3.3. Fat (%): Fat content was found to be influenced by season and geographic location [41]. In SDS and SDS+T shoal fish products, fat content was found 5.10%, and 4.79%, in fresh-process condition and after completing the duration of storage period it was 3.81% (165 days) and 3.26 (% (180 days) respectively (Fig. 2 & 3). It is clear from the present results that fat content was decreased significantly ($p<0.05$). This might be due

to oxidative deterioration, thereby affecting lipid extraction [42].A decrease in the level of crude protein and fat of small and large salted Bouri fish muscle (Mugil cephalus)was found [43].

3.4. Ash (%): In SDS and SDS+T shoal fish products, ash content was found 22.80%, and 22.41 %, in fresh-process condition and after completing the duration of storage period it was found 21.44 % (165 days) and 21.07 (% (180 days) respectively (Fig. 2 & 3). The higher value of total ash content in freshly processed SDS and SDS+T shoal fish than fresh fish was attributed to high salt content. Similar levels of ash content in salted fish were noticed by several workers [44].

3.5. Changes in pH value: pH is an indicator of the extent of microbial spoilage in fish and some proteolytic microbes produce acid after decomposition of carbohydrate, thereby increasing the acid level of the medium [45]. The pH value is a reliable indicator of the degree of freshness or spoilage.

The pH in fresh condition fresh- water fish flesh is almost neutral [46]. In the post-mortem period, decomposition of nitrogenous compounds leads to an increase in pH in the fish flesh [47]. The increase in pH indicates the loss of quality. The pH value of SDS and SDS+T shoal and fish-product was increased significantly ($P<0.05$) with storage period. pH value of fresh Shoal fish was 6.9 in our study . But when salt is added with the fish, pH value decrease due to increase of acidic compound and after that among shelf life study pH value increases in the time interval due to increase of basic compounds. In the present study pH value were found to vary from 6.2 (0 day) to 8.3 (165 days) for SDS and 6.3(0 day) to 8.2 (180 days) for SDS+T Shoal (Fig. 4).

The fish products are acceptable up to a pH of 6.8 but are considered to be spoiled above 7.0 of pH [48]. 6.8 to 7.0 is usually the limit of acceptability [49].While the initial pH values in the samples were similar to findings of other researchers, the increase in pH values during the storage of room temperature (27-30^{0}C) was higher than others. The probable reason of these differences is differences in fish species and different methods of salting.

Acknowledgement

The author acknowledges the scientists and technicians of Fish technology laboratory, IFST, BCSIR, Dhaka, Bangladesh for their technical supports and continuous assistance to carry out the present study.

IV. Conclusion

The present study reveals that different fish drying methods have a significant role on the proximate composition of Shoal fish. It was observed that the hygienically dried samples using salt and salt- turmeric comparatively good nutritional value and textural aspects. Salt and turmeric is also useful to reduce the time of drying of fish. The saving of time and improvement of the quality in the dry fish process will help the poor Fisher folks getting better price for their products but also enhance consumer preference in the local market. In case, poor coastal people are not able to afford to have solar dryers, they can take this process of fish drying.

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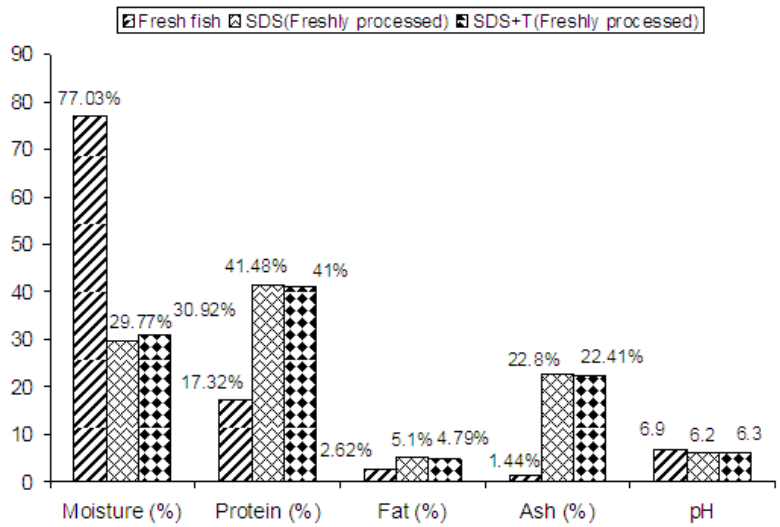

Figure.1. Initial proximate composition and pH value of the fresh experimental fish, freshly processed Sun-dried salted (SDS) and Turmeric treated Sun-dried salted (SDS+T) Shoal (Channa striatus).

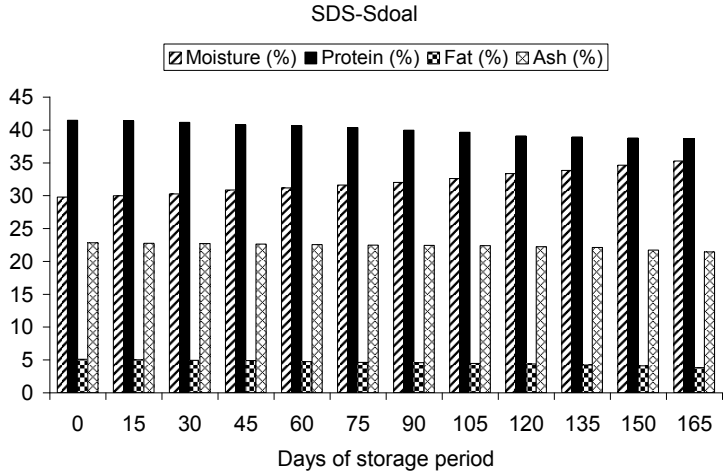

Figure. 2. Changes in Proximate Composition of Sun-dried-Salted (SDS) Shoal fish during Storage at Room Temperature.

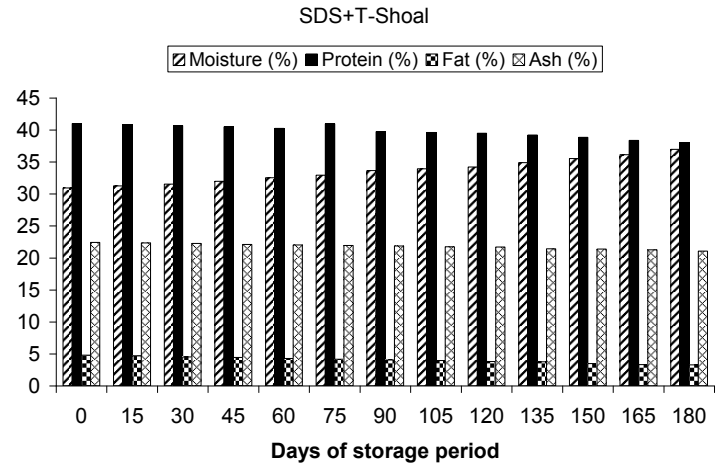

Figure. 3. Changes in Proximate Composition of Turmeric treated Sun-dried-Salted (SDS+T) Shoal fish during Storage at Room Temperature.

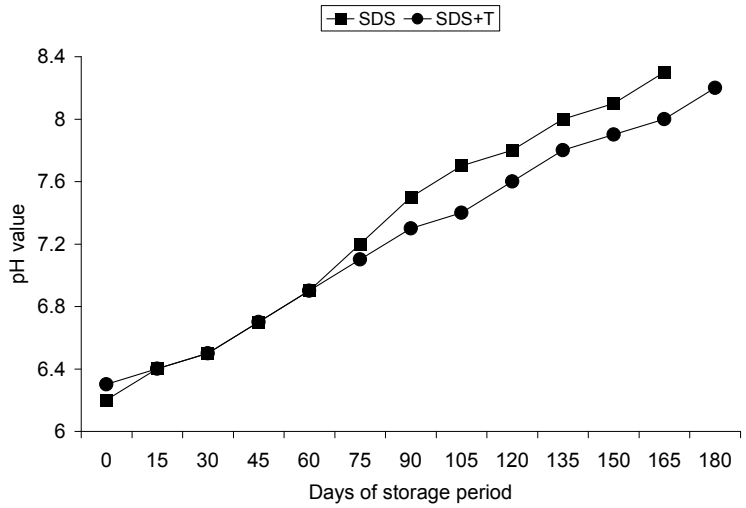

Figure. 4. Changes in pH value of Sun-dried-salted (SDS) and Turmeric treated Sun-dried-Salted (SDS+T) Shoal fish during Storage at Room Temperature.

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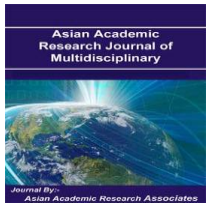

INVESTIGATION ON THE SHELF-LIFE QUALITY OF DRY-SALTED AND SUN-DRIED SALTED SNAKE-HEAD SHOAL FISH (CHANNA STRIATUS BLOCH, 1801) STORED AT REFRIGERATOR TEMPERATURE (4^0C)

FARZANA BINTE FARID[1]; DR. GULSHAN ARA LATIFA[2]; MOSARRAT NABILA NAHID[3]; MOHAJIRA BEGUM[4]

[1]Department of Zoology, University of Dhaka, Dhaka-1000, Bangladesh
[2]Department of Zoology, University of Dhaka, Dhaka-1000, Bangladesh
[3]Department of Zoology, University of Dhaka, Dhaka-1000, Bangladesh
[4]Institute of Food Science and Technology, Bangladesh Council of Scientific and industrial Research (BCSIR), Dhaka-1205, Bangladesh,

Abstract

This study was designed to evaluate and compare the shelf-life quality of Dry salted (DS) and Sun-dried salted (SDS) Shoal fish-product by analyzing physical changes such as color, texture, odor and visual appearance by acceptability technique and changes in chemical index of TVB-N, pH and FFA value at refrigerator temperature (4^0C). In fresh-process condition, values of TVB-N, pH and FFA were 5.25 mgN/100g, 6.0, 1.5% in case of DS shoal and 4.89 mgN/100g, 5.9 and 2.3% in case of SDS shoal respectively. TVB-N, pH and FFA value increased significantly ($p<0.05$) with the time of storage period and between these two salted products, these parameters rapidly increased in sun-dried salted fish than DS shoal fish and at the end of 24 months SDS Shoal fish product became spoiled whereas DS shoal fish still in fresh condition. Experimentally it has been proved that the DS shoal fish has longer shelf life (26 months) and has found better way for preservation in refrigerator temperature (4^0C).

Key Words: DS, SDS, Shoal, sensory-score, quality analysis

1. Introduction

About 8 million tons of fish (25-30%) of the world catch are being used for human consumption as dried, salted, smoked or treated by some combination of these processes **(**Kamruzzaman, 1992). Curing is a simple and cheapest method of processing requiring least technical expertise, but it has great significance and relevance in the socio-economic system of small-scale fisherfolk. About 14% of the marine landings are reported to be processed by curing (Sanjeev and Surendran, 1996).

Fish processing has been practiced in Bangladesh for a long time. One of the shelf life promoting strategy involves salting with sodium chloride (Ravishankar and Juneja, 2000). The presence of sufficient salt (sodium chloride) in fish prevents or drastically reduces the action of bacteria. It removes moisture by the process of osmosis, creating medium unsuitable for microbial growth. The rate of salt uptake and moisture loss is influenced by such factors as temperature, thickness of the flesh, fattiness of the flesh, freshness of the fish and the chemical purity of the salt used for curing. Another method of preserved fish uses this research is traditional open sun drying. Drying in the sun may accompany salting.

Salting and sun drying of fish is a traditional method of seafood preservation employed in many countries, however, some such methods may also destroy or remove some essential nutrients or decrease their digestibility.

Different processing methods have different effects on the nutritional compositions of fish. This is because heating, freezing and exposure to high concentration of salt lead to chemical and physical changes and therefore digestibility is increased, due to protein denaturation, but the content of thermolabile compounds and polyunsaturated fatty acids is often reduced (Chukwu and Shaba, 2009).

Therefore the quality of fish processed by the various methods cannot be the same and hence its subsequent effect on the fish's shelf life also varies.

Being a popular air-breathing fish the shoal fish (*Channa striatus*, Channidae) has been widely used as animal protein throughout the country. However, this fish species is being threatened day by day. In order to use it in different seasons and different parts of the country it is deemed important to preserve it for longer period.

Quality assessments are necessary to ensure food safety of any processed products. Therefore in present study, analysis of different parameters was carried out to assess the qualities of DS and SDS shoal fish stored in refrigerator temperature.

2. Material and Methods

2.1. Collection of the fishes and location of the experiment

Fresh experimental shoal fish (*Channa striatus*) had been collected from the river Meghna in the early hours of day and the fishes were brought to the Fish Technology Section, IFST, BCSIR, Dhaka for conducting the research activities, starts in the month of January, 2011. The whole experimental period covered 26 months of duration started from January, 2011 to March, 2013.

2.2. Preparation of fish

Fishes were carefully washed using cooled tap water. Head, scales, fins, gills and viscera were removed and again washed with tap water to remove blood, slime and unnecessary flesh. Due to the presence of hard shield like bony elements, bones of head are discarded as waste.

2.3. Fresh sample

A fresh sample of shoal fish species was taken to the laboratory for quality analysis of fresh experimental fish. About 6 or 7 slices of fishes were taken randomly that represented parts from whole body. The slices then were chopped with skin and bone and finally ground with an electric blender to make a homogenous sample before being sampled for analysis.

Therefore, the total cleaned fishes were grouped into 2 batches.

2.4. Method of salting

Being a safe, antimicrobial and incidental food additive (Klaassen, 1986), toxic for some microorganisms (Leroi *et al.* 2000), depressor of water activity (a^w) of the food (Turan *et al.* 2007), sodium chloride has been used as a seasoning and flavor enhancer as well as a preservative or curing agent.

2.4.1. Dry salting (DS) method

Fresh fishes were enrolled by dry commercial salt (NaCl) of about 30% by weight of the dressed fish (fish weight: salt weight 3 : 1), stacked in containers and stored for a salting or curing period, at room temperature. In this method, the extracted water of the fish due to salt action had been removed from the container. Thus the fishes are always allowed to remain in dry condition for the production of dry salt cured fish.

2.4.2. Sun-dried-salting (SDS) method

During this experiment the fresh fishes were enrolled by dry commercial salt (NaCl) of about 30% by weight of the dressed fish (fish weight: salt weight 3 : 1). They were kept on a plastic bade basket in the sun. They were kept in sun regularly during day time (12 a.m. to 3 p.m.)

for 2-7 days as sometimes the sky was cloudy and until the ripening period was over. At the same time, temperature and relative humidity were also recorded. During sun-drying, they were kept covered by dense meshed nylon or mosquito net to avoid outside contamination and prevent bird attack and fly infestation.

During dry-salting and sun-dried salting process, moisture content decreased and salt content increased considerably during the first 6 to 7 days which is called ripening period. The ripening of the product was determined by observing the changes in sensory characteristics such as color, texture, flavor etc. and changes in moisture content and salt penetration rate.

2.5. Storage of the product

At the end of the ripening period, dry- salted (DS) and sun-dried salted (SDS) shoal fish-products was packaging with plastic bag maintaining aseptic condition as far as possible and was stored at refrigerator temperature (4^0C). The preservation period of product is linked to the amount of salt added; therefore a straight proportion is present between the amount of salt used and the preservation period (Bahri *et al*. 2006).

2.6. Sampling procedures

Investigation of the shelf-life quality of dry salted and sun-dried salted fishes were carried out 2 month interval for refrigeration temperature (4^0C), until the fish become spoil or inedible condition. The experiment was done for second time at regular intervals during salting period. Salt crystal was removed from the dry-salted and sun-dried salted fish-product using dry tissue paper before being sampled for analysis.

To determine the quality of salted fishes during storage period some parameters, *viz.* freshness test by sensory scores, TVB-N value, pH, FFA, etc. were analyzed.

2.7. Estimation of sensory score

Determination of the sensory score of dry-salted (DS) and sun-dried salted (SDS) shoal fish was made by trained panel of six judges in BCSIR, IFST Lab. following 9-point hedonic scales. Comparison was carried out in terms of sensory characteristics, such as color, flavor, texture and general appearance. The panel was requested to rate each sensory feature of products. The average score of 5 was considered to be the borderline of acceptability according to Peryan and Pilgrim (1957), (9. Like extremely; 8. Like very much; 7. Like moderately; 6. Like slightly ;< 5. Bad).

2.8. Estimation of TVB-N (Total Volatile Base Nitrogen)

TVB-N was determined by Conway modified micro-diffusion technique (Conway and Byrene, 1993). TVB-N has been used as an index for the determination of freshness of fish (Stansby *et al.* 1944, Huss, 1988). Volatile nitrogenous bases increase in concentration during the spoilage of fish (Vynke and Marlevede, 1963). The TVB-N measurement can be used as a parameter for the determination of microbiological and enzymatic spoilage of fish product. The TVB-N value that helps for the determination of level of fish spoilage, has an inverse relationship with the sensory score of salted and sun dried fishes. When the sensory score decreased then the TVB-N value increases and vice versa.

TVB-N value was determined by using Conway modified micro-diffusion technique (Conway and Byrne, 1963). Samples that were in the different levels of acceptability from highly acceptable condition to unacceptable condition had been selected for TVB-N analysis. 25 ml of 10% Trichloro Acetic Acid (TCA) was added to 2 gm of ground fish sample and kept overnight and then filtrated with known volume. 2% boric acid, TCA, K_2CO_3 and the solutions made from the fish samples were taken into the Conway dishes in the following way.

Table: Showing the amount of chemicals taken in the inner and outer chamber of the Conway dishes.

Conway dishes	Inner Chamber		Outer chamber	
	Chemical	Amount	Chemical	Amount
Blank	2% Boric acid	1 ml	K_2CO_3	1 ml
With Sample	2% Boric acid	1 ml	Sample Solution	1 ml
			K_2CO_3	1 ml

After the addition of Potassium Carbonate (K_2CO_3), each dish was covered by a piece of glass that was stacked with glue (Paraffin soft white) initially. Then it was kept for 24 hours. The samples and Potassium Carbonate (K_2CO_3) reacts to form NH_3 which was absorbed by the boric acid and then the solution of each Conway dish had been titrated by N/70 H_2SO_4 with the help of a micro-burette.

Finally TVB-N was calculated.

Calculation

$$\text{TVB-N} = (\text{titration reading} - \text{blank reading}) \times \text{Strength of acid} \times 0.2 \times \frac{\text{Volume of extract}}{\text{Volume of extract taken}} \times \frac{100}{\text{Weight of sample taken}}$$

2.9. pH value measurement

A 1 g sample of the fish flesh was homogenized in 10 ml of distilled water and the mixture was filtered. The pH of the filtrate was measured according to the method of Vyncke (1981) using a pH meter (Mettler Toledo 320-s, Shanghi, China).

2.10. Free Fatty Acid (FFA) estimation

Oil sample used throughout the work was prepared by extracting the salted fish by Folch reagent (chloroform and methanol in the ratio of 2 : 1 v/v).The salted fish was first cut into small pieces and then ground. The ground material was then mixed with Folch reagent in a large wide mouthed stopper glass bottle for 24 hours at room temperature after 15 minutes stirring with glass rod.

Extraction was facilitated by occasional shaking. The mixture was filtered through a Buchner funnel and the filtrate was evaporated in batches under heater or oven at 60^0 C. Seven gram of well-mixed oil was taken into 250 ml flask and 50 ml ethanol was added, previously neutralized by adding 2 ml phenolphthalein solution. Titration was done using 0.25N sodium hydroxide. Vigorous shaking was done until permanent final faint pink colour appeared and persisted for at least one minute. The value was reported as percentage (%) of free fatty acid expressed as oleic acid. Millilitre of 0.25 N NaOH required for titration corresponds to the percentage of free fatty acid.

Data were analysed using SPSS for windows-20 statistical programme. Significance was established at $p< 0.05$ (Sokal and Rohlf 1987).

3. Results

TVB-N, pH value, FFA (Quality parameters) of fresh Shoal fish was 4.41 mgN/100g, 6.9 and 0.6% respectively (Figure 1).

The initial score of the sensory evaluation of dry-salted (DS) and sun-dried salted (SDS) shoal was 9. But during storage period this score rapidly decreased and at the end of the storage period, the score was 5 in case of DS (26 month) and SDS (24 month) Shoal (Figure 2).

Changes in TVB-N of DS and SDS shoal fish samples during the entire storage period are shown in Figure 3. TVB-N values were found to vary from 5.25 (0 day) to 19.06 mgN/100g (26 month) for DS and 4.89 (0 day) to 19.15 mgN/100g (24 month) for SDS. TVB-N values of the products storage at refrigeration temperature showed linearly increasing pattern

throughout storage period but neither of the value exceeded the recommended value set for fish regarded as acceptable condition. Significant statistical differences were found between the initial product and end product ($P < 0.05$) after storage period.

pH value of fresh Shoal fish was 6.9, but when salt is added with fish, pH value decreased and after that among shelf life study pH value increased during interval. In the present experiment, pH value was found to vary from 6.3 to 7.1 for DS and 6.2 to 7.1 for SDS (Figure 4).

FFA value is a measure of the extent of oxidative deterioration in oily fish, but it can fall further at latter stages of fish spoilage (FAO/SIFAR, 2001). The initial FFA values were 1.5% (oleic acid percentage) and 2.3% for DS and SDS samples. FFA values increased with storage time (Figure 5); At the end of the storage period values of FFA were found to be 10.1% (26 month)) for DS and 11.0% (24 month) for SDS respectively. Significant statistical differences were found between the initial product and end product ($P < 0.05$) during storage period.

4. Discussion

According to the panel's evaluation, the sensory properties of Dry-salted (DS) and Sun-dried salted (SDS) shoal fish-products were in acceptable condition throughout storage period though statistically there was significant difference ($p<0.05$) in the sensory evaluation during storage period based on the panel's score (Figure 2). Yu (1985) applied this hedonic rating scale to evaluate the acceptability of sun dried fishes by their external morphological and quality changes. Morshed (2005) applied the hedonic rating scale by using 9-points for the sensory evaluation of the dried and dehydrated fish.

Total volatile base nitrogen (TVB-N) is important compound provide a measure of the progress of spoilage that is dependent of sensory assessment. Gram (2002) opined that, the level of TVB-N in fish and fish products are mostly used as spoilage indicator through bacterial activity, the same result have been evident in the present study. The TVB-N of fish is an indicator of the freshness of the raw material (Zhou *et al.* 2011). According to Connell (1995) the limiting level for rejection of TVB-N is 20 mgN/100g for storage at refrigerator temperature. In present experiment, TVB-N values of all samples were lower than 25 mgN/100 g which was considered as the threshold for a good-quality fish product (Figure 3). High TVB-N values are unacceptable and are associated with unpleasant smell in the meat (Limbo *et al*. 2009). Assumably, this is because of the impact of the various treatments of

TVB-N, which primarily includes nitrogen from ammonia, TMA, and dimethylamine which reflects the extent of degradation of proteins and non protein nitrogenous compounds which can be explained by proteolysis, due to enzymatic and microbial activities in the samples on storage (Erkan and Ozden, 2008). In early storage, spoilage rate become slower than later storage time, it would appear from the Figure. This is also supported by Reed *et al*. (1929) and Muslemuddin (1970). The stage perhaps autolysis is mainly to the tissue explained (Tomiyasu and Zenateni, 1957) as a result TVB-N does not increase highly.

The pH in fresh condition fresh-water fish flesh is almost neutral (Virta, 2009). In the post-mortem period, decomposition of nitrogenous compounds leads to an increase in pH in the fish flesh (Shenderyuk and Bykowski, 1989). The increase in pH indicates the loss of quality. The pH value of DS and SDS shoal fish-product was increased significantly ($P<0.05$) with storage period (Figure 4). The limit of acceptability is usually 6.8 to 7.0 (Erkan *et al.* 2011).

The result of free fatty acids (FFA) (Figure 5) indicated that the salting and drying conditions accelerate lipid oxidation and this is in agreement with the results as shown by Smith 1988; Smith *et al*. 1988. Chakraborty *et al.* 1997 who found that FFA value ranged from 2.9% of total lipid on the start day to 7.58, 7.15 and 8.56% on the 60 days of observation in dry salting, wet salting and sun-dry salted fishes respectively. The present study on the contents of free fatty acid values were similar to that of the studies mentioned earlier.

A high level of FFA is characteristics of product that have undergone both microbial and biochemical spoilage (Horner 1997; Tungkawachara, 2003). This may indicate that greater proportions of unsaturated fatty acids were liberated and were subjected to oxidative splitting at the double bonds. The resulting substances, mostly ketones and aldehydes, appear to be largely responsible for flavor, odor and taste of the salted fish product (El-Sebahy, 1988). The same result was found in the present study.

5. Conclusion

However, Dry salted and Sun-dried Salted Snakehead shoal fishes are not popularly consumable in Bangladesh, but from this study it has been assessed that the salted samples have longer shelf life than Sun-dried salted samples storage at refrigerator temperature. Thus, these fish, if commercially preserved, can easily consume in other periods when this fish is not available.

