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).

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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

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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

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

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.
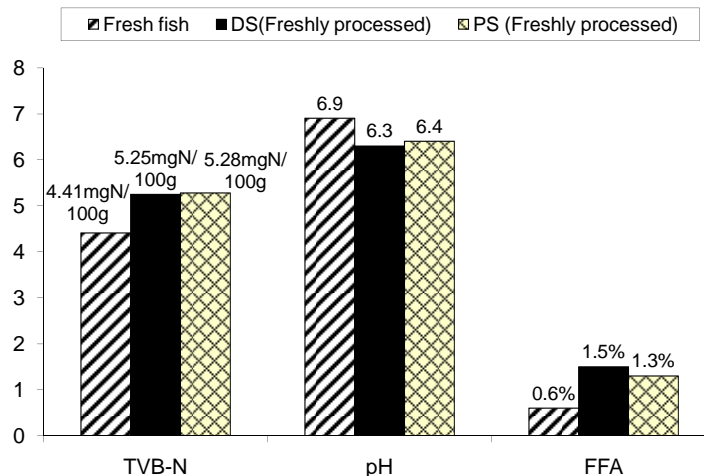

Fig 1: Chemical composition of fresh fish, freshly processed dry (DS) and pickle (PS) salted shoal fish.
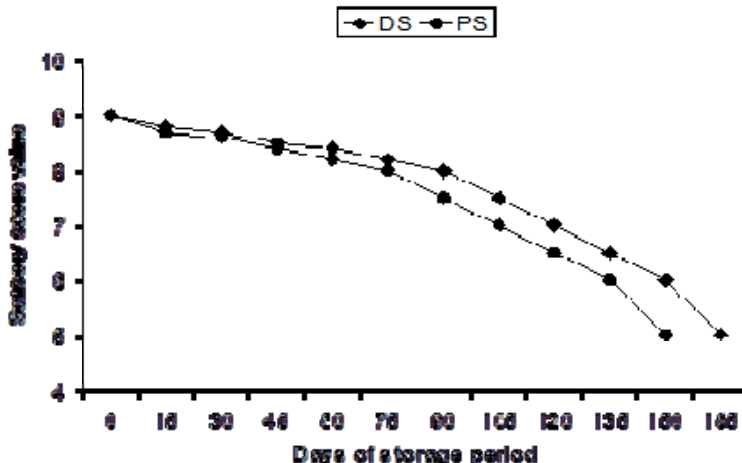

Fig 2: Changes in sensory score of DS and PS Shoal at Room temperature during different days of observation.
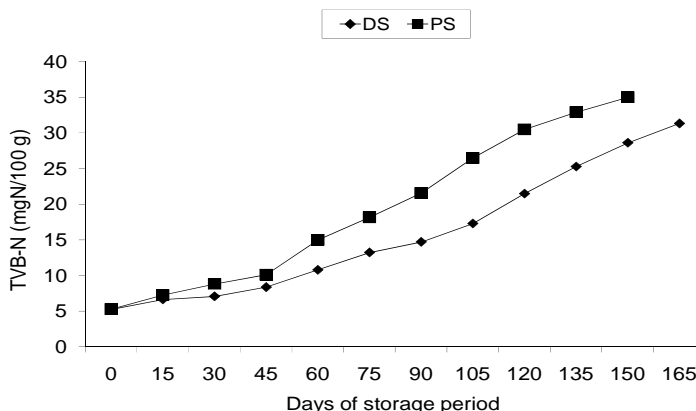