6. Acknowledgement

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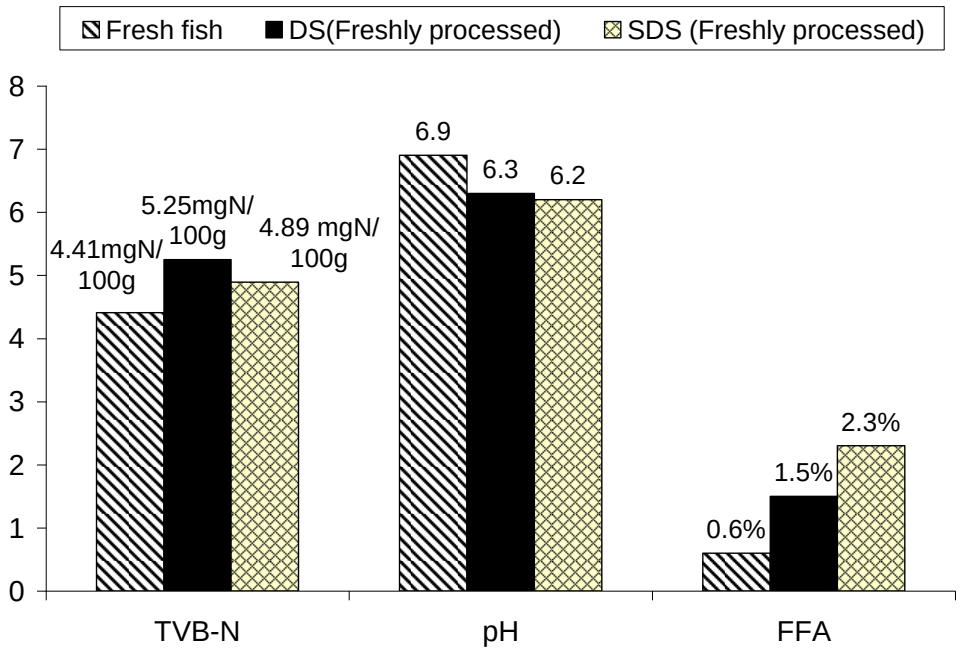

Figure 1. Chemical composition of fresh experimental fish, freshly processed dry salted (DS) and Sun-dried salted (SDS) shoal fish.

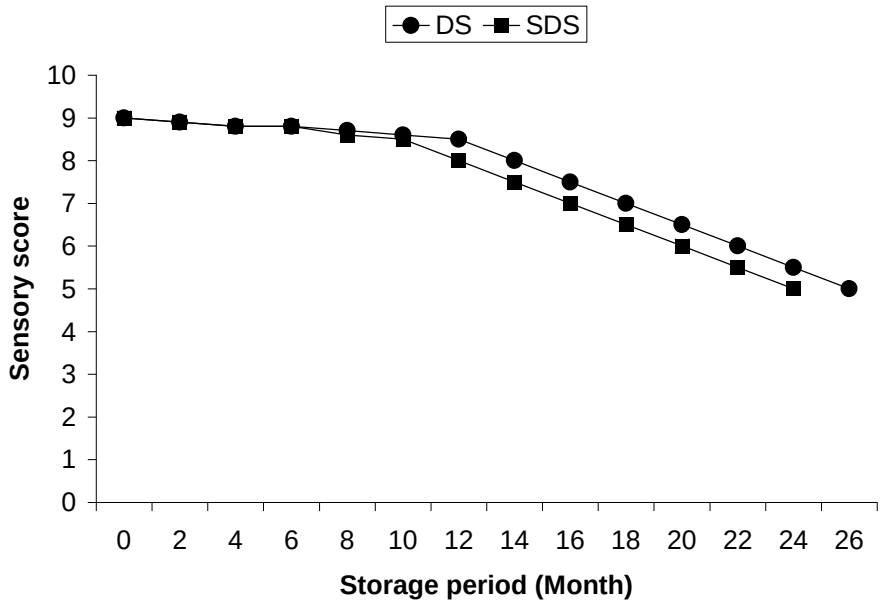

Figure 2. Changes in sensory score of dry-salted (DS) and sun-dried salted (SDS) Shoal fish during different duration of storage at Refrigerator Temperature (4^0C).

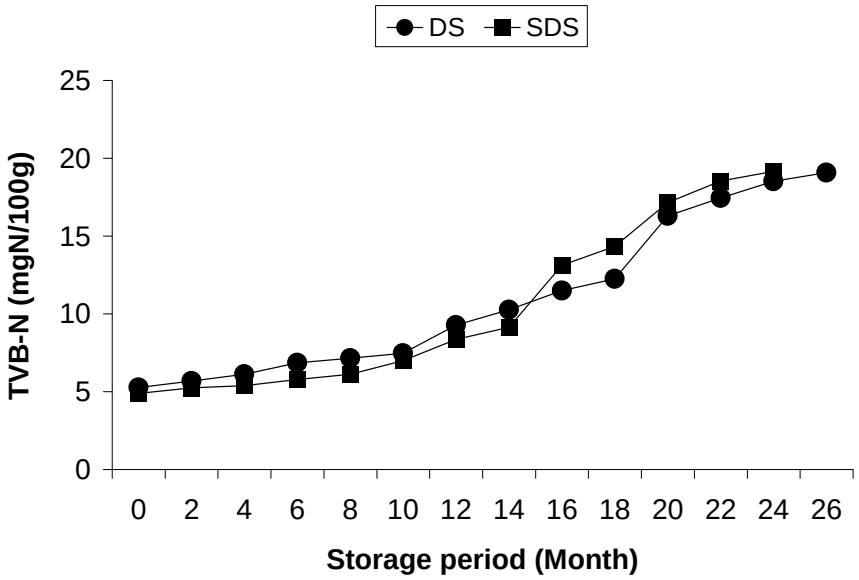

Figure 3. Changes in TVB-N (mgN/100g) contents of dry-salted (DS) and sun-dried salted (SDS) Shoal fish during different duration of storage at Refrigerator Temperature (4^0C).

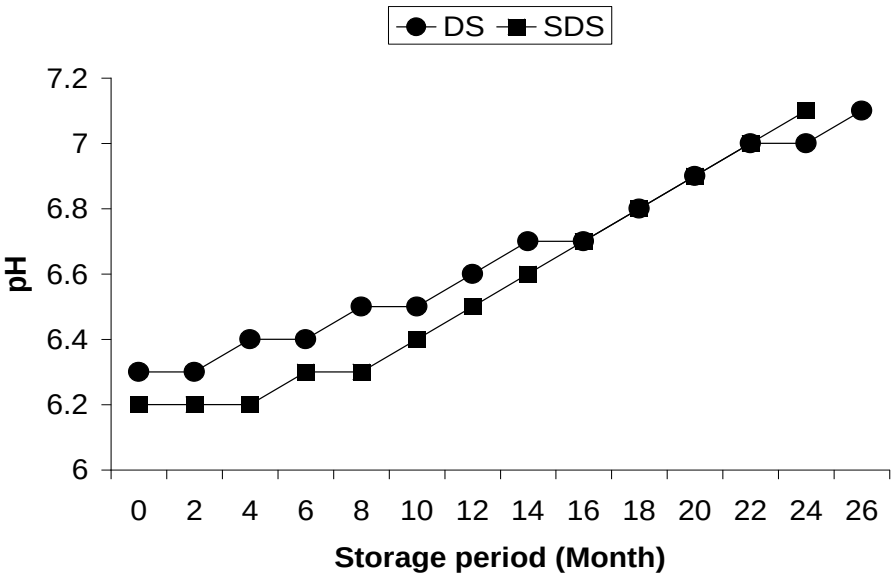

Figure 4. Changes in pH value of dry-salted (DS) and sun-dried salted (SDS) Shoal fish during different duration of storage at Refrigerator Temperature (4^0C).

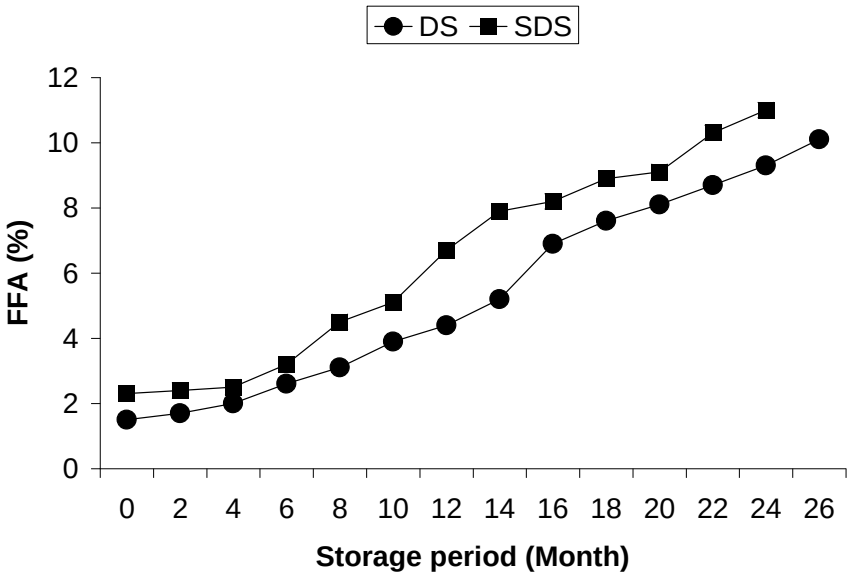

Figure 5. Changes in FFA (Free Fatty Acid) value of dry-salted (DS) and sun-dried salted (SDS) Shoal fish during different duration of storage at Refrigerator Temperature (4^0C).

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Comparative study on shelf life quality of Brine Salted Taki (Channa punctatus Bloch, 1793) and Shoal (Channa striatus Bloch, 1801) at Refrigerator Temperature (4^0C)

Farzana Binte Farid[1*], Dr. Gulshan Ara Latifa[1], Dr. Subhash Chandra Chakraborty[2], Mosarrat Nabila Nahid[1] and Mohajira Begum[3]
[1]*Department of Zoology, University of Dhaka, Dhaka 1000, Bangladesh*
[2]*Department of Fisheries Technology, Bangladesh Agricultural University, Mymensingh, Bangladesh*
[3]*Institute of food Science and Technology, BCSIR, Dhaka-1205, Bangladesh.*

Abstract: *A clear understanding on the difference between qualities and shelf lives of brine salted taki and shoal fish product stored at refrigerator temperature (4^0C) has been assessed by analyzing physical changes such as color, texture, odor and visual appearance by acceptability technique and changes in chemical index of TVB-N, pH and FFA value (quality parameters). In processed condition (after 7 days of ripening period in brine salting), values of TVB-N, pH and FFA were 5.7 mg N/100g, 6.8 and 2.6% in case of taki and 3.64 mg N/100g, 6.5 and 1.8% in case of shoal fish respectively. TVB-N, pH and FFA value increased significantly ($p<0.05$) with time of storage period and between two brine salted products, TVB-N, pH and FFA value rapidly increased in taki fish than shoal fish and at the end of 9 months brine salted taki fish product became spoiled whereas brine salted shoal fish still remained fresh. Experimentally it has been proved that brine salted shoal fish has longer shelf life (18 months) and is better to preserve in Refrigerator temperature (4^0C).*
Key Words: *Brine-Salting, Quality-analysis, Shoal, Shelf-life, Taki*

I. Introduction

Fish represents an important source of food for mankind throughout the world and is a very important source of animal protein [1, 2, 3, 4]. The important constituents of fish are water (70.0-85.0%), protein (15.0-20.0%), lipid (1.0-10.0%), ash (1.0-1.5%) and carbohydrate (0.3-1.0%) [4, 5, 6].

During the last decades in this century, healthy eating habits have received increased attention, and it is widely recognized that regular fish consumption is one possible health improving practice [7, 8].

Fish is also a very good source of vitamins and minerals [6, 9]. Most noticeably, the high nutrient containing biochemical composition classified fish as highly perishable food.

An estimated 50% of the fish produced in the remote coastal settlements and hinterland perish before reaching the consumers, as a result of poor handling, preservation and processing and for this; the fish loses its organoleptic characteristics and becomes progressively more unacceptable for human consumption [10].

Most kinds of fishes are readily decomposed by microorganisms unless special methods are used for their preservation. Spoilage of fish not only occurs at room temperature but also at the refrigeration temperature (4-7^0C). The goals of fish preservation methods are to increase the shelf life of the food and ensure the safety for human consumption. There is various fish preservation methods include-Drying, salting, smoking, freezing, use of low temperature, high temperature, etc. Among them, salting of fish is probably one of the oldest methods of fish preservation. Brine-salting is one kind of salt curing method. It is well known that brine-salting is a process which aims to reach the saline equilibrium between fish muscle and the surrounding salt solution in a specific time [11]. Salting reduces water activity (a_w) and inhibits the growth of many spoilage microorganisms and pathogen. In Bangladesh, India, Burma, Thailand and Sri Lanka salting of fish is popular. Different processing methods have different effects on the nutritional compositions of fish. This is because heating, freezing and exposure to high concentration of salt lead to chemical and physical changes and therefore digestibility is increased, due to protein denaturation, but the content of thermolabile compounds and polyunsaturated fatty acids is often reduced [12].

Therefore the quality of fish processed by the various methods cannot be the same and hence its subsequent effect on the fish's shelf life also varies.

Among the freshwater fish species Taki (C. punctatus) and Shoal (C. striatus) are very popular air-breating fish and also important due to commercial purpose. These fishes bear high market price and are delicious, nutritious and popular to the consumers. So it is necessary to take some steps for their proper preservation and marketing and during this period maintain proper quality.

[*] To whom communication should be made, e-mail: farzanafarid79@gmail.com

The people of Bangladesh are habituated and preferred to take fish in fresh condition. But it is difficult to reach the fish in fresh condition to the consumers all over the season. More over the peak of snake-headed taki and shoal fish catch in Bangladesh is seasonal. During catching season, the catch is much higher than the consumers need. So these need to store and transport carefully and safely to keep in good, acceptable condition. Due to high population growth there is an ever ending gap between supply and demand of fish and fisheries products in Bangladesh. Narrowing the gap not only requires increasing production but also improvement in all aspects of marketing and distribution system is also important [13]. For this the harvested fish and fisheries products should be processed properly to reduce the gap between supply and demand.

Quality assessments are necessary to ensure food safety of any processed products. Therefore in present study, analysis of different parameters was carried out to assess the qualities of brine-salted Taki and Shoal fish stored at refrigerator temperature.

II. Materials and Methods

2.1. Collection of the fishes and location of the experiment:

The fresh fish (Taki and Shoal) had been collected from the river Meghan in the early hours of the day and the fishes were brought to the Fish Technology Section, IFST, BCSIR, Dhaka for conducting the research activities, starts in the month of January, 2012.The whole experimental period covered 18 months of duration started from January, 2012 to July, 2013.

2.2. Preparation of fish:

The fishes were carefully washed with cooled tap water. Head, scales, fins, gills and viscera were removed and again washed with tap water to remove blood, slime and unnecessary flesh. Due to the presence of hard shield like bony elements, bones of head are discarded as waste.

2.3. Fresh Sample:

A few fresh fish-samples of Taki and Shoal fish species was taken to the laboratory for quality analysis. 6 or 7 slice was taken randomly which represented the parts from whole body of the fish. Then the slices were chopped with skin and finally ground with an electric blender to make a homogenous sample before being sampled for analysis.

2.4. Brine salting (BS) method:

Being a safe, antimicrobial and incidental food additive, toxic for some microorganisms, depressor of water activity (a^w) of the food, sodium chloride has been used as a seasoning and flavor enhancer as well as a preservative or curing agent [14, 15, 16].

During this experiment A 30% salt solution is prepared (30 gm salts in 100 ml water) which is called brine. The fishes are kept at this saturated brine solution stacked in containers and stored for a salting or curing period, at room temperature (26^0C-30^0C) for the production of brine-salted fish. The fish in brine were kept immersed by putting a glass weight.

During brine-salting process moisture content decreased and salt content increased considerably during the first 6 to 7 days which is called ripening period.

2.5. Storage of the product:

At the end of the ripening period, brine-salted product of two fishes was packaging with plastic bag maintaining aseptic condition as far as possible and was stored at refrigeration temperature (4^0C). The preservation period of product is linked to the amount of salt added; therefore a straight proportion is present between the amount of salt used and the preservation period [17].

2.6. Sampling Regime and Procedures:

Evaluation of quality changes in brine salted Taki and Shoal fishes were carried out 3 month interval for refrigeration temperature (4^0C), until the fish become spoil or inedible condition. The experiment was done for second time at regular intervals during salting period.

There are some parameters which determine the quality of salted fish during storage condition, such as-freshness test by sensory scores, TVB-N value, pH, FFA etc. Freshness test of the fishes indicate the quality test in term of odor, color and appearance in different species. TVB-N value, pH, FFA of fishes is important indicator to determine the quality of fish.

2.7. Estimation of sensory score:

Determination of the quality of Brine salted taki and shoal fish was made by trained panel of six judges in BCSIR, IFST Lab. following 9-point hedonic scales. Comparison was carried out in terms of sensory

characteristics, such as color, flavor, texture and general appearance. The panel was requested to rate each sensory feature of the end products. The average score of 5 was considered to be the borderline of acceptability (9. Like extremely; 8. Like very much; 7. Like moderately; 6. Like slightly; < 5. Bad.) [18].

2.8. TVB-N (Total Volatile Base Nitrogen) Estimation:

TVB-N was determined by Conway modified micro-diffusion technique [19]. 25 ml of 10% Trichloro Acetic Acid (TCA) was added to 2 gm of ground fish sample and kept overnight and then filtrated with known volume. 2% boric acid, K_2CO_3 and the solutions made from the fish samples were taken into the Conway dishes. After that, each dish was covered by a piece of glass that was stacked with glue (Paraffin soft white) initially. Then it was kept for 24 hours. The samples and Potassium Carbonate (K_2CO_3) reacts to form NH_3 which was absorbed by the boric acid and then the solution of each Conway dish had been titrated by N/70 H_2SO_4 with the help of a micro-burette.

Table: Showing the amount of chemicals taken in the inner and outer chamber of the Conway dishes:

Conway dishes	Inner Chamber		Outer Chamber	
	Chemical	Amount	Chemical	Amount
Blank	2% Boric acid	1 ml	K_2CO_3	1 ml
With Sample	2% Boric acid	1 ml	Sample Solution	1 ml
			K_2CO_3	1 ml

Finally TVB-N was calculated.

Calculation

$$\text{TVB-N} = (\text{titration reading - blank reading}) \times \text{Strength of acid} \times 0.2 \times \frac{\text{Volume of extract}}{\text{Volume of extract taken}} \times \frac{100}{\text{Weight of sample taken}}$$

2.9. Estimation of pH:

A 1 g sample of the fish flesh was homogenized in 10 ml of distilled water and the mixture was filtered. The pH of the filtrate was measured using a pH meter (Mettler Toledo 320-s, Shanghi, China) [20].

2.10. FFA (Free fatty acid) estimation:

Oil sample used throughout the work was prepared by extracting the brine salted taki and shoal fish by Folch reagent (chloroform and methanol in the ratio of 2:1 v/v).The brine-salted fish was first cut into small pieces and then ground. The ground material was then mixed with folch reagent in a large wide mouthed stopper glass bottle for 24 hours at room temperature after 15 minutes stirring with glass rod. Extraction was facilitated by occasional shaking. The mixture was filtered through a Buchner funnel and the filtrate was evaporated in batches under heater or oven at 60^0 C.

Seven gram of well-mixed oil was taken into 250 ml flask and 50ml ethanol was added, previously neutralized by adding 2 ml phenolphthalein solution. Titration was done by 0.25 N sodium hydroxide (NaOH) with vigorous shaking until permanent final faint pink color appeared and persisted for at least one minute. The value was reported as percentage (%) of free fatty acid expressed as oleic acid. Milli liter of 0.25 N NaOH required for titration corresponds to the percentage of free fatty acid.

Data were analyzed by using SPSS for windows-20 statistical programme with five percent level of significance [21].

III. Results and Discussion

TVB-N, pH, FFA (Quality parameters) of fresh Taki fish was 3.43mgN/100g, 7.0 & 0.5% and Shoal fish was 4.41mgN/100g, 6.9 and 0.6% respectively (Fig. 1).

3.1. Sensory evaluation (Score):

According to the panel's evaluation, the sensory properties of Brine salted taki and shoal fish products were in acceptable condition throughout storage period though, statistically there was significant difference ($p<0.05$) in the sensory evaluation during storage period based on the panel's score (fig. 2). The initial score of the sensory evaluation of BS taki and shoal was 9. But during storage period this score rapidly decreased and at the end of the storage period, the score was 5 in case of BS taki (9 month) and BS shoal (18 month). This hedonic rating scale was applied to evaluate the acceptability of the sun dried fishes by their external morphological and quality changes [22]. This hedonic rating scale was also applied by using 9- points for the sensory evaluation of the dried and dehydrated fish [23].

3.2. Changes in TVB-N value during storage period:

TVB-N has been used as an index for the determination of freshness of fish [24, 25]. Volatile nitrogenous bases increase in concentration during the spoilage of fish [26].

Samples that were in the different levels of acceptability from highly acceptable condition to unacceptable condition had been selected for TVB-N analysis. Gram opined that, the level of TVB-N in fish & fish products are mostly used as spoilage indicator through bacterial activity, the same result have been evident in the present study [27].

TVB-N values were found to vary from 5.70 (o day) to 24.48 mg N/100g (9 month) for BS Taki and 3.64 (o day) to 21.14 mg N/100g (18 month) for BS Shoal. Significant statistical differences were found between the initial product and end product ($P < 0.05$) after storage period (fig. 3). TVB-N values of the products storage at refrigeration temperature showed linearly increasing pattern throughout storage period but neither of the value exceeded the recommended value set for fish regarded as acceptable condition. According to Connell the limiting level for rejection of TVB-N is 30-40mgN/100g for storage at ambient temperature and 20mgN/100g for storage at refrigerator temperature [28]. The present findings are in close association with him. The rate of spoilage increases with the time and is closely interrelated. The same result had been evident from several researchers [29, 30]. The stage perhaps due to autolysis in the tissue [31].

3.3. Changes in pH value:

pH is an indicator of the extent of microbial spoilage in fish and some proteolytic microbes produce acid after decomposition of carbohydrate, thereby increasing the acid level of the medium. The pH value is a reliable indicator of the degree of freshness or spoilage [32].

The pH in fresh condition freshwater fish flesh is almost neutral [33]. In the post-mortem period, decomposition of nitrogenous compounds leads to an increase in pH in the fish flesh [34]. The increase in pH indicates the loss of quality. The pH value of BS taki and shoal fish-product was increased significantly ($P<0.05$) with storage period. pH value of fresh Taki and Shoal fish were found 7.0 and 6.9 respectively in the present study. But when salt was added with fish, pH value decreases rapidly while in the study of shelf life it was evident that the pH value increases with time. In the present study pH value were found to vary from 6.8 (0 day) to 7.8 (9 month) for BS Taki and 6.5 (0 day) to 7.2 (18 month) for BS Shoal (Fig. 4). The fish products are acceptable up to a pH of 6.8 but are considered to be spoiled above a pH of 7.0 [25]. This was similar to the present findings. The limit of acceptability is usually 6.8 to 7.0 [35].

3.4. Changes in FFA value:

Among the various parameters to assess the extent of deterioration in fish, determination of free fatty acid (FFA) content has been widely used. FFA a tertiary product of rancidity, increased during storage. The FFA is a measure of hydrolytic rancidity-the extent of lipid hydrolysis by lipase action. In most fish oils, rancidity is noticeable when the FFA (calculated as oleic acid) is in between 0.5%-1.5% [32]. It produced as a result of fat oxidation (rancidity). FFA value is a measure of the extent of oxidative deterioration in oily fish, but it can fall further at latter stages of fish spoilage [36]. A high level of FFA is characteristics of product that have undergone both microbial and biochemical spoilage [37].This may indicate that greater proportions of unsaturated fatty acids were liberated and were subjected to oxidative splitting at the double bonds. The resulting substances, mostly ketones and aldehydes, appear to be largely responsible for flavor, odor and taste of the salted fish product [38]. The same result was found in the present study.

The FFA value of salted shoal increased gradually with the passing of storage period (Fig. 5). Significant statistical differences were found between the initial product and end product ($P < 0.05$) during storage period and it was vary from 2.6% (o day) to 12.8% (9 month) for BS Taki and 1.8 % (o day) to 12.2% (18 month) for BS Shoal respectively. After 6 and 24 months of storage (at-13^0C to -25^0C) the appearance of considerable contents of FFA with certain bad odors and off flavors presumably derived from the degradation of fats, greatly reduced the consumer appeal of trout [39]. Free fatty acid (FFA) value ranged from 2.9% of total lipid on the start day to 7.58, 7.15 and 8.56% on the 60 days of observation in dry salting, wet salting and sundry salted fishes respectively [40]. The initial value of raw pre-spawning and post-spawning hilsa was 1.15% and 1.21% respectively which after dry salting increased to 7.86% and 8.35% respectively on 18^{th} days of storage at 28^0C to 32^0C [41]. FFA contents increased from 4.55-5.12% within 4 days of drying and then gradually increased up to 10 days of drying (6.86%) [42].

The present study denoted that the contents of free fatty acid values are similar with the above mentioned studies. The FFA value increased in a characteristics pattern to a certain level of storage period.

IV. Conclusion

From the present study, it was evident that, ~~the quality analysis of~~ the Brine salted Shoal has longer shelf life than Brine salted taki storage at refrigerator temperature (4^0C). The wide range of storage period indicates the diversity in the final quality and can be largely attributed to the effect of various conditions upon the salting agents and activities. It is seen that the main factor affecting the quality is time of storage and storage condition.

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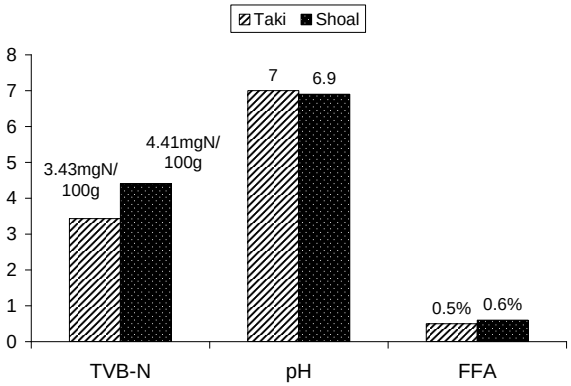

Figure 1. Initial quality of the fresh experimental fish, Taki (Channa punctatus) and Shoal (Channa striatus).

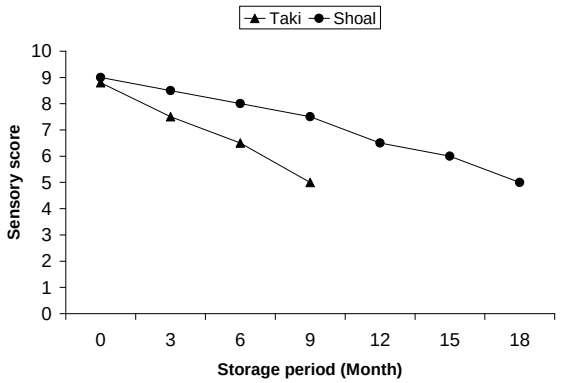

Figure 2. Changes in sensory score of brine salted Taki and Shoal fish during different duration of storage at Refrigerator Temperature.

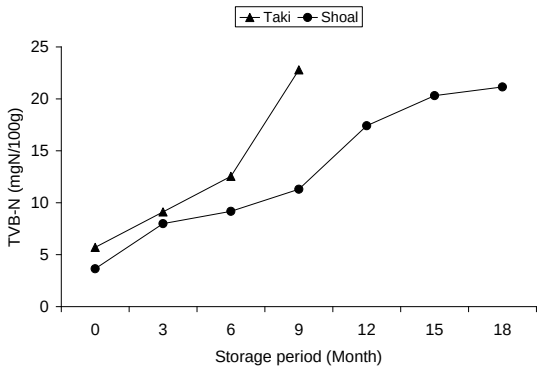

Figure 3. Changes in TVB-N (mgN/100g) contents of brine salted Taki and Shoal fish during different duration of storage at Refrigerator Temperature.

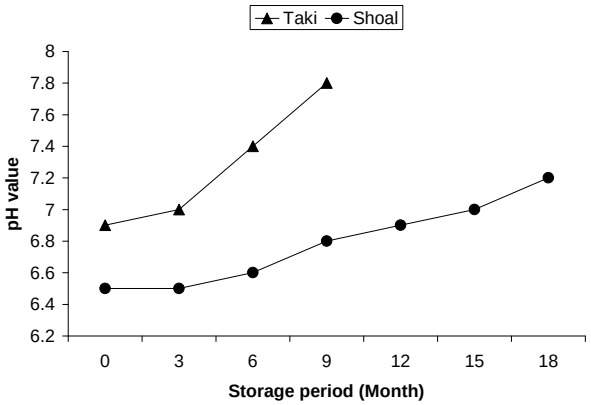

Figure 4. Changes in pH value of brine salted Taki and Shoal fish during different duration of storage at Refrigerator Temperature.

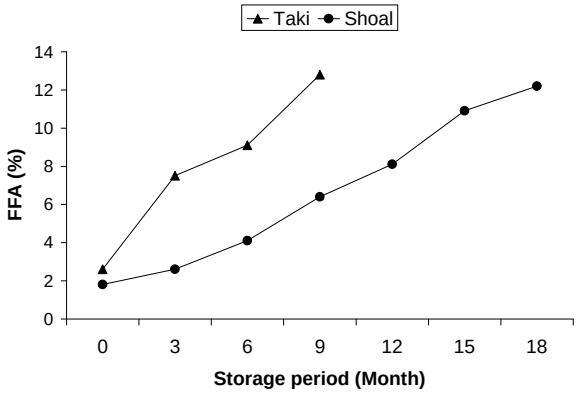

Figure 5. Changes in FFA (%) value of brine salted Taki and Shoal fish during different duration of storage at Refrigerator Temperature.

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Nutritional Quality Analysis of Bangladeshi Fish Species, *M. Tengra* (Hamilton-Buchanan, 1822) Preserved with Different Salt Curing Methods in Laboratory Condition

Gulshan Ara Latifa[1], Subhash Chandra Chakraborty[2], Mohajira Begum[3], Mosarrat Nabila Nahid[4], Farzana Binte Farid[5,*]

[1]Department of Zoology, University of Dhaka, Dhaka-1000, Bangladesh
[2]Department of Fisheries Technology, Bangladesh Agricultural University, Mymensingh, Bangladesh
[3]Institute of Food Science and Technology (IFST), Bangladesh Council of Scientific and Industrial Research (BCSIR), Dhaka-1205, Bangladesh
[4]Department of Zoology, University of Dhaka, Dhaka-1000, Bangladesh
[5]Organization Name: Department of Zoology, University of Dhaka, Dhaka-1000, Bangladesh
*Corresponding author: farzanafarid79@gmail.com

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Abstract Salt-cured fishes are highly appreciated because of their characteristic taste, texture and storage stability. This piece of work was done in an attempt to evaluate the issue of the traditional fish salting by using NaCl which are easily available and cheaper cost wise and evaluate the difference between biochemical composition (moisture, protein, fat, ash, TVB-N, pH and FFA) of Dry-salted (DS) and pickle-salted (PS) *M. tengra* fish-products in laboratory condition using standard methods of analyses. In processed condition (after salting) the values of moisture (%), protein (%), fat (%), ash (%), TVB-N, pH and FFA were 41.41%, 22.05%,10.65%, 26.15%, 3.90 mg N/100 gm, 6.0 and 2.8% respectively in case of DS *M. tengra* fish and 45.88%, 20.43%, 9.40%, 24.62%, 4.92 mg N/100 g, 6.0 and 3.2% respectively in case of PS *M. tengra* fish-product. During storage period, moisture (%), TVB-N, pH and FFA value were increased significantly ($p<0.05$) whereas total protein, lipid and ash contents were significantly ($p<0.05$) decreased. The values of moisture (%) content were increased 44.82 (7 month) and 49.09 (6 month), in DS and PS *M. tengra* respectively. The values of protein (%), fat (%) and ash (%) content were decreased 20.99%, 9.59% and 25.00% respectively in case of DS (7 month) and 19.28%, 8.68% and 23.41% (6 month) respectively in case of PS *M. tengra* fish. There were no significant ($p<0.05$) different among the samples and between this two salted products, TVB-N, pH and FFA value rapidly increased in PS than DS *M. tengra* fish-products and at the end of 6 month, pickle salted (PS) *M. tengra* fish-product became spoiled whereas dry salted tengra fish-product still remained fresh. Experimentally it has been proved that the fishes preserved in Dry-salt (DS) has longer shelf life (7 month) and has found better way for preservation.

Keywords: *Dry-salting, pickle-salting, M. tengra, proximate-composition, chemical analysis*

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1. Introduction

There is a popular saying that "Fish and Rice makes a Bangali". This popular saying reminds us the importance of fish in our life. It is a good source of protein and lipid for the development of our body [1]. Fish also rich in vitamin and minerals for both young and old age consumers [2,3,4]. Fisheries items are the major protein source of Bangladesh which contributing 58% of the nation's animal protein demands [5]. Fish oil is not health hazardous rather it contain *Omega 3* which helps to reduce the cholesterol level, thereby, reducing the risk of cardiac diseases. The current fish consumption rate is 17.52 kg/people/year whereas the demand is 20.44 kg/people/year and is 29.74 MT per year [6,7]. Being a riverine country, Bangladesh is rich in both close and open water bodies with high potential of producing freshwater and marine fish. Each year several thousand metric tons of fishes are captures from these water bodies. Besides there are culture fisheries resources too. During post harvest period large amount of fish are spoiled and wasted due to lack of proper measure for processing and preservation because of the fact that neither we can consume all the fishes caught nor can we transport to other places wherever necessary due to our insufficient handling and transportation system. In other words, proper handling, processing and preservation during post harvest period are a prerequisite for minimizing the spoilage loss [8]. The

quality of fish in generally decreases after death due to chemical reactions (changes in protein and lipid fractions, the formation of biogenic amines and hypoxanthine) and microbiological spoilage. Bangladesh is climatically a paradise for breeding and spreading of flora and fauna. Therefore, spoilage agents like microorganism, insects and other pests multiply tremendously and causes considerable damage to our fish resources during handling, transportation and storage. Preservation of a small quantity of loss will greatly improve the protein intake of our people. So to avoid fish spoilage and solve the fish deficit problem as well as uniform supply of fish in the market and even to the remote areas of the country throughout the season, development and utilization of proper scientific preservation techniques are very much essential for processors to produce a good quality of fish and fishery products. But it is not easy task to preserve fish scientifically as well as to maintain its nutritional value and flavor like the fresh one. The principal aim of preservation is to avoid spoilage of fish, avoid loss of protein component and lengthen shelf life. There are various types of preservation methods, such as icing, freezing, drying, salting, etc. in our country. Among them, salting process is considered as one of the oldest method of fish preservation. Salting is used to reduce water activity (aw) to obstruct or destroy the growth of microorganisms as well as inactive autolytic enzymes, where in this end the fish meat gets its way to durability [9,10]. Salted fish products are popular in many countries around the globe [11,12]. As these have been proven to be safe for millenniums, even in developed countries [13]. The aim of salting is not only to prolong the shell life of fresh fish but also to provide desirable sensorial changes [14,15]. In a developing country like Bangladesh, where freezing facilities are inadequate and there is an ever increasing energy crisis, energy intensive fish preservation procedures are not affordable. Furthermore, freezing does not eliminate pathogens which thus pose a health hazard.

Mystus tengra is one of the sole species of family Bagridae. This species very widely distributed in rivers, canals, khals, beels, ditches, inundated fields and other freshwater areas and is one of the most common catfish of the commercial catches of Bangladesh.

Proximate and biochemical analysis provides information on the nutritional value of a particular organism used as a source of food [16]. A number of studies on proximate composition of dried fishes were found in the literature [17-25]. But salting of freshwater fish species has a little in number compared to the dry fish.

The aim of this research is to carry on a comparative study on performance and quality assessment of 2 different type of salt curing method (Dry salting and Pickle salting) of *M. tengra* fish so that we can come to the conclusion, which one will be suitable and give better shelf life to create public awareness about the better process of salting.

2. Material and Methods

2.1. Collection of the Fishes and Location of the Experiment

Fresh experimental *Mystus tengra* fish had been collected from the river Meghna in the early hours of the day and the fishes were brought to the Fish Technology Section, IFST, BCSIR, Dhaka for conducting the research activities, starts in the month of June, 2013. The whole experimental period covered 7 months of duration started from June, 2013 to January, 2014.

2.2. Preparation of Fish

The fishes were carefully washed with cooled tap water. Fins, gills and viscera were removed and again washed with tap water to remove blood, slime and unnecessary flesh.

2.3. Fresh Sample

A few fresh tengra fish species was taken for quality analysis of fresh experimental fish and were chopped with head and bones and finally ground with an electric blender to make a homogenous sample before being sampled for analysis.

Therefore, the total cleaned fishes were grouped into 2 batches.

2.4. Method of Salting

Being a safe, antimicrobial and incidental food additive [26], toxic for some microorganisms [27], depressor of water activity (a^w) of the food [13], sodium chloride has been used as a seasoning and flavor enhancer as well as a preservative or curing agent.

2.4.1. Dry Salting (DS)

The raw fishes were enrolled by dry commercial salt (NaCl) of about 30% by weight of the dressed fish (fish weight: salt weight 3:1), stacked in containers and stored for a salting or curing period, at room temperature. In this method, the extracted water of the fish due to salt action had been removed from the container. Thus the fishes are always allowed to remain in dry condition for the production of dry salt cured fish.

2.4.2. Pickle Salting (PS)

The raw fishes were enrolled by dry commercial salt (NaCl) of about 30% by weight of the dressed fish (fish weight: salt weight 3:1), stacked in containers and stored for a salting or curing period, at room temperature. The salt reacts with the fish and water is extracted out from the fish-body and a salt-solution is formed. Thus in this method, the fishes are always, allowed to remain in such solution for the production of pickle-cured fish.

During dry-salting and pickle-salting process, moisture content decreased and salt content increased considerably during the first 6 to 7 days which is called ripening period. The ripening of the product was determined by observing the changes in sensory characteristics such as color, texture, flavor etc. and changes in moisture content and salt penetration rate.

2.5. Storage of the Product

At the end of ripening, 2 types of salted fish-product was packaging with plastic bag maintaining aseptic condition as far as possible and was stored at room

temperature (°C). The preservation period of product is linked to the amount of salt added; therefore a straight proportion is present between the Amount of salt used and the preservation period [28].

2.6. Sampling Procedures

Evaluation of the performance of 2 type's salted *M. tengra* fish-product was carried out 1 month interval in laboratory condition, until the fish become spoiling or inedible condition. The experiment was done for second time at regular intervals during salting period. Salt crystal was removed from the dry salted product using dry tissue paper before being sampled for analysis.

Analytical methods were applied for the determination of biochemical composition of the raw fish as well as of processed fish products on experimental basis. There are some bio-chemical parameters which determine the quality of salted fish during storage condition such as proximate composition (moisture, protein, fat, ash), Chemical composition (TVB-N value, pH, FFA) etc.

2.7. Estimation of Proximate Composition

Using conventional method of AOAC (Association of Official Analytical Chemicals), the proximate composition of fish was determined [29].

2.7.1. Estimation of Moisture

About 5 gram of previously prepared fairly minced samples were taken into each known weight basin and weighed in a digital balance (Toledo, Switzerland). The samples were allowed to dry into the oven (Memmet 854 Schwabach) at 105°C for 24 hours in order to remove the moisture until constant weight. After that, the basins are taken out of the oven, cooled in a desiccators and were weighed in a digital balance.

Calculation

$$\%\text{of Moisture} = \frac{\text{Weight Loss}}{\text{Original Weight of Sample Taken}} \times 100$$

2.7.2. Estimation of Protein

The protein content was estimated using conventional micro-kjeldahl method [30].

Calculation

$$\frac{\%\ \text{of } N_2 (*A) \times \text{strength of acid} \times 0.002 \times \frac{100}{5} \times 100}{\text{weight of sample taken}}$$

*A = titration reading - blank reading

For most routine purposes the percent of protein in the sample is then calculated by multiplying the % of N_2 with an empirical factor of 6.25 for fish.

$$\%\ \text{of protein} = \%\ \text{of total } N_2 \times 6.25$$

2.7.3. Estimation of Fat

About 5 g of the homogenous sample was taken into conical flasks and 10 ml of folch reagent (Chloroform: Methanol = 2:1) was added into the sample and homogenized properly and kept in air-tight condition for 24 hours. Fat contents of the fish muscle react with that solvent and remains in the solution. After 24 hours the solution of the flask was filtered in another weighed conical flask through a filter paper. Then these flasks were given in a hot water bath to dry up and removed the solvent. After that the flasks were kept into an oven for an hour to get the actual fat content. Then the flasks were weighed in an electronic balance to get the amount of fat content.

Calculation

$$\%\ \text{of Fat} = \frac{\text{Weight of the residue}}{\text{Weight of sample taken}} \times 100$$

2.7.4. Estimation of Ash

About 4-5 g fish sample was weighed into a pre-weighed crucible. The crucible with the contents was heated first over a long flame till all the material was completely churned. Then it was transferred in the Muffle Furnace held at dark red at a rate of 600°C for 5 hours until the residue become white. The crucible were cooled in desiccators and weighed. Finally the % of ash content was calculated.

Calculation

$$\%\ \text{of ash} = \frac{\text{Weight of fish}}{\text{Weight of sample taken}} \times 100$$

2.8. Estimation of Chemical Composition

To determine the quality of salted fishes during storage period some parameters, *viz.* TVB-N value, pH, FFA, etc. were analyzed.

2.8.1. Estimation of TVB-N (Total Volatile Base Nitrogen)

TVB-N has been used as an index for the determination of freshness of fish [31,32]. Volatile nitrogenous bases increase in concentration during the spoilage of fish [33]. The TVB-N measurement can be used as a parameter for the determination of microbiological and enzymatic spoilage of fish product. The TVB-N value that helps for the determination of level of fish spoilage has an inverse relationship with the sensory score of salted fishes. When the sensory score decreased then the TVB-N value increases and vice versa.

TVB-N value was determined by using Conway modified micro-diffusion technique [34]. Samples that were in the different levels of acceptability from highly acceptable condition to unacceptable condition had been selected for TVB-N analysis. 25 ml of 10% Trichloro Acetic Acid (TCA) was added to 2 gm of ground fish sample and kept overnight and then filtrated with known volume. 2% boric acid, TCA, K_2CO_3 and the solutions made from the fish samples were taken into the Conway dishes.

After the addition of Potassium Carbonate (K_2CO_3), each dish was covered by a piece of glass that was stacked with glue (Paraffin soft white) initially. Then it was kept for 24 hours. The samples and Potassium Carbonate (K_2CO_3) reacts to form NH_3 which was absorbed by the boric acid and then the solution of each Conway dish had been titrated by N/70 H_2SO_4 with the help of a micro-burette.

Finally TVB-N was calculated.

Calculation

$$\text{TVB-N} = (*A) \times \text{Strength of acid} \times 0.2 \times \frac{\text{Volume of extract}}{\text{Volume of extract taken}} \times \frac{100}{\text{Weight of sample taken}}$$

(*A = titration reading - blank reading)

2.8.2. Estimation of pH

pH value of the sample was determined with the help of a pH meter (Mettler Toledo 320-s, Shanghi, China) following standard method [35].

2.8.3. Free Fatty Acid (FFA) Estimation

Oil sample used throughout the work was prepared by extracting the salted fish by Folch reagent (chloroform and methanol in the ratio of 2:1 v/v).The salted fish was first cut into small pieces and then ground. The ground material was then mixed with folch reagent in a large wide mouthed stopper glass bottle for 24 hours at room temperature after 15 minutes stirring with glass rod.

Extraction was facilitated by occasional shaking. The mixture was filtered through a Buchner funnel and the filtrate was evaporated in batches under heater or oven at 60°C. Seven gram of well-mixed oil was taken into 250 ml flask and 50 ml ethanol was added, previously neutralized by adding 2 ml phenolphthalein solution. Tritation was done by 0.25 N sodium hydroxide with vigorous shaking until permanent final faint pink color appeared and persisted for at least one minute. The value was reported as percentage (%) of free fatty acid expressed as oleic acid. Milli litre of 0.25 N NaOH required for titration corresponds to the percentage of free fatty acid.

To calculate significance at $p < 0.05$ level all data was analyzed with the help of SPSS for windows, version 20 statistical software [36].

3. Results and Discussion

Determination of the biochemical-composition of experimental Tengra (*M. tengra*) fish in fresh condition as well as dry-salted (DS) and pickle-salted (PS) condition (storage at room temperature) were done. It has been established that the proximate composition of fish may vary in different species and even within the same species from one individual to another is mainly due to age, sex, season, size, species, starvation of the day, energy spending procedure and so on [37,38,39,40].

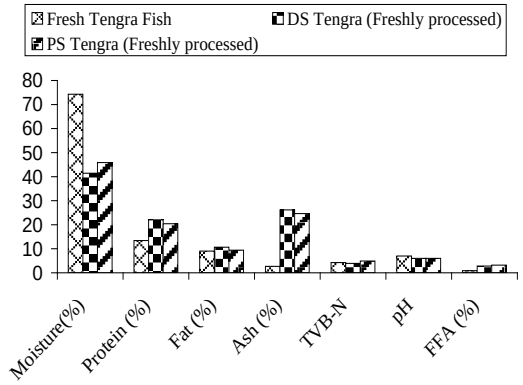

Figure 1. Bio-chemical composition of the fresh experimental fish, freshly-processed dry-salted (DS) and pickle-salted (PS) *M. tengra* fish-products

In case of fresh *M. tengra* fish the percentage of moisture, protein, fat, ash (Proximate composition) was 74.27%, 13.43%, 9.04% and 2.67% and chemical composition (TVB-N, pH, FFA) was 4.27 mgN/100 g, 7 and 0.9% respectively (Figure 1).

In present experiment, fresh *M. tengra* fish recorded a high moisture and low protein content, similar to previous report [41]. Generally, lipid content varies within species (1.46 to 5.77%) and is affected by the catching season (1.2 to 18.4%) [42,43]. The moderate lipid level in this small indigenous fish are similar than those found in other species. [42,44,45,46]. Ash content of fresh *M. tengra* fish was higher than those found in other species [47,48]. The higher ash content could be explained by the presence of the bones in the samples.

3.1. Proximate Analysis

3.1.1. Moisture (%)

Moisture content varies from species to species and temperature to temperature and at different time duration of its preservation. In present experiment, moisture content was found 41.41% and 45.88% in fresh processed DS and PS *M. tengra* fish and after completing the duration of storage period moisture content was increased 44.82% (7 month) and 49.09% (6 month) in DS and PS *M. tengra* fish respectively (Figure 2 and Figure 3). The moisture uptake during the storage period was significant in the products stored in room-temperature. Moisture absorption in such products is obvious during monsoon due to high relative humidity difference.

3.1.2. Protein (%)

Protein content was found 22.05% and 20.43% in fresh process DS and PS *M. tengra* fish and after completing the duration of storage period it was decreased into 20.99% (7 month) and 19.28% (6 month) in DS and PS *M. tengra* respectively (Figure 2 & Figure 3). The decrease of protein level was found to be significantly proportional ($P<0.05$). Protein decreased with storage of cured meat was attributed to some changes during storage that caused by 'maillard reaction and changes in pH [49]. Salt causes the proteins in fish muscle to swell and salt lead the protein become denatured if increases in the muscle [50].

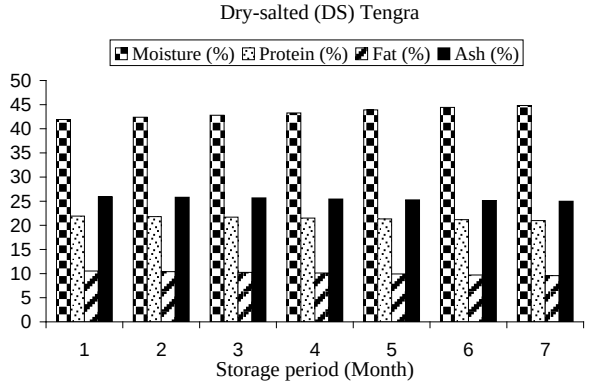

Figure 2. Changes in proximate composition of dry-salted (DS) *M. tengra* fish during storage at room temperature

3.1.3. Fat (%)

Fat content is found to be influenced by season and geographic location [51]. In DS and PS *M. tengra* fish-

products fat content was found 10.65% and 9.40% in fresh process condition and after completing the duration of storage period it was decreased into 9.59 % (7 month) and 8.68 % (6 month) respectively (Figure 2 & Figure 3). It is clear from the present results that fat content was decreased significantly ($p<0.05$). This might be due to oxidative deterioration, thereby affecting lipid extraction [52]. Decrease in the level of crude protein and fat contents of small and large salted Bouri fish muscle (*Mugil cephalus*) were reported [53].

3.1.4. Ash (%)

In DS and PS *M. tengra* fish products ash content was found 26.15% and 24.62% in fresh process condition and after completing the duration of storage period it was found 25.0% (7 month) and 23.41% (6 month) respectively (Figure 2 & Figure 3). The higher value of total ash content in freshly processed DS and PS *M. tengra* fish than fresh fish was attributed to high salt content. Similar levels of ash content in salted fish were noticed by several workers [54].

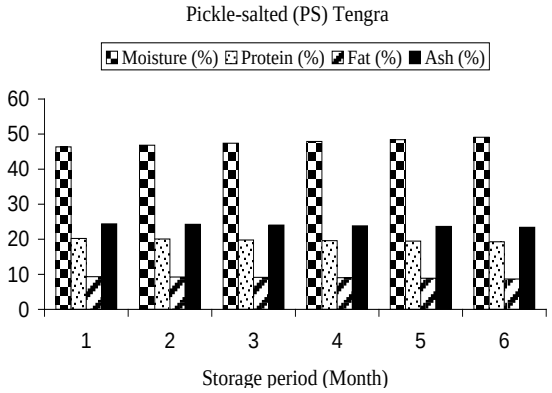

Figure 3. Changes in proximate composition of pickle-salted (PS) *M.tengra* fish during storage at room temperature

3.2. Chemical Analysis

3.2.1. Changes in TVB-N

TVB-N values were found to vary from 3.9 (0 day) to 30.22 mg N/100 g (7 month) for DS *M. tengra* and 4.92 (0 day) to 31.04 mg N/100 g (6 month) for PS *M tengra* fish-products. Significant statistical differences were found between the initial product and end product ($P < 0.05$) during storage period (Figure 4).

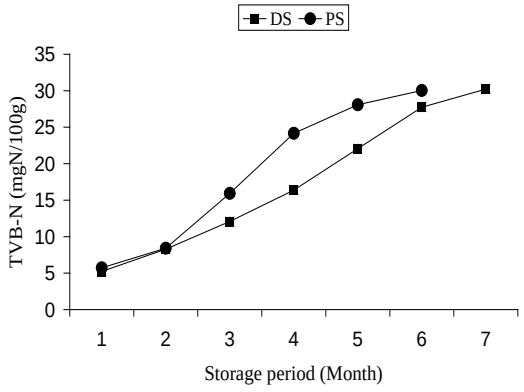

Figure 4. Changes in TVB-N contents of dry-salted (DS) and pickle-salted (PS) *M.tengra* during storage at room-temperature

TVB-N values of the products stored at room temperature showed linearly increasing pattern throughout storage period but neither of the value exceeded the recommended value set for fish regarded as acceptable condition. According to Connell the limiting level for rejection of TVB-N is 30-40 mgN/100 g for storage at ambient temperature. The present findings are in close association with him [55].