Fig 3: Changes in TVB-N (mgN/100 g) contents of 2 types of salt treated Shoal fish during different duration of storage at Room Temperature.
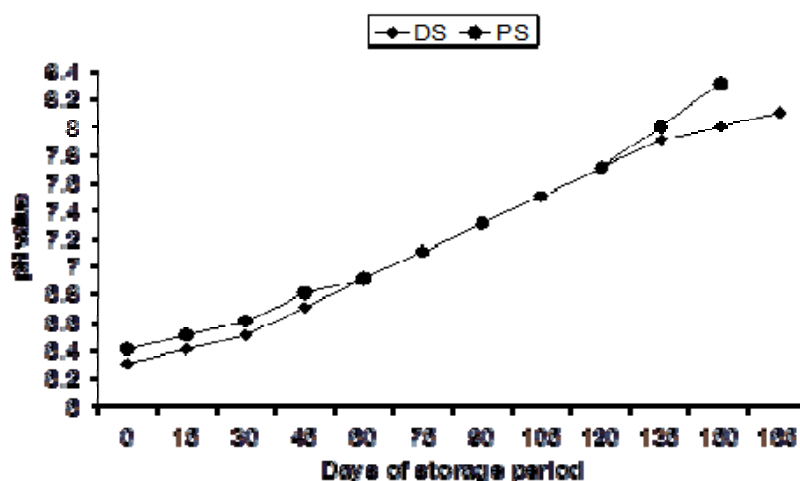
Fig 4: Changes in pH value of Dry and pickle salted Shoal fish during different duration of storage at Room Temperature.
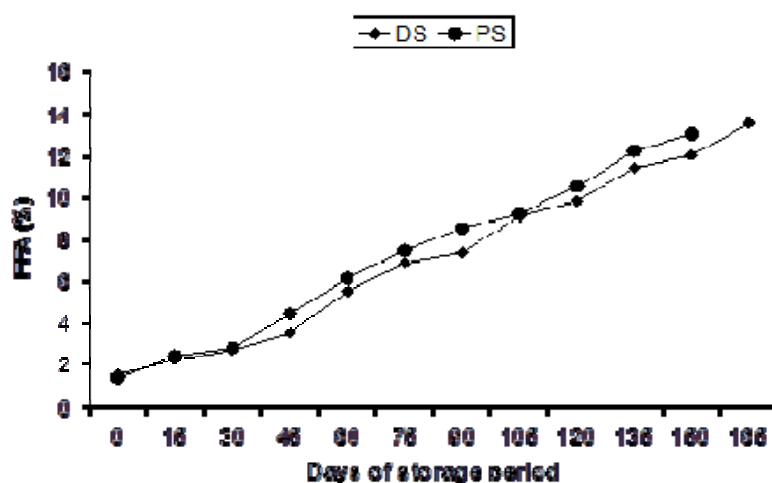

Fig 5: Changes in FFA (Free Fatty Acid) value of 2 types of salt treated Shoal fish during different duration of storage at Room Temperature.

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.