Total volatile base nitrogen (TVB-N) is important compound provide a measure of the progress of spoilage that is dependent of sensory assessment. The level of TVB-N in fish & fish products are mostly used as spoilage indicator through bacterial activity [56]. The same result has been evident in the present study. The TVB-N of fish is an indicator of the freshness of the raw material [57]. High TVB-N values are unacceptable and are associated with unpleasant smell in the meat [58]. Assumably, this is because of the impact of the various treatments of TVB-N, which primarily includes nitrogen from ammonia, TMA, and dim ethylamine which reflects the extent of degradation of proteins and non protein nitrogenous compounds which can be explained by proteolysis, due to enzymatic and microbial activities in the samples on storage [59]. In early storage, spoilage rate become slower than later storage time, it would appear from the Figure. This is also supported by other researchers [60,61]. The stage perhaps autolysis is mainly to the tissue as a result TVB-N does not increase highly [62].

3.2.2. Changes in pH

pH is an indicator of the Extent of microbial spoilage in fish and some proteolytic microbes produce acid after decomposition of carbohydrate, thereby increasing the acid level of the medium [63]. pH value is a reliable indicator of the degree of freshness or spoilage.

The pH in fresh condition fresh- water fish flesh is almost neutral. [64]. In the post-mortem period, decomposition of nitrogenous compounds leads to an increase in pH in the fish flesh [65]. The increase in pH indicates the loss of quality. The pH value of Dry-salted (DS) and Pickle-salted (PS) *M. tengra* fish-product was increased significantly ($P<0.05$) with storage period. pH value of fresh Shoal fish was 6.9 in our study. But when salt is added with the fish, pH value decrease due to increase of acidic compound and after that among shelf life study pH value increases in the time interval due to increase of basic compounds. In the present study pH value were found to vary from 6.0 (0 day) to 7.9 (7 month) for DS *M. tengra* and 6.0 (0 day) to 8.0 (6 month) for PS *M. tengra* fish-products (Figure 5).

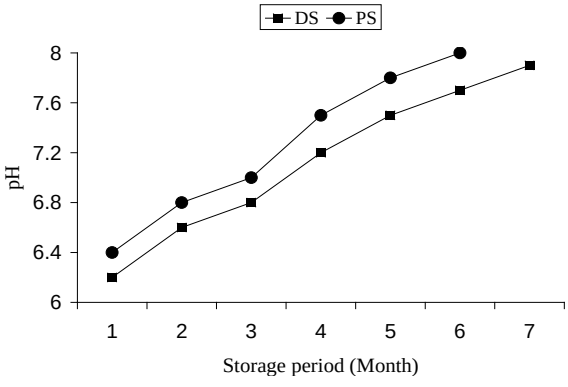

Figure 5. Changes in pH value of dry-salted (DS) and pickle-salted (PS) *M.tengra* during storage at room-temperature

The fish products are acceptable up to a pH of 6.8 but are considered to be spoiled above a pH of 7.0 [32]. The limit of acceptability is usually 6.8 to 7.0 [66]. While the initial pH values in the samples were similar to findings of other researchers; the increase in pH values during the storage of room temperature (27-31°C)was higher than others. The probable reason of these differences is differences in fish species and different methods of salting.

3.2.3. Changes in FFA (Free Fatty Acid)

Among the various parameters to assess the extent of deterioration in fish, determination of free fatty acid (FFA) content has been widely used.

Free fatty acid (FFA), a tertiary product of rancidity, increased during storage. The FFA is a measure of hydrolytic rancidity-the extent of lipid hydrolysis by lipase action. In most fish oils, rancidity is noticeable when the FFA (calculated as oleic acid) is in between 0.5%-1.5% [63]. It produced as a result of fat oxidation (rancidity). FFA value is a measure of the extent of oxidative deterioration in oily fish, but it can fall further at latter stages of fish spoilage [67]. A high level of FFA is characteristics of product that have undergone both microbial and biochemical spoilage [10,68]. This may indicate that greater proportions of unsaturated fatty acids were liberated and were subjected to oxidative splitting at the double bonds. The resulting substances, mostly ketones and aldehydes, appear to be largely responsible for flavor, odor and taste of the salted fish product [53]. The same result was found in the present study.

The FFA value of dry and pickle salted *M. tengra* increased gradually with the passing of storage period (Figure 6). Significant statistical differences were found between the initial product and end product ($P < 0.05$) after storage period. It was vary from 2.8% (o day) to 12.1% (7 month) for DS and 3.2 % (o day) to 12.7% (6 month) for PS *M. tengra* respectively.

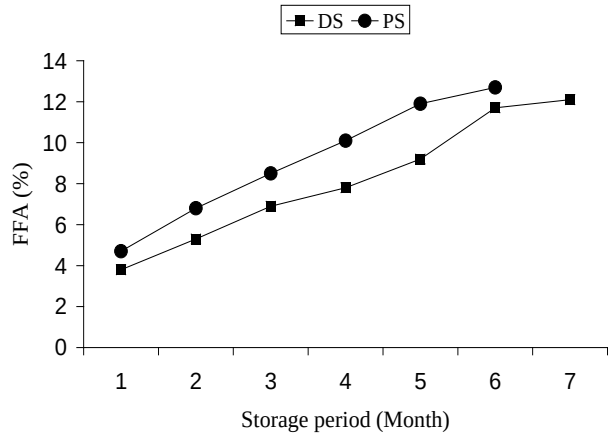

Figure 6. Changes in FFA contents of dry-salted (DS) and pickle-salted (PS) *M.tengra* during storage at room-temperature

Free fatty acid (FFA) value ranged from 2.9% of total lipid on the start day to 7.58, 7.15 and 8.56% on the 60 days of observation in dry salting, wet salting and sundry salted fishes respectively [69]. While the initial value of raw pre-spawning and post-spawning hilsa was 1.15% and 1.21% respectively which after dry salting increased to 7.86% and 8.35% respectively on 18th days of storage at 28°C to 32°C [70].

The present study denoted that the contents of free fatty acid values are similar with the above mentioned studies. The FFA value increased in a characteristics pattern to a certain level of storage period.

High level of FFA is an indication of microbial spoilage activity [32].Most fat acidity begins to be noticeable to the palate when the FFA value calculated as Oleic acid is about 0.5 -1.5 % [71].The result of free fatty acids (FFA) indicated that the salting conditions accelerate lipid oxidation and this is in agreement with the results as shown by other researchers [72,73].

4. Conclusion

It is evident from different test that, fish kept in dry salt maintains the longer period of shelf life and also maintains best quality in protein and fat content than other keeping condition.

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PROTECTIVE EFFECT OF BRINE-SALT CURING ON PHYSICO-CHEMICAL ATTRIBUTES ON THE TAKI FISH (*CHANNA PUNCTATUS*) AND THE TENGRA FISH (*MYSTUS TENGRA*) AT ROOM TEMPERATURE

Farzana Binte Farid*, Gulshan Ara Latifa, Mosarrat Nabila Nahid and Mohajira Begum[1]

Department of Zoology, University of Dhaka, Dhaka-1000, Bangladesh

Abstract: Brine salt curied (BS) Taki fish and the small Tengra fish were studied to observe such salting procedure on their bio-chemical composition stored at room temperature (26-32^{0}C). In fresh-processed condition, moisture, protein, fat, ash, pH and free fatty acid were 62.28%, 18.02%, 2.76%, 17.24%, 6.8 and 0.5% in BS taki fish and 57.35%, 15.3%, 6.84%, 20.8%, 5.9 and 0.9%, respectively in tengra. The value of moisture, pH and FFA increased significantly ($p<0.05$) with the time of storage and these values rapidly increased in cured taki than BS tengra and at the end of 75 days BS taki became spoiled whereas BS tengra still remained in fresh condition. It was observed that BS tengra fish-product had longer shelf life (120 days) and was found better for preservation in laboratory condition. This work also showed that the effect of the treatment on a fish sample dependent on the fish species.

Key words: Brine-salting, taki, tengra, biochemical-composition, room temperature

INTRODUCTION

Fishes ususlly spoil within 12 - 20 hours depending on species and the methods of harvesting. If they are not processed immediately after harvesting, certain irreversible spoilage and deterioration of meat quality begin to take place (Conne 1995). Most of the processing or preservation operations are intended to control the rate of spoilage by reducing water activity of fishes (Eyo 1986).

Processing is applied to fishes from the time of harvesting to the consumption by a customer. Commonly used methods for processing include salting/brining, sun-drying, freezing and smoking, which also increase fish availability to the consumers (Abolagba *et al.* 1996). Adequate information, however, available on the quality of various frozen and dried marine fishery products are available but very little is known about the nutritional quality of brine-salted fresh-water fish-products in Bangladesh.

Taki fish is a medium size air breathing fish while tengra is a small cat fish, both of these fishes live in freshwater environment and are easy available, tasty and nutritious as well as popular fishes. The present work was thus conducted to assess the effect of Brine salt curing on the macronutrient content of two commonly consumed fished of Bangladesh.

*Author for correspondence: <farzanafarid79@gmail.com>. [1]Institute of Food Science and Technology (IFST), BCSIR, Dhaka-1205, Bangladesh.

MATERIAL AND METHODS

The freshwater fish taki (*Channa punctatus*) and tengra (*Mystus tengra*) were collected from the river Meghna in early morning and brought to the Fish Technology Section, Institute of food Science and Technology (IFST), Bangladesh Council of Scientific and industrial Research (BCSIR), Dhaka, using sterile polythene, where fishes were carefully washed with cool tap water. Fins, gills, viscera, head and scales of taki were removed and washed to remove blood, slime and unnecessary flesh. The experiment was conducted for a period of 4 months between May 2013 and August 2013. Fresh fish-specimens were taken randomly and ground with an electric blender to make a homogenous sample before being sampled for analyses.

Brine salting method (BS): In the present experiment brine was prepared using 30 g salt in 100 ml water. Fishes were kept at this brine solution stacked in containers and stored for a salting or curing period, at room temperature (26^0C-32^0C) for the production of brine salted fish. The fishes in brine were kept immersed by putting a glass weight.

Storage of the product: After salting procedure, brine-salted products of the two fishes were preserved in plastic bag maintaining aseptic condition as far as possible and were stored at room temperature (26^0-32^0C). The preservation period of product was linked to the amount of salt added; therefore a straight proportion was present between the amount of salt used and the preservation period (Bahri 2006).

Sampling procedures: Evaluation of phisico-chemical characteristics in brine-salted taki and tengra fishes were carried out for 15 days interval for room temperature, until the fish became spoil or inedible condition. Two duplicate experiments were conducted at regular time intervals during salting period.

Biochemical analysis: Analytical methods were applied for the determination of biochemical composition of the raw fishes as well as of processed fish products on experimental basis. Moisture, fat, ash and FFA value of the fishes were determined by AOAC conventional method (AOAC 1990). The crude protein of the fish was determined by Micro-Kjeldal method (Pearson 1999). pH value of the sample was determined with the help of a pH meter (Mettler Toledo 320-s, Shanghi, China) follwing standard method (Vynke 1981).

To calculate the significance at $p< 0.05$ level all data was analyzed with the help of SPSS for windows, version 20 statistical software.

RESULTS AND DISCUSSION

Determination of the bio-chemical composition of taki (*C.punctatus*) and tengra (*M.tengra*) fish in fresh condition and brine-salted condition (storage at room temperature) were made.

Proximate composition: Moisture, protein, fat and ash were 78.65, 15.89, 3.02 and 1.16%, in case of fresh taki fish and 74.27, 13.43, 9.04 and 2.67%, in case of fresh tengra fish (Fig. 1). Fresh fish samples presented a high moisture and low protein content, similar to previous report (Eyo 1998).

After the end of brining, moisture, protein, fat and ash content were 62.28, 18.02, 2.76 and 17.24% in freshly processed BS taki fish and 57.35, 15.3, 6.84 and 20.8% in freshly processed BS tengra fish-product (Fig.1).

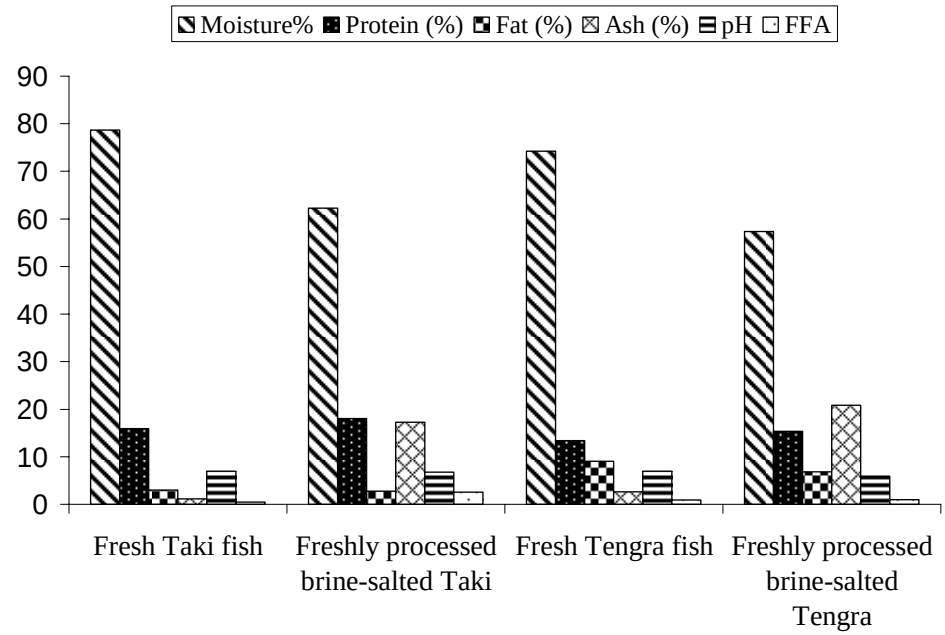

Fig. 1. Comparison of the Bio-chemical composition of fresh Taki (*Channa punctatus*) and Tengra (*Mystus tengra*) fish.

After completing the duration of storage period, moisture, protein, fat and ash contents were found as 65.08, 16.92, 2.48 and 15.99% in case of BS taki (75 days) and 60.7, 14.28, 6.01 and 19.4% in case of BS tengra (120 days) (Figs. 2 and 3).

Moisture uptake and decrease of protein, fat and ash content during the storage period were significant in the products stored in room-temperature. Moisture absorption in such products is obvious during monsoon due to high

relative humidity difference. Decrease of fat might be due to oxidative deterioration, thereby affecting lipid extraction (Gandotra *et al.* 2012). Decrease in the level of crude protein and fat contents of small and large salted Bouri fish muscle (*Mugil cephalus*) were reported (El-Sebahy 1988).

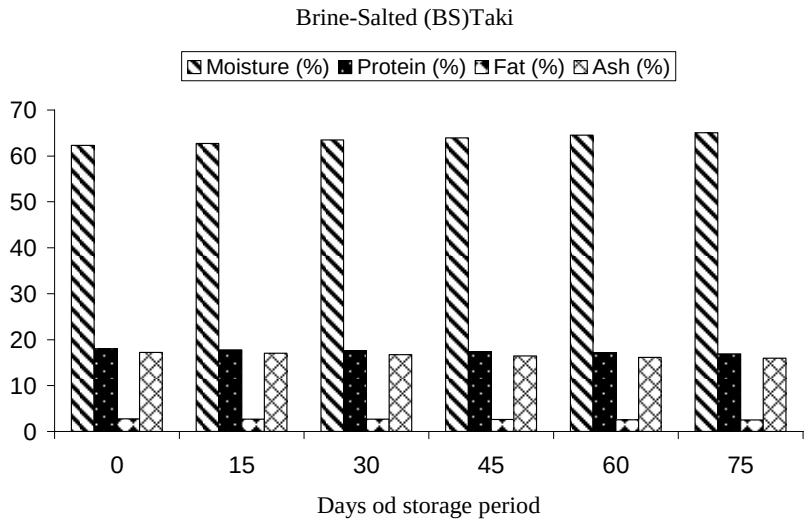

Fig. 2. Changes in Proximate Composition of Brine-salted (BS) Taki fish (*Channa punctatus*) during storage at Room Temperature.

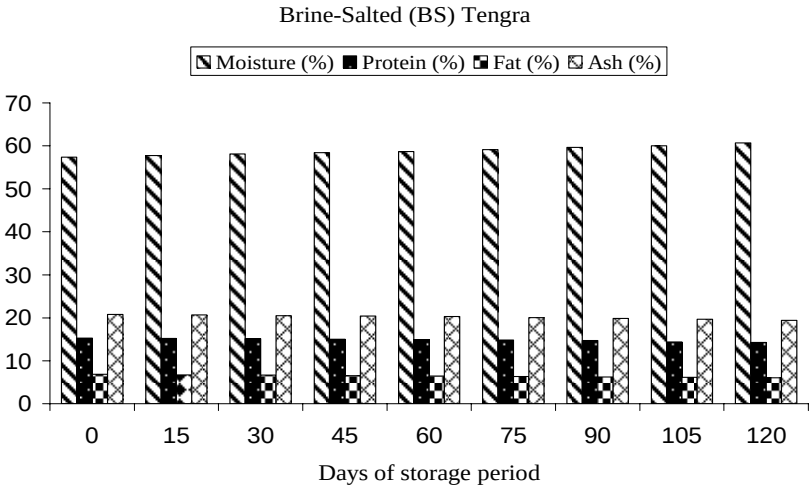

Fig. 3. Changes in Proximate Composition of Brine-salted (BS) Tengra fish (*Mystus tengra*) during storage at Room Temperature.

Changes in pH value: The pH of freshwater fish flesh at fresh-condition is almost neutral (Virta 2009). Increase in pH indicates the loss of quality in fishes. The pH value of BS taki and tengra fish-products was significantly ($P<0.05$)

increased during storage time. In the present study, pH values of both fresh taki and tengra fish were 7 and varied from 6.8 (0 day) to 8.3 (75 days) for BS taki and 5.9 (0 day) to 7.9 (120 days) for BS tengra (Fig. 4). The initial pH values in the samples were similar but the increase in pH values during the storage of room temperature (27-31^{0}C) was higher than reported in other researches (Huss 1988, Shenderyuk 1989, Eyo 1993 and Erkan *et al*. 2011). The probable reason behind these differences was due to differences in fish species and different methods of salting used.

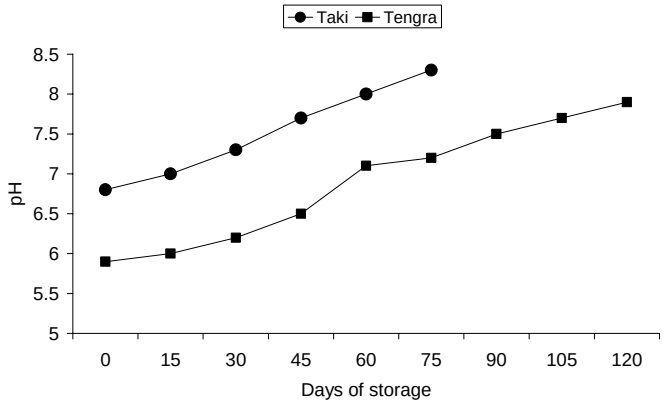

Fig. 4. Changes in pH value of Brine-salted (BS) Tengra (*Mystus tengra*) fish during storage at Room Temperature.

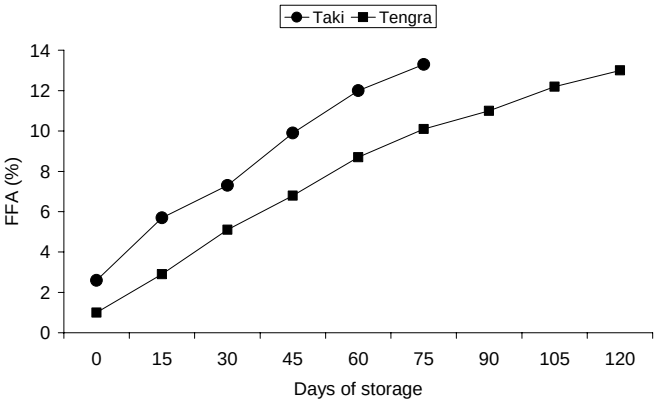

Fig. 5. Changes in FFA (%) value of Brine-salted (BS) Tengra (*Mystus tengra*) fish during storage at Room Temperature.

FFA value: FFA value is a measure of the extent of oxidative deterioration in fish, increased during storage but it can fall further at latter stages of fish spoilage (FAO/SIFAR, 2001).The same result was found in the present study. It

varied from 2.6% (o day) to 13.3% (75 day) for BS taki and 1 % (o day) to 13% (120 day) for BS tengra respectively (Fig. 5).

The above result showed that brine-salted tengra fish-product has grater nutritive value and has longer shelf life (120 days) and has better way for preservation at laboratory condition.

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Comparison of the changes in phisico-chemical characteristics of Dry Salted Snake-head Shoal (*Channa striatus* Bloch, 1801) and Taki (*Channa punctatus* Bloch, 1793) at Room temperature (27^0-31^0C)

Farid F.B.[1*], Latifa G.A.[1], Nahid M.N.[1] and Begum M.[2]
[1]Dept. of Zoology, University of Dhaka, Dhaka 1000, BANGLADESH
[2]Institute of Food Science and Technology, BCSIR, Dhaka 1205, BANGLADESH

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Abstract

Proximate composition (moisture, protein, fat and ash) and chemical analysis (pH) of fresh and dry-salted (DS) fish samples-Shoal (C. striatus) and Taki (C. punctatus) were determined using standard methods of analysis at room temperature (27^0-31^0C) for shelf life study. In processed condition, moisture(%), protein (%), fat (%), ash (%) and pH were 48.84%, 28.49%, 5.63%, 18.97% and 6.3 respectively in case of DS Shoal and 46.21%, 23.58%, 4.03%, 27.27% and 6.5 respectively in case of DS Taki fish-product. During different days of storage, moisture and pH was significantly increased ($p<0.05$) whereas total protein, lipid and ash contents were decreased significantly ($p<0.05$). Moisture (%) and pH was increased 53.69% and 8.1 in case of DS Shoal (165 days) and 53.02% and 7.9 in case of DS Taki (150 days) respectively. Protein (%), fat (%) and ash (%) content were decreased 26.52%, 3.00% and 17.28% in case of DS Shoal (165 days) and 20.41%, 3.24% and 23.45% in case of DS Taki (150 days) fish-product. Experimentally it has been proved that the dry-salted shoal fish-product has longer shelf life (165 days) and has found better way for preservation at laboratory condition.

Keywords: Dry-salting, shoal, taki, proximate-composition, pH, Room-temperature.

Introduction

Being one of the richest sources of proteins, vitamins and minerals fishes are widely used in Bangladesh as essential source of nutrients required for people. Moreover, as supplementary source fishes alone contribute about 80% to the nation's animal protein. But the availability of this valuable nutrients source largely depends on the extent of salting, drying, smoking and freezing and many other types of preservation methods[1,2].

Fishes are very much susceptible to spoilage factors and stat to spoil just after being caught. The spoilage is mainly due to microbial flora, sun, poor handling methods and so on[3]. So immediate care should be taken just after caught in order to prevent possible damaged. There is increased pressure from countries engaged in fish processing to establish effective quality assurance systems in their plants and companies must devote both management and technical resources to meet this objective.

The purpose of the present study is to develop an efficient and effective model for curing of various fish species using cheapest ingredients (like salt) for the production of high quality end products, such as Dry-salted products and transfer the technology to the rural small-scale fisher folks allover Bangladesh. Among the freshwater fish species, Shoal (*C. striatus*) and Taki (*C. punctatus*) are delicious, nutritious and popular to the consumers as well as bear high market price. So it is necessary to take some steps for their proper preservation and marketing and during this period maintain proper quality.

Material and Methods

The fresh fish (Shoal and taki) were collected from the river Meghna in early morning and the fishes were brought to the Fish Technology Section, IFST, BCSIR, Dhaka, using sterile polythene where fishes were carefully washed with cooled tap water. Head, scales, fins, gills and viscera were removed and washed to remove blood, slime and unnecessary flesh. The experiment was conducted for a period of 6 months between March, 2013 and August 2013.

A fresh flesh sample of shoal and Taki fish specimens (6 to 7 slices) were taken randomly which represented the parts from whole body of the fish. Then the slices were chopped with skin and bone and finally ground with an electric blender to make a homogenous sample before being sampled for analysis.

Dry-salting (DS) method: Sodium chloride (NaCl), also called common salt, and table salt, is generally recognized as a safe, antimicrobial and incidental food additive[4]. Salt has been used as a seasoning and flavor enhancer as well as a preservative or curing agent, had been purchased from the local market. Salt is a powerful depressor of water activity (a^w) of the food[5]. It has also been indicated that chloride ions are toxic for some

microorganisms[6]. The fresh fishes were enrolled by dry commercial salt (NaCl) of about 30% by weight of the dressed fish (fish weight: salt weight 3:1), stacked in containers and stored for a salting or curing period, at room temperature. In this method, the extracted water of the fish due to salt action had been removed from the container. Thus the fishes were always allowed to remain in dry condition for the production of dry salt-cured fish.

Storage of the product: At the end of salting process, Dry-salted product of two fishes was packaging with plastic bag maintaining aseptic condition as far as possible and was stored at room temperature (27^0-31^0C). The preservation period of product is linked to the amount of salt added; therefore a straight proportion is present between the amount of salt used and the preservation period[7].

Sampling procedures: Evaluation of phisico-chemical characteristics in Dry salted Shoal and Taki fishes were carried out 15 days interval for room temperature, until the fish become spoil or inedible condition. Two duplicate experiments were conducted at regular time intervals during salting period. Salt crystal was removed from the dry-salted products by tissue paper before being sampled for analysis.

Proximate Analysis: Using conventional method of AOAC, the proximate composition of fish was determined[8].

Estimation of moisture content: About 5 gram of previously prepared fairly minced samples were taken into each known weight basin and weighed in a digital balance (Toledo, Switzerland). The samples were allowed to dry into the oven (Memmet 854 Schwabach) at 105^0C for 24 hours in order to remove the moisture until constant weight. After that, the basins are taken out of the oven, cooled in a desiccators and were weighed in a digital balance.

Calculation

$$\text{Moisture (\%)} = \frac{\text{Weight Loss}}{\text{Original Weight of Taken Sample}} \times 100$$

Estimation of protein content: The protein content was estimated using conventional micro-kjeldahl method[9].

Calculation

$$\frac{\%\ N_2(\text{titration reading - blank reading}) \times \text{strength of acid} \times 0.002 \times \frac{100}{5} \times 100}{\text{weight of taken sample}}$$

% of protein = % of total N_2 × 6.25 (For most routine purposes the percent of protein in the sample is then calculated by multiplying the % of N_2 with an empirical factor of 6.25 for fish).

Estimation of fat content: About 5 g sample was taken into conical flasks and 10 ml of folch reagent (Chloroform: Methanol = 2: 1) was added into the sample and homogenized properly and kept in air-tight condition for 24 hours. Fat contents of the fish muscle react with that solvent and remains in the solution. After 24 hours the solution of the flask was filtered in another weighed conical flask through a filter paper. Then these flasks were given in a hot water bath to dry up and removed the solvent. After that the flasks were kept into an oven for an hour to get the actual fat content. Then the flasks were weighed in an electronic balance to get the amount of fat content.

Calculation

$$\text{Fat (\%)} = \frac{\text{Weight of the residue}}{\text{Weight of taken sample}} \times 100$$

Estimation of ash content: About 4-5 g fish sample was weighed into a pre-weighed crucible and heated over a long flame till all the material was completely churned and transferred in a Muffle Furnace (600^0C) for 5 hours. Then the crucible was cooled in desiccators. Finally ash (%) was estimated.

Calculation

$$\text{ash (\%)} = \frac{\text{Weight of fish}}{\text{Weight of taken sample}} \times 100$$

Estimation of pH value: pH value of the sample was determined with the help of a pH meter (Mettler Toledo 320-s, Shanghi, China) follwing standard method[10].

Statistical analysis: To calculate significance at $p < 0.05$ level all data was analyzed with the help of SPSS for windows, version 20 statistical software.

Results and Discussion

Determination of the proximate composition and pH of Shoal and Taki fish in fresh condition and dry-salted condition (storage at room temperature) were done. It has been established that the proximate composition of fish may vary in different species and even within the same species from one individual to another is mainly due to age, sex, season, size, species, starvation of the day, energy spending procedure and so on[11-14].

Moisture, protein, fat, ash and pH value was 77.03%, 17.32%, 2.62%, 1.44% and 6.9 in case of fresh shoal fish and 78.65%, 15.89%, 3.02%, 1.16% and 7 in case of fresh taki fish respectively (figure 1). Fresh fish samples presented a high moisture and low protein content, similar to previous report[15]. Fish species with low levels of fat are suitable to be processed[16].

Changes in proximate composition in dry-salted shoal and taki fish products during different days of observation period through shelf life study were shown in figure 2 and 3.

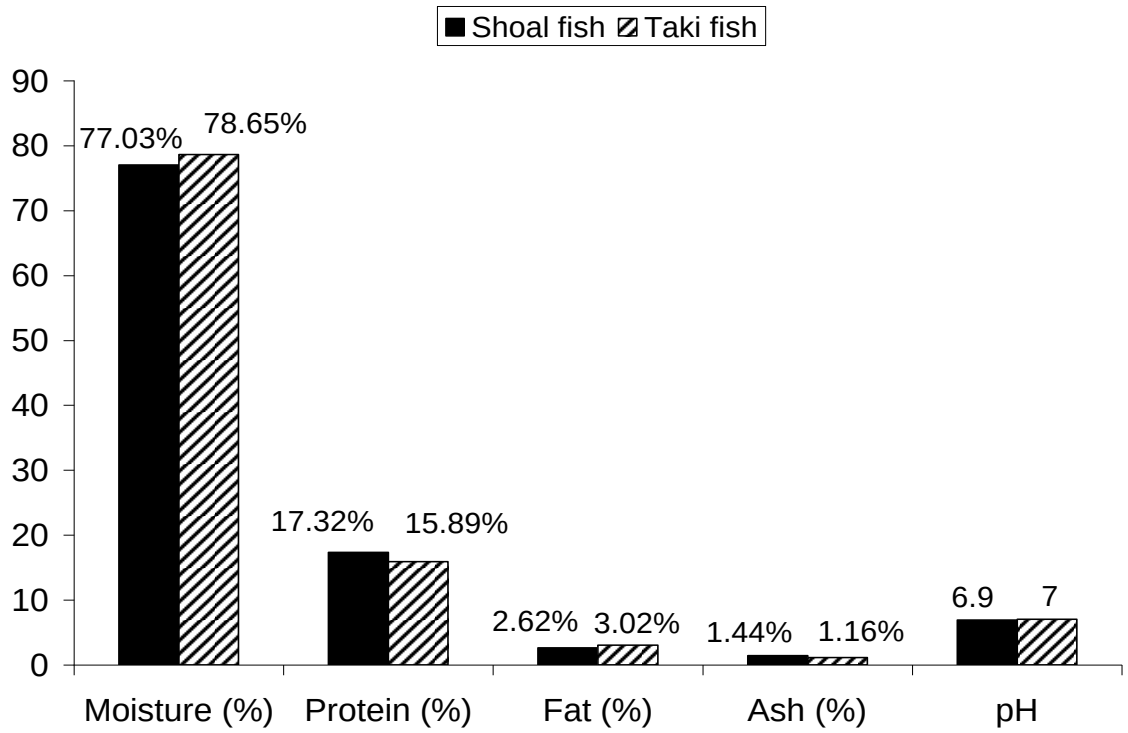

Figure-1
Comparison of the proximate composition and pH value of fresh Shoal (*C.striatus*) and Taki (*C. punctatus*) fish

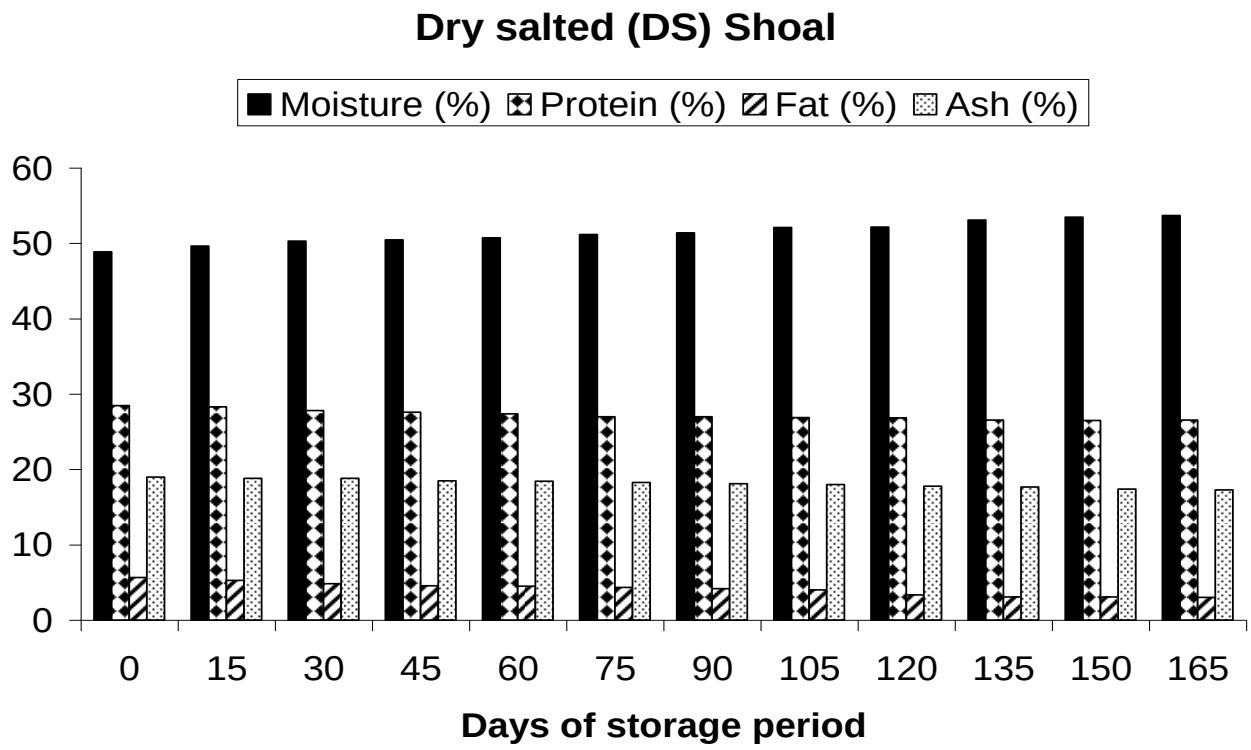

Figure-2
Changes in Proximate Composition of Dry-Salted Shoal fish during Storage at Room Temperature

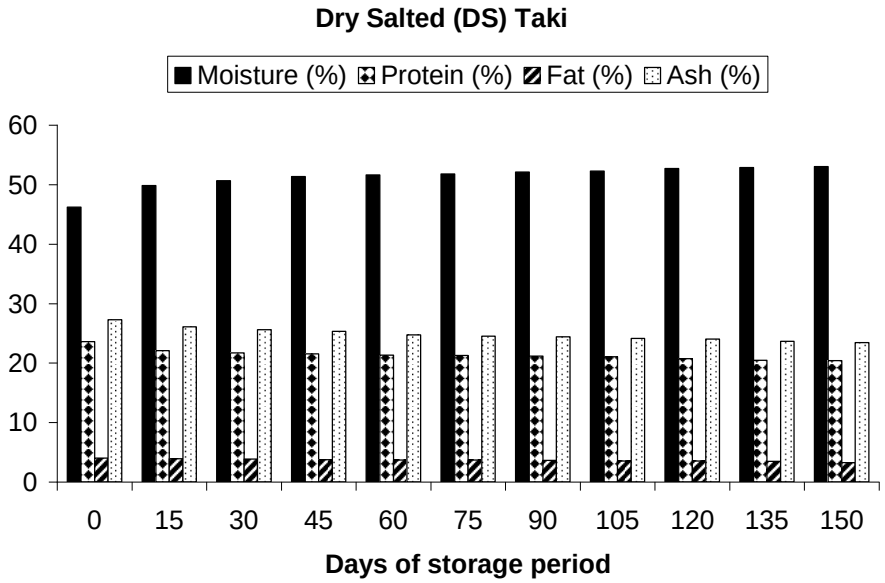

Figure-3
Changes in Proximate Composition of Dry-Salted Taki fish during Storage at Room Temperature

Moisture (%): In present experiment, moisture content was 48.84% and 46.21% in freshly processed dry-salted shoal and taki fish and after completing the duration of storage period, it was found 53.69% (165 days) and 50.92% (150 days) respectively (figure 2 and 3). The moisture uptake during the storage period was significant in the products stored in room-temperature. Moisture absorption in such products is obvious during monsoon due to high relative humidity difference.

Protein (%): In dry-salted shoal and taki fish products, protein content was found 28.21%, and 23.58%, in fresh-process condition and after completing the duration of storage period it was decreased into 25.52% (165 days) and 21.53 (% (150 days) respectively (figure 2 and 3). The decrease of protein level was found to be significantly proportional ($P<0.05$). Protein decreased with storage of cured meat was attributed to some changes during storage that caused by 'maillard reaction and changes in pH[17]. Salt causes the proteins in fish muscle to swell and salt lead the protein become denatured if increases in the muscle[18].

Fat (%): Fat content may be influenced by season and geographic location[19]. In dry-salted shoal and taki fish products, fat content was found 3.99%, and 3.93%, in fresh-process condition and after completing the duration of storage period it was 3.30% (165 days) and 3.49 (% (150 days) respectively (Figure 2 & 3). It is clear from the present results that fat content was decreased significantly ($p<0.05$). This might be due to oxidative deterioration, thereby affecting lipid extraction[20]. Decrease in the level of crude protein and fat contents of small and large salted Bouri fish muscle (*Mugil cephalus*) were reported[21].

Ash (%): In dry-salted shoal and taki fish products, ash content was found 18.98%, and 26.37 %, in fresh-process condition and after completing the duration of storage period it was found 17.58 % (165 days) and 24.55 (% (150 days) respectively (figure 2 and 3). The higher value of total ash content in freshly processed dry-salted shoal and taki fish than fresh fish was attributed to high salt content. Similar levels of ash content in salted fish were noticed by several workers[22].

Changes in pH value: The pH of freshwater fish flesh at fresh-condition is almost neutral[23]. In post-mortem period, decomposition of nitrogenous compounds often tends to increase in pH level in fish flesh[24]. Increase in pH indicates the loss of quality in fishes. The pH value of DS shoal and taki fish-product was significantly ($P<0.05$) increased during storage time. pH value of fresh Shoal and taki fish was 6.9 and 7 in our study . But when salt is added with the fish, pH value decrease due to increase of acidic compound and after that among shelf life study pH value increases in the time interval due to increase of basic compounds. In the present study pH value were found to vary from 6.3 (0 day) to 8.1 (165 days) for DS shoal and 6.5 (0 day) to 7.9 (150 days) for DS taki (figure 4).

The acceptable range of fish pH is 6. 8 but are considered to be spoiled above 7[25]. The initial pH values in the samples were similar with other researchers. The increase in pH values during the storage of room temperature (27-31^{0}C) was higher than reported in other researches. The probable reason behind these differences was due to differences in fish species and different methods of salting used.

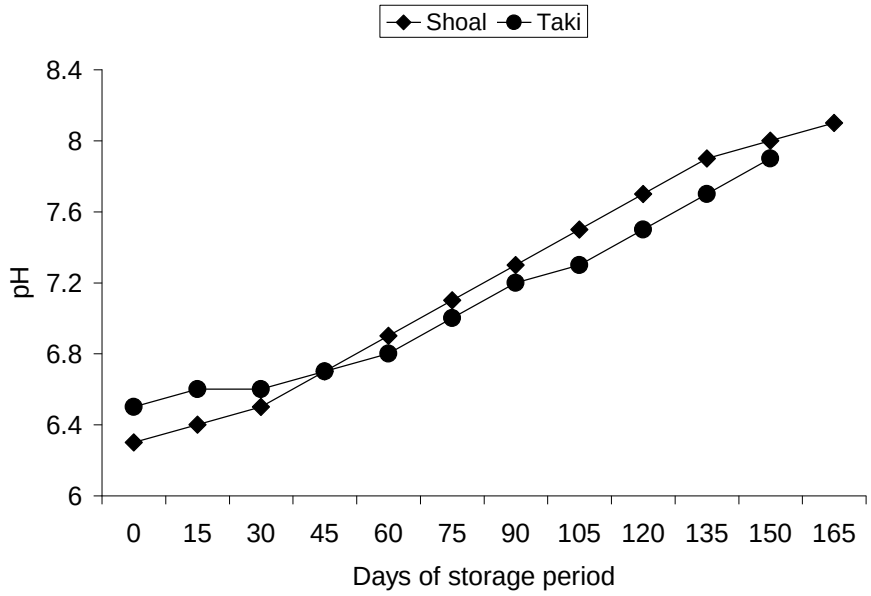

Figure-4
Changes in pH value of Dry-Salted Shoal and Taki fish during Storage at Room Temperature

Conclusion

From the result of this study, the proximate analysis and pH value showed that dry salted shoal product have grater nutritive value in terms of percentage crude protein and experimentally it has been proved that this product has longer shelf life (165 days) and has found better way for preservation at laboratory condition.

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Protective effect of Salt (sodium chloride) and Turmeric (*Curcuma longa*) on Physicochemical attributes of sun-dried Tengra fish *(Mystus tengra*; Hamilton-Buchanan, 1822) at Laboratory condition

Farid F.B*[1], Latifa G.A[1], Chakraborty S.C[2]. Nahid M.N[1] and Begum M[3]
[1]Department of Zoology, University of Dhaka, Dhaka 1000, BANGLADESH
[2]Department of Fisheries Technology, Bangladesh Agricultural University, Mymensingh, BANGLADESH
[3]Institute of Food Science and Technology, BCSIR, Dhaka 1205, BANGLADESH

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Abstract

Sun-drying is one of the most important low cost methods of fish preservation and the products provide nutrients to all categories of people through the world including Bangladesh. The experiment was subjected to the difference between biochemical-composition and quality-analysis of sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) Tengra fish product for making a better flavored product with a view to preserve it in laboratory level for a long time. In fresh-process condition the values of total volatile base nitrogen , pH and free fatty acid were 1.9 mgN/100g, 6.3, 1.8% in case of SDS and 2.52 mgN/100g, 6.4 and 1.6% in case of SDS+T Tengra respectively. This value increased significantly ($p<0.05$) with the time of storage and between this two products, these values rapidly increased in SDS tengra than SDS+T Tengra fish-product and at the end of 12 months, the SDS Tengra fish-product became spoiled whereas SDS+T Tengra fish-product still in fresh condition. The TVB-N value had been found to have inverse relationship with the sensory score of both dried products. From the overall performance, it has been proved that this fish was highly acceptable level in salt-turmeric condition and also maintain best quality.

Keywords: Salt, turmeric, sun-drying, tengra, bio-chemical-composition, quality-analysis.

Introduction

In Asia, Bangladesh is ranked as third largest aquaculture producing country after Chaina and India[1]. Fisheries items are the major protein source of Bangladesh which contributing 58% of the nation's animal protein demands[2]. The current fish consumption rate is 17.52 kg/people/year whereas the demand is 20.44 kg/people/year and is 29.74 MT per year[3,4] .

The freshwater small indigenous fish species provide food and nutrition substance and supplemental income to great majority of people. The rural people have easy access to flood plain, reservoir and natural water bodies where small fishes are abundantly caught by traditional gear. It is well known that small fishes has high nutritional value in terms of both protein content and presence of micronutrients, vitamins and minerals for both young and old age consumers because these fishes can be consumed with their bones and heads[5]. These fishes are acceptable in all classes of peoples as fresh as well as dried products.

Catfish being one of the most valued and very diverse groups of bony fishes. Popular lean catfish tengra (*Mystus tengra*) is selected for the present study which is one of the sole species of family Bagridae. This species very widely distributed in rivers, canals, khals, beels, ditches, inundated fields and other freshwater areas and is one of the most common catfish of the commercial catches of Bangladesh.

In our country, small indigenous fish in a fresh condition is not always available. Major fishing grounds are far away from the cities and the consuming centers which are not easily accessible. Fishing is also seasonal. Seasonal abundance in certain places and a dearth of fish in others stimulates fisherman to preserve their catch.

Fish is a low acid food and is therefore very susceptible to the growth of food spoilage bacteria. Fish begin to deteriorate as soon as they leave the water. The preservation of fish is therefore considered to be a major hindrance to its production and utilization especially in the tropical countries like Bangladesh where spoilage is rapid at ambient temperature. Due to perishable nature of fish, traditional methods of preservation have been developed over the years which including Salting, drying, smoking etc[6]. Preservation process starts when it is harvested and become complete when reaches the consumer's table[7]. Among the different fish products, dried fish is an important source of animal protein in Bangladesh. Sun drying is an important and low cost method of fish preservation. It is practiced in Bangladesh as well as throughout the world since the time of immemorial and regarded as a traditional and primitive means of fish preservation. Fishery industry of Bangladesh is mainly processing high value items such as frozen shrimps and dried products.

The main purpose in drying is to prolong the shelf life of the product. It is a slow process (usually it takes 5-7 days to dry) that makes the product spoiled and unhygienic due to partial destruction of protein content of the fish through hydrolysis oxidation. One of the major problems associated with the lengthy sun-drying of fish is the infestation of the products by the blowfly and beetle larvae. To avoid such infestations and microbial contaminations salt and salt-turmeric was used combined in order to achieve the desire product. Being a safe, antimicrobial and incidental food additive, toxic for some microorganisms, depressor of water activity (a^w) of the food, sodium chloride has been used as a seasoning and flavor enhancer as well as a preservative or curing agent[8,9]. Brining reduces the microorganisms count on dry fish[10]. The active ingredient of turmeric (*Curcuma longa*) having pesticidal action is curcumin[11]. It is also known to have antibacterial properties and have a range of pharmacological activities[12-14]. It is used as a food additive, preservative and coloring agent in many other Asian countries, including China and South East Asia[15]. The suitability of herbal products like turmeric in repelling dry fish insect[16].

In the traditional storage of dried fish in Bangladesh, no proper measures are normally taken to protect the fish against unfavorable environmental conditions. In order to ensure micronutrient supply for the growing population and to enable the poor fishermen and processors to produce high quality marketable products, the improvement of traditional fish drying is an urgent necessity.

In Bangladesh very little is known about the production and quality aspects of traditional dried fresh water fishery products. With this view in mind the present study was undertaken.

Material and Methods

Collection of the fishes and location of the experiment: Fresh experimental Tengra Fish (*Mystus tengra*) had been collected from the river Meghna in the early hours of day and the fishes were brought to the Fish Technology Section, IFST, BCSIR, Dhaka for conducting the research activities, starts in the month of January, 2013. The whole experimental period covered 14 months of duration started from January, 2013 to March, 2014.

Preparation of fish: Fishes were carefully washed using cooled tap water. Fins, gills and viscera were removed and again washed with tap water to remove blood, slime and unnecessary flesh.

Fresh sample: Few fresh samples of experimental fish species was taken to the laboratory for quality analysis. Fishes were taken randomly and ground with an electric blender to make a homogenous-sample before being sampled for analysis.

Therefore, the total cleaned fishes were grouped into 2 batches.
Sun-dried-salting (SDS) method: During this experiment the fresh tengra fishes were enrolled by dry commercial salt (NaCl) of about 30% by weight of the dressed fish (fish weight: salt weight 3 : 1). They were kept on a plastic bade basket in the sun. They were kept in sun regularly during day time (12 a.m. to 3 p.m.) for 2-7 days as sometimes the sky was cloudy and until the ripening period was over. At the same time, temperature and relative humidity were also recorded. During sun-drying, they were kept covered by dense meshed nylon or mosquito net to avoid outside contamination and prevent bird attack and fly infestation.

Turmeric treated sun-dried-salt curing method: During this method the fresh tengra fishes were enrolled by dry commercial salt (NaCl) of about 30% by weight of the dressed fish (fish weight: salt weight 3:1) and Turmeric powder of about 1% of dressed fish weight. They were kept on a plastic bade basket in the sun. They were kept in sun regularly during day time (12 a.m. to 3 p.m.) for 2-7 days until the ripening period was over. At the same time, temperature and relative humidity were also recorded. During sun-drying, they were kept covered by dense meshed nylon or mosquito net to avoid outside contamination and prevent bird attack and fly infestation.

Storage of the product: At the end of the drying period, sun-dried salted (SDS) and turmeric treated sun-dried-salted (SDS+T) tengra fish-products was packaging with plastic bag maintaining aseptic condition as far as possible and was stored at room temperature.

Sampling procedures: To determine the bio-chemical composition of fresh as well as dried fish and quality of dried fishes during storage period some parameters, *viz.* freshness test by sensory scores, TVB-N value, pH, FFA, etc. were analyzed 2 month interval until the fish become spoil or inedible condition. The experiment was done for second time at regular intervals during salting period. Salt crystal was removed from both salted-dried fish-products using dry tissue paper before being sampled for analysis.

Biochemical analysis: Analytical methods were applied for the determination of biochemical composition of the fresh fish and shelf-life quality of processed fish products on experimental basis. The analytical methods are: Moisture, fat and ash contents of the fish were determined by AOAC method[17], The crude protein of the fish was determined by Micro-Kjeldal method[18], Sensory score evaluation has been done by using 9-point hedonic scales as described by Peryan and Pilgrim (9. Like extremely; 8. Like very much; 7. Like moderately; 6. Like slightly ;< 5. Bad)[19]. TVB-N was determined by Conway modified micro-diffusion technique[20]. pH was used to measure quality deterioration of Tengra fish using a pH meter (Mettler Toledo 320-s, Shanghi, China)[21], FFA of the fish was determined by AOAC method[17].

Data were analyzed using SPSS for windows-20 statistical programme. Significance was established at $p< 0.05$.

Results and Discussion

Biochemical analysis: The experimental fish tengra (*Mistus tengra*) is small in size and rich in nutrients. In case of fresh tengra fish the percentage of moisture, protein, fat, ash (Proximate composition) was 74.27%, 13.43%, 9.04% and 2.67% and chemical composition (TVB-N, pH, FFA) was 4.27 mgN/100g, 7 and 0.9% respectively (Fig.1). Generally, lipid content varies within species (1.46 to 5.77%) and is affected by the catching season (1.2 to 18.4%)[22]. In present experiment, fresh tengra fish recorded a high moisture and low protein content, similar to other report[23]. Ash content of fresh tengra fish was higher than those found in other species[24]. The higher ash content could be explained by the presence of the bones in the samples.

Values of Moisture, protein, fat, ash, TVB-N, pH and FFA was 4.9%, 43.00%, 15.99%, 36.20%, 1.9 mgN/100g, 6.3 and 1.8% in case of sun-dried salted (SDS) and 6.08%, 42.60%, 15.58%, 35.60%, 2.52 mgN/100g, 6.4 and 1.6% in case of turmeric treated sun-dried-salted (SDS+T) tengra respectively.

Quality assessment and storage stability: Sensory evaluation (Score): According to the panel's evaluation, the sensory properties of sun-dried salted (SDS) and turmeric treated sun-dried-salted (SDS+T) tengra fish-products were in acceptable condition throughout storage period though, statistically there was significant difference ($p<0.05$) in the sensory evaluation during storage period based on the panel's score. The initial score of the sensory evaluation of SDS and SDS+T tengra was 9. But during storage period this score rapidly decreased and at the end of the storage period, the score was 5 in case of SDS (12 month) and SDS+T (14 month) Tengra. Figure-2, 3. This hedonic rating scale was applied by using 9- points for the sensory evaluation of the dried and dehydrated fish[25]. The sensory analysis of salted- sun dried (SDS) and turmeric treated salted sun- dried (SDS+T) fishes were done and reported that the quality of turmeric treated salted sun- dried product was much better.