5. Acknowledgement

6. References

1. Abdullahi SA, Abolude DS. Ega RA. Nutrient Quality of four Oven Dried Freshwater Catfish in Northern Nigeria. J Trop Biosci 2001, 70.
2. FAO. Fish for Food and Development. 1991, 49.
3. Minkin SE, Rahman MM. Halder S. Fish biodiversity, human nutrition and environmental restoration in Bangladesh. In: Openwater Fisheries of Bangladesh (eds. Tsai C and Ali MY) 75-88. The University Press Limited. Dhaka, Bangladesh, 1997.
4. Hels O, Hassan N, Tetens H. Thilsted SH. Food consumption energy and nutrient intake and nutritional status in rural Bangladesh: Changes from 1981-82 to 1995-96. Eur j Clon-Nutr 2002; 57:586-594.
5. Eyo AA. Fish processing Technology in the Tropics. Published by National Institute for Freshwater Fisheries Research (NIFFR), P.M.B. 2001, New Bussa, Niger State, 2001; 37-39:130-138-153-160-164.
6. Geoff A, Clucas I, Paul SS. Post-Harvest Losses of Fish in the tropics. Natural Resources Institute, 1991, 1-5.
7. Igene JO. Drying of fish, factors to consider. Fisheries Society of Nigeria (FISON) 3rd annual Proceedings. 1983.
8. Cihazala S. New Packaging Technology for Seafood Preservation, Shelf-life Extension and Pathogen Control. In: Fisheries Processing Biotechnological Applications, Martin, A.M. (Eds.), Chapman and Hall, London, ISBN: 9780412584602, 1994, 83-110.
9. Bellagha S, Sahli A, Farhat A, Kechaou N. Glenza A. Studies on salting and drying of sardine (*Sardinella aurita*): Experimental kinetics and modeling. J Food Eng 2007; 78:947-952.
10. Horner WFA. Preservation of fish by curing (drying, salting and smoking). In Fish Processing Technology. Blackie Academic & Professionals, New York, 1997, 31-71.
11. Martínez-Alvarez O, Gómez-Guillén MC. The effect of brine composition and pH on the yield and nature of water-soluble proteins extractable from brined muscle of cod (*Gadus morhua*). Food Chem, 2005; 92:71-77.
12. Klaassen CD, Amdur MO, Doull JA. Principles of Toxicology. In Casarett and Doull's Toxicology: The Basic Science of Poisons. New York: Macmillan, 1986.
13. Turan H, Sonmez G, Celic MY. Yalcin M. Effects of different salting process on the storage quality of Mediterranean Muscle (*Mystus Galloprovincialis* L. 1819) J Muscle Foods, 2007; 18:380-390.
14. Leroi F, Joffraud JJ, Chevalier F. Effect of salt and smoke on the microbiological quality of cold- smoked salmon during storage at 5 ^{0}C as estimated by the factorial design method. J Food Prot 2000; 63:502-508.
15. Bahri P, Ayse GI, Gulsum O, Irfan I. Microbiological and chemical qualities of salted grey mullet. Int J Sci Techno 2006; 2:91-98.
16. Peryan DR, Pilgrim FJ. The Methology of sensory testing, IFST sykp. Pittsburg, U.S.A. Food Technology Champaign 1957; 2:9-14.
17. Sokal RR, Rohlf F J. Introduction to biostatistics. New York: W. H. Freeman and Company, 1987, 363.
18. Conway EJ, Byrne A, Micro-diffusion analysis of TVN. Biochem J 1933; 27:419-429.
19. Vyncke W. pH of fish muscle comparison of methods, Western European Fish Technologists' Association (WEFTA). Copenhagen, Denmark, 1981.
20. Yu SY. Comparison on sun-dried and oven dried salted fish. FAO Fish Rep 1985; Supple: 317:328-330.
21. Morshed M. Sun dried products of Bombay duck and its quality assessment at different storage condition. Department of Zoology, University of Dhaka, 2005, 29.
22. Stansby ME, Harrison RW, Dassow J, Sater M. Ind. Eng. Chem. Analyst, Edn 1944; 16(9):593.
23. Huss HH. Fresh Fish-quality and quality changes, FAO Fihs Series 29, FAO DANINAQ, Rome. Italy. 1988, 132.
24. Vynke W, Marlevede E. Arch. Beges. Med. Ttavais. Med. Legale. 1963; 21(3):147-160.
25. Gram L, Ravn L, Rasch M, Bruhn JB, Christensen AB, Givskov M. Food spoilage-interactions between food spoilage bacteria. International Journal of Food Microbiology 2002; 78:79-97.
26. Connell JJ. Control of fish quality. Fourth edition published by fishing Books, a division of Blackwell Scientific Ltd, 1995.
27. Reed GB, Rice EE, Sinclair RJ. Comparative study of analysis and bacterial decomposition in haddock, Lobster and Clamb muscle. cntribs. Can Bi Fisheries 1929; 4(18):229-235.
28. Muslemuddin M. Some aspects of the determination of fish muscle during storage. A department of Fd. Sci. National college of Fd. Tech Uni of Fd Technology 1970, 84.
29. Tomiyasu Y, Zenitani B. Spoilage of fish and its preservation by chemical agents, Advances in food Research Academic Press inc. M.Y and London, 1957; 7:42-74.
30. Eyo AA. Traditional and improved fish handling, preservation and processing techniques. Paper presented at national workshop on fish processing storage, marketing and utilization, 1993.
31. Virta S. Bachelor's Thesis, Isolation and Identification of Rainbow Trout spoiling Microbiota. Biotechnology and Food Technology, Turku University of Applied Science 2009, 8. (Unpublished).
32. Shenderyuk VI, Bykowski PJ. Salting and Marinating of fish. In: Seafood Resources. Nutritional Composition and preservation, Sikorski, Z.E. (Ed.) CRC Press Inc. Boca Raton Florida, 1989.
33. Erkan N, Tosun SY, Ulusoy S, Uretener G. The use of thyme and laurel essential oil treatments to extend the shelf life of bluefish (*Pomatomus saltatrix*) during storage in Ice. Journal für Verbraucherschutz und Lebensmittelsicherheit 2011; 6(1):39-48.
34. FAO/ SIFAR, Non-Sensory Assessment of Fish quality. (FAO in partnership with support unit for international Fisheries and Aquatic Research, SIFAR, Torry Advisory Note No. 92. 2001.

35. Tungkawachara S, Park J, Choi Y. Biochemical properties and consumer acceptance of Pacific whiting fish sauce. J Food Sci 2003; 68:855-860.
36. El-Sebahy L, Metwali M. Changes in some Chemical Characteristics and Lipid Composition of salted Fermented Bouri fish Muscle (Mugil cephalus). Food Chem 1988; 31:41-50.
37. Chakraborty SC, Islam MN, Khan AS. Study on the evaluation of the quality aspects of dry salted, wet salted and sun dried Hilsa fish (*Hilsa ilisha*). Bangladesh J Agri Sci 1997; 24(2):71-79.
38. Mansur MN, Chakraborty SC, Uddin MN, Khan MNA. Influence of spawning on the sensory, biochemical and bacteriological characteristics of dry salted hilsa (*Hilsa ilisha*) fish at ambient temperature. Bangladesh J Fish 1998; 21(1):65-72.
39. Pearson D. The chemical Analysis of foods, the edition, Churchill Livingston, Edinburgh London and New York, 1976; 2:387-497.