Changes in TVB-N (Total Volatile Base Nitrogen) value: The TVB-N of fish is an indicator of the freshness of the raw material[26]. TVB-N values were found to vary from 1.9 (0 day) to 30.15 mgN/100g (12 month) for sun-dried salted (SDS) tengra and 2.52 (0 day) to 30.24 mgN/100g (14 month) for turmeric treated sun-dried-salted (SDS+T) tengra. TVB-N values of the products storage at room temperature showed linearly increasing pattern throughout storage period but neither of the value exceeded the recommended value set for fish regarded as acceptable condition. Significant statistical differences were found between the initial

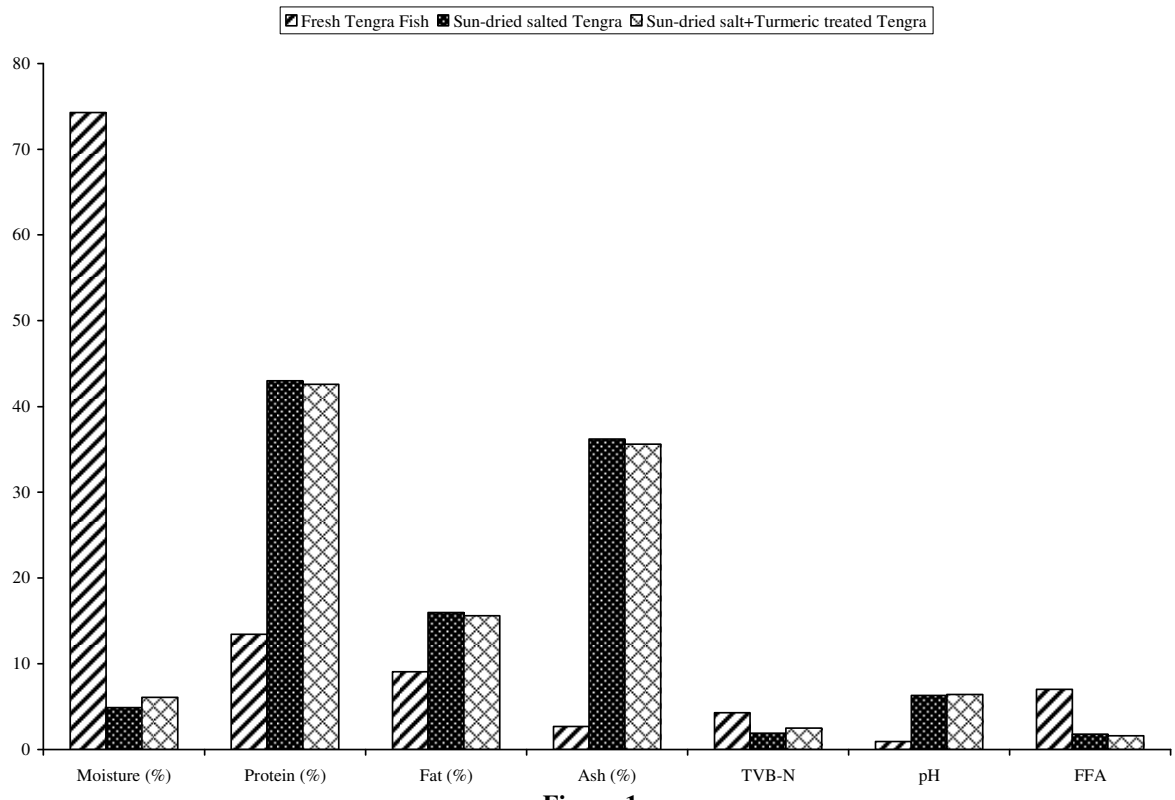

Figure-1
Biochemical composition of fresh, freshly processed sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) Tengra fish-products

product and end product ($P < 0.05$) during storage period. Total volatile base nitrogen (TVB-N) is important compound provide a measure of the progress of spoilage that is dependent of sensory assessment. TVB-N contents of mirror carp fish fillets were found mean value 11.67 mg/100 g, and TVB-N contents in treated fillets did not show an important increase till days 14 of storage, on the following days, reached to 17.9-20.07 mg/ 100g levels on days 28, increasing rapidly[27]. The level of TVB-N in fish and fish products are mostly used as spoilage indicator through bacterial activity[28]. The same result has been evident in the present study. TVB-N is mainly contributed by ammonia in the muscle produced by determination of muscle proteins[29]. A value of 35 mg/100 g of TVB-N has been suggested as border line[30]. In present experiment, TVB-N values of all samples were lower than 35 mgN/100 g which was considered as the threshold for a good-quality fish product (figure-2, 3). In early storage, spoilage rate become slower than later storage time, it would appear from the Figure. TVB-N value increases when decrease of sensory score value with the increase of storage period.

Changes in pH value: pH value is a reliable indicator of the degree of freshness or spoilage. The pH in fresh condition fresh-water fish flesh is almost neutral[31]. Because of the decomposition of nitrogenous compounds, pH in the fish flesh increase in the post-mortem period. The increase in pH indicates the loss of quality. The pH value of sun-dried salted (SDS) and turmeric treated sun-dried-salted (SDS+T) tengra fish-product was increased significantly ($P<0.05$) with storage period. pH value of fresh tengra fish was 7.0 in present study . But when salt is added with the fish, pH value decrease due to increase of acidic compound and after that among shelf life study pH value increases in the time interval due to increase of basic compounds. In the present study pH value were found to vary from 6.3 (0 day) to 7.1 (12 month) for SDS and 6.4 (0 day) to 7.2(14 month) for SDS+T tengra.

The limit of acceptability of fish products is usually 6.8 to 7.0[32]. While the initial pH values in the samples were similar to findings of other researchers; the increase in pH values during the storage of room temperature (30-34^{0}C) was higher than others. The probable reason of these differences is differences in fish species and different methods of salting.

Changes in FFA (Free Fatty Acid) value: FFA value is a measure of the extent of oxidative deterioration in oily fish, but it can fall further at latter stages of fish spoilage[33]. The initial FFA values were 1.8% (oleic acid percentage) and 1.6% for sun-dried salted (SDS) and turmeric treated sun-dried-salted (SDS+T) tengra fish-products. FFA values increased with storage time.

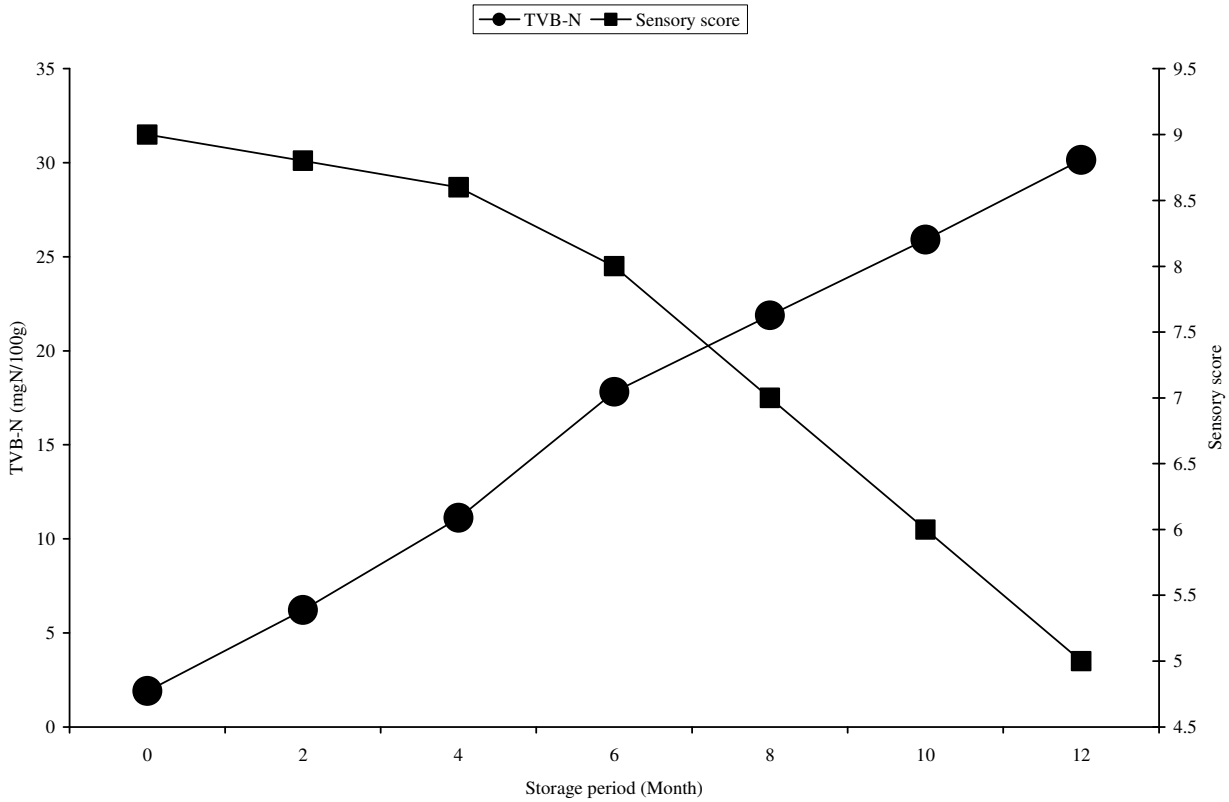

Figure-2
Inverse relationship between TVB-N and sensory score of sun-dried salted (SDS) Tengra fish-product during storage at room temperature

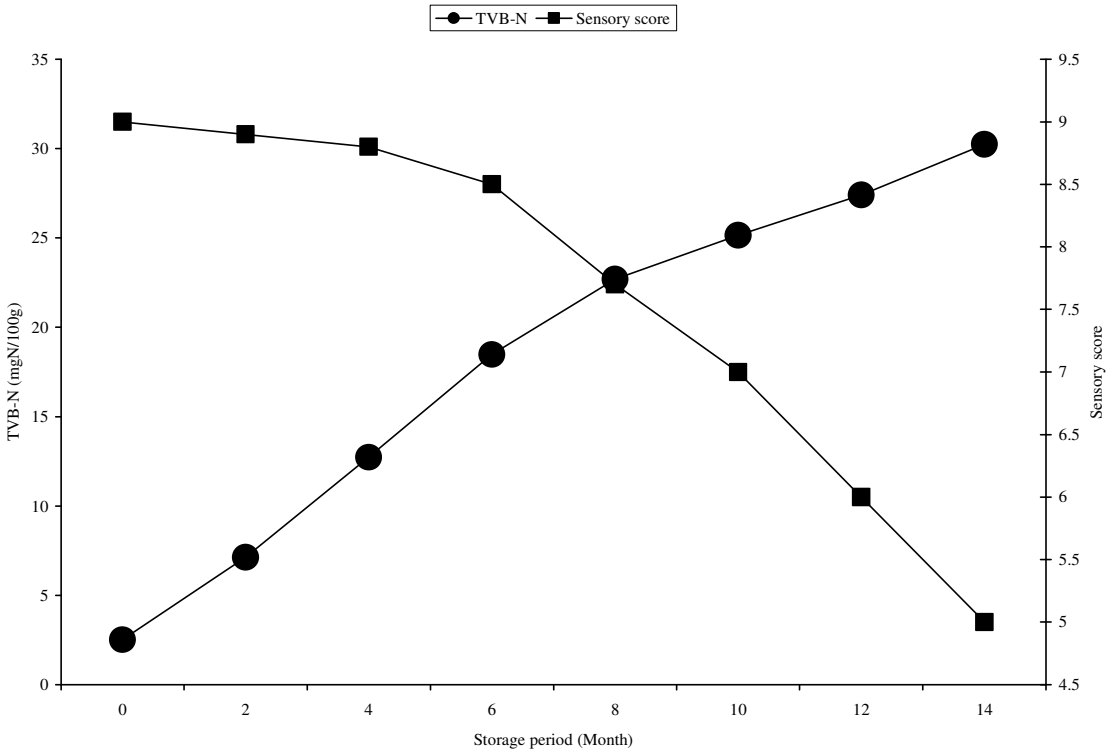

Figure-3

Inverse relationship between TVB-N (mg N/100g) and sensory score of turmeric treated sun-dried salted (SDS+T) Tengra fish-product during storage at room temperature

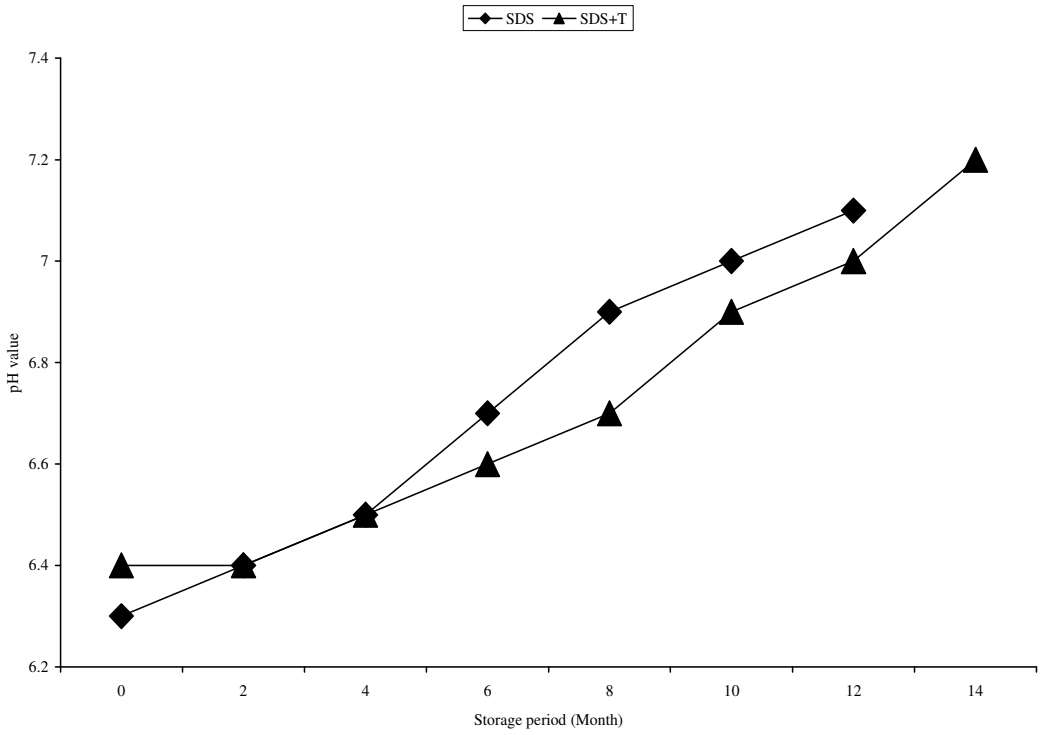

Figure-4

Changes in pH value of sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) Tengra fish-products during storage at room temperature

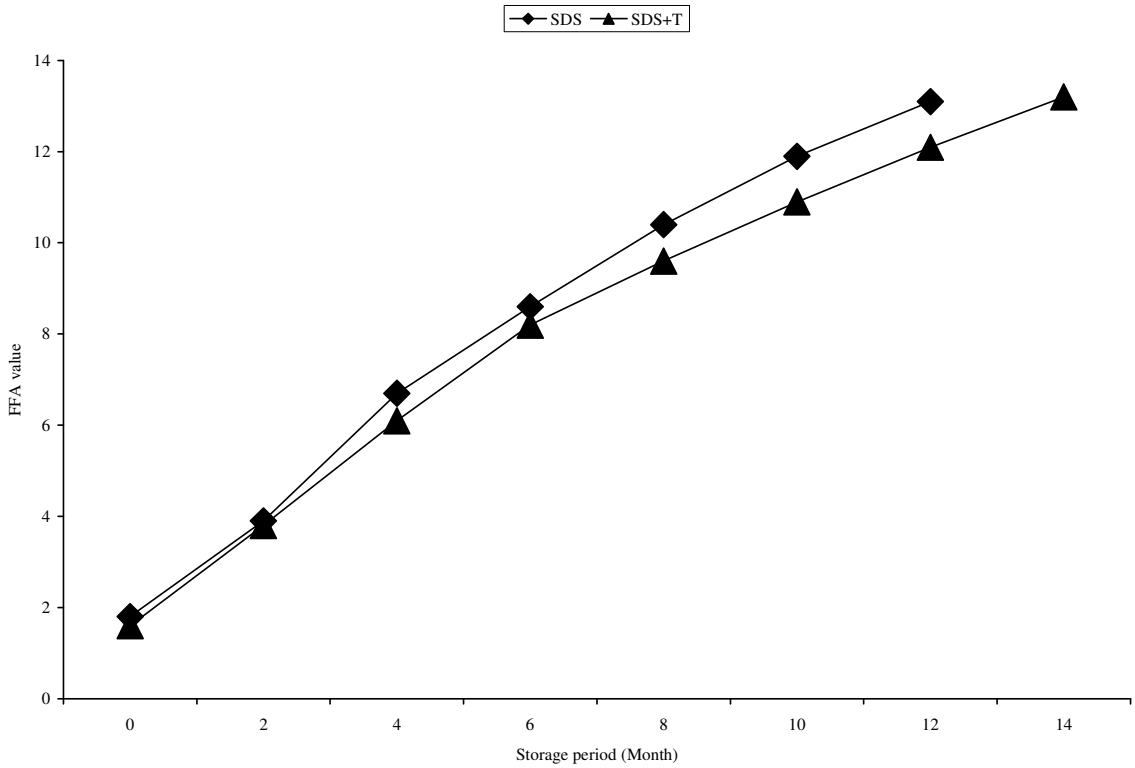

Figure-5
Changes in FFA (%) value of sun-dried salted (SDS) and turmeric treated sun-dried salted (SDS+T) Tengra fish-products during storage at room temperature

At the end of the storage period values of FFA were found to be 13.1% (12 month) for SDS tengra and 13.2% (14 month) for SDS+T tengra respectively. Significant statistical differences were found between the initial product and end product ($P < 0.05$) during storage period. Lipid hydrolysis by itself has no nutritional significance but the accumulation of free fatty acids (FFA) in fish oils in undesirable amount due to secondary reaction catalyzed, such as increased susceptibility to oxidation and consequent development of off flavors[34]. The result of free fatty acids (FFA) (Figure 5) indicated that the salting and drying conditions accelerate lipid oxidation and this is in agreement with the results as shown by other researchers. A high level of FFA is characteristics of product that have undergone both microbial and biochemical spoilage[35].

Conclusion

In the present study, quality and shelf life of dried Tengra fish using two different treatments were analyzed and found organoleptically excellent quality. The main hypothesis of the present study is to understand the effect of different treatments used for drying and to find out the best method of drying for the consumer's safety and economic benefit of the coastal society who mainly depend on the dry fish for livelihood. Commercial traders those who produce market dry fish in our country may be asked to follow the suggestions made over here on the basis of the findings of the present study.

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Biochemical and mineral composition and bacteriological study in some selected fresh water fishes in Meghna river of Bangladesh

[1]Farzana Binte Farid, [2]Gulshan Ara Latifa, [3]Shubhash Chandra Chakraborty, [4]Mosarrat Nabila Nahid, [5]Mohajira Begum

[1]Department of Zoology, University of Dhaka, Dhaka, Bangladesh.

[2]Department of Zoology, University of Dhaka, Dhaka, Bangladesh.

[3]Department of Fisheries Technology, Bangladesh Agricultural University, Mymensingh, Bangladesh.

[4]Department of Zoology, University of Dhaka, Dhaka, Bangladesh.

[5]Institute of Food Science and Technology, Bangladesh Council of Scientific and industrial Research (BCSIR), Dhaka, Bangladesh.

Abstract
Fish play an important role in the Bangladeshi diet, constituting the main and often irreplaceable animal-source food in poor rural households. Study on the biochemical, mineral-compositions and bacteriology of three different freshly-caught fishes were conducted using standard procedures. The fishes are *Channa striatus* (shol), *Channa punctatus* (taki) and *Mystus tengra* (tengra). The results revealed the mean moisture contents were 77.03%, 78.05% and 74.27%; protein contents were 19.52%, 16.89% and 16.96%; fat contents were1.93%, 2.50% and 6.04%; ash contents were 1.44%, 1.36 and 2.67% respectively for *C. striatus*, *C. punctatus* and *M. tengra*. Chemical-parameters and Total bacterial count were in acceptable limits. Minerals included calcium (11.2-22.02 mg/g) and magnesium (9.4-10.4 mg/g) while iron, zinc, copper and manganese were present in trace amounts. The data has proved with strong evidence that the fishes undertaken for the present study are rich in most of the nutrients essential for proper health maintenance of humans.

Keywords: Freshwater fishes, bio-chemical composition, minerals, bacteriological-study

1. Introduction
Fish is a non-tetrapod chordate, i.e., an animal with a backbone that has gills throughout its life and has limbs in the shape of fins [1]. It is one of the best sources of proteins, vitamins and minerals and are essential nutrients required for supplementing both infant and adult diets [2].
It is well known fact that Rice and Fish dominate the diet of Bangladeshi peoples. In terms of weight of food consumed, fish ranks third after rice and vegetables [3, 4]. In Asia, Bangladesh is ranked third largest aquaculture producing country after China and India [5].According to DoF, Fisheries provide 63% of animal protein along with other nutrients like vitamins, minerals etc., and contributed 3.74% GDP and 2.46% foreign exchange earnings in 2011-2012 fiscal year [5].
Among the freshwater fish species, *Channa striatus* (Shol) and *Channa punctatus* (Taki) are delicious, nutritious and popular to the consumers as well as bear high market price. Another popular lean catfish *Mystus tengra* (Tengra) is selected for the present study which is one of the sole species of family Bagridae and a common catfish of the commercial catches of Bangladesh. These fishes have unique test and high demand from all corners of the country as these are economical in price and full of nutrients especially animal protein and fats.
The Bio-chemical composition is an important aspect of fish quality and its influences both the keeping quality and technological characteristics of fish. The knowledge of fish composition is essential for its maximum utilization. The nutritional composition of fish varies greatly from one species and individual to another, depending on age, feed intake, sex and sexual changes connected with spawning, the environment and season [6]. The chemical composition could influence the postharvest processing and storage and could assist in determining the suitability of the different fishes to specific processing and storage techniques.
Now-a-days, large group of consumers have become more health conscious and interested in convenience food. The changing pattern of life style and increasing number of households in the rural area have an impact on the market demands since consumers now a days insist that the product should be acceptable in respect both quality and safety.
In Bangladesh, very little work has been done on the presence of macro and micro elements in freshwater fish, despite such data are important to assess the quality and safety of fish and fishery products for domestic consumption as well as for export. For above reason, the present investigation was carried out to determine the biochemical and mineral composition of three freshwater fishes and study their total bacterial load.

2. Materials and methods
2.1. Collection of experimental fishes: Fresh mature fish samples were collected from fishermen of Meghna River in the early hours of the day.

2.2. Handling of experimental fish in laboratory: Being air breathing fish, *Channa striatus* (Shol) and *Channa punctatus* (Taki) fish were transported to the research laboratory in dram full with water. In case of *Mystus tengra* (Tengra) fish, they were carried in clean, good quality polythene bag with ice in order to keep the fish fresh.

2.3. Place/location of the experiment: Biochemical and Microbial analysis was carried out at the 'Fish Technology Section' and 'Food Microbiology section' of the Institute of Food Science and Technology (IFST) of Bangladesh Council of Scientific and Industrial Research (BCSIR), Dhanmondi, Dhaka and from center for Advanced Research in Sciences (CARS) for Minerals.

2.4. Preparation of fish: The fishes were carefully washed with cooled tap water. Fins, scales, gills and viscera were removed and again washed with tap water to remove blood, slime and unnecessary flesh. Because of hard and large bones of the head, the bones and head of *C. striatus* and *C. punctatus* are included as the waste.

2.5. Sampling procedure: 6 or 7 slice of 3 experimental fish species was taken randomly which represented the parts from whole body of the fish. Then the slices were chopped with skin and bone and finally ground with an electric blender to make a homogenous sample before being sampled for analysis. Triplicate experiments were conducted for this analysis.
Analytical methods were applied for the determination of biochemical composition of the raw fishes on experimental basis.

2.6. Proximate composition analysis: Proximate compositions of fish were determined by conventional method of AOAC (Association of Official Analytical Chemicals) on weight basis [7].

2.6.1. Estimation of moisture: About 5 gram of previously prepared fairly minced samples were taken into each known weight basin and weighed in a digital balance (Toledo, Switzerland). The samples were allowed to dry into the oven (Memmet 854 Schwabach) at 105^0C for 24 hours in order to remove the moisture until constant weight. After that, the basins are taken out of the oven, cooled in a desiccators and were weighed in a digital balance.

Calculation

$$\% \text{ of Moisture} = \frac{\text{Weight Loss}}{\text{Original Weight of Sample Taken}} \times 100 \quad (1)$$

2.6.2. Estimation of protein: The protein content of the fish was determined by micro-kjeldahl method [8]. It involves conversion of organic nitrogen to ammonium sulphate by digestion with concentrated sulphuric acid in a microkjeldahl flask. The digest was diluted, made alkaline with sodium hydroxide and distilled. The liberated ammonia was collected in a boric acid solution and was determined titrametrically.

Calculation
The percentage of nitrogen in the sample was calculated by the following equation:

$$\frac{\%\text{ of } N_2(\text{titration reading - blank reading}) \times \text{strength of acid} \times 0.002 \times \frac{100}{5} \times 100}{\text{weight of sample taken}} \quad (2)$$

For most routine purposes the percent of protein in the sample is then calculated by multiplying the % of N_2 with the protein conversion factor of 6.25 for fish.
% of protein = % of total $N_2 \times 6.25$

2.6.3. Estimation of fat: About 5 g of the homogenous sample was taken into conical flasks and 10 ml of folch reagent (Chloroform: Methanol = 2:1) was added into the sample and homogenized properly and kept in air-tight condition for 24 hours. Fat contents of the fish muscle react with that solvent and remains in the solution. After 24 hours the solution of the flask was filtered in another weighed conical flask through a filter paper. Then these flasks were given in a hot water bath to dry up and removed the solvent. After that the flasks were kept into an oven for an hour to get the actual fat content. Then the flasks were weighed in an electronic balance to get the amount of fat content.

Calculation

$$\% \text{ of Fat} = \frac{\text{Weight of the residue}}{\text{Weight of sample taken}} \times 100 \quad (3)$$

2.6.4. Estimation of ash: About 4-5 g fish sample was weighed into a pre-weighed crucible. The crucible with the contents was heated first over a long flame till all the material was completely churned. Then it was transferred in the Muffle Furnace held at dark red at a rate of 600^0C for 5 hours until the residue become white. The crucible were cooled in desiccators and weighed. Finally the % of ash content was calculated.

Calculation

$$\% \text{ of ash} = \frac{\text{Weight of fish}}{\text{Weight of sample taken}} \times 100 \quad (4)$$

2.7. Estimation of Chemical Composition: To determine the quality of fishes, some parameters, *viz.* TVB-N value, pH, FFA, etc. were analyzed.

2.7.1. Estimation of TVB-N (Total Volatile Base Nitrogen): TVB-N has been used as an index for the determination of freshness of fish [9, 10]. Volatile nitrogenous bases increase in concentration during the spoilage of fish [11]. The TVB-N measurement can be used as a parameter for the determination of microbiological and enzymatic spoilage of fish product. TVB-N value was determined by using Conway modified micro-diffusion technique [12]. Samples that were in the different levels of acceptability from highly acceptable condition to unacceptable condition had been selected for TVB-N analysis. 25 ml of 10% Trichloro Acetic Acid (TCA) was added to 2 gm of ground fish sample and kept overnight and then filtrated with known volume. 2% boric acid, TCA, K_2CO_3 and the solutions made from the fish samples were taken into the Conway dishes. After the addition of Potassium Carbonate (K_2CO_3), each dish was covered by a piece of glass that was stacked with glue (Paraffin soft white) initially. Then it was kept for 24 hours. The samples and Potassium Carbonate (K_2CO_3) reacts to form NH_3 which was absorbed by the boric acid and then the solution of each Conway dish had been titrated by N/70 H_2SO_4 with the help of a micro-burette. Finally TVB-N was calculated.

Calculation

$$TVN = (\text{titration reading} - \text{blank reading}) \times \text{Strength of acid} \times 0.2 \times \frac{\text{Volume of extract}}{\text{Volume of extract taken}} \times \frac{100}{\text{Weight of sample taken}} \quad (5)$$

2.7.2. Estimation of pH: A 1 g sample of the fish flesh was homogenized in 10 ml of distilled water and the mixture was filtered. The pH of the filtrate was measured using a pH meter (Mettler Toledo 320-s, Shanghi, China) [13].

2.7.3. Free Fatty Acid (FFA) Estimation: Oil sample used throughout the work was prepared by extracting the fish by Folch reagent (chloroform and methanol in the ratio of 2:1 v/v).The fish was first cut into small pieces and then ground. The ground material was then mixed with folch reagent in a large wide mouthed stopper glass bottle for 24 hours at room temperature after 15 minutes stirring with glass rod. Extraction was facilitated by occasional shaking. The mixture was filtered through a Buchner funnel and the filtrate was evaporated in batches under heater or oven at 60°C. Seven gram of well-mixed oil was taken into 250 ml flask and 50 ml ethanol was added, previously neutralized by adding 2 ml phenolphthalein solution. Tritation was done by 0.25 N sodium hydroxide with vigorous shaking until permanent final faint pink color appeared and persisted for at least one minute. The value was reported as percentage (%) of free fatty acid expressed as oleic acid. Milli litre of 0.25 N NaOH required for titration corresponds to the percentage of free fatty acid.

2.8. Mineral determination: Samples for mineral analysis were prepared according to recommendations of Perkin Elmer's procedures of Atomic Absorption Spectrometer [14].

2.9. Estimation of total bacterial load: Enumeration of bacterial load was done using plate count agar by pour plate techniques. 10 g of the sample was mixed with 90 ml of previously sterile ringer solution. Appropriate dilutions of homogenate-fish samples was then transferred to petri dish and mixed with plate count agar and incubated at 37°C for 24 hours and the colonies were counted for total Plate count and the count was expressed as CFU/g [15].

Statistical analysis: Data were analyzed by using SPSS for windows-20 statistical programme. Significance was established at $p< 0.05$ [16].

3. Results & Discussion

3.1 Bio-chemical Composition: Bio-chemical composition of the fresh experimental fish, *Channa striatus* (shol), *Channa punctatus* (taki) and *Mystus tengra* (tengra) is given Table-1.

Table 1: Mean initial bio-chemical composition of the fresh experimental fish, *Channa striatus* (shol), *Channa punctatus* (taki) and *Mystus tengra* (tengra)

Parameters	*C. striatus*	*C. punctatus*	*M. tengra*
Moisture (%)	77.03±.12^{a}	78.65±.07^{b}	74.27±.07^{c}
Protein (%)	19.52±.07^{a}	16.89±.10^{b}	16.96±.07^{b}
Fat (%)	1.93±.07^{a}	2.50±.06^{b}	6.04±.06^{c}
Ash (%)	1.44±.11^{a}	1.36±.11^{a}	2.67±.08^{c}
TVB-N	4.41±.01^{a}	3.43±.02^{b}	4.27±.02^{c}
FFA (Free fatty acid)	0.6±.06^{a}	0.5±.1^{ba}	0.9±.21^{ca}
pH	6.9±.06^{a}	7.0±.06^{a}	7.0±.1^{a}

Values are shown as mean±standard deviation of triplicate measurements; a, b, c: Means significant differences between groups. Common superscript letter within a row (horizontal) are not significantly different ($p>0.05$) from each other using LSD test.

The major component of fish muscle was found to be moisture.The moisture contents of *C. striatus, C. punctatus* and *M. tengra* fish in fresh raw condition was found 77.03±.12, 78.65±.07 and 74.27±.07% respectively which resembles with the findings that moisture content of fresh water fish ranged from 70-80% [17].

The protein (%) of *C. striatus, C. punctatus* and *M. tengra* fish in fresh raw condition was 19.52±.07, 16.89±.10 and 16.96±.07% which have got more or less similarities with the findings of the results of *M. aculeatus,G. chapra and P. chola*[18, 19]. Salam reported that, the protein in *H. fossilis* was 18.25% which was similar with findings of *C. striatus,* but higher compared with the findings of *C. punctatus* and *M. tengra* and this might be to species variation [20]. All the fish species examined belonged to high-protein group of fish which resembles with the findings that protein content of fresh water fish ranged from 18-23% [21].

In fresh raw *C. striatus, C. punctatus* and *M. tengra* fish, fat content was 1.93±.07, 2.50±.06 and 6.04±.06% respectively which was more or less similar to the findings of Habashy in *C. carpio,* Nabi and Hossain in *P. gonionotus* and of Mazumder in *G. chapra,P. chola, A. coila* and *A.mola* [22, 18, 19].Salam estimated the highest fat content as 3.25% in *H. fossilis* which was less than that of *M. tengra*[20]. These differences might be due to the reason of species variation. According to Ackman, fish can be grouped into four categories according to their fat content: lean fish (< 2 %), low fat (2 to 4 %), medium fat (4 to 8%), and high fat (> 8%) [23]. Thus, *C. striatus* and *C. punctatus* can be grouped as low fat fish whereas *M. tengra* can be treated as medium fat fish.

The value of ash in *C. striatus, C. punctatus* and *M. tengra* fish was 1.44±.11, 1.36±.11and 2.67±.08% respectively which was nearer to the values obtained by Mazumderin *Aila coila* &*A. mola* andby Salamin *H. fossilis*[19, 20]. The present findings state that the ash contents of these three freshwater fishes might be a good source of minerals such as calcium, magnesium, iron, copper, zinc, manganese etc. Findings of this experimentin respect of fat and ash content of fresh raw fish have got similarities with the findings of Srivastava and Balachandran who has got percent of fat and ash ranged from 1.0-7.0 and 0.4-3.0 respectively [24, 25].From the table it is clear that fresh fish samples presented a higher moisture and low protein content [26].

TVB-N (total volatile base nitrogen) contents of fresh *C. striatus, C. punctatus* and *M. tengra* fish was 4.41±.01, 3.43±.02 and 4.27±.02 mgN/100g respectively. According to EEC the TVB-N value of raw fish was much lower than the acceptable upper limits of 25-35 mg/100 g for some fish species [27]. This is in agreement with the initial TVB-N values of these three fishes.

FFA (%) (Free fatty acid) value of fresh *C. striatus, C. punctatus* and *M. tengra* fish was 0.6±.06%, 0.5±.1% and 0.9±.21% respectively. The acceptable limit of FFA in fresh fish is about 0.5-1.5% [28]. The FFA (%) value in this experiment with three fish species showed the values within the range of acceptable limit.

pH value of fresh *C. striatus, C. punctatus* and *M. tengra* fish was 6.9±.06, 7.0±.06 and 7.0±.1 respectively. The pH in fresh condition fresh-water fish flesh is almost neutral [10]. This is in agreement with the initial pH values of these three fishes.

From the table it is clear that Moisture, fat and TVB-N content of *C. striatus, C. punctatus* and *M. tengra* fish are significantly different ($p<0.000$). But, in case of protein, no significant difference was shown between *C. punctatus* and *M. tengra* fish. No significant different between *C. striatus* and *C. punctatus* in case of ash content whereas in case of FFA value, *C. striatus* is insignificant with both *C. punctatus* and *M. tengra* but between *C. punctatus* and *M. tengra* they are significantly different. No significant different were present in *C. striatus, C. punctatus* and *M. tengra* in case of pH.

3.2 Mineral composition: Important mineral (mg/100g of Fish) composition of macro and micro elements in three experimental fish in fresh condition is reported in Table-2.

Table 2: Important mineral (mg/100g of Fish) composition of macro and micro elements in experimental fresh fish, *Channa striatus*, *Channa punctatus* and *Mystus tengra*

Experimental Fishes	Macroelements		Micro/Trace elements			
	Ca	Mg	Fe	Cu	Zn	Mn
C. striatus	11.2	10.125	1.475	0.7	0.25	0.1
C. punctatus	16.35	9.425	1.275	0.65	0.425	0.05
M. tengra	22.025	11.4	2.25	0.55	1.275	0.125

The macro elements Ca, Mg were abundant in all the fishes examined while micro elements Cu, Fe, Zn, Mn were present in trace amounts. Their abundant presence may be due to the facts that the body needs these macro elements in more amounts than the micro elements in the structure and function of the body and that the concentrations in the water body is very low.

Fresh *Channa striatus*, *Channa punctatus* and *Mystus tengra* fish had calcium (Ca) and magnesium (Mg) content of 11.2, 16.35 & 22.025mg/100g and 10.125, 9.425, and 11.4 mg/100g respectively. Small fishes are mostly eaten with bones, so the amount of available calcium is high in *Mystus tengra*fish than in other two fishes. Ca is essential in human body for the formation of bones,teeth, muscle tone and nervous impulse [29, 30]. It has been reported that, fractional Ca absorption in human body is 24% [31].Comparatively higher concentration of Mg was observed in *Mystus tengra* fish than other fish, which could be linked to the Mg deposit in the fish bone. In that case, *Mystus tengra*fish as a whole one may be considered as one of the good source of Ca and Mg in the diet.Normal extra cellular calcium concentrations are necessary for blood coagulation and for the integrity, intracellular cement substances [32].

The trace element contents in the fishes examined recorded very low in trace amounts. Fe content among the trace elements was dominant in these three fishes. Fe content was highest (2.25 mg/100g) in *Mystus tengra*and lowest (1.275 mg/100g) in *Channa punctatus.* (Fe) Iron has the longest and best history among all the micronutrients. It is key elements in the metabolism of almost all living organisms.Iron is necessary component for the formation of hemoglobin, which is required to form red blood cells [33].Fresh *Channa striatus*, *Channa punctatus* and *Mystus tengra* fish had Zn content of 0.25, 0.425 & 1.275; Cu content of 0.7, 0.65 & 0.55; Mn content of 0.1, 0.05 & 0.125 mg/100g respectively. The presence of zinc in the fishes could mean that the fishes can play valuable roles in the management of diabetes, which result from insulin malfunction [32].

From this study mineral elements detected were in the order Ca>Mg>Fe>Cu>Zn>Mn for *C. striatus* and *C. punctatus* whereas Ca>Mg>Fe>Zn>Cu>Mn for *M. tengra*.Similar order of magnitude of the trace elements was reported in fillets of several species of fresh water fishes [34]. Fawole reported decreasing order of Zn>Fe>Ni>Cu>As in the studies of some fresh water species [35]. Gopakumar reported the values of major elements of Indian fishes in the above same order in most fishes [36].

The mineral contents of each species is a function of the availability of these elements in their local environment, diet absorptive capability and as well as their preferential accumulation. It is therefore become important to equally consider the minerals status of fish and the persistence food safety of the fish prior to consumption in addition to the prevailing choice of taste, size, type and external morphology of fish.

3.3 Bacteriological study: The total bacterial count (SPC) was found to be 2×10^5, 1.1×10^5 and5×10^5 CFU/g in *C. striatus, C. punctatus* and *M. tengra* fish which is presented in Table- 3.

Table 3: Total Bacterial count (CFU/g) in experimental fresh fish, *Channa striatus*, *Channa punctatus* and *Mystus tengra*

Channa striatus(CFU/g)	*Channa punctatus*(CFU/g)	*Mystus tengra*(CFU/g)
2×10^5	1.1×10^5	5×10^5

Although it is widely accepted that the initial microbial load of freshwater fish varies depending on water conditions and temperature, most of the available literature on different freshwater and marine water species (Tilapia, Striped bass, Rainbow trout, Silver perch and Sea bream) reports bacterial counts of 2 to 7 Log CFU/g [37]. The acceptable limit for bacterial count is 5×10^5/g for fresh fish [38]. The bacterial count in this experiment with three fish species showed the values within the range of acceptable limit.

4. Conclusions

Fish is a highly proteinous food consumed by the populace. A larger percentage of consumers eat fish because of its availability, flavors, palatability while fewer percentage do so because of it's nutritional value. This present work has elucidated more on the importance of freshwater fishes as good sources of protein and minerals. It has also broadened our knowledge on the nutritional value of some freshwater fish speciesand in this point of view, it can be suggested that taste, size, freshness and other related external appearances should not be the only factors to be considered in making choice for marketing and consumption of fishes.However, it was discovered that, some microbes present but their count was in low range. Also Regarding the suitable amount of lipids and

nutrient contents, it appears that *C. striatus*, *C.punctatus* and *M. tengra* of river Meghna could be used as human diet.

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Shelf-life Study of Pickle-Salted *Channa striatus*, *Channa punctatus* and *Mystus tengra* during refrigeration (4^0C) storage

Farzana Binte Farid [1], Gulshan Ara Latifa [2], Subhash Chandra Chakraborty[3], Mosarrat Nabila Nahid[4] and Mohajira Begum[5]

[1] Department of Zoology, University of Dhaka,
Dhaka 1000, Bangladesh
farzanafarid79@gmail.com

[2] Department of Zoology, University of Dhaka,
Dhaka 1000, Bangladesh
gulshan_al@yahoo.com

[3] Department of Fisheries Technology, Bangladesh Agricultural University,
Mymensingh, Bangladesh
subhash55chakraborty@yahoo.co.uk

[4]Department of Zoology, University of Dhaka,
Dhaka 1000, Bangladesh
mnabilanahid@yahoo.com

[5]Institute of Food Science and Technology (IFST), Bangladesh Council of Scientific and Industrial Research (BCSIR)
Dhaka-1205, Bangladesh
mohajira10@yahoo.com

Abstract: *This study evaluated shelf-life quality of pickle salted three different fishes; C. striatus (Shol), C. punctatus (Taki) and M. tengra (Tengra) by analyzed their biochemical composition, sensory evaluation and bacteriological load during storage at refrigeration temperature (4°C). There was a general decline in sensory score of these three pickle-salted fish products during storage. The initial value of moisture, TVB-N, pH, FFA, SPC and HBC of freshly processed pickle-salted C. striatus was 50.29%, 5.28 mgN/100gm, 6.4, 1.3%, 3.6×10^3 and 4.1×10^2 ; C. punctatus was 52.71% 3.79 mgN/100gm, 6.6, 2.4%, 3.0×10^4 and 3.3×10^3 and M. tengra was 45.88% 4.92 mgN/100gm, 6.0, 3.2%, 1.2×10^4 and 1.4×10^3 respectively. But, with the laps of storage period, these values increased significantly. Among these three pickle-salted fish products, C. striatus became spoiled at the end of 22 month whereas C. punctatus and M. tengra still remain in good condition. The shelf-life of pickle-salted C. punctatus and M. tengra fish product was 24 month and 28 month respectively. No yeast or mould was detected in these three pickle-salted fish products. Therefore, it can be inferred that pickle salted C. striatus, C. punctatus and M. tengra fish products have long shelf life when they are stored at refrigeration temperature (4°C) and among these three fishes, pickle salted M. tengra fish product has highest shelf life.*

Keywords: Pickle-salted fish, shelf-life study, refrigeration storage**.**

1. Introduction

From time immemorial, 'Rice' and 'Fish' dominate the diet of Bangladeshi peoples. Fish (including shrimp and prawn) is the second most valuable agricultural crop in Bangladesh [1]. Fisheries contributed 4% GDP and 2.09% foreign exchange earnings in 2013-2014 fiscal year and the quantity of exported salted and dehydrated fish products was 261 metric tons which valued Taka 21.65 crores [2]. Fish as perishable and low in amino acid food and is susceptible to pathogenic and enzymatic spoilage [3]. During periods of oversupply and to enhance long storage, it is necessary to adopt appropriate as well as affordable processing and preservation techniques for fish and during this period maintain proper quality.

Although salting process is one of the oldest methods of fish preservation but it is still being used in various countries. Salt is effective to reduce water activity (aw), to destroy the growth of microorganisms and to increase the shelf life [4]. It also provides desirable sensorial changes and ensures the safety for human consumption [5]. This is done because it works as cheaper and easily available protection against insect, pest, fungus and other pathogens.

Among freshwater fish species, *Channa striatus* (shol), *Channa punctatus* (taki) and *Mystus tengra* (tengra) are delicious, nutritious and popular to consumers. These fishes

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have unique test and high demand from all corners of the country as these are economical in price and full of nutrients especially animal protein and fat.

Usually fatty fishes are used for salt-curing in Bangladesh. But in the present research work, the above mentioned fishes are selected because these fishes considered widely accepted and preferred by the peoples and they need these fishes when these are not available. To make these fishes available in off seasons, attempts have been made to cure them using commercial salt. Besides, these highly accepted fishes are not yet tried to preserve using salting or in any other forms.

Several works have been done in biochemical and microbial qualities of different kinds of freshwater fishes, but no works on the shelf life study of pickle-salted fish has yet been done. Considering these facts, the present investigation was carried out to determine the overall shelf life qualities of three freshwater fishes (*Channa striatus*, *Channa punctatus* and *Mystus tengra*) treated with pickle-salting process.

2. Materials and Methods

2.1. Collection and handling of experimental fishes

Fresh fish had been collected from the river Meghan in the early hours of the day. Being air breathing fish, *C. striatus* and *C. punctatus* were transported to the research laboratory in dram full with water. In case of *M. tengra*, they were carried in clean, good quality polythene bag with ice in order to keep the fish fresh.

2.2. Preparation of fish

Fishes were carefully washed with cooled tap water. Head, scales, fins, gills and viscera were removed and again washed with tap water to remove blood, slime and unnecessary adherents. Because of hard and large bones of the head, the bones and head of *C. striatus* and *C. punctatus* were included as the waste.

2.3. Method of Pickle-salting

In this method, the fresh experimental fishes were salted by dry commercial salt (NaCl) of about 30% by weight of the dressed fish stacked in containers and stored at room temperature. The salt entered the fish and water extracted out from the fish-body and a salt-solution is formed. Thus in this method, the fishes are always, allowed to remain in such solution for the production of pickle-cured fish.

2.4. Storage of the product

At the end of salting, pickle salted fishes were packaging with plastic bag maintaining aseptic condition as far as possible and was stored at refrigeration temperature (4^0C).

2.5. Sampling procedures

Evaluation of quality changes in pickle salted fishes were carried out 2 months interval until the fish became inedible for consumption. 6 or 7 slice was taken randomly from fish and were chopped with skin and bone and finally ground with an electric blender to make a homogenous sample before being sampled for analysis.

2.6. Parameters of shelf-life study

There are some parameters which determine the shelf-life quality of pickle-salted fish products during storage condition, such as-

- The moisture content of the pickle-salted fish was determined by AOAC method [6].
- Sensory score evaluation has been done by using 9-point hedonic scales (9. Like extremely; 8. Like very much; 7. Like moderately; 6. Like slightly ;< 5. Bad) [7].
- TVB-N was determined by Conway modified micro-diffusion technique [8].
- pH was determined using a pH meter [9].
- FFA of the fish was determined by AOAC method [10].
- Bacteriological study (SPC and HBC) was done according to the Standard methods of AOAC [11].

3. Results

3.1. Changes in moisture content

Moisture content was found to vary from 50.29 to 52.85% (22 months), 52.71 to 55.71% (24 months) and 45.88 to 48.79% (28 months) in pickle salted *C. striatus*, *C. punctatus* and *M. tengra* respectively during storage at refrigeration temperature (figure-1).

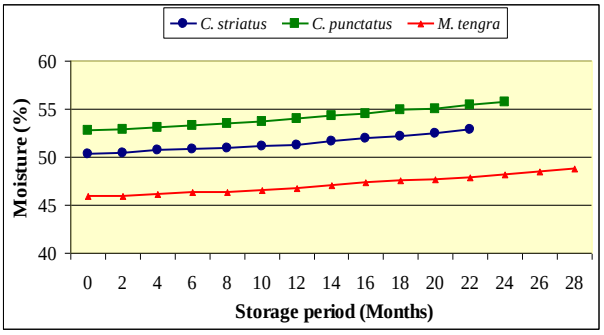

Figure 1: Changes in moisture (%) content of pickle salted *C. striatus*, *C. punctatus* and *M. tengra* during storage at refrigeration temperature

3.2. Sensory score value

The initial score of the sensory evaluation of pickle salted *C. striatus*, *C. punctatus* and *M. tengra* were 9. But during storage at refrigeration temperature (4^0C), this score decreased slowly and at the end of the storage period, the score was 5 in case of pickle salted *C. striatus* (22 months), *C. punctatus* (24 months) and *M. tengra* (28 months) (figure-2).

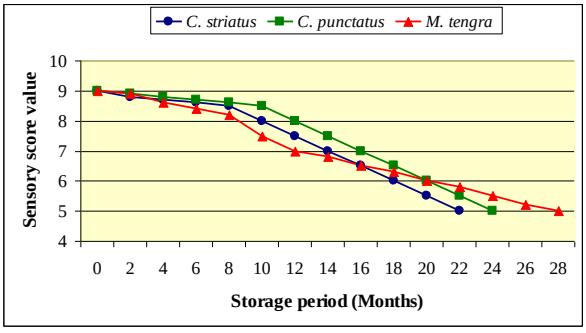

Figure 2: Changes in sensory score value of pickle salted *C. striatus*, *C. punctatus* and *M. tengra* during storage at refrigeration temperature

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3.3. Changes in TVB-N value

The range of TVB-N value of pickle salted *C. striatus*, *C. punctatus* and *M. tengra* fish products during storage at refrigeration temperature were 5.28 to 19.74 (22 months), 3.79 to 20.58 (24 months) and 4.92 to 20.59 (28 months) mgN/100g of fish (figure-3).

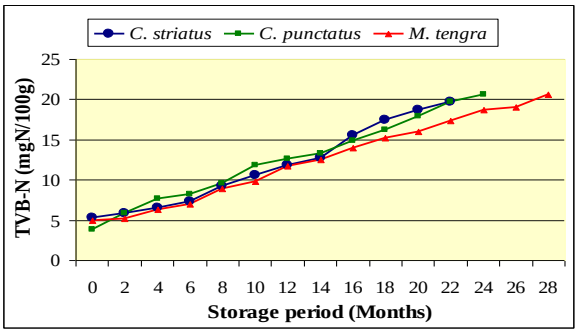

Figure 3: Changes in TVB-N (mgN/100g) value of pickle salted *C. striatus*, *C. punctatus* and *M. tengra* during storage at refrigeration temperature

3.4. Changes in pH value

In case of pickle salted *C. striatus*, *C. punctatus* and *M. tengra* fish, during storage at refrigerated temperature pH value was varied in the range of 6.4 to 7.2 (22 months), 6.6 to 7.8 (24 months) and 6.0 to 6.9 (28 months) respectively (figure-4).

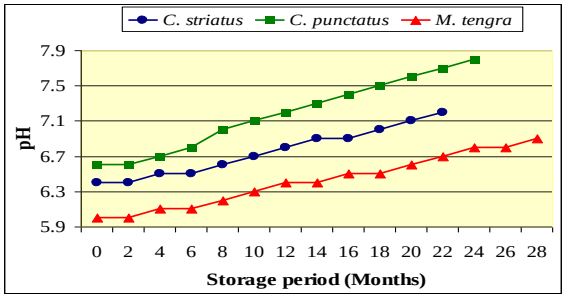

Figure 4: Changes in pH value of pickle salted *C. striatus*, *C. punctatus* and *M. tengra* during storage at refrigeration temperature

3.5. Changes in FFA value

FFA values were found to vary from 1.3 to 11.9% (22 months) in pickle salted *C. striatus*, 2.4 to 12.7% (24 months) in pickle salted *C. punctatus* and 3.2 to 11.4% (28 months) in pickle salted *M. tengra* respectively (figure-5).

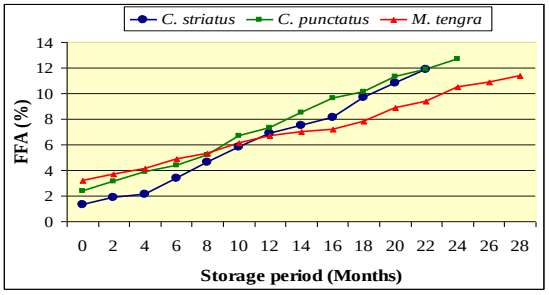

Figure 5: Changes in FFA (%) value of pickle salted *C. striatus*, *C. punctatus* and *M. tengra* during storage at refrigeration temperature

3.6. Changes in Total bacterial Load (SPC)

In this study, the range of SPC of pickle salted *C. striatus*, *C. punctatus* and *M. tengra* were 3.6×10^3 to 2.2×10^6, 3.0×10^4 to 4.2×10^6 and 1.2×10^4 to 3.3×10^6 CFU/g of fish respectively during storage at refrigeration temperature (Table-1).

3.7. Changes in halophilic bacterial load (HBC)

In case of pickle salted *C. striatus*, *C. punctatus* and *M. tengra* fish, during storage at refrigerated temperature HBC count were varied in the range 4.1×10^2 to 3.1×10^5 (22 months), 3.3×10^3 to 3.5×10^5 (24 months) and 1.4×10^3 to 3.1×10^5 (28 months) respectively (Table-1).

Table 1: Changes in total bacterial load (SPC) and halophilic bacterial load (HBC) of pickle salted *C. striatus*, *C. punctatus* and *M. tengra* during different duration of storage at refrigerator temperature (4^0C)

Storage period (months)	*C. striatus*		*C. punctatus*		*M. tengra*	
	SPC	HBC	SPC	HBC	SPC	HBC
0*	3.6×10^3	4.1×10^2	3.0×10^4	3.3×10^3	1.2×10^4	1.4×10^3
4	5.2×10^3	1.1×10^3	4.9×10^4	8.2×10^3	2.6×10^4	3.6×10^3
8	1.0×10^4	4.4×10^3	7.5×10^4	1.3×10^4	5.1×10^4	7.1×10^3
12	2.4×10^4	1.7×10^4	1.1×10^5	2.4×10^4	8.7×10^4	1.6×10^4
16	6.6×10^4	4.3×10^4	2.2×10^5	5.1×10^4	1.6×10^5	3.4×10^4
20	8.0×10^5	1.6×10^5	5.5×10^5	2.0×10^5	6.2×10^5	5.1×10^4
24	-	-	4.2×10^6	3.5×10^5	9.0×10^5	1.8×10^5
28		-	-	-	3.3×10^6	3.1×10^5

*Just after completion of pickle-salting

4. Discussions

4.1. Changes in moisture content

Moisture content varies from species to species and temperature to temperature and at different time duration of its preservation. The moisture content is an exact indicator of the susceptibility of a product to undergo microbial spoilage. Moisture also affects the stability and shelf life of the food product. The moisture content increased as storage progressed in refrigeration temperature, which agreed with earlier work, which finding showed an increase in moisture content from an initial value of 79.35 to 82.60% for 3 weeks of storage at refrigeration temperature of C. *capio* [12].

4.2. Sensory score value

There was a general decline in sensory score of these three pickle-salted fish products during storage. The reduction in the sensory qualities with increase in the storage period of processed fish could be attributed to higher activities of the spoilage agents. Similar trend was observed in variously processed catfish and Telapia stored at ambient temperature [13].

4.3. Changes in Total Volatile Base Nitrogen (TVB-N) value

The TVB-N measurement can be used as a parameter for the determination of microbiological and enzymatic spoilage of fish product. The limiting level for rejection of TVB-N is 20mgN/100g for storage at refrigerator temperature [14]. In present investigation, TVB-N values of the products stored at refrigeration temperature showed linearly increasing pattern throughout storage period but neither of the value exceeded the recommended value set by different researchers for various fish and fish products as acceptable condition. In the present study, increase in TVB-N throughout the storage period may be due to

microbial activity, absorption of moisture. Increase in TVB-N value is related to bacterial spoilage.

4.4. Changes in pH value

The increase in pH indicates the loss of quality. pH is an indicator of the extent of microbial spoilage in fish and some proteolytic microbes produce acid after decomposition of carbohydrate, thereby increasing the acid level of the medium. The pH value is a reliable indicator of the degree of freshness or spoilage. Changes in pH value of 20% brine salted red mullet stored in 4^0C (refrigerator temperature) and found that, this value slightly increased in a range of 6.51 to 6.59 during 11 days of storage, which indicate that pH value of salted fish also increased during refrigeration storage[15]. The pH is an important factor that affects microbial growth and spoilage of foods. A good relationship between changes in pH and organoleptic qualities of fish was observed, where the quality greatly decreased along with the increase of pH. This finding is similar with present study [16].

4.5. Changes in Free Fatty Acid (FFA) value

The FFA value which indicates the rancidity of fat, increased gradually with the passing of storage period. A high level of FFA is characteristics of product that have undergone both microbial and biochemical spoilage [17].

4.6. Changes in Total bacterial load (SPC)

Determination of the microbial load of fishes and their products considered as the most important tests to determine the flesh quality and storage period. Gradual increase of SPC count of pickle salted *C. striatus*, *C. punctatus* and *M. tengra* was observed during storage at refrigeration temperature due to increase in moisture content of the product.

In this study, the total viable count of salted samples was varied during storage time but evaluation of these salted products were within the limits of 10^7cfu/g specified for quality grading of fish by the International Commission of Microbiological Standards for Foods [18]. There is a positive relationship between moisture content and bacterial growth in fish. A correlation was found between bacterial count and total volatile base nitrogen. The sample with high total volatile base nitrogen showed maximum bacterial count [19].

4.7. Changes in halophilic bacterial load (HBC)

Although salt prevents the growth of spoilage bacteria, but other microorganisms such as high salt tolerant and halophiles are not affected by the presence of salt. Halophilic bacterial populations increased in 3.59 to 7.12 log CFU/g in dry salted whole sardine and 3.59 to 6.11 log CFU/g in dry salted fillet sardine when 5 months stored in 4^0C (refrigeration temperature) [20]. This finding is in accordance with present results.

5. Conclusion

Pickle-salting is anticipated to provide a simple, cheaper, healthier and safer method of fish preservation in developing countries. Thus the present study provides information about the suitability of pickle-salting method of three commercially important fishes to produce a very stable and safe product with long storage life. By assessing the shelf-life quality as well as the feasibility of the method, it can be recommended to explore the pickle-salting method in commercial scale which further will contribute in national economy of Bangladesh.

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Author Profile

Farzana Binte Farid obtained B.Sc and M.Sc degree**s** in Zoology (Fisheries) from National University, Bangladesh; in 2000 and 2001 respectively. She worked on different indigenous fish of Bangladesh. She is a Ph D researcher in University of Dhaka (Zoology Department) Bangladesh.

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RESEARCH ARTICLE

Effects of Dry, Pickle and Brine Salting on biochemical and mineral composition and bacterial load of freshwater snakehead fish Taki (*Channa punctatus*)

Farzana Binte Farid[1], Dr. Gulshan Ara Latifa[2], Dr. Subhash Chandra Chakraborty[3], Mosarrat Nabila Nahid[4] and Mohajira Begum[5]

1. Department of Zoology, University of Dhaka, Dhaka-1000, Bangladesh
2. Professor, Department of Zoology, University of Dhaka, Dhaka-1000, Bangladesh
3. Dean, Faculty of Fisheries, Bangladesh Agricultural University, Mymensingh, Bangladesh
4. Department of Zoology, University of Dhaka, Dhaka-1000, Bangladesh
5. SSO, Institute of Food Science and Technology, Bangladesh Council of Scientific and industrial Research (BCSIR), Dhaka-1205, Bangladesh.

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Corresponding Author

Farzana Binte Farid.

Abstract

Salt-cured fishes are highly appreciated because of their characteristic taste, texture and storage stability. In present study, effects of dry, pickle and brine salting on the production of high quality salted fish-products from taki fish (*Channa punctatus*) and their nutritive value was investigated. In fresh, dry salted (DS), pickle salted (PS) and brine salted (BS) taki fish-products, moisture (%) content were 78.65±.07%, 46.21±.04%, 52.71±.06% and 62.28±.02%; protein (%) content were 16.89±.10%, 23.58±.01%, 21.39±.02% and 18.02±.01%; fat (%) content were 2.50±.06%, 3.93±.01%, 3.40±.01% and 2.76±.01%; ash (%) content were 1.36±.11%, 26.37±.02%, 22.96±.01% and 17.24±.01%; salt (%) content were -, 16.06±.06%, 16.22±.04% and 15.50±.04%; TVB-N content were 3.43±.02, 4.18±.00, 3.79±.02 and 5.70±.01 mgN/100g of fish; FFA (%) value were 0.5±.1%, 1.9±.15%, 2.4±.21% and 2.6±.15%; pH value were 7.0±.06, 6.5±.06, 6.6±.15 and 6.8±.10; Ca content were 16.35, 600, 497.5 and 123 mg/100g of fish; Mg content of 9.425, 147.75, 65.5 and 52.5 mg/100g of fish; Fe content were 1.275, 3.75, 2.95 and 2.07 mg/100g of fish; Cu content were 0.65, 0.425, 0.25 and 0.2 mg/100g of fish; Zn content were 0.425, 1.525, 1.43 and 1.175 mg/100g of fish and Mn content were 0.05, 0.625, 0.60 and 0.55 mg/100g of fish respectively. The bacteriological examination revealed that fresh and dry, pickle and brine salted taki fish-products are within the acceptable levels of the specified bacteriological limits for fish and fishery-products.

Introduction:-

Fish is a highly proteinous food consumed by a larger percentage of populace because of its availability and palatability, while fewer percentages do so because of its nutritional value **(Foran *et al.*, 2005)**. Fish is especially important in the developing world. In some countries of Asia (Bangladesh and Cambodia) obtained as much as 75% of their daily protein from fish. Often fish provides essential nourishment, especially high quality proteins and fats (omega 3 and 6 fatty acids), vitamins and minerals. In the economy of Bangladesh fishery products play a vital role.

Fish processing has been practiced in Bangladesh for a long time. The methods commonly used for preservation, are the traditional techniques such as salting or brining, sun-drying and smoking, which also increase fish availability to the consumers **(Abolagba *et al.*, 1996).** Traditional preserving methods of fish products have still wide acceptance around the world due to their accustomed taste and aroma **(Kose, 2010).**

Salting is a popular procedure for preserving fish. Salted fish products are popular in many countries around the globe **(Basti *et al.*, 2006; Lakshmanan *et al.*, 2002).** Sodium chloride (NaCl), also called salt, is generally recognized as a safe, antimicrobial and incidental food additive **(Klaassen, 1986).** Salt has been used as a seasoning and flavor enhancer as well as a preservative or curing agent and is a powerful depressor of water activity (a^w) of the food **(Turan *et al.*, 2007).** Moreover, it is known that chloride ions are toxic for some microorganisms **(Leroi *et al.*, 2000).**

The presence of sufficient salt in fish prevents or drastically reduces the action of bacteria. Salting involves the application of salt to fish either directly or as brine. It removes moisture by the process of osmosis, creating medium unsuitable for microbial growth. The rate of salt uptake and moisture loss is influenced by such factors as temperature, thickness of the flesh, fattiness of the flesh, freshness of the fish and the chemical purity of the salt used for curing. Length of salting period as well as salt concentration depends on the expected final product **(Bellagha et *al.*, 2007).** Salting fish is likely to remain in good demand by those who value tradition and taste but it has also gained acceptance in innovative products that provide convenience. The principle of salting and storage is also to control or minimize the fish deterioration by killing or reducing the growth of microorganisms in the fish. The high concentration of salt has been shown to prevent microbial spoilage in similar products **(Andersen *et al.*, 2007).**

Biochemical analysis provides information on the nutritional value of a particular organism used as a source of food. The measurement of some proximate profiles such as protein contents, carbohydrates, lipids, moisture contents and ash percentage is often necessary to ensure that they meet the requirements of food regulations and commercial specifications **(Watermann, 2000).** In the developed world, people are more concern about the risk and health issues **(Redmond and Griffith, 2005).** On the other hand, in the developing countries due to social inequality some consumers have higher purchasing power also conscious about health issues regarding intake of food **(Petrovici *et al.*, 2004).** Nutrition is an important influencing factor of fish product consumption **(Olsen, 2004 and Ahamed, 2009).** At present, people are aware about health and nutritional issues **(Hossain *et al.*, 2008)** and they concern about the nutritional value of the food items when they buy food items for their household. A number of studies found that higher income people are more concern about harmful and health hazardous food intake in Bangladesh **(Siddique, 2011).**

A number of study on biochemical composition of dried fishes were found in the literature but salting of freshwater fish species has a little in number compared to the dry fish.

Among the freshwater fish species, Taki (*C. punctatus*) fish is delicious, nutritious and popular to the consumers as well as bear high market price. So it is necessary to take some steps for their proper preservation and marketing and during this period maintain proper quality.

The purpose of the present study is to develop an efficient and effective model for salt-curing of freshwater fish species Taki (*C. punctatus*) for the production of high quality Dry-salted, Pickle-salted and Brine-salted products and transfer the technology to the rural small-scale fisher folks allover Bangladesh.

Materials and Methods:-

Collection of the fishes:

Fresh taki fish (*C. punctatus*) had been collected from the river Meghna in the early hours of the day.

Handling of experimental fish in laboratory:

Being air breathing fish, Taki fish was transported to the research laboratory in dram full with water.

Place of the experiment:

Biochemical analysis and Microbial analysis were carried out at the 'Fish Technology Section' and 'Food Microbiology Section' of the Institute of Food Science and Technology (IFST) of Bangladesh Council of Scientific and Industrial Research (BCSIR), and from center for Advanced Research in Sciences (CARS) Dhaka, Bangladesh for Minerals.

Preparation of fish:
The fishes were carefully washed with cooled tap water. Head, scales, fins, gills and viscera were removed and again washed with tap water to remove blood, slime and unnecessary flesh. Because of hard and large bones of the head, the head and bones of taki were included as the waste.

For salt curing, total cleaned fishes were grouped into 3 batches.

Methods of salting:-

Dry salting (DS) method: Fresh taki fishes were enrolled by dry commercial salt (NaCl) of about 30% by weight of the dressed fish (fish weight: salt weight 3:1), stacked in containers and stored for a salting or curing period, at room temperature. In this method, the extracted water of the fish due to salt action had been removed from the container. Thus the fishes are always allowed to remain in dry condition for the production of dry salt cured fish.

Pickle-salting (PS) method: In this method, fresh taki fishes were enrolled by dry commercial salt (NaCl) of about 30% by weight of the dressed fish (fish weight: salt weight 3:1), stacked in containers and stored for a salting or curing period, at room temperature. The salt reacts with the fish and water is extracted out from the fish-body and a salt-solution is formed. Thus in this method, the fishes are always, allowed to remain in such solution for the production of pickle-salted fish.

Brine salting (BS) method: During this experiment, 30% salt solution is prepared (30 gm salt in 100 ml water) which is called brine. Fresh taki fishes were kept at this saturated brine solution stacked in containers and stored for a salting or curing period, at room temperature (26^0C-30^0C) for the production of brine salted fish. The brine solution was changed with new solution once every week for keeping nearly constant saturation outside the fish. The fish in brine were kept immersed by putting a glass weight on it.

Sampling procedures:
6 or 7 slice was taken randomly which represented the parts from whole body of the taki fish. Salt crystals (if any) were removed from salted fishes by using tissue paper. Then the slices were chopped and finally ground with an electric blender to make a homogenous sample before being sampled for analysis.

Parameters of quality assessment:
The analytical methods used in this experiment are given below**:**

- The moisture, fat, ash and salt contents of the fish were determined by AOAC method **(AOAC, 1990).**
- The crude protein of the fish was determined by Micro-Kjeldhal method as described by **Pearson (1999).**
- TVB-N was determined by Conway modified micro-diffusion technique as described by **Conway (1933).**
- pH was determined using a pH meter **(Vyncke, 1981).**
- FFA of the fish was determined by AOAC method **(AOAC, 2000).**
- Bacteriological study (SPC and HBC) was done according to the standard methods of **AOAC (1995)** and **FDA BAM (2001).**
- Samples for mineral analysis were prepared according to recommendations of **Perkin Elmer's** procedures of Atomic Absorption Spectrometer **(1996).**

Statistical analysis: Data were analyzed by using SPSS for windows-20 statistical programme. Significance was established at $p< 0.05$.

Results & Discussion:-

Bio-chemical composition:
Bio-chemical composition of fresh taki fish and freshly processed Dry salted (DS), Pickle salted (PS) and Brine salted (BS) Taki fish-products are shown in **Table 1.**

In present study, fresh, freshly processed dry salted (DS), pickle salted (PS) and brine salted (BS) **taki** fish-products, **moisture (%)** content were 78.65±.07%, 46.21±.04%, 52.71±.06% and 62.28±.02%**; protein (%)** content were 16.89±.10%, 23.58±.01%, 21.39±.02% and 18.02±.01%**; fat (%)** content were 2.50±.06%, 3.93±.01%, 3.40±.01% and 2.76±.01%**; ash (%)** content were 1.36±.11%, 26.37±.02%, 22.96±.01% and 17.24±.01%**; salt (%)** content were -, 16.06±.06%, 16.22±.04% and 15.50±.04%; **TVB-N** content were 3.43±.02, 4.18±.00, 3.79±.02 and 5.70±.01

mgN/100g of fish; **FFA (%**) value were 0.5±.1%, 1.9±.15%, 2.4±.21% and 2.6±.15%**; pH** value were 7.0±.06, 6.5±.06, 6.6±.15 and 6.8±.10 respectively.

In comparison of fresh fish, the salting process resulted in a significant decrease ($P < 0.05$) in moisture content and a significant increase ($P < 0.05$) in protein, fat and ash content of Dry-salted, pickle-salted and brine-salted Taki fish samples. This is consistent with the observations by **Abraham-Olukayode *et al.* (2012).** Nutritional components, such as protein, lipid, and ash, were increased due to the loss of water in fish muscle in the salting process **(Bras & Costa, 2010; Chaijan, 2011).**

Significant increased in protein levels ($p<0.05$) in all salted fish-products when compared with the fresh fish, suggested that protein nitrogen was not lost during salting **(Nahid *et al.*, 2014).** The findings in the protein and fat contents were also in agreement with related studies by **(Thorarinsdottir *et al.*, 2004)** where quite related trends were observed with indications that the amount of protein was a function of salt concentration and water content.

The higher value of total ash content in freshly processed Dry-salted, pickle-salted and brine-salted Taki fish than fresh fish was attributed to high salt content which added more ash components to the products. These results were in accordance with **El-Bassir *et al.* (2015).**

The FFA (%) value recorded in this work is low and hence there is no fear of rancidity. This agrees with the work of **Frazier and Westhoff (1998).**

The salting process caused a decrease in pH values in all salted fish samples. **Goulas (2005)** observed that pH values of mackerel (*Scomber japonicus*) decreased after salting.

According to **Hernandez-Herrero *et al.* (1999),** after salting of Anchovies, salt content increased from 0.32 to 19.3%, ash content increased from 1.6% to 21% and pH decreased from 6.13 to 5.72 which are in harmony with present findings.

Monsur (2007) observed that, 30% dry salted, pickle salted and brine salted punti fish had salt content of 24.21%, 24.68% and 17.28% respectively and chapila fish had salt content of 25.11%, 21.43% and 15.94% respectively which is higher from present study.

Mineral contents:-

The mineral composition of fresh taki fish and freshly processed Dry salted (DS), Pickle salted (PS) and Brine salted (BS) taki fish-products are shown in **Table 2.**

In case of fresh, dry salted (DS), pickle salted (PS) and brine salted (BS) **taki** fish-products, **Ca** (calcium) content were 16.35, 600, 497.5 and 123 mg/100g of fish; **Mg** (magnesium) content were 9.425, 147.75, 65.5 and 52.5 mg/100g of fish; **Fe** (iron) content were 1.275, 3.75, 2.95 and 2.07 mg/100g of fish; **Cu** (Cooper) content were 0.65, 0.425, 0.25 and 0.2 mg/100g of fish; **Zn** (zinc) content were 0.425, 1.525, 1.43 and 1.175 mg/100g of fish and **Mn** (manganese) content were 0.05, 0.625, 0.60 and 0.55 mg/100g of fish respectively.

From **table 2.** it is clear that three types of salting methods raised the mineral composition of dry salted, pickle salted and brine salted taki fish products.

There is very little information on mineral contents of salted fish-products. However, the present values are comparable with the results reported for some dry freshwater fish (**Ahmed *et al.*, 2011; Mohammed, 2013).** According to **Begum *et al.* (2012),** freshly processed salted-dried punti (*Puintius sophore*) fish have calcium (Ca) and Iron (Fe) was in a range of 320-330 mg/100g and 3.1-3.9mg/100g of fish respectively.

Bacteriological study:-

Standard plate count (SPC) (CFU/g) or total bacterial count (TVC) and halophilic bacterial count (HBC) (CFU/g) of fresh, dry salted (DS), pickle salted (PS) and brine salted (BS) **taki** fish-products are given in **Table 3.**

The Standard plate count (SPC) in fresh, dry salted (DS), pickle salted (PS) and brine salted (BS) **taki** fish-products were recorded in 1.1×10^5, 4.0×10^3, 3.0×10^4 and 4.3×10^4 CFU/g whereas halophilic bacterial count was recorded in -, 3.0×10^2, 3.3×10^3 and 4.1×10^3 CFU/g respectively.

According to **Surendran (2006)** the acceptable limit for bacterial count is 5×10^5/g for fresh fish which is in accordance with present findings of fresh taki fish. Freshly process dry salted (DS), pickle salted (PS) and brine salted (BS) **taki** fish-products also indicated an acceptable microbial load ($<10^5$ CFU/g^{-1}) **(ICMSF, 1988).**

From **Table 3.** it was observed that in comparison with the fresh fish, there was a decrease in total bacterial count which may be due to the presence of high salt concentration, so the pathogenic microorganism growth is controlled. This result is in agreement with the findings of **Abu Giddeire (2001).**

According to **Abbas Bakhiet and Khogalie (2011),** salting process reduces total bacterial count of *Hydrocynus spp.* fish and found that TVC of fresh fish was 58.1×10^3 and it reduces in freshly processed 15%, 20% and 25% salted fish-products in 10. 0×10^3, 7.8×10^3 and 4×10^3 respectively which is in agreement with the present study.

Ames *et al.* (1991) reported that when the water activity is considerably reduced, most microorganisms become inactive but haploidic microorganisms become the major causes of microbial spoilage. The high salt concentration leaves only salt tolerant microorganisms to survive **(Horner, 1997).**

Table 1: Biochemical composition of fresh, Dry-salted (DS), Pickle-salted (PS) and Brine-salted (BS) Taki fish:

Samples	**Moisture** (%)	**Protein** (%)	**Fat** (%)	**Ash** (%)	**Salt** (%)	**TVB-N** mgN/100g	**FFA** (%)	**pH**
Fresh Taki	78.65±.07	16.89±.10	2.50±.06	1.36±.11	-	3.43±.02	0.5±.1	7.0±.06
DS Taki	46.21±.04	23.58±.01	3.93±.01	26.37±.02	16.06±.06	4.18±.00	1.9±.15	6.5±.06
PS Taki	52.71±.06	21.39±.02	3.40±.01	22.96±.01	16.22±.04	3.79±.02	2.4±.21	6.6±.15
BS Taki	62.28±.02	18.02±.01	2.76±.01	17.24±.01	15.50±.04	5.70±.01	2.6±.15	6.8±.10

Table 2: Mineral composition of fresh, Dry-salted (DS), Pickle-salted (PS) and Brine-salted (BS) Taki fish:

Samples	**Macro elements**		**Micro / Trace elements**			
	Ca Mg/100g	**Mg** Mg/100g	**Fe** Mg/100g	**Cu** Mg/100g	**Zn** Mg/100g	**Mn** Mg/100g
Fresh Taki	16.35	9.425	1.275	0.65	0.425	0.05
Dry-salted Taki	600	147.75	3.75	0.425	1.525	0.625
Pickle-salted Taki	497.5	65.5	2.95	0.25	1.43	0.60
Brine-salted Taki	123	52.5	2.07	0.2	1.175	0.55

Table 3: Standard plate count (CFU/g) and halophilic bacterial count (CFU/g) in fresh, Dry-salted (DS), Pickle-salted (PS) and Brine-salted (BS) Taki fish

Samples	**SPC (CFU/g)**	**HBC (CFU/g)**
Fresh Taki	1.1×10^5	-
DS Taki	4.0×10^3	3.0×10^2
PS Taki	3.0×10^4	3.3×10^3
BS Taki	4.3×10^4	4.1×10^3

Conclusion:-

The present study reveals that different fish salting methods have a positive significant role on the biochemical and mineral composition of freshwater Taki (*C. punctatus*) fish and reduces bacterial load as well as makes them nutritionally suitable for all.

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